

Influence of silicon on some components of resistance to anthracnose in susceptible and resistant sorghum lines

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Abstract This study aimed to evaluate the effect of silicon (Si) rates on some components of sorghum resistance to anthracnose. Two 2×5 factorial experiments, consisting of two sorghum lines (BR005 and BR009, resistant and susceptible, respectively) and five Si application rates (0, 0.06, 0.12, 0.24 and 0.30 g Si kg⁻¹ of soil) were arranged in a completely randomised design with three replications. Plants from both lines were inoculated with a conidial suspension of *Colletotrichum sublineolum* (1×10⁶ conidia ml⁻¹) 30 days after emergence. The incubation period (IP), latent period (LP₆₀), area under relative infection efficiency progress curve (AURIEPC), area under anthracnose index progress curve (AUAIPC), final disease severity (FDS), percentage of pigmented leaf area (PLA), and percentage of necrotic leaf area (NLA) were evaluated. Silicon and calcium (Ca) content in leaf tissue of both lines was also determined. The content of Si in leaf tissue increased relative to the control by 55 and 58%,

respectively, for the susceptible and resistant lines. There was no significant change in Ca content in leaf tissue for either of the lines; therefore the variations in Si accounted for differences in the level of disease response. The IP for the resistant line was not affected by Si application rates. The LP₆₀ was not evaluated in the resistant line due to the absence of acervuli. For the resistant line, Si application rates had no significant effect on AUAIPC, FDS, percentage of PLA, and percentage of NLA. On the susceptible line, a quadratic regression model best described the effect of Si application rates on IP, LP₆₀, AURIEPC, AUAIPC, FDS, percentage of PLA, and percentage of NLA. The correlation between Si content in leaf tissue of the susceptible line and the AURIEPC, AUAIPC, FDS, PLA, and NLA was negatively significant ($r = -0.57, -0.37, -0.40, -0.67, \text{ and } -0.77$, respectively). There was no correlation between Si content and IP or LP₆₀. The correlation between the percentage of PLA with the percentage of NLA was negatively significant ($r = -0.74$). In conclusion, the results from this study underscore the importance of Si in sorghum resistance to anthracnose particularly for the susceptible line.

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Introduction

Anthrachnose, caused by *Colletotrichum sublineolum* (Sutton 1980; Sherrif et al. 1995), is the most destructive fungal disease of sorghum (*Sorghum bicolor*), particularly in warm and humid regions worldwide (Ali and Warren 1992). The disease dramatically reduces yield when susceptible sorghum cultivars are grown under favourable conditions for severe epidemics (Harris et al. 1964). The fungus is able to infect leaves, leaf sheaths, stalks, peduncles, panicles, and grains (Ali and Warren 1992). Particularly on leaves, symptoms of anthrachnose appear as circular-elliptical small tan to orange-red to black lesions especially in the midvein, but lesion size and colour is closely associated with sorghum genotype (Warren 2000). Expanded lesions are usually tan in the centre with a distinct reddish-purple to brown border (Warren 2000). Several acervuli containing many conidia with dark setae interspersed among them are formed in the centre of the tan lesions (Ali and Warren 1992).

Cultivars with race-specific resistance to anthrachnose are unstable under field conditions; therefore the use by growers of cultivars with a level of partial resistance is more plausible to slow the rate of disease progress. According to Parlevliet (1979), a reduced infection frequency, a lower epidemic rate, and fewer conidia produced by lesions are very important components associated with this form of resistance. Crop rotation with dicots, removal of crop residues, and fungicide application may also be used to decrease disease intensity (Ali and Warren 1987, Gwary and Asala 2006).

Other methods for disease control need to be urgently investigated. Some economically important diseases in rice, wheat, barley, rye, corn, cucumber, grape and strawberry are effectively controlled by supplying silicon (Si) to the plants (Datnoff et al. 2007). Many components of resistance to certain foliar pathogens of rice have been negatively impacted by Si application. For example, Seebold et al. (2001) found that although the latent period of *Pyricularia grisea* did not differ between some rice cultivars with different levels of partial resistance to blast, the incubation period lengthened with increasing calcium silicate application rates in the soil and there was a significant decrease in infection efficiency, lesion size, rate of lesion expansion, sporulation per lesion, and diseased leaf area. The incubation period of *Rhizoctonia solani* causing sheath

blight in rice was not affected by increasing Si application rates in the soil, but the total number of sheath blight lesions, total area under the relative lesion extension progress curve, disease severity, and the highest relative lesion height on the main tiller were reduced (Rodrigues et al. 2003). Silicon application may also be a viable method of enhancing sorghum resistance to anthrachnose considering that plants can contain about 1 to 2% of Si in the shoot, although this is less than rice, which is known as a Si accumulator plant with Si content up to 5% (Rodrigues et al. 2001a, b; Hattori et al. 2005).

Research in the literature showing the effect of Si on anthrachnose development on sorghum, especially closely examining some components of host resistance, is missing. Therefore, the current research aimed to investigate the effect of Si application rates on some components of sorghum resistance to anthrachnose in resistant and susceptible lines.

Materials and methods

Soil amendment with Si application

The soil type used in the experiments was a Si-deficient typical Acrustox red yellow latosol collected at the 'Triângulo Mineiro' savanna area with 530 g kg⁻¹ of clay; pH in KCl = 4.8; P (Mehlich-1) = 0.5 mg dm⁻³; K (Mehlich-1) = 13 mg dm⁻³; Al³⁺, Ca²⁺, Mg²⁺, H+Al³⁺ = 0.1, 0.0, 0.0 and 3.8 cmol_c dm⁻³, respectively; base saturation = 2%, and organic matter = 2.3 dag kg⁻¹. The concentration of available Si (extraction in CaCl₂) was 11.8 mg dm⁻³. Each plastic pot was filled with 1 kg of air-dried, sieved (5 mm) soil. Wollastonite, used as the Si source (CaSiO₃; Vansil, EW-10, Ipiranga Chemical Co., São Paulo, Brazil) is composed of 24.2% Si and 31% Ca. Wollastonite was incorporated into each pot at the rates of 0, 0.25, 0.5, 1.0, and 1.25 g kg⁻¹ of soil, which corresponded, respectively, to 0, 0.06, 0.12, 0.24, and 0.30 g of elemental Si per pot. Calcium carbonate (40% Ca, Sigma-Aldrich, St. Louis, MO) was added at the rates of 0.97, 0.77, 0.58, and 0.19 g to pots that received 0, 0.25, 0.5 and 1 g of wollastonite, respectively, to equilibrate the amount of Ca in these treatments with the amount present in pots that received 1.25 g of wollastonite. The amount of Ca among the treatments was fixed at 0.39 g per pot. Soil

in each pot was incubated for 60 days with relative humidity (RH) of around 65%.

Plant growth

Sorghum seeds from lines CMSXS116 [BR005 (SC326-6)-Texas] and CMSXS142 [BR009 (Tx623)-Texas], resistant and susceptible to *C. sublineolum*, respectively, were surface-sterilised in 10% (v v⁻¹) NaOCl for 1.5 min, rinsed in sterilised water for 3 min and sown at the rate of 5 seeds of each line per pot. Five days after emergence, each pot was thinned to two plants. Soil in each pot was fertilised before sowing 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). Magnesium was supplied as MgSO₄ (Sigma-Aldrich, St. Louis, MO) at the rate of 1 cmol_c dm⁻³ of soil. The Ca:Mg ratio in the soil was fixed at 2:1. The nutrient solution was applied at the amount of 50 ml per pot every 2 days after seedling emergence. Plants were watered as required.

Inoculation procedure

A pathogenic isolate of *C. sublineolum* (CNPMS-12), obtained from symptomatic sorghum plants and provided by EMBRAPA-National Centre for Sorghum and Maize Research, was used to inoculate the plants. This isolate was preserved on glass vials containing potato-dextrose-agar (PDA) covered with mineral oil at 4°C. Pieces of PDA containing fungal mycelia were transferred to Petri dishes containing oat media (Dhingra and Sinclair 1995). After 3 days, oat plugs containing fungal mycelia were transferred to new Petri dishes containing oat media. These Petri dishes were kept in a growth chamber at 25°C with a 12 h photoperiod for 10 days. After this period, mycelia were carefully removed from the Petri dishes with a rubber policeman. Petri dishes were returned to the growth chamber at 25°C under continuous light to induce sporulation. Plants were inoculated with a conidial suspension of *C. sublineolum* (1×10⁶ conidia ml⁻¹) at 30 days after emergence (growth stage 30) (Frederiksen 2000). A volume of 20 ml of suspension was applied as a fine mist to the adaxial leaf blades of each plant until run-off, using a VL Airbrush atomiser (Paasche Airbrush Co., Chicago, IL). Immediately after inoculation, plants were transferred to a mist chamber at 25±2°C,

RH of approximately 90±2%, and an initial 18 h dark period. After this period, plants were transferred to a growth chamber at 26°C with a 12 h photoperiod of approximately 225 μE m⁻² s⁻¹ provided by cool-white fluorescent lamps and RH of approximately 70±5%. Plants were kept inside this chamber for the duration of the experiments.

Quantification of some components of sorghum resistance to anthracnose

The following components of resistance were evaluated: incubation period (IP), latent period (LP₆₀), relative infection efficiency (RIE), anthracnose severity, final disease severity (FDS), percentage of pigmented leaf area (PLA), and percentage of necrotic leaf area (NLA). The fourth and fifth leaves of each plant were marked and used to evaluate the components of resistance mentioned above. The IP was scored for the appearance of lesions by examining the marked leaves every 24 h starting from 18 h after inoculation (HAI). Five lesions on each marked leaf were randomly selected and examined every 24 h with a hand-held microscope (x30) to determine the beginning of acervulus formation. LP₆₀ was achieved when acervuli were found on 60% of the examined lesions on each marked leaf. The RIE was evaluated based on the number of acervuli cm⁻² of leaf area at 8, 9, 11, and 19 days after inoculation (DAI). A total of five randomly chosen places on each leaf were examined and the number of acervuli counted. Area under RIE progress curve (AURIEPC) for each leaf in each plant was computed using the trapezoidal integration of RIE progress curve over time, following the formula proposed by Shaner and Finney (1977). Anthracnose severity on marked leaves of each plant was scored at 3, 5, 7, 9, and 11 DAI with a scale of 0–5 (Harris and Sowell 1970) with a few modifications as follows: 0 = no visible symptoms; 1 = pigmented flecks covering <5% of the leaf area; 1.5 = pigmented flecks covering from 5 to 10% of the leaf area; 2 = pigmented flecks covering from 10 to 35% of the leaf area and absence of necrotic lesions; 2.5 = many pigmented flecks on the leaf and <5% of lesions with acervuli; 3 = many pigmented flecks on the leaf and lesions with acervuli covering up to 10% of the leaf area; 3.5 = lesions with acervuli covering from 10 to 35% of the leaf area; 4 = lesions with acervuli covering up to 50% of the leaf area; 4.5 = lesions with numerous acervuli covering

up to 75% of the leaf area; and 5 = lesions with numerous acervuli covering >75% of the leaf area. Data from anthracnose severity were used to calculate the anthracnose index (AI) based on the formula proposed by McKinney (1923) where: $AI = (\text{rate on the disease scale} \times \text{number of plants with this rate}) / (\text{total number of plants} \times \text{the highest rate on the disease scale}) \times 100$. The AI values obtained were used to calculate the area under AI progress curve (AUAIPC) similarly as determined for AURIEPC. FDS was evaluated at 11 DAI using the scale mentioned above. The fourth and fifth leaves of each plant were harvested, scanned at 400 dpi resolution, and the images were processed using the software QUANT (Liberato 2003) to obtain the percentage of PLA (red) and NLA (brown).

Plant tissue analysis for Si and Ca content

After the experiment, all leaves of each plant, per replication and treatment were collected, washed in deionised water, dried for 72 h at 65°C and ground to pass through a 40 mesh screen with a Thomas-Wiley mill. Si in leaf tissue was determined by a colorimetric analysis on 0.1 g of dried and alkali digested tissue (Korndörfer et al. 2004). Dried leaf tissue was digested with a nitric-perchloric solution (3:1, v v⁻¹) and the content of Ca was determined by atomic absorption spectrophotometry.

Experimental design and data analysis

Two 2x5 factorial experiments, consisting of two sorghum lines (BR005 and BR009) and five Si application rates were arranged in a completely randomised design with three replications. Each replication consisted of one plastic pot containing 1 kg of soil and two plants. The experiment was repeated once. A Cochran's test for homogeneity of variance (Gomez and Gomez 1994) indicated that the data from the two experiments could be pooled, so the data from the two trials were pooled for data analysis. The experiment-treatment interactions were not significant ($P \geq 0.05$) when compared to the main effects of treatments. Data were subjected to analysis of variance (ANOVA) and linear and polynomial regression procedures (SAS Institute Inc., 1989, Cary, NC). Means of the six replications were used to fit the regression models. Treatment mean comparisons for

Ca content in leaf tissue were made using Tukey's test ($P \leq 0.05$). Data from Si content in leaf tissue was correlated with the components of host resistance evaluated. The percentage of PLA was also correlated with the percentage of NLA.

Results

Silicon and calcium contents in sorghum leaf tissue

There was a significant interaction between sorghum lines and Si application rates ($P \leq 0.05$). The Si content in leaf tissue of both lines was directly related to the amount of Si applied to the soil (Fig. 1). There was no significant change ($P \geq 0.05$) in Ca content in leaf tissue of both lines as a result of wollastonite and/or calcium carbonate rates used to equilibrate the amount of Ca in the soil. The values for Ca content in leaf tissue ranged from 5.13 to 6.57 g kg⁻¹ and from 6.03 to 7.58 g kg⁻¹, respectively, for lines BR005 and BR009.

Development of anthracnose symptoms

The pattern of anthracnose symptom development on the adaxial epidermis of leaves from sorghum plants of the lines BR005 (resistant) and BR009 (susceptible), at Si application rates ranging from 0 to 0.30 g kg⁻¹ of soil, is illustrated in Fig. 2. On leaves from plants of the resistant line, there was absence of necrotic lesions with acervuli, but pigmented reddish to purple flecks were formed on some, mainly between leaf veins with

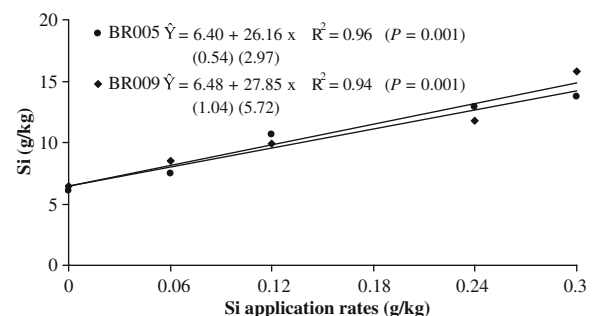


Fig. 1 Relationship between silicon (Si) content on leaf tissue of sorghum lines BR005 and BR009 with increasing silicon (Si) application rates. The standard error of each regression model parameter is shown in parentheses. Data are from two pooled experiments ($n=6$)

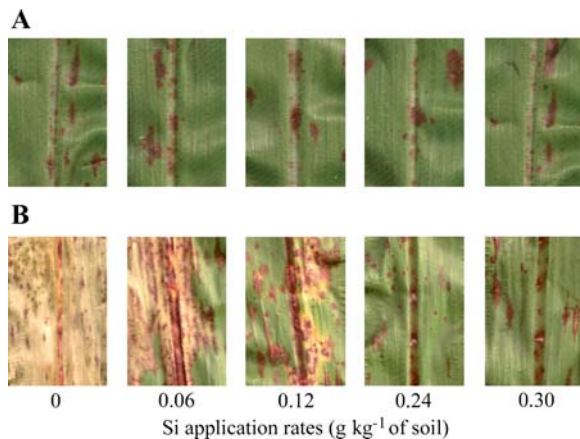


Fig. 2 Symptoms of anthracnose on leaves of sorghum lines BR005 **A** and BR009 **B** with increasing silicon (Si) application rates

some coalescence of lesions. Visually, there was no difference in the size and number of these pigmented flecks between Si application rates. By contrast, on leaves of the susceptible line at Si application rates of 0 and 0.06 g kg⁻¹ of soil, there were many circular-elliptical necrotic lesions showing intense chlorosis and coalescence. Their centres became greyish to dark straw coloured with wide margins that varied in colour from red to tan to blackish purple. Numerous acervuli with prominent black to dark brown setae were seen on these lesions. As the Si application rates increased, the number and size of these necrotic lesions were reduced, and showed a decrease in the intensity of chlorosis and lesion coalescence. Pigmented flecks were observed on leaves from plants at all Si application rates; however, at the two highest Si application rates, they resembled those observed on leaves of the resistant line at the same Si application rate.

Incubation period

There was a significant interaction between sorghum lines and Si application rates ($P \leq 0.05$). The IP of *C. sublineolum* on leaves of the resistant line BR005 ranged from 73 to 84 h, but there was no effect of Si application rates on this component of resistance. A second order regression curve best described ($P \leq 0.05$) the effect of Si application rates on the IP for the susceptible line (Fig. 3). The IP at the highest Si rate was 13% less than the control.

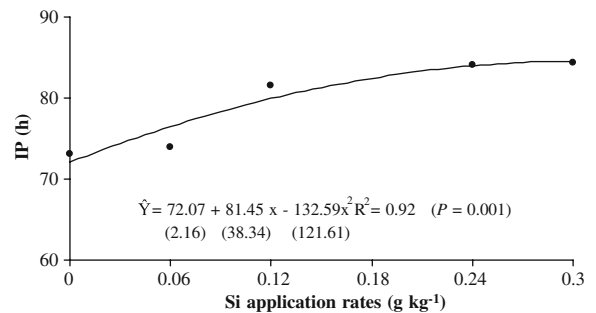


Fig. 3 Relationship between incubation period (IP) of anthracnose on leaves of sorghum line BR009 with increasing silicon (Si) application rates. The standard error of each regression model parameter is shown in parentheses. Data are from two pooled experiments ($n=6$)

Latent period

The LP₆₀ was not evaluated on the resistant sorghum line BR005 due to the absence of acervuli. For the susceptible line, a quadratic model best described ($P \leq 0.05$) the relationship of Si application rates and LP₆₀ (Fig. 4). The LP₆₀ increased >24% as the Si application rates increased from 0 to 0.3 g kg⁻¹ of soil.

Area under relative infection efficiency progress curve

This component of resistance was evaluated only on the susceptible sorghum line because the LP₆₀ cannot be determined on the resistant line. The AURIEPC decreased ($P \leq 0.05$) by >75% as the Si application rates increased from 0 to 0.3 g Si kg⁻¹ of

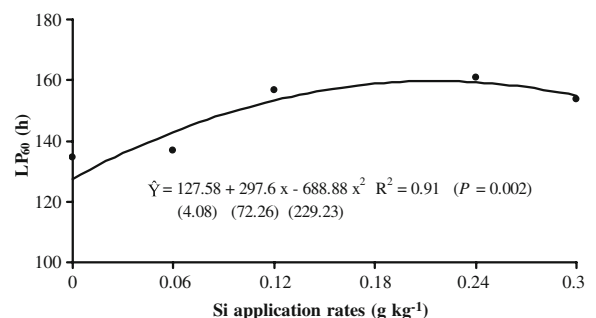


Fig. 4 Relationship between latent period (LP₆₀) of anthracnose on leaves of sorghum line BR009 with increasing silicon (Si) application rates. The standard error of each regression model parameter is shown in parentheses. Data are from two pooled experiments ($n=6$)

soil. The lowest AURIEPC was obtained at the Si rate of 0.25 g kg⁻¹ of soil (Fig. 5).

Area under anthracnose index progress curve

There was a significant interaction between sorghum lines and Si application rates ($P \leq 0.05$). The AUAIPC on the resistant sorghum line was not affected by Si application rates. The response of the susceptible line in AUAIPC to Si application rates was quadratic ($P \leq 0.05$) with the lowest value occurring at the Si rate of 0.27 g kg⁻¹ of soil (Fig. 6). The AUAIPC decreased by >31% as the Si application rates increased from 0 to 0.30 g kg⁻¹ of soil.

Final disease severity

There was a significant interaction between sorghum lines and Si application rates ($P \leq 0.05$). There was no significant effect of Si application rates on the FDS for the resistant line. A quadratic model best described ($P \leq 0.05$) the effect of Si application rates on the FDS for the susceptible line (Fig. 7). The FDS at the highest Si rate was 27% less than the control.

Pigmented and necrotic leaf area

There was a significant interaction between sorghum lines and Si application rates ($P \leq 0.05$). Silicon application rates only influenced the percentage of PLA and NLA on the susceptible line. The relationship of Si application rates and the percentage of PLA

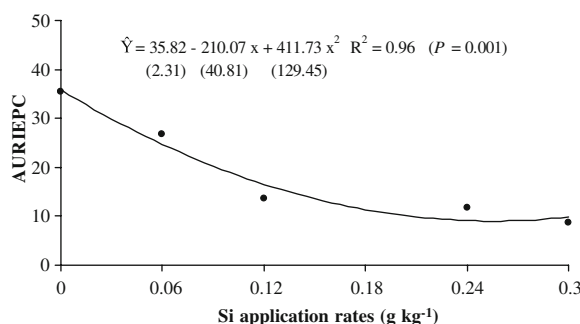


Fig. 5 Relationship between area under relative infection efficiency progress curve (AURIEPC) on leaves of sorghum line BR009 with increasing silicon (Si) application rates. The standard error of each regression model parameter is shown in parentheses. Data are from two pooled experiments ($n=6$)

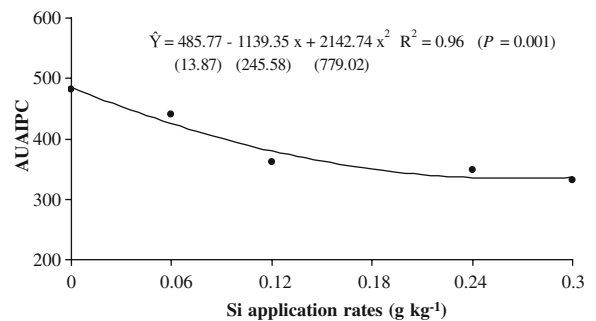


Fig. 6 Relationship between area under anthracnose index progress curve (AUAIPC) on leaves of sorghum line BR009 with increasing silicon (Si) application rates. The standard error of each regression model parameter is shown in parentheses. Data are from two pooled experiments ($n=6$)

(Fig. 8a) and NLA (Fig. 8b) was quadratic ($P \leq 0.05$). The lowest percentage of PLA and NLA was 8 and 12%, respectively, at the Si application rates of 0.24 and 0.27 g kg⁻¹ of soil.

Pearson correlation

The correlation between Si content on leaf tissue of the susceptible line and the AURIEPC, AUAIPC, FDS, PLA, and NLA was negatively significant ($P \leq 0.05$) ($r = -0.57, -0.37, -0.40, -0.67$, and -0.77 , respectively). There was no correlation between Si content and IP or LP₆₀. The correlation between the percentage of PLA with the percentage of NLA was negatively significant ($r = -0.74, P \leq 0.01$).

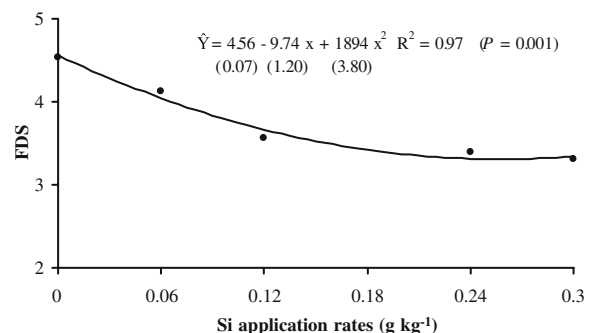


Fig. 7 Relationship between final disease severity (FDS) on leaves of sorghum line BR009 with increasing silicon (Si) application rates. The standard error of each regression model parameter is shown in parentheses. Data are from two pooled experiments ($n=6$)

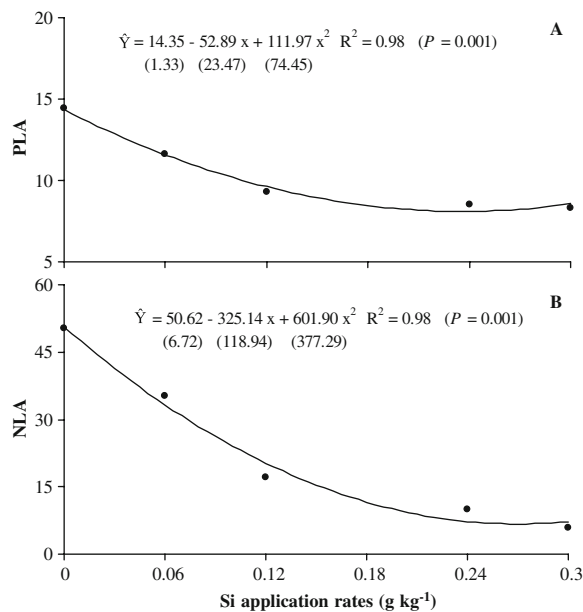


Fig. 8 Relationship between the percentage of pigmented leaf area (PLA) and percentage of necrotic leaf area (NLA) on leaves of sorghum lines BR005 **A** and BR009 **B** with increasing silicon (Si) application rates. The standard error of each regression model parameter is shown in parentheses. Data are from two pooled experiments ($n=6$)

Discussion

Economically important diseases in rice, wheat, barley, rye, corn, cucumber, grape, and strawberry are efficiently controlled by supplying plants with Si (Datnoff et al. 2007). However, in the sorghum-*C. sublineolum* pathosystem, to our knowledge, no study to date has investigated whether Si could increase host resistance to anthracnose. Therefore, this study attempted to fill this gap by providing novel information on how Si affects some components of resistance in sorghum lines resistant and susceptible to anthracnose. Sorghum can contain about 1 to 2% of Si in the shoot, but this is less than rice, which is known as a Si accumulator plant with Si content up to 5% (Rodrigues et al. 2001a, b; Hattori et al. 2005). Si content in leaf tissue of the resistant and susceptible lines was, on average, 13.7 and 16.7 g kg⁻¹, respectively, at the highest Si rate. This Si content seemed to be quite sufficient, based on the innate physiological capacity of sorghum plants to uptake this element from the soil solution, to negatively impact disease progress. The fact that sorghum plants having high Si content in their leaf tissue became

more resistant to anthracnose was more evident in the susceptible than in the resistant line. Since Ca content in sorghum leaf tissue did not change, it can be concluded that variations in Si accounted for differences in the level of disease response observed in the present study. Rodrigues et al. (2003) found that levels of Si in tissue of six rice cultivars, 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 and Okasaka 1995). Silicon uptake also may depress the absorption of Ca by the rice plant, resulting in decreased Ca in shoot tissues (Ma and Takahashi 2002).

In the resistant line, Si application rates had no apparent effect on the IP, AUAIPC, FDS, PLA, and NLA. There was an absence of acervuli on the pigmented flecks formed on leaf blades, regardless of the presence of Si. This clearly indicates that fungal infection was not successful, which suggests a possible hypersensitive type resistant reaction. According to Ali and Warren (1987), when this type of resistance occurs, small necrotic lesions appear on leaf blades, but the fungus fails to sporulate. Rodrigues et al. (2005) found that a cultivar named Katy responded to an avirulent race of *P. grisea* through the development of a hypersensitive response along with a strong induction of PR-1 and peroxidase transcripts, independent of the presence of Si. The results of their study proved an active participation, distinct from single-gene-defined resistance, for Si in the defence of a rice susceptible cultivar against blast. Rodrigues et al. (2001b) also reported that Si had more effect on reducing the severity of sheath blight and the area under the vertical lesion extension progress curve in completely susceptible and moderately susceptible rice cultivars than in cultivars with a high level of partial resistance.

All components of resistance evaluated on plants from the susceptible line were affected by Si. The longest IP and LP₆₀ occurred at the highest Si application rate. Any slight increase in both IP and LP can slow the epidemic rate, which consequently,

decreases the number of pathogen secondary cycles (Zadoks 1971). Several sorghum cultivars with resistance to anthracnose exhibited an increase in the LP and this variable had a negative correlation with the area under anthracnose progress curve (Casela et al. 1993). Seebold et al. (2001) found that although the latent period of *P. grisea* did not differ between some rice cultivars with different levels of partial resistance to blast, the incubation period lengthened with increasing Si application rates in the soil. The incubation period of *R. solani* causing sheath blight in rice was not affected by soil amendment with Si application rates (Rodrigues et al. 2003). Increased resistance of rice plants supplied with Si to blast disease has been attributed to the deposition of this element below the cuticle (Kim et al. 2003). However, probing more deeply into how Si affects blast disease in rice, it was found that plants supplied with this element respond more promptly to fungus infection by increasing the production of phenolic compounds and phytoalexins associated with a strong activation of some PR-genes such as peroxidase and PR-1 (Rodrigues et al. 2005; Datnoff et al. 2007). The reduction in AUAIPC, FDS, and the percentage of NLA at the highest Si application rate indirectly indicates that even though the pathogen still gains full access to host tissue, its colonisation can be affected by some mechanism of resistance which will dictate the final lesion size. A lower infection frequency accumulated during host growth results in an overall lower level of disease severity (Parlevliet 1979). Highest Si application rates had a greater impact in reducing the AURIEPC. Seebold et al. (2001) reported that the relative infection efficiency, expressed as the number of rice blast sporulating lesions mm^{-2} of leaf area, decreased significantly in a linear manner with increasing Si application rates. Brecht et al. (2004) found that the number of grey leaf spot lesions on leaves of St. Augustine grass was significantly affected by Si.

Silicon application rates had no effect on the percentage of leaf area with pigmented flecks in the resistant line. In contrast, on the susceptible line, the percentage of leaf area with pigmented flecks decreased with increasing Si application rates. Interestingly, the percentage of leaf area with pigmented flecks and their pattern on leaf blades of plants from the susceptible line was very similar to those observed on leaves of the resistant line at the two highest Si

application rates. A plausible explanation for a reduction in the percentage of leaf area with pigmented flecks on leaf blades of the susceptible line can be attributed to the reduced number and size of the necrotic lesions. Even though the number and size of necrotic lesions was not evaluated, the reduction in NLA can be an indirect indicator of a decrease in these variables. In sorghum, it is very common to observe production of anthocyanidins after applying elicitors, but many of the pigmented compounds do not exhibit toxicity against pathogens (Nicholson and Hammerschmidt 1992). The pigmented flecks formed on leaf blades of sorghum plants, regardless of being resistant or susceptible to *C. sublineolum*, are known to contain 3-deoxyanthocyanidin phytoalexins known as luteolinidin, 5-methoxyluteolinidin, apigeninidin, and a caffeic acid ester of 5-O-abrabinosyl-apigeninidin (Nicholson et al. 1987). In the incompatible interaction, their production is more rapid, intense, and complex in composition when compared to that in the compatible interaction (Lo et al. 1999).

Methods used to protect economically important crops such as sorghum against devastating pathogens like *C. sublineolum* are mainly focused on using genetic resistance. New strategies need to be developed considering the high variability in the pathogen population. In line with this approach, results provided in this study support the conclusion that application of Si increases sorghum resistance against infection by *C. sublineolum* by affecting some components of host resistance. This is, to our knowledge, the first study to report a reduction in a sorghum disease after supplying Si to plants. This information may prove to be invaluable in future basic research aiming to determine the exact mechanisms behind sorghum resistance to anthracnose facilitated by Si.

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References

- Ali, M. E. K., & Warren, H. L. (1987). Physiological races of *Colletotrichum graminicola* on sorghum. *Plant Disease*, 71, 402–404.

- Ali, M. E. K., & Warren, H. L. (1992). Anthracnose of sorghum. In W. A. J. De Milliano, R. A. Frederiksen, & G. D. Bengston (Eds.), *Sorghum and millets diseases: A second world review* (pp. 203–208). Patancheru, India: ICRISAT.
- Brecht, M. O., Datnoff, L. E., Kucharek, T. A., & Nagata, R. T. (2004). Influence of silicon and chlorothalonil on the suppression of gray leaf spot and increase plant growth in St. Augustine grass. *Plant Disease*, 88, 338–344.
- Casela, C. R., Frederiksen, R. A., & Ferreira, A. S. (1993). Evidence for dilatory resistance to anthracnose in sorghum. *Plant Disease*, 77, 908–911.
- Datnoff, L. E., Rodrigues, F. Á., & Seebold, K. W. (2007). Silicon and plant disease. In L. E. Datnoff, W. H. Elmer, & D. M. Huber (Eds.), *Mineral nutrition and plant disease* (pp. 233–246). St. Paul, MN: The American Phytopathological Society.
- Dhingra, O. D., Sinclair, J. B. (1995). *Basic plant pathology methods* (p. 434). 2nd edition. CRC Press Inc.
- Frederiksen, R. A. (2000). *Compendium of sorghum diseases* p. 129. St. Paul, MN: The American Phytopathological Society.
- Gomez, K. A., & Gomez, A. A. (1994). *Statistical procedures for agricultural research* (2nd ed.). USA, New York: Wiley.
- Gwary, D. M., & Asala, S. W. (2006). Cost-benefit of fungicidal control of anthracnose on sorghum in Northern Nigeria. *International Journal of Agriculture & Biology*, 8, 306–308.
- Harris, H. B., & Sowell, G. (1970). Incidence of *Colletotrichum graminicola* on *Sorghum bicolor* introductions. *Plant Disease Reporter*, 54, 60–62.
- Harris, H. B., Johnson, B. J., Dobson, J. W., & Luttrell, E. S. (1964). Evaluation of anthracnose on grain sorghum. *Crop Science*, 4, 460–462.
- Hattori, T., Inanaga, S., Araki, H., An, P., Morita, S., Luxova, M., & Lux, A. (2005). Application of silicon enhanced drought tolerance in *Sorghum bicolor*. *Physiologia Plantarum*, 123, 459–466.
- Inanaga, S., & Okasaka, A. (1995). Calcium and silicon binding compounds in cell walls of rice shoots. *Japanese Journal of Soil Science and Plant Nutrition*, 41, 103–110.
- Kim, S. G., Kim, K. W., Park, E. W., & Choi, D. (2003). Silicon-induced cell wall fortification of rice leaves: a possible cellular mechanism of enhanced host resistance to blast. *Phytopathology*, 92, 1095–1103.
- Korndörfer, G. H., Pereira, H. S., & Nolla, A. (2004). *Análise de silício: solo, planta e fertilizante* p. 24. Uberlândia, MG: Universidade Federal de Uberlândia (Boletim Técnico).
- Liberato, J. R. (2003). Desenvolvimento e avaliação do Software Quant para quantificação de doenças de plantas por análise de imagens. Viçosa, Brasil: Universidade Federal de Viçosa, Ph.D thesis.
- Lo, S. C., De Verdier, K., & Nicholson, R. L. (1999). Accumulation of 3-deoxyanthoxanidin phytoalexins and resistance to *Colletotrichum sublineolum* in sorghum. *Physiological and Molecular Plant Pathology*, 55, 263–273.
- Ma, J. F., & Takahashi, E. (2002). *Soil, Fertilizer, and Plant Silicon Research in Japan* (p. 274, 1st ed.). Amsterdam, The Netherlands: Elsevier Science.
- McKinney, H. H. (1923). Influence of soil temperature and moisture on infection of wheat seedlings by *Helminthosporium sativum*. *Journal of Agricultural Research*, 26, 195–218.
- Nicholson, R. L., & Hammerschmidt, R. (1992). Phenolic compounds and their role in disease resistance. *Annual Review of Phytopathology*, 30, 369–389.
- Nicholson, R. L., Kollipara, S. S., Vincent, J. R., Lyons, V. P., & Cadena-Gomez, G. (1987). Phytoalexin synthesis by the sorghum mesocotyl in response to infection by pathogenic and non pathogenic fungi. *Proceedings of the National Academy of Science of the USA*, 84, 5520–5524.
- Novais, R. F., Neves, J. C. L., & Barros, N. F. (1991). Ensaio em ambiente controlado. In A. L. Oliveira, W. E. Garrido, J. D. Araújo, & S. Lourenço (Eds.), *Métodos de pesquisa em fertilidade do solo* (pp. 153–189). Brasília, DF: EMBRAPA-SEA.
- Parlevliet, J. E. (1979). Components of resistance that reduce the rate of epidemic development. *Annual Review of Phytopathology*, 17, 203–222.
- Rodrigues, F. Á., Datnoff, L. E., Korndörfer, G. H., Seebold, K. W., & Rush, M. C. (2001a). Effect of silicon and host resistance on sheath blight development in rice. *Plant Disease*, 85, 827–832.
- Rodrigues, F. Á., Jurick, W. M., Datnoff, L. E., Jones, J. B., & Rollins, J. A. (2005). Silicon influences cytological and molecular events in compatible and incompatible rice-*Magnaporthe grisea* interactions. *Physiological and Molecular Plant Pathology*, 66, 144–159.
- Rodrigues, F. Á., Korndörfer, G. H., Corrêa, G. F., Buki, G. B., Silva, O. A., & Datnoff, L. E. (2001b). Response of six gramineae species to application of calcium metasilicate. In L. E. Datnoff, G. H. Snyder, & G. H. Korndörfer (Eds.), *Silicon in agriculture* vol. 8, (p. 378). Amsterdam, The Netherlands: Elsevier Science B. V. Studies in Plant Science.
- Rodrigues, F. Á., Vale, F. X. R., Korndörfer, G. H., Prabhu, A. S., Datnoff, L. E., Oliveira, A. M. A., & Zambolim, L. (2003). Influence of silicon on sheath blight of rice in Brazil. *Crop Protection*, 22, 23–29.
- Seebold, K. W., Kucharek, T. A., Datnoff, L. E., Correa-Victoria, F. J., & Marchetti, M. A. (2001). The influence of silicon on components of resistance to blast in susceptible, partially resistant, and resistant cultivars of rice. *Phytopathology*, 91, 63–69.
- Shaner, G., & Finney, R. E. (1977). The effect of nitrogen fertilization on the expression of slow-mildewing resistance in Knox wheat. *Phytopathology*, 67, 1051–1056.
- Sheriff, C., Whelan, M. J., Arnold, G. M., & Bailey, J. A. (1995). rDNA sequence analysis confirms the distinction between *Colletotrichum graminicola* and *C. sublineolum*. *Mycological Research*, 99, 475–478.
- Snyder, G. H., Jones, D. B., & Gauch, G. J. (1986). Silicon fertilization of rice on Everglades Histosols. *Soil Science Society of American Journal*, 50, 1259–1263.
- Sutton, B. C. (1980). *The Coelomycetes: Fungi Imperfecti with Pycnidia, Acervuli and Stromata* p. 696. Kew, London: Commonwealth Mycological Institute.
- Warren, H. L. (2000). Leaf anthracnose. In R. A. Frederiksen (Ed.), *Compendium of sorghum diseases* (pp. 10–11). St. Paul, MN: The American Phytopathological Society.
- Zadoks, J. C. (1971). Systems analysis and the dynamics of epidemics. *Phytopathology*, 61, 600–610.