PHYSIOLOGY AND MORPHOLOGY OF RICE PLANTS WITH SILICON SUPPLEMENTATION AND DIETHOLATE SEED TREATMENT UNDER WATER DEFICIT

Jéssica Cezar Cassol, Sidinei José Lopes, Sylvio Henrique Bidel Dornelles, Mariane Peripolli, Luciane Almeri Tabaldi
Universidade Federal de Santa Maria, Brazil

Abstract

Silicon is an enzyme stimulator that promotes signaling for the production of antioxidant, osmoprotective compounds and attenuates interference in photosynthesis in rice plants subjected to water deficit. The aim of this study was to evaluate the possible effects of silicon as a stress reliever in rice plants grown from seeds treated with dietholate under of water deficit conditions. The experimental design was fully randomized with three replicates, 144 experimental units consisting of pots containing 4.4 pounds soil, and a 3x2x2x4 factorial arrangement: three soil water conditions (50% and 100% of soil water retention capacity and water blade of 5.0 cm); two cultivars (IRGA 424 RI and Guri INTA CL); two sources of Si (sodium and potassium metasilicate); and four Si rates (0; 4.0; 8.0 and 16 g L\textsuperscript{-1}). Silicon boosted stomatal density; induced an increase in the maximum photochemical efficiency of photosystem II (PSII) under both water deficit and optimal conditions, boosting photosynthesis; and increased effective quantum yield of PSII and levels of total dry mass. Thus, silicon attenuated the effects of water deficit in plants grown from seeds treated with dietholate.

Keywords: abiotic stress; Oryza sativa L.; Safener.

1. Introduction

Irrigated rice (Oryza sativa L.) is grown on over 167 million hectares of land (FAO, 2018) in ecosystems with varying temperatures and water regimes. Annual worldwide yield is estimated at over 487 million tonnes, with 11 million tonnes produced in Brazil (EMBRAPA, 2018). Rio Grande do Sul is the largest rice producer in Brazil, responsible for approximately 71% of production (CONAB, 2017). Almost all areas cultivated with rice are irrigated by controlled flooding, representing more than 90% of Brazil’s total national production (EMBRAPA, 2018). The high costs of electric energy, the seasonality of rains and the increase in temperatures, makes it the option to rationalize the use of water, however, it can cause several morphological, biochemical and physiological changes in the plants, directly interfering in the final productivity (Carneiro et al., 2011).

Around 40% of the total crop losses are because of the single abiotic stress i.e., drought (Fabregas et al.,
2018), diminishing yield and inducing biochemical and physiological changes in plants, including higher stomatal density (Luković et al., 2009), impaired stomatal opening and a drop in the photosynthetic rate (Bota et al. 2004). Plants also regulate stomatal closure and their development to control water loss as one of the stress adaptation strategies (Yadav et al., 2019). Furthermore, these adverse responses to water deficiency are aggravated in plants grown from seeds treated with the dietholate safener (0.0-diethyl 0 phenyl phosphorothioate) (Mauad et al., 2011).

Dietholate is a seed protector, which gives culture, tolerance to the herbicide clomazone, even in low doses (Sancho et al., 2010). However, the enzymatic compounds inhibited by dietholate are also responsible for reducing the harmful effects of reactive oxygen species (ROS) formed during cellular metabolism (Gill et al., 2013) under biotic and abiotic stress conditions. Thus, the use of biostimulants (Inoue et al., 2012), such as silicon (Si) (Mauad et al., 2011) has been recommended to minimize the phytotoxic effects of dietholate in the presence of water deficit.

Silicon (Si) is an enzymatic stimulant (Taiz et al., 2017) and can affect plant growth and development by initiating signaling for the production of antioxidant compounds (Etesami et al., 2017) and osmoprotectants, such as proline and glycine betaine (Etesami et al., 2017). The accumulation of silicon in plants can mitigate the stresses that result in interference in rice plant photosynthesis under water deficit conditions (Artigiani et al., 2012) or due to anaerobic conditions and the formation of ROS (Zia et al., 2017). By increasing stomatal conductance and maintaining elevated electron transfer, high rates of photosynthesis are possible, together with high levels of total chlorophyll that affect photosynthetic rates. Si is also associated with the prevention of leaf senescence by maintaining continued photosynthesis and protecting chlorophyll distribution, especially under high air temperature and water deficit conditions (Agarie et al., 1998). According to the authors, silicon promotes the thermal stability of lipids in cell membranes and, under stress conditions, prevents rice cell membranes from structural and functional deterioration. Thus, the aim of this study was to evaluate the possible effects of Si as a water drought reliever in rice plants grown from seeds treated with dietholate.

2. Material and methods

The experiment was conducted in the greenhouse (6 x 20 m, headroom 5 m) from February to April 2018. The experimental design was fully randomized with 48 treatments and three replicates, organized in a 3x2x4x2 factorial arrangement: three soil water regimes (50% and 100% of soil water retention capacity (WRC) and water blade of 5.0 cm); two cultivars (IRGA 424 RI and Guri INTA CL); four Si rates applied to the furrow (0; 4.0; 8.0 and 16 g L⁻¹) and two sources of Si (sodium metasilicate (composition: Na₂O ≅ 28%; SiO₂ ≅ 27%; Fe ≅ 0.02%) and potassium metasilicate (composition: N ≅ 3%; P₂O₅ ≅ 2%; K₂O ≅ 15%; SiO₂ ≅ 25%)).

The 144 experimental units each consisted of a pot containing 4.4 pounds de soil. The fertilization of the experimental units was based according to the soil physical analyses report of the Soil Physics Laboratory – UFSM. The soil belonging to sandy loam textural class (SBSCS), with base saturation < 50, 60.6 of sand, 22.8% of silt and 16.6% of clay. Ten seeds treated with dietholate seed protector (Permit Star®) at
a rate of 6 mL per kg seed were sown in each pot at a depth of two centimeters, and after emergence the six most uniform seedlings in each pot were retained.

Irrigation was carried out daily through the weighing method, using an electronic scale (ACS System) with precision of 5 g, by adding water until reaching the total predetermined mass (pot + dry soil + water volume to reach 100 or 50% soil WHC). To determine soil water conditions (50 and 100% soil WHC), the following adapted formulas were used (Schwab, 2011):

\[
\text{MP50\%} = (\text{MPWHC} - \text{MPdry})0.5 + \text{MPdry} \quad (1)
\]

\[
\text{MP100\%} = (\text{MPWHC} - \text{MPdry})1.0 + \text{MPdry} \quad (2)
\]

Where: MPn\% - mass of pot for each treatment; MPWHC - mass of pot at water holding capacity; and, MPdry - mass of pot filled with dry soil.

Treatments related to soil water conditions were begun 15 days after sowing, simulating water ingress conditions in an irrigated rice crop. Physiological and morphological variables were evaluated when the rice plants had reached vegetative stage, scale 39 of the BBCH (MEIER, 2001).

The physiological variables of chlorophyll “a” fluorescence, including the ratio of variable fluorescence to maximum fluorescence (maximum photochemical efficiency of PSII) (Fv/Fm), effective quantum yield of PSII (YII\(_{25}\)) and the electron transport rate (ETR\(_{1500}\)), were measured with a JUNIOR-PAM modulated pulse fluorometer (Walz, Germany) between 3:00 am and 8:00 am. Measurements were taken using the fourth leaf fully expanded of three different plants from three experimental units for each treatment. Before measuring, the leaves to be analyzed were pre-adapted in the dark for a period of 30 minutes to determine the initial fluorescence (Fo) and subsequently subjected to a pulse of saturating light (10,000 \(\mu mol \ m^{-2} \ s^{-1}\)) for 0.6 s to determine the maximum fluorescence (Fm). The maximum photochemical efficiency of PSII (Fv/Fm) was calculated using the fluorescence ratio variable (Fm-Fo).

The electron transport rate (ETR\(_{1500}\)) was determined using a light curve (electron transport rate x light intensity - PAR), which was constructed by subjecting each sample to radiation levels of 285\(\mu\)mol electrons m\(^{-2}\) s\(^{-1}\) every 10 s. The measurements were adjusted using the equation ETR = ETRmax \[1– e^{\chi_kQ}\], where k is a fitting constant and Q represents the light intensity (PAR), according to the method described by Rascher et al. (2000).

Stomatal density (nº/mm\(^2\)) was determined based on the epidermal impression of the abaxial and adaxial surfaces of the leaf on glass slides using instant adhesive (Super Bonder®). This was done by collecting one fully expanded leaf, in three experimental units different, for each treatment. The slides were then photographed under a microscope and the images processed using Image Pro Plus software to obtain the area and stomata count. To evaluate plant dry weight, three plants were removed from each pot, placed in porous paper bags and dried in a forced air oven at 65° C until a constant dry weight (g) was reached.

Response variables were subjected to analysis of variance, and factors analyzed by the Tukey test at 0.05 error probability, except for the silicon rates which were fit to polynomial models using Sisvar® 5.3 statistical software (Ferreira, 2014).
3. Results and discussion

Total dry weight, ratio of variable fluorescence to maximum fluorescence and adaxial surface stomatal density showed significant interaction for all factors (soil water conditions × cultivar × Si rate × Si source). Abaxial surface stomatal density showed a significant interaction for soil water × Si rate × cultivar, and no effect was produced for the Si source in the F-test (p <0.05). Thus, for this variable, regression models and tables were obtained using the means of the two Si sources. For effective quantum yield of PSII (YII125) and electron transport rate (ETR1500), there was a significant effect only for the Si rate, with no effect for the cultivar, Si source and soil water in the F-test (p <0.05). Thus, regression models were obtained for the means of the cultivar, Si source and soil water.

It can be seen from an examination of soil water conditions, that, for adaxial surface stomatal density (Table 1), that of the sixteen comparisons between the water conditions of the soil, within the levels of the other factors, 81% of the cases the plants under water deficit (50% WRC) did not differentiate the number of stomata of the adaxial face per mm², in relation to the plants under 100% WRC and immersed plants. These results may indicate that transpiration control can be done more by stoma closure than by the presence of fewer stomata.

Comparing the results for Si sources, in terms of adaxial surface stomatal density (Table 1), of the twenty-four comparisons between the Si sources, given the levels of the other factors, 79% of cases did not show any statistical difference between sources. In terms of the cultivar variable (Table 1), in general, of the twenty-four comparisons between cultivars, given the levels of the other factors, 79% of cases did not show any statistical difference between cultivars, possibly due to their genetic similarity.

Table 1 – Comparison of means for adaxial surface stomatal density in rice leaves for water conditions and Si sources at different Si rates and for different cultivars. Santa Maria, RS, 2019.
The regression analysis of Si rates for adaxial surface stomatal density (Figure 1) show that at the highest rate of sodium metasilicate (16 g L\(^{-1}\)), there was a linear increase of 32% in the number of stomata for the immersed Guri INTA CL cultivar compared to non-supplemented plants (0.0 g L\(^{-1}\)), whereas none of the other soil water conditions fit the polynomial models (Figure 1a). For the IRGA 424 RI cultivar, there was a quadratic increase at 50% WRC (water deficit), reaching a maximum at a rate of 9.9 g L\(^{-1}\) sodium metasilicate; none of the other soil water conditions fit the polynomial models (Figure 1b). The presence of a greater number of stomata can be a useful strategy to increase atmospheric CO\(_2\) absorption or carbon exchange rate. For the potassium metasilicate source and both cultivars (Guri INTA CL and IRGA 424RI, Figures 1c and 1d), at 100% WRC there was a quadratic adjustment in adaxial surface stomatal density for the Guri INTA CL cultivar and linear increase of 85% in the number of stomata for the IRGA 424 RI cultivar at the highest rate of Si (16 g L\(^{-1}\)) compared to non-supplemented plants (0.0 g L\(^{-1}\)). In immersed plants, increasing the rate of potassium metasilicate resulted in a linear increase of 65% in adaxial surface stomatal density in the IRGA 424 RI cultivar at the highest rate of Si (16 g L\(^{-1}\)) and Guri INTA CL cultivar there was a quadratic increase in adaxial surface stomatal density, reaching a maximum at a rate 8 g L\(^{-1}\) compared to non-supplemented plants (0.0 g L\(^{-1}\)) (Figure 1c and 1d). At 50% of the WRC, there was not adjustment of polynomial models in both cultivars (Figure 1c and 1d). In rice (\textit{Oryza sativa} L.) seedlings, 2 mM Si treatment increased the stomatal density and decreased the stomatal size of the adaxial epidermis (Ju et al., 2017). These same authors suggest that higher stomatal densities and smaller stomatal sizes improved the sensitivity of the stomatal regulation of the plants and improved the stomatal function at similar leaf areas. Agarie et al. (1998) showed that Si applications significantly decreased stomatal transpiration by influencing stomatal movement in rice plants.
In general, 25% of the six soil water comparisons exhibited a statistical difference for the effect of soil water conditions on abaxial surface stomatal density, given the levels of the other factors (Table 2). The Guri INTA CL cultivar supplemented with 4.0 mg L\(^{-1}\) Si and at 50% WRC showed a higher stomatal density per mm\(^2\) than at 100% WRC and did not differ statistically from immersed plants. The IRGA 424 RI cultivar supplemented with 8.0 g L\(^{-1}\) at 100% WRC showed higher stomatal density, but there was no statistical difference compared to plants at 50% WRC.

Comparing the cultivars, the only statistical difference was for abaxial surface stomatal density at 100% WRC and 4.0 g L\(^{-1}\) Si, and the IRGA 424 RI cultivar exhibited a higher mean abaxial surface stomatal density than the Guri INTA CL cultivar (Table 2). This may indicate a greater capacity to absorb and fix CO\(_2\) in this cultivar and under these conditions.

### Table 2 – Comparison of means for abaxial surface stomatal density in rice leaves for cultivars under different water conditions and Si rates. Santa Maria, RS, 2019.

| RATES OF Si | CULTIVAR       | WATER SOIL CONDITIONS |
|-------------|----------------|-----------------------|
|             | 50 % WRC       | 100 % WRC             | Water blade |
| 0 g.L\(^{-1}\) | Guri INTA CL   | 09.16 Aa*             | 10.16 Aa    | 07.33 Aa |
|             | IRGA 424 RI    | 11.33 Aa              | 09.00 Aa    | 09.66 Aa |
In terms of regression analysis for the rates of Si, abaxial surface stomatal density (Figure 2a) for the IRGA 424 RI cultivar showed a quadratic increase at 100% WRC, rising to a maximum at an Si rate of 9.0 g L$^{-1}$. None of the other water conditions fit the polynomial models. Under water deficit (50% WRC) and water blade conditions (Figure 2b), the Guri INTA CL cultivar also showed a quadratic increase in abaxial surface stomatal density, reaching a maximum at a rate of 11 g L$^{-1}$ Si for both water conditions. At 100% WRC, there were no fits to any polynomial model. Research has shown that environmental factors (e.g. light) regulate the development of stomata in young leaves through a mechanism for detecting the levels of these factors in mature leaves on the same plant (Nadeau, 2002). The presence of a layer of silica on leaf surfaces is capable of altering the emissivity spectrum (Da Luz, 2006). Thus, higher rates of Si can induce a thicker layer on leaf surfaces, which can affect light transmittance and consequently the formation and density of stomata.

![Figure 2 – Relationship between abaxial surface stomatal density in IRGA 424 RI and Guri INTA CL rice cultivars. under different water conditions (50 and 100% WRC and water blade) and Si rates. Santa Maria. RS. 2019.](image-url)
WRC), increasing capacity to transfer excitation energy from the antenna pigments to the reaction centers. The fact that Fv/Fm values were maintained in 43% of comparisons for water deficit condition, given the other factors, may be related to supply of Si. Tatagiba et al., (2017) in a study with *Oryza sativa* L leaves, supplied or not with Si, they found increases in the values of Fv/Fm, in the plants that were supplied with Si. The Fv/Fm value differed between plants under water deficit (50% WRC) and water blade, in the other 19% of comparisons, with lower Fv/Fm values in plants under water deficit, evidence of an impairment of the photosynthetic apparatus. Decreases in the efficiency of PSII can be caused by a drop in the accumulation of acceptors, and by increased thermal dissipation of excess excited energy prior to reaching PSII reaction centers (Demmig-adams *et al*., 1995).

**Table 3** – Comparison of means for photochemical efficiency of PSII (Fv/Fm) in rice plants under different water conditions and for two cultivars and two Si sources in different Si rates. Santa Maria, RS, 2019.

| RATES OF Si | CULTIVAR   | WATER SOIL CONDITIONS AND SOURCES OF Si | 50 % WRC | 100 % WRC | Water blade |
|------------|------------|----------------------------------------|----------|-----------|------------|
|            |            | potassium metasilicate | sodium metasilicate | potassium metasilicate | sodium metasilicate | potassium metasilicate | sodium metasilicate |
| 0 g.L⁻¹    | Guri INTA CL | 0.66 Abβ* | 0.75 Aaα | 0.78 Aaα | 0.78 Aaα | 0.80 Aaα | 0.77 Aaα |
|           | IRGA 424RI  | 0.69 Aaα | 0.61 Bbβ | 0.76 Aaα | 0.74 Aaα | 0.75 Aaα | 0.81 Aaα |
| 4.0 g.L⁻¹  | Guri INTA CL | 0.75 Aaα | 0.77 Aaα | 0.82 Aaα | 0.79 Aaα | 0.79 Aaα | 0.81 Aaα |
|           | IRGA 424RI  | 0.76 Aaα | 0.74 Aaα | 0.76 Aaα | 0.80 Aaα | 0.80 Aaα | 0.77 Aaα |
| 8.0 g.L⁻¹  | Guri INTA CL | 0.81 Abα | 0.79 Abα | 0.88 Aabα | 0.86 Aabα | 0.89 Aaα | 0.88 Aaα |
|           | IRGA 424RI  | 0.73 Bbα | 0.76 Abα | 0.85 Aaα | 0.89 Aaα | 0.86 Aaα | 0.87 Aaα |
| 16 g.L⁻¹   | Guri INTA CL | 0.79 Abα | 0.79 Abα | 0.86 Aabα | 0.92 Aaα | 0.89 Aaα | 0.84 Aabα |
|           | IRGA 424RI  | 0.80 Abα | 0.82 Aaα | 0.90 Aaα | 0.88 Aaα | 0.94 Aaα | 0.87 Aaα |

Coefficient of variation = 5.43%

*Means not followed by the same uppercase letter in the column were statistically different between cultivars and the same lowercase letter and greek on the rows for soil water regimes and different for Si respectively sources in the Tukey test (p≤0.05). WRC: soil water retention capacity.

With regard to the effects of cultivar and Si source on maximum PSII photochemical efficiency (Fv/Fm), of the twenty-four comparisons for each factor, 92% showed no statistical differences for different Si sources and cultivars (Table 3).

In the regression analysis for Si rates and maximum PSII photochemical efficiency (Fv/Fm) (Figures 3a and 3b), both cultivars IRGA 424 RI and Guri INTA CL, supplemented with potassium metasilicate, showed a linear increase in the values of Fv/Fm under all three soil water conditions, with respective increases of 14, 18 and 25% at 50% and 100% WRC and for immersed plants at the highest rate of potassium metasilicate (16 g L⁻¹) compared to plants not supplemented with Si (0g L⁻¹). For the Guri INTA CL cultivar at 50% WRC, all rates of potassium metasilicate produced a positive quadratic increase, reaching a maximum at a rate of 11 g L⁻¹, whereas at 100% WRC and when immersed, there
was a respective linear increase of 10% and 11% in Fv/Fm values at the highest rate of potassium metasilicate compared to plants not supplemented with Si.

For the sodium metasilicate source, the IRGA 424 RI cultivar under soil water conditions of 50% and 100% WRC (Figure 3c) showed quadratic increases in the values of Fv/Fm, reaching respective maximums at rates of 14 and 12 g L\(^{-1}\), and a linear increase in water blade conditions, with a 7% increase in the Fv/Fm values at the highest rate of sodium metasilicate (16 g L\(^{-1}\)) compared to plants not supplemented with Si (0 g L\(^{-1}\)) (Figure 3c). For the Guri INTA CL cultivar, the rates of sodium metasilicate resulted in respective linear and quadratic increases at 100% WRC and water blade, with an 18% increase in the Fv/Fm values at 100% WRC at the highest rate of sodium metasilicate (16 g L\(^{-1}\)) compared to plants not supplemented with Si (0 g L\(^{-1}\)), and reaching a maximum at a rate of 10 g L\(^{-1}\) in immersed plants. There were no fits to polynomial models at 50% WRC.

Figure 3 – Relationship between maximum photochemical efficiency of PSII (Fv/Fm) and Si rates in IRGA 424 RI and Guri INTA CL rice cultivars under different water conditions (50 and 100% WRC and water blade) and for different Si sources (sodium and potassium metasilicate). Santa Maria. RS. 2019.

The relationship between Si rate and electron transport rate (ETR\(_{1500}\)) (Figure 4a) was quadratic, reaching a maximum at a rate of 16 g L\(^{-1}\), resulting in an increase in photosynthesis, possibly due to the Si boosting the photosynthetically active leaf area index (Kozlowski, 2002). The beneficial effect of Si on Fv/Fm and ETR\(_{1500}\) values can be attributed, at least in part, to anatomical variations produced by the
deposition of silica on the walls of epidermal cells, which keep the leaves upright and improve light interception, stimulating photosynthesis (MA & TAKAHASHI, 2002; SAVIO et al., 2011).

Figure 4 b shows that the regression analysis of Si rates for effective quantum yield of PSII (YII_{125}), which, as for an ETR\textsubscript{1500} resulted in a quadratic increase, reaching a maximum at 16 g L\textsuperscript{-1}, evidence of increased photosynthetic performance in plants supplemented with Si. Normally, the stress on the plant caused by any environmental factor can lead to an initial reduction in YII_{125} and in Fv/Fm, linked to impaired functionality of the photosynthetic apparatus (Kramer et al., 2004). The maximum efficiency photochemical efficiency of the FSII or the FV / FM ratio is a sensitive indicator of the photosynthetic performance of plants, with values ranging from 0.75 to 0.85 (Wagner & Merotto Junior, 2014). In plants not supplemented with Si and kept at 50% WRC, Fv/Fm values were below optimal, evidencing impairment of the photosynthetic apparatus during water drought. In the presence of Si, there was an increase in Fv/Fm in plants under water deficit and under optimal conditions, indicating the importance of this element for boosting photosynthesis. One possible explanation for the highest YII_{125} values observed in plants supplemented with Si is that, in the presence of Si, the plants began using a higher fraction of YII_{125} excitation energy instead of dissipating it in the form of heat, thus increasing photosynthetic yield (Klughamer and Schreiber, 2008). Therefore, according to the findings of this study, Si supplementation enhances photosynthetic yield in rice plants.

Figure 4 – Relationship between electron transport rate (ETR\textsubscript{1500}) (a) and effective quantum yield of PSII (YII) (b) and silicon rates in rice shoots (means for Si sources (sodium and potassium metasilicate). cultivars (IRGA 424 RI and Guri INTA CL) and water conditions (50 and 100% WRC and water blade). Santa Maria. RS. 2019.

Comparing the effect of soil water conditions on total dry mass (Table 4), in the sixteen comparisons, given the other factors, 56% showed higher total dry mass for immersed plants. The drop in total dry mass under the other soil water conditions was probably due to lower cell expansion and lower turgor pressure (Kramer, 1994), impairing plant growth and development and impacting dry mass accumulation. Similar results in rice cultivars were reported by Mauad (2003; 2006), Crisciol et al. (2003 a) and Crisciol et al. (2003b).
Table 4 – Comparison of means for rice plant total dry mass for different water conditions and cultivars at different Si rates and for different Si sources. Santa Maria. RS. 2019.

| RATES OF Si | CULTIVAR         | WATER SOIL CONDITIONS AND SOURCES OF Si |
|-------------|------------------|-----------------------------------------|
|             |                  | 50 % WRC                               | 100 % WRC                               | Water blade |
|             |                  | Potassium metasilicate                  | Sodium metasilicate                      | Potassium metasilicate | Sodium metasilicate |
|             |                  | 0.29 Abβ*                               | 0.67 Aaabα                               | 0.65 Abα            | 0.36 Abα            | 1.44 Aaα            | 0.82 Aaβ            |
| 0 g.L⁻¹     | Guri INTA CL     | 0.28 Abα                               | 0.26 Bba                               | 0.52 Aaabα          | 0.71 Aaα           | 0.76 Baα           | 0.92 Aaa            |
|             | IRGA 424RI      | 0.25 Abα                               | 0.57 Abα                               | 0.49 Aaabα          | 0.54 Abα           | 0.92 Aaβ           | 1.32 Baα            |
|             |                  | 0.32 Abα                               | 0.51 Abα                               | 0.75 Aaα            | 0.61 Abu           | 0.68 Aabβ          | 1.80 Aaα            |
| 4.0 g.L⁻¹   | Guri INTA CL     | 0.24 Abα                               | 0.49 Abα                               | 0.53 Abα            | 0.80 Abu           | 2.93 Aaa            | 2.35 Aaβ            |
|             | IRGA 424RI      | 0.31 Abα                               | 0.50 Abα                               | 0.65 Aaabα          | 0.73 Abu           | 0.99 Baβ           | 2.17 Aaα            |
| 8.0 g.L⁻¹   | Guri INTA CL     | 0.53 Abα                               | 0.53 Abα                               | 0.53 Bba            | 0.64 Abu           | 2.79 Aaa            | 2.55 Aaa            |
|             | IRGA 424RI      | 0.45 Aaα                               | 0.38 Abα                               | 0.94 Abα            | 0.51 Abβ           | 1.47 Bcβ           | 2.31 Aaα            |
| 16.0 g.L⁻¹  | Guri INTA CL     | 0.53 Abα                               | 0.53 Abα                               | 0.53 Bba            | 0.64 Abu           | 2.79 Aaa            | 2.55 Aaa            |
|             | IRGA 424RI      | 0.45 Aaα                               | 0.38 Abα                               | 0.94 Abα            | 0.51 Abβ           | 1.47 Bcβ           | 2.31 Aaα            |

Coefficient of variation = 24.75%

*Means not followed by the same uppercase letter in the column were statistically different between cultivars and the same lowercase letter and greek on the rows for soil water regimes and different for Si respectively sources in the Tukey test (p≤0.05). WR: soil water retention capacity.

In terms of the effects of cultivar and Si source on total dry mass, in the twenty-four comparisons, 75% showed no statistical difference between cultivars, indicating very similar Si action for both factors, possibly due to the genetic similarity of the cultivars (Table 4). For Si source (Table 4), in 16% of comparisons the sodium metasilicate source resulted in higher mean total dry mass compared to potassium metasilicate; in most cases, this statistical difference between Si sources was found in immersed plants.

In the Guri INTA CL cultivar supplemented with potassium metasilicate (Figure 5a), there was a quadratic adjustment for immersed plants, where for the soil water conditions of 50 and 100% of the WRC there were not adjustments of polynomial models. For the IRGA 424 RI cultivar, potassium metasilicate rates at 100% WRC and in immersed plants (Figure 5b) there was a linear fit, increase of 77 and 93%, respectively, at the highest rate of potassium metasilicate (16 g L⁻¹) compared to plants not supplemented with Si (0 g L⁻¹).

For the sodium metasilicate source, both cultivars when immersed (Figures 5c and 5d) showed a quadratic increase, reaching respective maxima at rates of 15 and 13 g L⁻¹. At 50% and 100% WRC, there was no significant polynomial fit.
Thus, we found that there was a relationship between the availability of Si and total dry mass of the plant, possibly due to improved use of the water available in the soil. Lee et al. (2010) reported that Si boosts growth by enhancing the absorption of mineral nutrients, possibly through a kinetic relationship with the absorption processes for some nutrients. Faria Junior et al. (2009), evaluating the effects of increased Si rates on rice cultivars (Conai and Curinga), found an increase in plant root dry matter. Sávio et al. (2011) reported that Brachiaria brizantha and Panicum maximum responded positively to Si in terms of dry weight yield. These results are not consistent with those reported by Melo et al. (2003), who studied the effect of Si on dry matter yield in two species of the genus Brachiaria under different soil water conditions. Melo et al. (2003) reported that, despite the accumulation of high quantities of Si in the shoot, there was no effect on the tolerance of both species to water deficiency in the presence of Si.

4. Conclusion

The physiological responses of rice plants to silicon supplementation were a) higher stomatal density, adapting them to water deficit; b) increased maximum photochemical efficiency of PSII (Fv/Fm) in plants under both water deficit and optimum conditions, boosting photosynthesis; and c) increased effective quantum yield of PSII (YII). In morphological terms, there was an increase in the total dry weight in plants supplemented with silicon under both water deficit and optimum conditions. Therefore, silicon attenuated water deficit stress in plants grown from seeds treated with dietholate.
5. Acknowledgements

To the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for the scholarship granted to first author.

6. References

Agarie, S., H. Uchida, W. Qgata, F. Kubota, & P. B. Kaufman. (1998). Effects of silicon on transpiration and leaf conductance in rice plants (Oryza sativa L). *Japanese Journal of Crop Science* 1: 89–95.

Artigiani, A.C.C, Crusciol, C.A.C, Arf, O, Alvarez, R.C.F & Nascente, A.S. (2012). Produtividade e qualidade industrial do arroz de terras altas em função da disponibilidade hídrica e adubação. *Pesquisa Agropecuária Tropical*. V.42, n.3, p-340-349. Goiânia. doi: 0.1590/S1983-40632012000300011

Carneiro, M.M.C.; Deuner, S.; Oliveira, P.V.; Teixeira, S.B.; Sousa, C. P.; Bacarin, M.A. & Moraes, D.M. (2011). Atividade antioxidante e viabilidade de sementes de girassol após estresse hídrico e salino. *Revista Brasileira de Sementes.*, v. 33, n. 4, p. 752 – 761.

CONAB: Companhia Nacional De Abastecimento. (2017). Acompanhamento da safra brasileira - Grãos. Safra 2016/17. Monitoramento agricola- Safra 2017, v. 4, n. 1, p. 1–98.

Crusciol, C. A. C., Arf, O., Soratto, R. P., Rodrigues, R. A. F. & Machado, J. R. (2003ª). Produtividade do arroz de terras altas em função de lâminas de água e da adubação mineral. *Científica*, Jaboticabal, v. 31, n. 1, p. 189-200. doi:10.1590/S0100-06832003000400009

Crusciol, C. A. C., Arf, O., Soratto, R. P., Rodrigues, R. A. F. & Machado, J. R. (2003b). Manejo de irrigação por aspersão com base no Kc e adubação mineral na cultura de arroz de terras altas. *Bragantia*, Campinas, v. 62, n. 4, p. 465-75. doi:10.1590/S0006-87052003000300013.

Da Luz, B. R. (2006). Attenuated total reflectance spectroscopy of plant leaves: A tool for ecological and botanical studies. *New Phytologist*. v. 172, p. 305-318. doi: 10.1111/j.1469-8137.2006.01823.x

Demmig-Adams, B., Adams, W.W. II, logan, B.A. & Verhoeven, A.S. (1995). Xanthophyll cycle-dependent energy dissipation and flexible PS II efficiency in plants acclimated to light stress. *Australian Journal of Plant Physiology*, Victoria, v.22, p.249-260. doi:10.1071/PP9950249

Embrapa arroz e feijão. (2019). *Dados conjunturais da produção de arroz (Oryza sativa L.) no Brasil (1986 a 2018): área, produção e rendimento*. Santo Antônio de Goiás: Embrapa Arroz e Feijão. Available at: <http:// http://www.cnptaf.embrapa.br/socioeconomia/index.htm >. Accessed: 04 de março de 2020.

Etesami, H. & Jeong, B.R. (2017). Silicon (Si): reviewand future prospectsontheactionmechanisms in alleviatingbioticandabiotictresses in plants. *Ecotoxicologyand Environmental Safety*. V. 147. p.881-896. doi:10.1016/j.ecoenv.2017.09.063

FAO. Base de dados Faostat. (2018). Available at http://www.fao.org/faostat/es/#data/QC. Accessed: 09 de Agosto de 2018.

Faria Junior, L. A. de, Carvalho, J. G. de, Pinho, P. J. de, Bastos, A. R. R. & Ferreira, E. V. (2009). Produção de matéria seca, teor e acúmulo de silício em cultivares de arroz sob doses de silício. *Ciência e Agrotecnologia*, Lavras, v. 33, n. 4, p. 1034-1040. doi:10.1590/S1413-70542009000400013.
Ferreira, D. F. (2014). Sisvar: a Guide for its Bootstrap procedures in multiple comparisons. *Ciência e Agrotecnologia*, v.38, n.2, p.109-112. doi:10.1590/S1413-70542014000200001.

Fábregas N., et al. (2018) Overexpression of the vascular brassinosteroid receptor BRL3 confers drought resistance without penalizing plant growth. *Nature Communications*, v.9 , p. 4680. doi:10.1038/s41467-018-06861-3

Gill S.S. & Tuteja N. (2010). Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plants. *Plant Physiol Biochem.* 48(12):909-930. doi:10.1016/j.plaphy.2010.08.016

Inoue, M.H., Araújo, T.D.C., Mendes, K.F., Ben, R. & Conciani, P.A. (2012). Eficiência do dietholuate e bioestimulantes isolados e associados no tratamento de sementes de algodoeiro adensado com clomazone aplicado em pré-emergência. *Revista de Ciências Agro-ambientais*. V.10; n.2, p. 163-172. doi:10.1016/j.plaphy.2010.08.016

Ju S., Wang L., Zhang C., Yin T. & Shao S (2017) Alleviatory effects of silicon on the foliar micromorphology and anatomy of rice (*Oryza sativa* L.) seedlings under simulated acid rain. PLoS ONE 12(10): e0187021. https://doi.org/10.1371/journal.pone.0187021

Karam, D., Carneiro, A.A., Albert, L.H., Da Cruz, M.B., Costa, G.T. & Magalhães P.C. (2003). Seletividade da cultura do milho ao herbicida clomazone por meio do uso de dietholuate. R. Bras. Milho Sorgo, v. 2, n. 1, p. 72-79. doi: 10.18512/1980-6477/rbms.v2n1p72-79

Klughamer, C. and Schreiber, U. (2008). Saturation pulse method for assessment of energy conversion in PS I. *PAM Application Notes* 1: 11-14.

Kramer, D. M., Johnson, G., Kiirats, O. & Edwards, G. E. (2004). New Fluorescence Parameters for the Determination of QARedox State and Excitation Energy Fluxes. *Photosynthesis Research*, 79(2), 209–218. doi:10.1023/b:pres.0000015391.99477.0d

Kozlowski, T. T. & Pallardy, S. G. (2002). Acclimation and Adaptive Responses of Woody Plants to Environmental Stresses. *The Botanical Review*, v. 68, n. 2, p.270–334. doi:10.1663/0006-8101(2002)068[0270:aaarow]2.0.co;2

Lee, S.K., Sohn, E.Y., Hamayun, M., Yoon, J.Y. & Lee, I.J. (2010). Effect of silicon on growth and salinity stress of soybean plant grown under hydroponic system. *Agroforest Systems*, v.80, p.333–340. doi:10.1007/s10457-010-9299-6

Luković, J. et. al. (2009). Histological characteristics of sugar beet leaves potentially linked to drought tolerance. *Industrial Crops and Products*, v.30, n.2, p 281-286. doi:10.1016/j.indcrop.2009.05.004

Ma, J.F. & Takahashi, E. (2002). Soil, Fertilizer, and Plant Silicon Research in Japan. Elsevier Science, Amsterdam. Cap. 6, p.73-106.

Mauad, M., Crusciol, C. A. C. & Filho, H. G. (2011). Dry matter and plant nutrition of upland rice under water deficit and silicon fertilization. *Semina: Ciências Agrárias*, Londrina, v. 32, n. 3, p. 939-948. doi:10.5433/1679-0359.2011v32n3p939

Meier, U. (2001). *Growth stages of mono-and dicotyledonous plants - BBCH*. 2. ed. Berlin: German 412 Federal Biological Research Centre for Agriculture and Forestry.

Melo, S. P. de, Korndörfer, G. H., Korndörfer, C. M., Lana, R. M. Q. & Santana, D. G. de. (2003). Silicon accumulation and water deficit tolerance in *Brachiaria* grasses. *Scientia Agricola*, 60(4), 755-759. doi:10.1590/S0103-90162003000400022
Pillitteri, L. J. and Dong, J. (2013). Stomatal Development in *Arabidopsis*. The *Arabidopsis* Book, 11, e0162. doi:10.1199/tab.0162

Sávio, F. L.; Silva, G. C.; da; Teixeira, I. T. & Borém, A. (2011). Produção de biomassa e conteúdo de silício em gramíneas forrageira sob diferentes fontes de silicato. Semina: Ciências Agrárias, Londrina, v. 32, n. 1, p. 103-110. doi:10.5433/1679-0359.2011v32n1p103

Sancho, D.M. et al. (2010) Phorate e dietholate protegem o arroz da fitotoxicidade do clomazone em doses elevadas. *Planta Daninha*, v.28, n.4, p.909-912.

Santana, J.R.F., Paiva, R.; Resende, R. K. S., Castro, E.M.; Pereira, F.D. & Oliveira, L. M.. (2008). Estímulo do comportamento fotoautotrófico durante o enraizamento *in vitro* de *Annona glabra* L. Aspectos da anatomia da folha antes da aclimatização. *Ciência agrotécnica*, v.32, n.2, p.640-4. doi:10.1590/S1413-70542008000200046

Schwab, N. T. (2011). Disponibilidade hídrica no cultivo de cravina em vasos com substrato de cinzas de casca de arroz. Santa Maria: UFSM. 2011. 82p. Dissertação Mestrado.

Sharma, P. & Dubey, R.S. (2005). Modulation of nitrate reductase activity in rice seedlings under aluminium toxicity and water stress: role of osmolytes as enzyme protectant. *Journal of Plant Physiology*, v. 162, p. 854-862. doi:10.1016/j.jplph.2004.09.011

Stone, L. F. (1984). Deficiência hídrica, vermiculita e cultivares. *Pesquisa Agropecuária Brasileira*, Brasília, v. 19, n. 6, p. 695-707.

Taiz L., Zeiger E., Moller I. M. and Murph A. (2017). *Fisiologia e Desenvolvimento Vegetal*. 6ª ed. Artmed Editora S.A. 120p.

Tatagiba S.D, Peloso A. F. & Rodrigue F. A. (2017). O silício potencializa o desempenho fotoquímico em folhas de arroz. *Revista Univap*, v. 22, n. 40, p. 750. doi:10.18066/revistaunivap.v22i40.1533

Wagner, J. F. & Merotto Junior, A. (2014). Parâmetros fisiológicos e nutricionais de cultivares de soja resistentes ao glifosato em comparação com cultivares isogênicas próximas. *Ciência Rural*, v.44, p.393-399. doi:10.1590/S0103-84782014000300002

Yadav D.K., Barik B.R. & Pradhan G., et al. (2019). Responses of crops plant to drought and its management for crop water availability: a review J. Pharmacogn. Phytochem., v.8, p. 167-172.

Zia, Z, Bakhat, H.F, Saqib, Z.A, Shah, G.M, Fahad, S. & Ashraf, M.R. (2017). Effect of water management and silicone on germination, growth, phosphorus and arsenic uptake in rice. *Ecotoxicology and Environmental Safety*. V.144. p.11-18. doi:10.1016/j.ecoenv.2017.06.004.