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Influence of silicon on sheath blight of rice in Brazil

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Abstract

The effect of silicon (Si) on sheath blight (*Rhizoctonia solani* Kühn) of rice was studied under greenhouse conditions. The predominant commercial rice cultivars 'BR-IRGA 409', 'Metica-1', 'EPAGRI-109', 'Rio Formoso', 'Javaé', and 'CICA-8', were grown in pots containing low-Si soil amended with 0, 0.48, 0.96, 1.44 or 1.92 g Si pot^{-1}. Plants were inoculated at the maximum tillering stage. For all cultivars, Si concentration in straw increased more than 60% as the rates of Si increased from 0 to 1.92 g pot^{-1}. Incubation period of *R. solani* was slightly prolonged with increasing Si rates and ranged from 53 to 64 h depending upon the cultivar. Total number of sheath blight lesions, total area under the relative lesion extension progress curve, severity of sheath blight, and the highest relative lesion height on the main tiller decreased by 37%, 40%, 52% and 24%, respectively, as the rate of Si increased from 0 to 1.92 g pot^{-1}. Silicon may offer a viable method to control sheath blight in areas where soil is deficient in Si and cultivars with sheath blight resistance are not commercially available.

Keywords: Calcium silicate; *Rhizoctonia solani*; Nutrition; *Oryza sativa*; Host resistance

1. Introduction

Brazil is a major rice (*Oryza sativa* L.) producing country with five million hectares grown annually under upland and irrigated conditions. Sheath blight (*Rhizoctonia solani* Kühn, *Thanatephorus cucumeris* (Frank) Donk, anastomosis group 1 IA (AG-1 IA)) (Ogoshi, 1987) and other diseases have caused great yield losses especially in the states of Tocantins and Mato Grosso do Sul in Brazil. The rotation of rice with soybean in these regions as well as the use of high rates of nitrogen fertilizer and planting of susceptible cultivars have been associated with an increased incidence of sheath blight. In most rice-growing regions worldwide, early season planting, double cropping, high plant density per unit area, and planting of early maturing, short-stature, high tillering, compact, susceptible cultivars are also factors responsible for severe sheath blight (Lee and Rush, 1983; Ou, 1985; Dath, 1990; Rush and Lee, 1992; Roy, 1996).

Strategies for sheath blight control are limited since cultivars with a significant level of resistance are not known (Li et al., 1995). Consequently, cultural and biological control methods are used to minimize the intensity of sheath blight (Mew and Rosales, 1986; Belmar et al., 1987; Jones and Belmar, 1988; Damicone et al., 1993; Roy, 1996; Willocquet et al., 2000). Sheath blight control, however, has mainly focused on fungicides and these pesticides are of environmental concern (Groth et al., 1990; Rush and Lee, 1992). Silicon fertilization may offer a viable method for sheath blight control, especially where soils are low or limiting in plant available Si. Some economically important diseases in rice such as blast (*Magnaporthe grisea* (Hebert) Barr), brown spot (*Cochliobolus miyabeanus* (Ito & Kuribayashi in Ito) Drechs ex Dastur), stem rot (*Magnaporthe salvinii* Catt.), scald (*Monographella albescens* Theum), and grain discoloration have been reduced by silicon fertilization (Elawad and Green,

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1979; Datnoff et al., 1991, 1992, 1997; Deren et al., 1994; Seebold et al., 2000, 2001; Korndörfer et al., 1999). According to Mathai et al. (1977), application of Si reduced sheath blight intensity, even though the difference between the high and low levels of Si was not statistically significant. Winslow (1992) reported that Si only reduced the severity of sheath blight in irrigated *indica* rice but not *japonica* upland rice and intermediate genotypes. Rodrigues et al. (2001) demonstrated that Si reduced sheath blight development of susceptible and moderately susceptible US rice cultivars to levels comparable to those observed in cultivars high in partial resistance to sheath blight but, not fertilized with Si.

Since little, if any, research is available on the influence of Si on rice diseases in Brazil, the purpose of this study was to determine the effect of Si fertilization on sheath blight in six Brazilian rice cultivars.

2. Material and methods

Experiments were conducted under greenhouse conditions in 1998 and 1999 at the Federal University of Viçosa, Viçosa, Minas Gerais, Brazil. The soil type used in the experiments was a Si-deficient typic acrustox red-yellow latosol, with 14.6 mg dm^{-3} plant-available Si from the 'Triângulo Mineiro' area. Each pot was filled with 2 kg of air dried, sieved (5 mm) soil. Wollastonite, used as the Si source ($CaSiO_3$; Vansil, EW-20, Ipiranga Chemical Co., São Paulo, Brazil) is composed of 24.3% Si, 42.1% Ca, and other minor elements such as Mn (0.2%), Mg (0.3%), and K (0.1%). The wollastonite was incorporated into each pot to obtain equivalent rates of 0, 0.48, 0.96, 1.44 or 1.92 g pot^{-1} of elemental Si. Dolomitic lime (38.9% CaO and 12.7% MgO) was added to pots treated with 0, 0.48, 0.96 or 1.44 g Si to equilibrate the amount of Ca present in these treatments with the treatment containing 1.92 g Si. Pots containing 1.44 or 1.92 g of Si received 1.44 or 2.88 g of $MgCl_2 \cdot 6H_2O$, respectively, to equilibrate magnesium among treatments. Soil in each pot was incubated 60 days (moist) before sowing 8 rice seeds and fertilized with 100 ml of a nutrient solution containing, in mg kg^{-1} soil, 100 N, 300 P, 150 K, 40 S, 0.81 B, 1.33 Cu, 1.55 Fe, 3.66 Mn, 0.15 Mo, and 4.00 Zn (Novais et al., 1991). Ten days after emergence, each pot was thinned to two plants. Plants were kept under flooded conditions until the end of the experiments. The most commonly cultivated Brazilian rice cultivars chosen for evaluation in this study were 'BR-IRGA 409', 'Metica-1', 'EPAGRI-109', 'Rio Formoso', 'Javaé', and 'CICA-8'.

A virulent isolate of *R. solani* (AG-1 IA) obtained from rice (cultivar 'Metica-1') in the state of Tocantins by the National Center for Research on Rice and Beans in Brazil was used to inoculate plants. The isolate of *R. solani* was grown on acid potato–dextrose–agar medium (APDA) for 10 days at room temperature to produce mycelia. Fifteen sterile wooden toothpicks, 1-cm in length, infused with potato broth and transferred to APDA plates along with five (5 mm diameter) agar plugs from the margin of an actively growing colony of *R. solani* were allowed to incubate for 8 days at room temperature so that *R. solani* could colonize the toothpicks.

Plants were inoculated at maximum tillering stage (65 days after emergence) by placing a *R. solani* colonized toothpick into the bottom lowest inner sheath of the main tiller. Immediately after inoculation, all plants were transferred to a mist chamber where they remained throughout the experiments. Plants were kept at 27°C and 75–90% relative humidity. The 12-h relative humidity per day inside the chamber was controlled using a humidifier (Herrmidifier 500 Co., Inc., Lancaster, PA). Plants received 12-h photoperiod of approximately 162 μE m^{-2} s^{-1} provided by cool-white fluorescent lamps.

Disease progress on the inoculated and upper three sheaths for the main tiller were evaluated as a ratio of vertical lesion extension (cm) to sheath length (cm). The relative lesion extension progress curve was constructed from sheaths, replication, treatment, and experiment data. Area under the relative lesion extension progress curve for each sheath in each plant, replication, treatment, and experiment was computed using the trapezoidal integration of relative lesion extension progress curve over time according to the formula proposed by Shaner and Finney (1977). The total area under the relative lesion extension progress curve represents the sum of the area under the relative lesion extension progress curve from all sheaths.

The severity of sheath blight was scored with a scale of 0–9 based on relative lesion height on the whole plant (IRRI, 1996) as follows: 0 = no infection; 1 = lesions limited to the lower 20% of plant height; 3 = lesions limited to the lower 20–30% of plant height; 5 = lesions limited to the lower 31–45% of plant height; 7 = lesions limited to the lower 46–65% of plant height; and 9 = lesions on the upper 35% of plant height.

The highest relative lesion height was determined by dividing the linear distance from the surface of the soil to the upper edge of the highest lesion on the culm of the main tiller by the height of the plant. Incubation period was scored as the time in hours from inoculation until the appearance of the first lesion on inoculated sheaths. Total number of sheath blight lesions was also counted on the four sheaths.

Plant material for all cultivars was harvested 25 or 29 days after inoculation, depending on the experiment. Leaves and culms from each replication were dried and ground to pass through a 40-mesh screen with a

Thomas-Wiley mill (Thomas Scientific, Swedesboro, NJ). Silicon in leaf tissue was determined by automated colorimetric analysis on 0.1 g of digested dried tissue (Elliot and Snyder, 1991). Results obtained were expressed in grams (g) of Si per kg of plant tissue. Calcium concentration ($g\,kg^{-1}$) in tissue was determined after percloric digestion by atomic absorption spectrophotometry (Johnson and Ulrich, 1959).

Two 5×6 factorial experiments, consisting of five Si rates and six rice cultivars, were arranged in a randomized design with four replications. The experiment was repeated twice and data for each variable were pooled because homogeneity of variance was confirmed by Bartlett's test (Gomez and Gomez, 1994). Data were analyzed using standard analysis of variance (ANOVA), and polynomial regression procedures were appropriate with SAS (SAS Institute, Inc., 1989, Cary, NC). Treatment mean comparisons were made using Fisher's protected least significant difference (FLSD, $P \leqslant 0.05$) test.

3. Results

There was no significant interaction between cultivars and rates of Si ($P \geqslant 0.05$) for any variable (data not shown); consequently, the main effects are presented by cultivar averaged across rates of Si (Fig. 1), and by rates of Si averaged across cultivars (Fig. 2).

The average Si concentration in straw ranged from 22.5 to 25.8 $g\,kg^{-1}$ (Fig. 1A). Javaé had significantly higher Si than BR-IRGA 409 and CICA-8. No significant differences in Si were found among EPAGRI-109, Metica-1, and Javaé. The concentration of Si in straw was directly related to the amount of Si applied to the soil (Fig. 2A). Silicon concentration in the straw of all cultivars increased 11.27 $g\,kg^{-1}$ per unit increase in Si applied. There was no significant change ($P \geqslant 0.05$) in Ca in the straw for any of the cultivars tested (Table 1).

Incubation period of *R. solani* ranged from 53.4 to 63.8 h depending upon the cultivar (Fig. 1B). No

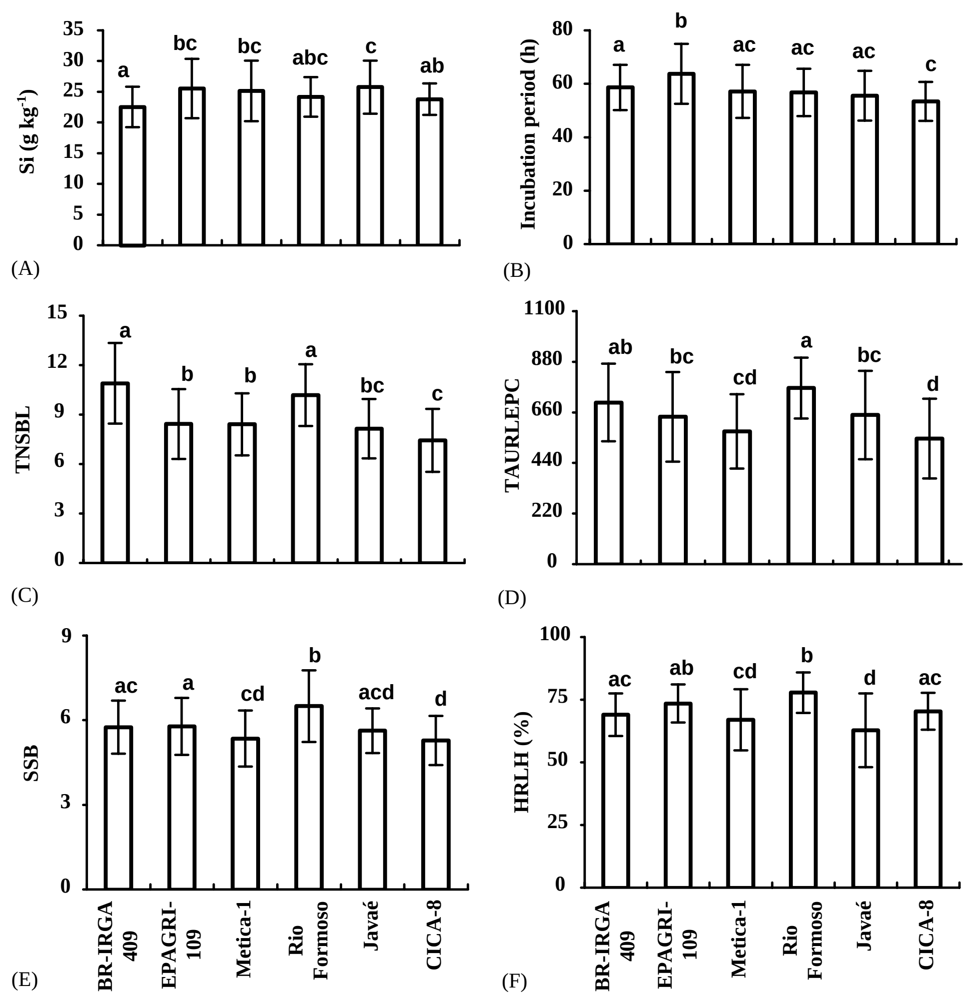

Fig. 1. Mean Si concentration in straw (A), incubation period (B), total number of sheath blight lesions (TNSBL) (C), total area under the relative lesion extension progress curve (TAURLEPC) (D), severity of sheath blight (E), and the highest relative lesion height (HRLH) on the main tiller (F) for each cultivar averaged across Si rates. Bars with the same letter do not differ significantly at $P = 0.05$ as determined by Fisher's protected LSD. Error bars represent standard deviations of means from two pooled experiments.

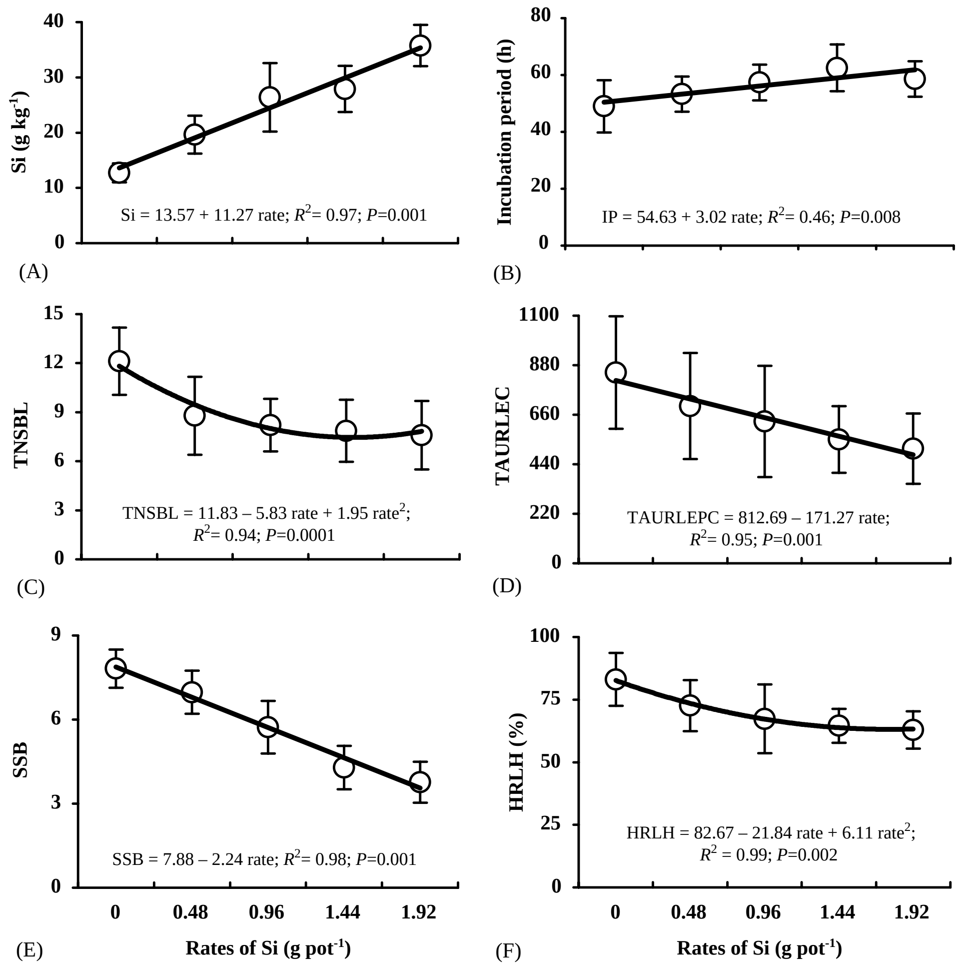

Fig. 2. Relationship between mean Si concentration in straw (A), incubation period (B), total number of sheath blight lesions (TNSBL) (C), total area under the relative lesion extension progress curve (TAURLEPC) (D), severity of sheath blight (E), and the highest relative lesion height (HRLH) on the main tiller (F) and Si rates for all cultivars. Bars represent standard deviations of means from two pooled experiments.

Table 1
Calcium concentration in the straw of six Brazilian rice cultivars after equilibrating calcium in the soil with rates of wollastonite and/or dolomitic lime

Cultivars	Calcium concentration ($g\,kg^{-1}$)[a]
BR-IRGA 409	2.77
EPAGRI-109	2.98
Metica-1	2.97
Rio Formoso	2.92
Javaé	2.98
CICA-8	2.89
FLSD ($P\leqslant 0.05$)	0.26

[a] Dolomitic lime was added to pots treated with 0, 0.48, 0.96 and 1.44 g Si so that Ca would be equivalent to the treatment containing 1.92 g Si.

significant differences in incubation period were found among BR-IRGA 409, Metica-1, Rio Formoso, and Javaé. The incubation period was significantly longer on EPAGRI-109 than the other cultivars. Cultivars BR-IRGA 409 and EPAGRI-109 had longer incubation period than Cica-8. Regardless of cultivar, incubation period increased linearly as the rate of Si increased ($P<0.05$) (Fig. 2B).

The total number of sheath blight lesions was lower on CICA-8 than on BR-IRGA 409, EPAGRI-109, Rio Formoso, and Metica-1 (Fig. 1C). More than twice as many lesions developed on BR-IRGA 409 and Rio Formoso than on CICA-8. CICA-8, Javaé, Metica-1, and EPAGRI-109 had 30%, 23%, 21%, and 20% fewer lesions than BR-IRGA 409 and Rio Formoso. A second order regression curve best described the effect of Si rate on total number of sheath blight lesions (Fig. 2C). Total number of sheath blight lesions at the highest Si rate were 37% less than the control.

The total area under the relative lesion extension progress curve did not differ significantly among BR-IRGA 409, EPAGRI-109, and Javaé (Fig. 1D). This variable was 21% and 26% lower on Metica-1 and CICA-8, respectively, than on BR-IRGA 409 and Rio Formoso. The relationship between the total area under the relative lesion extension progress curve and Si rates

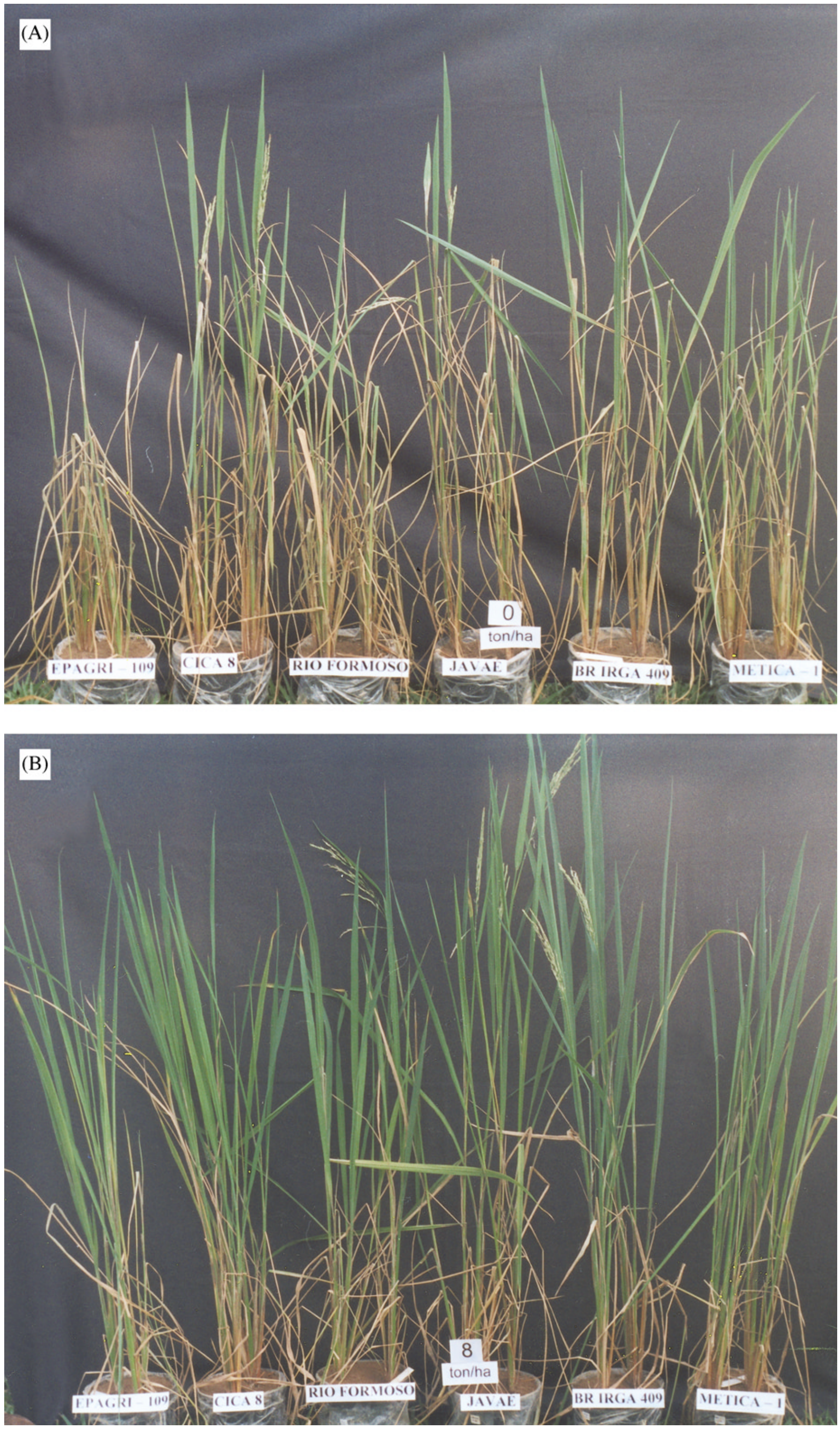

Fig. 3. Symptoms of sheath blight on six Brazilian cultivars of rice. (A) Nontreated control plants (0 g Si pot^{-1} or 0 ton ha^{-1}), and (B) plants grown at the highest rate of Si (1.92 g Si pot^{-1} or 8 ton ha^{-1}).

was linear (Fig. 2D). When the rate of Si was increased from 0 to 1.92 g pot^{-1}, the total area under the relative lesion extension progress curve decreased 40%, across all cultivars.

Average severity of sheath blight was significantly higher on BR-IRGA 409, EPAGRI-109, and Rio Formoso than on CICA-8 (Fig. 1E). Severity of sheath blight was 14%, 13%, and 8% lower on CICA-8, Metica-1, and Javaé, respectively, than on EPAGRI-109 and Rio Formoso. Significant differences in the severity of sheath blight were not found among BR-IRGA 409, EPAGRI-109, and Javaé. The linear regression model described the severity of sheath blight–Si rate relationship (Fig. 2E). The severity of sheath blight was reduced 52% at the highest rate of Si.

The highest relative lesion height on the main tiller was higher on BR-Irga 409, EPAGRI-109, Rio Formoso, and CICA-8 than on Javaé (Fig. 1F); but not among BR-IRGA 409, EPAGRI-109, and CICA-8. Highest relative lesion height on the main tiller was significantly reduced ($P<0.05$) by Si. A quadratic model ($P<0.05$) best described the relationship of rates of Si and highest relative lesion height on the main tiller (Fig. 2F). The highest relative lesion height on the main tiller decreased more than 20% as the rate of Si increased from 0 to 1.92 g pot^{-1}.

4. Discussion

Rice cultivars grown at highest Si rate had sheath blight intensities that were greatly reduced in comparison to cultivars grown in pots not amended with Si (Fig. 3). Although calcium is known to reduce the intensity of some diseases in different crops (Huber, 1980), the level of this element in the soil was carefully equilibrated in all treatments to avoid its potential interference with Si in the suppression of sheath blight. Since Ca did not change in the plant, it can be concluded that variations in Si accounted for differences in the level of disease response. Datnoff et al. (1991) found that tissue levels of Si, but not Ca, increased as the rate of calcium silicate increased in the soil. Silicon was the only element that increased significantly in rice tissue over a 3-year period in organic soil amended with calcium silicate slag (Snyder et al., 1986). Silicic acid may compete with Ca for binding sites on the cell wall because silicic acid can readily form complexes with polyhydric alcohols, organic acids, lignin and phenol-carbohydrate complexes in a manner similar to Ca (Inanaga et al., 1995). Silicon uptake also may depress the absorption of Ca by the rice plant, resulting in decreased Ca in shoot tissues (Ma and Takahashi, 1993).

The incubation period was only minimally affected by Si and increased only 5% at the highest Si rate over the control. Although the incubation period for *R. solani* on EPAGRI-109 was prolonged, there were no differences in incubation period among the other cultivars. It is important to note that *R. solani* can penetrate through stomatal openings as well as directly into epidermal cells (Ou, 1985). Placing the inoculum within the inner leaf sheath may have favored the early appearance of the first lesion on inoculated sheaths. Stomatal invasion seldom occurs on the outer surface of the sheath, but is quite common on the inner surface (Ou, 1985). These findings are similar to the report of Seebold et al. (2001) for *M. grisea* among susceptible and partially resistant rice cultivars amended with Si.

One mechanism of Si-reduced disease is reported to result from condensation of silicic acid into a hard, glass-like coating of polymerized SiO_2, known as plant opal, in the cuticle layer (Yoshida et al., 1962; Lanning, 1963). This physical barrier of SiO_2 in the leaf sheath should increase the incubation period and impede the penetration by *R. solani* to reduce both lesion numbers and lesion extension on sheaths. In contrast to formation of a physical barrier to initial penetration, Rodrigues et al. (2001) considered lesion extension as the most important component of resistance to sheath blight, especially in moderately susceptible and susceptible US rice cultivars. Volk et al. (1958) hypothesized that blast lesion size could be reduced by organosilicon compounds in the walls of epidermal cells, and Inanaga et al. (1995) later showed that Si forms complexes with organic compounds in these cells. Silicon-induced resistance would contribute to reduce sheath blight intensity as opposed to only a physical barrier of SiO_2 in the cuticle layer. In spite of extensive research, the exact mechanism(s) by which Si reduces sheath blight and other diseases such as blast is unknown. Other researchers have suggested that Si acts in crops such as cucumber and barley to stimulate host defense mechanisms to pathogenesis by increasing the level of inhibitory phenolic compounds and the activity of chitinases, β-1,3-glucanases, peroxidase, phenylalanine ammonia-lyase, and polyphenoloxidase (Carver et al., 1987; Menzies et al., 1991; Chérif et al., 1992; Fawe et al., 1998).

Results obtained in this study, even though the experiments were realized under greenhouse conditions, suggest that silicon has the potential to complement inherent host resistance and reduce sheath blight intensity. Among the Brazilian rice cultivars tested, CICA-8 and Metica-1 exhibited a relatively high level of resistance to sheath blight, especially in combination with Si. This information may prove to be invaluable in the field especially in areas where soils are known to be Si-deficient.

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