# Silicon In the Growth of Rice Seedlings Pretreated with Dietholate and Subjected to Cold Stress

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## Abstract

*Silicon (Si) is an enzyme stimulator that can promote signaling for the production of antioxidant compounds, important in cellular detoxification of excess ROS accumulated during stress. The aim of the study was to evaluate the effects of Si on post-germination rice seeds in the mitigation of cold stress combined with stress induced by seed treatment with the dietholate protector. The experimental design was fully randomized with three replicates and a 3x2x2x4x2 factorial arrangement: three temperatures (5, 10 and 20 °C), two cultivars (IRGA 424 RI and Guri INTA CL), two seed pretreatments (with and without dietholate), four rates of Si (0; 4.0; 8.0 and 16 mg.L$^{-1}$) and two sources of Si (sodium and potassium metasilicate). Seed pretreatment with dietholate reduced shoot and radicle length, especially at the lower temperatures of 5 and 10 °C. Sodium metasilicate as the source of Si was more efficient in boosting shoot and radicle length, both with and without pretreatment, regardless of temperature. Si was found to attenuate low-temperature stress and the impairment of shoot and radicle growth in rice seedlings grown from seeds pretreated with dietholate.*


**Keywords:** abiotic stress, sodium metasilicate, potassium metasilicate.

## 1. Introduction

Rice is the second most cultivated cereal in the world. Average production in Brazil reached 11.6 million metric tons over the last five years. The state of Rio Grande do Sul (RS) is the biggest Brazilian producer, with average production of 8,000 metric tons over the last five years (CONAB, 2019). However, this figure is below the estimated yield potential of up to 12 metric tons ha-1, for the main cultivars used, such as Guri INTA CL, Puitá INTA CL and IRGA 424 RI.

Several factors can lead to a drop in average grain yield and are related to both biotic and abiotic stresses. Stress due to low temperature can be particularly problematic, affecting rice plants in 25 countries, with negative impacts on productivity (Cruz et al., 2013). Biotic stresses include competition from poaceous weeds (grasses) (Silva et al., 2006).

The main herbicide widely used in rice areas in the state of RS is clomazone (Andres and Machado, 2004), which at excessive rates can cause phytotoxicity, negatively impacting the development of post-germination seedlings and tiller production (Sanchotene et al., 2010). Safener dietholate is used to protect seedlings from the phytotoxic effects of clomazone. It allows seedling to withstand higher doses of clomazone (Karam et al., 2003) by inhibiting the action of some enzymes. However, these enzymes are also responsible for reducing the damaging effects of reactive oxygen species (ROS), formed during cellular metabolism (Gill et al., 2013) under biotic and abiotic stress conditions.

In RS, rice growers have reported the adverse effects of dietholate combined with cold stress on the Guri INTA CL cultivar, affecting the development of post-germination seedlings and tiller production. The ideal air temperature for crop development is between 25 ºC and 30 ºC (Yoshida, 1981). However, air temperatures below 15 °C at night are very common in RS during post-germination seedling development, delaying initial development and causing leaf depigmentation, wilting and low growth rates (Song et al., 2011). In addition, low temperatures can increase the levels of reactive oxygen species (ROS) and malondialdehyde (MDA), resulting in oxidative cell damage (Theocharis et al., 2012).

To reduce these effects, formulated products containing some essential minerals and phytohormones have been used (Inoue et al., 2012), as well as silicon (Si) (Mauad et al., 2011). Si has positive effects on plants subjected to exogenous stresses. It is an enzyme stimulator (Detmann et al., 2013; Taiz et al., 2017) and can affect plant growth and development by promoting signaling for the production of antioxidant compounds, such as superoxide dismutase (SOD), catalase (CAT) and ascorbate peroxidase (APX) (Etesami et al., 2017). There are a number of Si-mediated stress improvement mechanisms, but the main stress-combating strategies triggered by Si are improved photosynthesis in stressed plants (Gong and Chen, 2012; Muneer et al., 2014),and the stimulation of signaling for the production of osmoprotective compounds, such as proline, which promotes osmotic adjustment in plants, helping them tolerate the cold by maintaining membrane integrity and preventing cellular dehydration caused by osmotic pressure (Etesami et al., 2017 and Huang et al., 2012).

Thus, the aim of this study was to evaluate the effects of Si in the mitigation of cold stress associated with dietholate seed pretreatment in post-germination rice seeds.

## 2. Material and methods

The experiment was carried out "in vitro", in a fully randomized design, with three replicates. Each Petri dish (diameter 150 mm) formed an experimental unit with two layers of germitest paper, containing 20 seeds per plate.

The 96 treatments were arranged in a 3x2x2x4x2 factorial scheme: three germination temperatures (5, 10 and 20 °C), two cultivars (IRGA 424 RI and Guri INTA CL), two seed treatments (with and without dietholate - Permit Star® - at a rate of 6 mL per kg of seeds), four rates of Si (0; 4.0; 8.0 and16 $mg.L^{-1}$) 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%), totaling 288 experimental units.

Si solutions from both sources (sodium and potassium metasilicate) were prepared at rates of 0.0; 4.0; 8.0 and 16 $mg.L^{-1}$, diluted in distilled water, with the pH adjusted to 5.8 ± 1.

The size of the seedlings to be treated with Si was previously standardized by inoculating the rice seeds in Petri dishes containing two sheets of Germitest paper pre-soaked in 20 ml distilled water, and kept in a growth chamber at 25 °C ± 1 for 72 h, with a 16-hour photoperiod. Seeds with a 1.0 mm radicle were then transferred to Petri dishes with two sheets of Germitest paper pre-soaked in 20 ml of the appropriate silicon rate, and taken to different BOD germination chambers set to the three temperatures of 5, 10 and 20 °C, for seven days, with a 16-hour photoperiod. They were then transferred to a growth room with a 16-hour photoperiod (RFA of ~ 73 $\mu mol\ m^{-2}\ s^{-1}$) and temperature of 25 °C ± 1, for a further seven days. At the end of this procedure, data on the initial growth of the seedlings were collected, and the length (cm) of the radicle and shoot measured using a millimeter ruler.

Shoot and radicle length data were subjected to analysis of variance, and the means compared using the Tukey test at 0.05 % error probability, except for the silicon rates which were fitted to polynomial models using Sisvar® 5.3 statistical software (Ferreira, 2014).

## 3. Results and discussions

Shoot and radicle length variables showed significant interaction for all factors (temperature x cultivar x seed pretreatment x Si rate x Si source).

Comparison of diethloate pretreated and non-pretreated seeds with the other factors showed that, for each of the variables in the sixteen comparisons performed for each of the three temperatures (5, 10 and 20 °C), with respective results of 87, 94 and 69% for shoot length and 81, 75 and 56% for radicle length, dietholate seed pretreatment significantly reduced shoot and radicle length, with deleterious effects especially at the lower temperatures (5 and 10 °C), compared to seedlings without pretreatment (Table 1). Dietholate inhibits antioxidant enzymes (Sacandalios, 2005), mainly associated with low temperature stress (Rosa et al., 2017), which neutralize or eliminate reactive oxygen species (ROS) (Shah et al., 2001) resulting from abiotic or biotic stresses against which the seedlings must be protected. These ROS peroxidize lipids and adversely affect cell division, delaying or impairing seed germination and/or seedling vigor (Inoue et al., 2012; Rosa et al., 2017), prolonging the crop cycle by increasing emergence time and affecting tiller production.

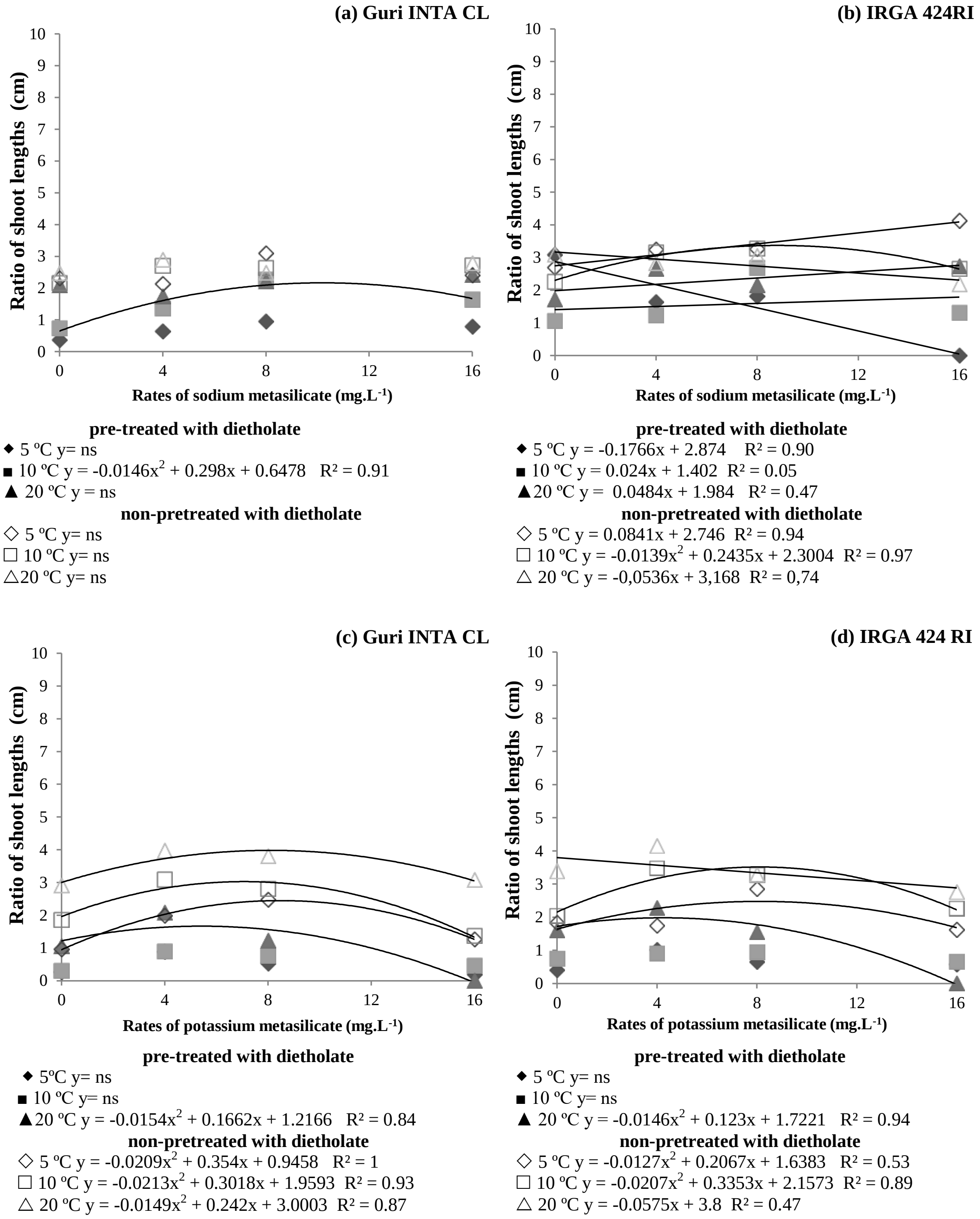
Table 1. Comparison of means for shoot and radicle length in rice seedlings with and without dietholate pretreatment for different temperatures, cultivars, sources and rates of silicon.

| | | | | Ratio of shoot lengths | | | | Ratio of radicle lengths | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Rates of Si (mg.L$^{-1}$) | | | | | | | |
| Temperatures | Sources of Si | Cultivars | Seed pretreatments | 0 | 4 | 8 | 16 | 0 | 4 | 8 | 16 |
| 5 °C | potassium metasilicate | Guri INTA CL | with dietholate | 0.27 b | 0.89 b | 0.54 b | 0.19 b | 0.42 a | 0.87 b | 0.76 b | 0.11 a |
| | | | without dietholate | 0.97 a | 1.99 a | 2.48 a | 1.26 a | 0.95 a | 2.42 a | 2.41 a | 0.23 a |
| | | Irga 424RI | with dietholate | 0.40 b | 1.03 b | 0.65 b | 0.59 b | 1.07 b | 1.37 b | 0.61 b | 0.22 a |
| | | | without dietholate | 1.83 a | 1.75 a | 2.87 a | 1.62 a | 3.40 a | 2.76 a | 2.42 a | 0.58 a |
| | sodium metasilicate | Guri INTA CL | with dietholate | 0.39 b | 0.65 b | 0.97 b | 0.80 b | 0.98 b | 1.20 b | 1.8 b | 1.34 b |
| | | | without dietholate | 2.32 a | 2.14 a | 3.10 a | 2.41 a | 3.15 a | 3.07 a | 4.09 a | 4.54 a |
| | | Irga 424RI | with dietholate | 2.70 a | 1.64 b | 1.83 b | 0 b | 2.15 b | 0 b | 0 b | 0 b |
| | | | without dietholate | 3.09 a | 3.25 a | 3.26 a | 4.17 a | 4.34 a | 4.66 a | 5.04 a | 6.20 a |
| 10 °C | potassium metasilicate | Guri INTA CL | with dietholate | 0.31 b | 0.89 b | 0.76 b | 0.46 b | 0.39 b | 1.40 b | 0.93 b | 0.29 a |
| | | | without dietholate | 1.87 a | 3.09 a | 2.81 a | 1.36 a | 3.69 a | 4.99 a | 3.17 a | 0.81 a |
| | | Irga 424RI | with dietholate | 0.75 b | 0.91 b | 0.95 b | 0.66 b | 1.64 b | 1.42 b | 1.58 b | 0.18 a |
| | | | without dietholate | 2.04 a | 3.48 a | 3.29 a | 2.27 a | 3.80 a | 5.19 a | 3.85 a | 1.29 a |
| | sodium metasilicate | Guri INTA CL | with dietholate | 0.74 b | 1.37 b | 2.29 a | 1.65 b | 1.67 b | 2.51 b | 3.02 a | 3.38 b |
| | | | without dietholate | 2.16 a | 2.71 a | 2.65 a | 2.73 a | 4.03 a | 4.15 a | 3.71 a | 5.02 a |
| | | Irga 424RI | with dietholate | 1.06 b | 1.23 b | 2.68 a | 1.32 b | 1.96 b | 3.51 a | 3.63 b | 1.17 b |
| | | | without dietholate | 2.26 a | 3.16 a | 3.28 a | 2.67 a | 4.58 a | 4.67 a | 5.54 a | 4.57 a |
| 20 °C | potassium metasilicate | Guri INTA CL | with dietholate | 1.05 b | 2.08 b | 1.24 b | 0 b | 2.24 a | 3.35 b | 2.06 b | 0 b |
| | | | without dietholate | 2.91 a | 3.98 a | 3.81 a | 3.08 a | 3.58 a | 5.21 a | 3.70 a | 1.65 a |
| | | Irga 424RI | with dietholate | 1.61 b | 2.28 b | 1.55 b | 0 b | 3.29 a | 3.69 a | 1.82 b | 0 b |
| | | | without dietholate | 3.40 a | 4.15 a | 3.29 a | 2.76 a | 4.46 a | 4.90 a | 3.32 a | 1.36 a |
| | sodium metasilicate | Guri INTA CL | with dietholate | 2.11 a | 1.74 b | 2.22 a | 2.42 a | 5.22 a | 3.80 a | 3.93 a | 4.54 a |
| | | | without dietholate | 2.47 a | 2.91 a | 2.49 a | 2.79 a | 3.78 b | 4.44 a | 3.67 a | 4.70 a |
| | | Irga 424RI | with dietholate | 1.72 b | 2.67 a | 2.16 b | 2.19 a | 4.33 a | 5.59 a | 6.11 a | 5.51 a |
| | | | without dietholate | 3.10 a | 2.83 a | 3.06 a | 2.75 a | 4.10 a | 4.04 b | 4.71 b | 3.89 b |
| | | | | CV (%) = 20.54 | | | | CV (%) = 26.51 | | | |

* Means for shoot and radicle length with and without dietholate not followed by the same letter in the column differed statistically in the Tukey test ($p < 0.05$).

Regression analysis of the rates of Si and shoot length showed that both cultivars were influenced by the rates of potassium metasilicate. At a temperature of 20 °C, the Guri INTA CL cultivar pre-treated with dietholate (Figure 1c) showed a quadratic increase in the length of the shoot up to a rate of 5.53 $mg.L^{-1}$ potassium metasilicate. At the other temperatures (5 and 10 °C), there was no significant fit to the polynomial models. However, Guri INTA CL cultivar seeds not pre-treated and kept at 5, 10 and 20 °C showed a quadratic increase in shoot length at all three temperatures, peaking at respective rates of 8.75; 7.54 and 12 $mg.L^{-1}$ potassium metasilicate (Figure 1c).

Figure 1. Ratio of shoot lengths for IRGA 424 RI and Guri INTA CL rice cultivars, with and without dietholate seed pretreatment, at different temperatures and for Si rates from different sources (sodium and potassium metasilicate).



For the IRGA 424 RI cultivar pretreated at the seed stage with dietholate and kept at 20 °C, there was a quadratic fit peaking at a rate 6,12 mg.$L^{-1}$ potassium metasilicate. There were no polynomial fits at 5 and 10 °C (Figure 1d). However, without pretreatment, the IRGA 424 RI cultivar supplemented with potassium metasilicate and kept at 5 and 10 °C fit a quadratic polynomial model with shoot length peaking at rates of 10 and 8.25 mg.$L^{-1}$, with a negative linear fit at 20 °C and at the highest rate of potassium metasilicate (16 g$L^{-1}$) reducing shoot length by 18.6% compared to a rate of 0 mg.$L^{-1}$ (no Si supplement) (Figure 1d).

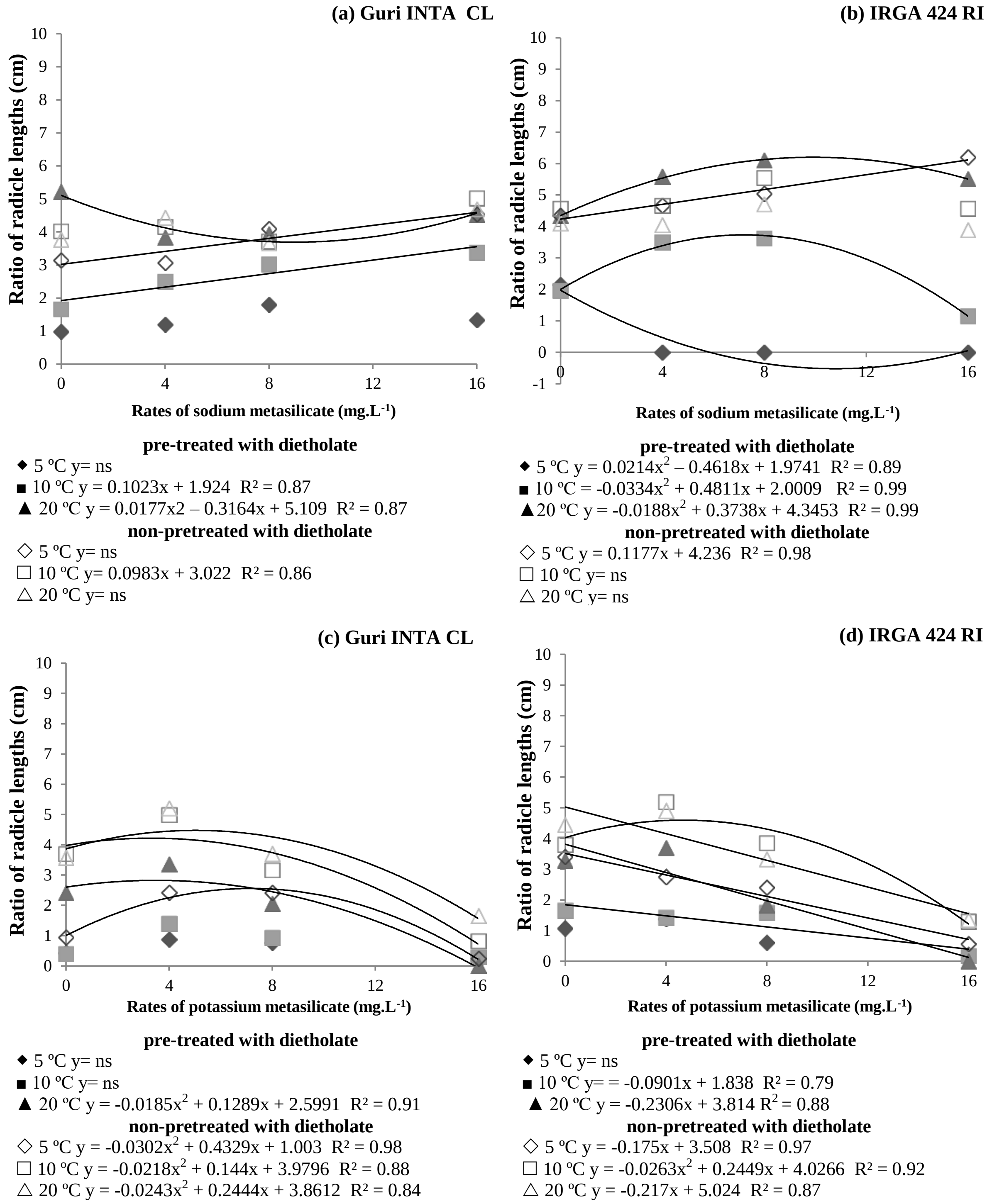
In the regression analysis of sodium metasilicate Si rates (Figures 1a and 1b), the shoot length of the Guri INTA CL cultivar with no dietholate seed pretreatment (Figure 1a) did not fit the polynomial models for all three temperatures (5, 10 and 20 °C). For pretreated seeds, a quadratic polynomial fit was found only at a temperature of 10 °C, peaking at a rate of 9.6 mg.$L^{-1}$ (Figure 1a).

Compared to the IRGA 424 RI cultivar (Figure 1b) with dietholate pretreatment and supplemented with sodium metasilicate, there was a negative linear fit at a temperature of 5 °C, with a 100% reduction in shoot length at a rate of 16 mg.$L^{-1}$ sodium metasilicate compared to plants not supplemented with Si (0 mg.$L^{-1}$), and a positive linear fit at temperatures of 10 and 20 °C, with a respective increase in shoot length of 31 and 59% at the highest rate (16 g.$L^{-1}$) of sodium metasilicate compared to plants not supplemented with Si (0 mg.$L^{-1}$) (Figure 1b). Without seed pretreatment and at 10 °C, there was a quadratic fit peaking at a rate of 12 mg.$L^{-1}$, and at 5 and 20 °C a respective positive and negative linear fit, with an increase of 53% at 5 °C and a drop of 30% at 20 °C in shoot length at a rate of 16 mg. $L^{-1}$ sodium metasilicate compared to plants not supplemented with Si (0 mg.$L^{-1}$) (Figure 1b).

In general, shoot length decreased for both rice cultivars when pretreated with dietholate alone, and increased when the dietholate was combined with Si, showing that rice seeds pretreated solely with the dietholate suffer delays in initial growth, but when associated with Si there is an increase in shoot length. Plants that grow in silicon-rich environment differ from those grown under deficiency conditions mainly in terms of tolerance to abiotic factors (Rodrigues et al. 2011).

In the Si rate and radicle length regression analysis for the Guri INTA CL cultivar with dietholate pretreated seeds, increasing the potassium metasilicate rate at a temperature of 20 °C resulted in a quadratic relationship peaking at a rate of 4.3 mg.$L^{-1}$. At temperatures of 5 and 10 °C, there were no significant fits to polynomial models (Figure 2c). However, for Guri INTA CL cultivar seeds with no pretreatment, increasing the rate of potassium metasilicate at temperatures of 5, 10 and 20 °C was found to boost radicle length quadratically, peaking at respective rates of 7, 2, 3.6 and 6.1 mg.$L^{-1}$ (Figure 2c).

Figure 2. Ratio of radicle lengths for IRGA 424 RI and Guri INTA CL rice cultivars, with and without dietholate seed pretreatment, at different temperatures and for Si rates from different sources (sodium and potassium metasilicate.




For pretreated IRGA 424 RI cultivar seeds, the potassium metasilicate source resulted in a linear drop in radicle length at temperatures of 10 (drop of 66%) and 20 °C (70%), at the highest rate of potassium metasilicate (16 mg.L$^{-1}$) compared to plants not supplemented with Si (0 mg.L$^{-1}$), and at a temperature of 5 ° C there were no fits to polynomial models (Figure 2d). For IRGA 424 RI cultivar seeds with no pretreatment, increasing the potassium metasilicate rate resulted in a linear reduction of radicle length at temperatures of 5 and 20 °C and a quadratic reduction at 10 °C, peaking at a rate of 6.1 mg.L$^{-1}$ (Figure 2d). In terms of sodium metasilicate rates, dietholate pretreated seeds of the IRGA 424 RI cultivar exhibited a quadratic relationship at all three temperatures (5, 10 and 20 °C) peaking at respective rates of 11.5, 8.0, 12.3 mg.L$^{-1}$ (Figure 2b). With no pretreatment, there was a linear polynomial fit only at 5 °C, with a 43% increase in seedling radicle length at the highest rate of sodium metasilicate (16 gL$^{-1}$) compared to plants not supplemented with Si (0.0 gL$^{-1}$) (Figure 2b).

For pretreated Guri INTA CL cultivar seeds, rates of sodium metasilicate resulted in a linear increase at 10 °C, with a 103% increase in radicle length at the highest rate (16 mg.L$^{-1}$) compared to plants not supplemented with Si (0 mg.L$^{-1}$). At a temperature of 20 °C, there was a quadratic increase peaking at a rate of 10.3 mg.L$^{-1}$ and at a temperature of 5 °C there were no fits to the polynomial models (Figure 2a). With no seed pretreatment, the Guri INTA CL cultivar at 10 °C showed a linear fit with a 24% increase in seedling radicle length at the highest rate of sodium metasilicate (16 mg.L$^{-1}$) compared to plants not supplemented with Si (0 mg.L$^{-1}$). At 5 and 20 °C there were no fits to the polynomial models (Figure 2a).

Seedling length is an important physiological parameter. The longer the roots, the greater the surface area for finding and absorbing nutrients and water (Taiz and Zeiger, 2012). The findings of our study corroborate those of Lima Filho et al. (2005), who grew wheat plants under hydroponic conditions in solution with 100 mg.L$^{-1}$ of Si, and the addition of Si was found to increase root biomass.

In terms of temperature (Table 2), as a general rule the higher the temperature (20 °C), the longer the seedling shoot, with and without pretreatment with dietholate. However, at temperatures of 5 and 10 °C (severe stress), dietholate pretreated seedling shoot length was lower than that exhibited by non-pretreated seeds, showing that at lower temperatures it increases the stressor effect and influences germination and initial growth, corroborating reports from rice farmers. Thus the stress caused by extreme temperatures can be minimized by supplementing with silicon. This may be because Si stimulates osmoprotective compounds that adjust the osmotic pressure of the plant, helping it withstand the cold by maintaining the integrity of the cell membrane, preventing cell dehydration caused by osmotic pressure (Huang et al., 2012). Our findings corroborate those of Habibi (2016) and He et al. (2010) who reported that, at low temperature, the Si applied to maize plants and grass made them more resistant to stress than plants not treated with Si.

Table 2. Comparison of means for the effect of temperature on shoot and radicle length in rice seedlings with and without dietholate pretreatment for different cultivars and silicon rates and sources.

| | | | | Ratio of shoot lengths | | | Ratio of radicle lengths | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | | Temperatures | | | | | |
| Rates of Si | Seed pretreatments | Sources of Si | Cultivars | 5 °C | 10 °C | 20 °C | 5 °C | 10 °C | 20 °C |
| 0 (mg.L$^{-1}$) | with dietholate | potassium metasilicate | Guri INTA CL | 0.27 b | 0.30 ab | 1.05 a | 0.42 b | 0.39 b | 2.40 a |
| | | | IRGA 424 RI | 0.40 b | 0.75 b | 1.61 a | 1.07 b | 1.64 b | 3.29 a |
| | | sodium metasilicate | Guri INTA CL | 0.39 b | 0.74 b | 2.10 a | 0.98 b | 1.67 b | 5.22 a |
| | | | IRGA 424 RI | 3.09 a | 1.06 b | 1.72 b | 2.15 b | 1.96 b | 4.33 a |
| | without dietholate | potassium metasilicate | Guri INTA CL | 0.97 c | 1.87 b | 2.91 a | 0.95 b | 3.69 a | 3.58 a |
| | | | IRGA 424 RI | 1.83 b | 2.04 b | 3.40 a | 3.40 a | 3.80 a | 4.45 a |
| | | sodium metasilicate | Guri INTA CL | 2.31 a | 2.16 a | 2.47 a | 3.15 a | 4.03 a | 3.78 a |
| | | | IRGA 424 RI | 2.70 ab | 2.26 b | 3.10 a | 4.34 a | 4.58 a | 4.10 a |
| 4,0 (mg.L$^{-1}$) | with dietholate | potassium metasilicate | Guri INTA CL | 0.89 b | 0.89 b | 2.08 a | 0.87 b | 1.40 b | 3.35 a |
| | | | IRGA 424 RI | 1.02 b | 0.91 b | 2.28 a | 1.37 b | 1.42 b | 3.69 a |
| | | sodium metasilicate | Guri INTA CL | 0.64 b | 1.37 ab | 1.74 a | 1.20 b | 2.05 ab | 3.83 a |
| | | | IRGA 424 RI | 1.64 b | 1.23 b | 2.67 a | 0 c | 3.51 b | 5.59 a |
| | without dietholate | potassium metasilicate | Guri INTA CL | 1.99 c | 3.09 b | 3.98 a | 2.42 b | 4.99 a | 5.21 a |
| | | | IRGA 424 RI | 1.75 b | 3.48 a | 4.15 a | 2.76 b | 4.90 a | 5.19 a |
| | | sodium metasilicate | Guri INTA CL | 2.14 a | 2.71 a | 2.90 a | 3.07 a | 4.15 a | 4.47 a |
| | | | IRGA 424 RI | 3.25 a | 3.16 a | 2.83 a | 4.66 a | 4.67 a | 4.05 a |
| 8,0 (mg.L$^{-1}$) | with dietholate | potassium metasilicate | Guri INTA CL | 0.53 a | 0.76 a | 1.23 a | 0.76 a | 0.93 a | 2.05 a |
| | | | IRGA 424 RI | 0.65 b | 0.95 ab | 1.55 a | 0.61 a | 1.58 a | 1.82 a |
| | | sodium metasilicate | Guri INTA CL | 0.97 b | 2.22 a | 2.29 a | 1.80 b | 3.02 ab | 3.93 a |
| | | | IRGA 424 RI | 1.83 b | 2.68 a | 2.16 ab | 0 c | 3.64 b | 6.11 a |
| | without dietholate | potassium metasilicate | Guri INTA CL | 2.48 b | 2.81 b | 3.80 a | 2.41 a | 3.17 a | 3.70 a |
| | | | IRGA 424 RI | 2.87 a | 3.29 a | 3.29 a | 2.42 a | 3.85 a | 3.32 a |
| | | sodium metasilicate | Guri INTA CL | 3.10 a | 2.65 a | 2.49 a | 4.09 a | 3.71 a | 3.67 a |
| | | | IRGA 424 RI | 3.26 a | 3.28 a | 3.06 a | 5.04 a | 5.54 a | 4.71 a |
| 16 (mg.L$^{-1}$) | with dietholate | potassium metasilicate | Guri INTA CL | 0.19 a | 0.46 a | 0 a | 0.11 a | 0.29 a | 0 a |
| | | | IRGA 424 RI | 0.59 a | 0.66 a | 0 a | 0.22 a | 0.18 a | 0 a |
| | | sodium metasilicate | Guri INTA CL | 0.80 b | 1.65 a | 2.42 a | 1.34 b | 3.39 a | 4.54 a |
| | | | IRGA 424 RI | 0 c | 1.32 b | 2.74 a | 0 b | 1.17 b | 5.51 a |
| | without dietholate | potassium metasilicate | Guri INTA CL | 1.26 b | 1.36 a | 3.09 a | 0.23 a | 0.81 a | 1.65 a |
| | | | IRGA 424 RI | 1.62 b | 2.27 ab | 2.76 a | 0.58 a | 1.29 a | 1.36 a |
| | | sodium metasilicate | Guri INTA CL | 2.41 a | 2.73 a | 2.79 a | 4.54 a | 5.02 a | 4.70 a |
| | | | IRGA 424 RI | 4.13 a | 2.70 b | 2.19 b | 6.20 a | 4.57 b | 3.89 b |
| | | | | CV (%) = 20.54 | | | CV (%) = 26.51 | | |

* Means for shoot and radicle length for temperatures not followed by the same letter in the row differed statistically in the Tukey test ($p < 0.05$).

In terms of temperature as a factor influencing mean radicle length, it was generally the case that in most comparisons made, the seedlings produced by seeds not pretreated with dietholate showed no statistical difference at the three temperatures (5, 10 and 20 °C), not even when treated with Si. However, the radicle length for dietholate pretreated seeds was observed to drop as the cold stress increased, but as the rate of silicon increased, this made no difference to radicle length for the three temperatures. It has been suggested that the phytotoxic effects of dietholate triggered by cold stress, causing a drop in radicle length, are not exhibited as Si rates increase, indicating that Si is beneficial as a mitigator of the stress induced by the combination of dietholate treatment and low temperatures, and affecting post-germination radical growth (Table 2). This means that the use of silicon could be recommended as a strategy to reduce the problems caused by pretreating rice seeds with dietholate.

Comparing the cultivars (Table 3), it was found that for shoot length, of the sixteen comparisons made between the Guri INTA CL and IRGA 424 RI cultivars at each of the three temperatures (5, 10 and 20 °C), in a respective 37, 6 and 6% of cases the seedlings of IRGA 424 RI exhibited longer shoots than those of Guri INTA CL. This may be due to the fact that IRGA 424 RI adapts better to colder conditions (Sosbai, 2018). In terms of radicle length, there were no significant differences between the cultivars tested (Table 3), indicating very similar Si action in both cultivars.

Table 3. Comparison of cultivar means for shoot and radicle length of rice seedlings with and without dietholate seed pretreatment for different temperatures and Si rates and sources.

| | | | | Ratio of shoot lengths | | | | Ratio of radicle lengths | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Rates of Si (mg.L$^{-1}$) | | | | | | | |
| Temperatures | Tratamento de semente | Sources of Si | Cultivars | 0 | 4 | 8 | 16 | 0 | 4 | 8 | 16 |
| 5 ° C | with dietholate | potassium metasilicate | Guri INTA CL | 0.27 a | 0.87 a | 0.54 a | 0.19 a | 0.42 a | 0.87 a | 0.76 a | 0.10 a |
| | | | IRGA 424 RI | 0.40 a | 1.03 a | 0.65 a | 0.59 a | 1.07 a | 1.37 a | 0.61 a | 0.22 a |
| | | sodium metasilicate | Guri INTA CL | 0.39 b | 0.65 b | 0.97 b | 0.8 a | 0.98 a | 1.20 a | 1.8 a | 1.34 a |
| | | | IRGA 424 RI | 3.10 a | 1.64 a | 1.83 a | 0 b | 2.15 a | 0 a | 0 b | 0 b |
| | without dietholate | potassium metasilicate | Guri INTA CL | 0.97 b | 1.99 a | 2.48 a | 1.26 a | 0.95 b | 2.42 a | 2.41 a | 0.23 a |
| | | | IRGA 424 RI | 1.83 a | 1.75 a | 2.87 a | 1.62 a | 3.4 a | 2.76 a | 2.42 a | 0.58 a |
| | | sodium metasilicate | Guri INTA CL | 2.32 a | 2.14 b | 3.10 a | 2.41 b | 3.15 a | 3.07 b | 4.09 a | 4.54 b |
| | | | IRGA 424 RI | 2.70 a | 3.25 a | 3.26 a | 4.13 a | 4.34 a | 4.66 a | 5.04 a | 6.20 a |
| 10 ° C | with dietholate | potassium metasilicate | Guri INTA CL | 0.31 a | 0.89 a | 0.76 a | 0.46 a | 0.39 b | 1.40 a | 0.93 a | 0.29 a |
| | | | IRGA 424 RI | 0.75 a | 0.91 a | 0.95 a | 0.66 a | 1.64 a | 1.42 a | 1.58 a | 0.18 a |
| | | sodium metasilicate | Guri INTA CL | 0.74 a | 1.37 a | 2.29 a | 1.32 a | 1.67 a | 2.51 a | 3.02 a | 3.39 a |
| | | | IRGA 424 RI | 1.06 a | 1.23 a | 2.68 a | 1.65 a | 1.96 a | 3.51 a | 3.63 a | 1.17 b |
| | without dietholate | potassium metasilicate | Guri INTA CL | 1.87 a | 3.09 a | 2.81 a | 1.36 b | 3.69 a | 4.99 a | 3.17 a | 0.81 a |
| | | | IRGA 424 RI | 2.04 a | 3.48 a | 3.29 a | 2.27 a | 3.80 a | 5.19 a | 3.85 a | 1.29 a |
| | | sodium metasilicate | Guri INTA CL | 2.16 a | 2.71 a | 2.65 a | 2.72 a | 4.03 a | 4.15 a | 3.71 b | 4.57 a |
| | | | IRGA 424 RI | 2.26 a | 3.16 a | 3.28 a | 2.67 a | 4.58 a | 4.67 a | 5.54 a | 5.02 a |
| 20 ° C | with dietholate | potassium metasilicate | Guri INTA CL | 1.05 a | 2.08 a | 1.24 a | 0 a | 2.40 a | 3.35 a | 2.06 a | 0 a |
| | | | IRGA 424 RI | 1.61 a | 2.28 a | 1.56 a | 0 a | 3.29 a | 3.69 a | 1.82 a | 0 a |
| | | sodium metasilicate | Guri INTA CL | 2.10 a | 1.74 b | 2.22 a | 2.42 a | 5.22 a | 3.83 b | 3.93 b | 4.54 a |
| | | | IRGA 424 RI | 1.72 a | 2.67 a | 2.16 a | 2.75 a | 4.33 a | 5.59 a | 6.11 a | 5.51 a |
| | without dietholate | potassium metasilicate | Guri INTA CL | 2.91 a | 3.98 a | 3.81 a | 3.09 a | 3.58 a | 5.20 a | 3.70 a | 1.36 a |
| | | | IRGA 424 RI | 3.4 a | 4.15 a | 3.29 a | 2.76 a | 4.46 a | 4.90 a | 3.32 a | 1.65 a |
| | | sodium metasilicate | Guri INTA CL | 2.47 a | 2.83 a | 2.49 a | 2.79 a | 3.78 a | 4.44 a | 3.77 a | 4.70 a |
| | | | IRGA 424 RI | 3.10 a | 2.83 a | 3.06 a | 2.18 a | 4.10 a | 4.05 a | 4.71 a | 3.89 a |
| | | | | CV (%) = 20.54 | | | | CV (%) = 26.51 | | | |

* Means for shoot and radicle length of cultivars not followed by the same letter in the column differed statistically in the Tukey test ($p < 0.05$).

Analyzing the sources of Si (Table 4) for length of shoot and radicle, it was generally the case that, of the sixteen comparisons between sodium and potassium metasilicate sources, for each of the variables and at each of the three temperatures (5, 10 and 20 °C), sodium metasilicate exhibited the highest average in a respective 42, 42 and 25% of cases for shoot length and in 42, 50 and 67 % of cases for radicle length, in seedlings with and without dietholate seed pretreatment. This difference in shoot and radicle length between the sources of Si may be due to differences in the composition of each source.

Table 4. Comparison of means for the effect of Si sources on shoot and radicle length of rice seedlings with and without dietholate seed pretreatment for different temperatures, cultivars and Si rates.

| | | | | Ratio of shoot lengths | | | | Ratio of radicle lengths | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Rates of Si (mg.L$^{-1}$) | | | | | | | |
| Temperatures | Seed pretreatments | Cultivars | Sources of Si | 0 | 4 | 8 | 16 | 0 | 4 | 8 | 16 |
| 5 °C | with dietholate | Guri INTA CL | sodium metasilicate | 0.39 | 0.65 a | 0.97 a | 0.80 a | 0.98 | 1.20 a | 1.80 a | 1.34 a |
| | | | potassium metasilicate | 0.27 | 0.89 a | 0.54 b | 0.19 a | 0.42 | 0.87 a | 0.76 a | 0.11 b |
| | | Irga 424RI | sodium metasilicate | 3.09 | 1.64 a | 1.83 a | 0.00 b | 2.15 | 0 b | 0 a | 0 a |
| | | | potassium metasilicate | 0.4 | 1.03 a | 0.65 b | 0.59 a | 1.07 | 1.37 a | 0.61 a | 0.22 a |
| | without dietholate | Guri INTA CL | sodium metasilicate | 2.32 | 2.14 a | 3.10 a | 2.41 a | 3.15 | 3.07 a | 4.09 b | 4.54 a |
| | | | potassium metasilicate | 0.97 | 1.99 a | 2.48 a | 1.26 b | 0.95 | 2.42 a | 2.41 a | 0.23 b |
| | | Irga 424RI | sodium metasilicate | 2.7 | 3.25 a | 3.27 a | 4.13 a | 4.34 | 4.66 a | 5.04 a | 6.20 a |
| | | | potassium metasilicate | 1.83 | 1.75 b | 2.87 a | 1.62 b | 3.4 | 2.75 b | 2.41 b | 0.58 b |
| 10 °C | with dietholate | Guri INTA CL | sodium metasilicate | 0.74 | 1.37 a | 2.27 a | 1.65 a | 1.67 | 1.40 a | 3.02 a | 3.39 a |
| | | | potassium metasilicate | 0.31 | 0.89 a | 0.76 b | 0.46 b | 0.39 | 2.50 a | 0.93 b | 0.29 b |
| | | Irga 424RI | sodium metasilicate | 1.06 | 1.23 a | 2.68 a | 1.32 b | 1.96 | 3.51 a | 3.63 a | 1.17 a |
| | | | potassium metasilicate | 0.75 | 0.91 a | 0.95 b | 0.66 a | 1.64 | 1.42 b | 1.58 b | 0.18 a |
| | without dietholate | Guri INTA CL | sodium metasilicate | 2.16 | 2.71 a | 2.65 a | 2.72 a | 4.30 | 4.15 a | 3.71 a | 5.02 a |
| | | | potassium metasilicate | 1.87 | 3.09 a | 2.81 a | 1.36 b | 3.69 | 4.99 a | 3.17 a | 0.81 b |
| | | Irga 424RI | sodium metasilicate | 2.26 | 3.16 a | 3.28 a | 2.67 a | 4.58 | 4.67 a | 5.54 a | 4.57 a |
| | | | potassium metasilicate | 2.04 | 3.48 a | 3.29 a | 2.26 a | 3.80 | 5.19 a | 3.85 a | 1.29 b |
| 20 °C | with dietholate | Guri INTA CL | sodium metasilicate | 2.1 | 1.74 a | 2.22 a | 2.42 a | 5.22 | 3.83 a | 3.93 a | 4.54 a |
| | | | potassium metasilicate | 1.05 | 2.08 a | 1.24 b | 0 b | 2.40 | 3.35 a | 2.06 b | 0 b |
| | | Irga 424RI | sodium metasilicate | 1.72 | 2.28 a | 2.17 a | 2.75 a | 4.33 | 5.58 a | 6.10 a | 5.51 a |
| | | | potassium metasilicate | 1.61 | 2.28 a | 1.57 a | 0 b | 3.29 | 3.69 b | 1.82 b | 0 b |
| | without dietholate | Guri INTA CL | sodium metasilicate | 2.47 | 2.91 b | 2.49 b | 2.79 a | 3.78 | 4.43 a | 3.67 a | 4.70 a |
| | | | potassium metasilicate | 2.91 | 3.98 a | 3.81 a | 3.09 a | 3.58 | 5.21 a | 3.70 a | 1.65 b |
| | | Irga 424RI | sodium metasilicate | 3.1 | 2.83 b | 3.06 a | 2.19 a | 4.1 | 4.05 a | 4.71 a | 3.89 a |
| | | | potassium metasilicate | 3.39 | 4.15 a | 3.29 a | 2.76 a | 4.46 | 4.90 a | 3.32 b | 1.36 b |
| | | | | CV (%) = 20.54 | | | | CV (%) = 26.51 | | | |

* Means for shoot and radicle length for Si sources not followed by the same letter in the column differed statistically in the Tukey test ($p < 0.05$).

## 5. Conclusion

Seed pretreatment with dietholate caused a decrease in shoot and radicle length, mainly at the lower temperatures of 5 and 10 °C. Sodium metasilicate was the most efficient source in terms of increasing seedling shoot and radicle length, with and without pretreatment with dietholate and regardless of temperature. Therefore, silicon can be recommended as an effective strategy for attenuating problems caused by the combination of dietholate pretreated seeds and low temperatures, in respect of shoot and radicle length.

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