

Effects of Si fertilization on Si in soil solution, Si uptake by rice, and resistance of rice to biotic stresses in Southern Vietnam

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Received: 1 April 2017 / Accepted: 19 July 2017 / Published online: 25 July 2017

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Abstract Silicon (Si) mitigates abiotic and biotic stresses for rice plants (*Oryza sativa* L.). Here, we test relationships between Si cycling, plant growth, and pest and fungal attacks in rice agroecosystems. We conducted a plot experiment on Si fertilization in a Southern Vietnamese paddy, where plant-available Si was inherently low. For two cropping seasons, we investigated the temporal dynamics of Si in soil solution, plant Si uptake, and the occurrence of leaf folders (*Cnaphalocrocis medinalis*) and rice blast caused by the fungus *Magnaporthe oryzae*. Silicon application increased Si concentrations in soil solutions collected in the field as expected from previous laboratory experiments. Soil solution Si concentrations were furthermore affected by Si uptake by plants and by recycling Si with rice straw ash. Silicon concentrations in rice leaves at tillering stage increased with increasing Si

application. However, surprisingly, no relationship between Si in soil solution and Si concentration in straw at maturity stage was found. The occurrences of leaf folders and rice blast disease were mitigated by increased Si uptake. However, rice biomass production was not affected, probably because the biotic stress level was generally low. Our field data emphasize the importance of recycling crop residues in rice fields for the Si supply to plants, especially in regions with low Si availability. They furthermore show that under field conditions, the relationship between dissolved Si in soil solution and Si uptake by rice plants is not as straightforward as expected and thus needs to be further investigated.

Keywords Dissolved silicon · Leaf folder · Rice · Rice blast · Silicon fertilization

Electronic supplementary material The online version of this article (doi:10.1007/s10333-017-0610-2) contains supplementary material, which is available to authorized users.

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Introduction

Rice plants (*Oryza sativa* L.) accumulate up to 10% silicon (Si), mainly in the form of amorphous Si dioxide particles (Jones and Handreck 1967; Ma and Takahashi 2002). Sufficient Si supply enhances the plants' strength and rigidity and improves defense against abiotic stresses (such as strong rain, wind, salinity, and drought) and biotic stresses (such as attacks by insect pests and fungi; Guntzer et al. 2012; Meharg and Meharg 2015). Hence, Si fertilization in paddy fields with inherently low plant-available Si might be an option to increase rice yields (Marxen et al. 2016) and at the same time to decrease the demand for pesticides (Guntzer et al. 2012).

Silicon availability in paddy soils can differ greatly between rice production regions; the major reason for these differences seems to be the weathering status of the soil

(Tsujiimoto et al. 2014; Klotzbücher et al. 2015a). Weathering of soil minerals and irrigation are the two main sources of plant-available dissolved Si (dSi) in paddies, while significant losses can occur due to water percolation and drainage of flooded fields before harvest (Desplanques et al. 2006; Klotzbücher et al. 2015b; Nguyen et al. 2016). Other main factors determining Si cycling in paddies are plant Si uptake, removal of Si due to plant harvest, and recycling of Si via the rice straw (Seyfferth et al. 2013; Klotzbücher et al. 2015b; Marxen et al. 2016). Hence, Si cycling and availability are determined by both natural soil conditions and agricultural management.

A number of Si fertilizer experiments were conducted under field conditions as reviewed by Guntzer et al. (2012), Haynes (2014), and Tubana et al. (2016). In these studies, Si was applied in the form of calcium (Ca) or magnesium (Mg) silicate, a residue from blast furnace of industrial phosphorous mining (slag), which is highly soluble and releases plant-available Si. The addition of these materials typically enhances plant growth, an effect ascribed to the benefits of Si. From a researcher's point of view, the use of such materials has the disadvantage that the effects of enhanced Si availability cannot be studied directly, as they might be superimposed by effects of Ca^{2+} and Mg^{2+} on plant growth. Furthermore, slags might contain considerable amounts of heavy metal contaminants and their dissolution releases $(\text{OH})^-$ ions causing an increasing soil pH (Haynes 2014).

An alternative Si source that can be used to directly address effects of Si availability on plant growth is silica gel (SiO_2), which does not import other elements or affect soil pH. We used silica gel as fertilizer in an experiment in Northern Vietnam, where plant-available Si was inherently low as soils are strongly weathered and de-silicified. We found in a field experiment that rice yields were increased by 34% for the extremely high application rate of $17.3 \text{ Mg Si ha}^{-1}$ (Marxen et al. 2016). For a low application rate of silica gel ($0.4 \text{ Mg Si ha}^{-1}$), yield was not affected, but Si uptake still increased. However, the experiment only lasted one cropping season and we did not assess whether increased Si uptake increased the plants' resistance against biotic stresses like pests and diseases.

A common pest in rice cultivation is the leaf folder (*Cnaphalocrocis medinalis*) which damages rice crops during its larval stage. The larva folds a leaf blade longitudinally with silk strands and feeds on mesophyll tissue inside the folded leaf, thereby creating longitudinal white and transparent streaks on the blade, disturbing photosynthesis and growth, and ultimately reducing rice yield (Han et al. 2015 and references therein). High Si concentration in rice plants is assumed to cause mandibular wear of leaf folder larvae (Reynolds et al. 2009). Han et al. (2015) showed in a pot experiment that increased Si concentration

in rice decreased the net reproduction rate of the rice leaf folder population and enhanced the resistance of rice plants to the pest because of reduced food quality and food conversion efficiencies, although consumption increased with Si concentration.

Also, the rice blast fungus (*Magnaporthe oryzae*) is common in rice cultivation and occurs mainly as leaf blast or neck blast (Bonman et al. 1989; Webster and Gunnell 1992). Leaf blast can cause severe damage before plants reach the productive growth phase, while neck blast is the most destructive in terms of yield loss (Ou 1985; Bonman et al. 1989). The fungus *M. oryzae* enters the plant via appressorial penetration through the epidermis. It was oftentimes shown in field experiments that Si mitigates rice blast disease (Datnoff et al. 1997; Seebold et al. 2000, 2004); the mechanism behind this effect is, however, still uncertain. It might be a physical effect, i.e., silicified cells in the leaf epidermis or the cuticle-Si double layer may act as a physical barrier for appressorial penetration. Another explanation might be that Si mediates physiological changes of rice plants to confer disease resistance (Seebold et al. 2004).

The present study was motivated by the shortage of field data on relationships between Si cycling, plant growth, and pest and fungal attacks in rice agroecosystems. We conducted a field plot experiment over the course of two cropping seasons in a paddy field in Southern Vietnam, where plant-available Si is low (Klotzbücher et al. 2015a). By applying easily soluble silica gel, we manipulated the release of potentially plant-available dSi. Our aims were to assess

- temporal changes in dissolved Si concentrations ([dSi]) during rice production cycles,
- effects of silica gel application on [dSi],
- how plant growth and uptake of Si and other nutrients relate to [dSi], and
- how differences in Si supply to plants affect the occurrence of leaf folders and of leaf and neck blast, which commonly cause problems in the study region.

Materials and methods

Setup of field plot experiment

The experiment was installed on a farmers' paddy field ($10^\circ 26' 39.76'' \text{N}$ $106^\circ 3' 32.24'' \text{E}$) in Tien Giang Province in Southern Vietnam. In the field, rice has been cultivated for at least 40 years. The soil was classified as Hydragic Anthrosol (Eutric, Clayic, Amphigleyic); basic soil parameters are given in Table 1. Industrially graded silica gel of 2–3 mm grain size (Anh Duc Co. Ltd, Vietnam) was

Table 1 Basic soil parameters of the Hydragric Anthrosol (Eutric, Clayic, Amphigleyic; IUSS Working Group 2014), horizons and texture were classified according to FAO (2006)

Depth (cm)	Horizon	Texture	pH (KCl)	TOC (g kg ⁻¹)	C/N	CEC (mmol _c kg ⁻¹)	Ca ²⁺ (mmol _c kg ⁻¹)	Mg ²⁺	K ⁺	Na ⁺	BS (%)	Fe _{oxalate} (g kg ⁻¹)
0–22	Arp	C	3.1	38	11	300	117	79	2	6	68	5.7
22–33	Ardp	SiC	3.1	12	16	190	86	77	3	6	90	2.6
33–66	Brl1	SiC	3.5	13	15	199	58	73	2	6	70	2.1
66–90	Brl2	SiC	3.5	16	18	223	57	74	3	6	63	2.5
>90	Brl3	SiC	3.4	10	15	204	56	78	3	7	70	2.1

TOC total organic carbon, CEC cation exchange capacity (extracted with ammonium acetate at pH 7), BS base saturation, C clay, SiC silty clay

applied to the plots once, in the beginning of the first cropping season on 28 of November 2013 at three levels: 0.1, 0.4, and 1.5 Mg Si ha⁻¹. The medium application rate of 0.4 Mg Si ha⁻¹ was used to compare the effects to our fertilization experiment in Northern Vietnam, where this application rate increased Si uptake by rice (Marxen et al. 2016). The fourfold higher application rate of 1.5 Mg Si ha⁻¹ was hypothesized to additionally increase rice yield, and the fourfold lower application rate of 0.1 Mg Si ha⁻¹ was used to test a fertilizer amount that is economically cheaper for farmers to apply. Additionally, a control (i.e., no silica gel) was established. All four treatments were spatially replicated five times. The plots had a size of 8 m × 8 m, but all samplings were done within an inner core of 6 m × 6 m in order to avoid ‘edge effects.’ Plots were arranged randomly and margined by bunds as shown in the picture (Online Resource 1) to minimize Si transport from/to the surroundings by the flood water. Silica gel was homogeneously distributed in the moist plots and mixed into the topsoil using a spade; directly afterward, rice (*Oryza sativa* L. cv. IR 50404) was seeded by hand which is the locally common practice. Nine days later, the plots were flooded for the first time. They were flooded two additional times during the cropping season in order to maintain continuous submerged soil conditions until they were drained 10 days before harvest. Fertilization was done by the farmer according to the locally common practice; 80 kg N ha⁻¹, 14 kg P ha⁻¹, and 73 kg K ha⁻¹ were applied during the cropping season. Pesticides were not applied to the experimental field but to the surrounding paddies; active substances are provided in the table (Online Resource 2). The first cropping season lasted 85 days. After harvest, the rice straw was returned to the respective plots (except for a small amount, which was kept for analyses; see paragraph on plant sampling and analyses) and burned within the plots. Rice straw burning is the common practice during that season in the study area; during the wet seasons, straw is incorporated into the soil without prior burning. After a fallow period of 10 days, the

new rice crop (*Oryza sativa* L. cv. IR 50404) was seeded. Flooding and drainage of the experimental plots were carried out in the same way as during the first cropping season. Fertilizer amounts equaled 95 kg N ha⁻¹, 16 kg P ha⁻¹, and 65 kg K ha⁻¹ during the second cropping season. The second rice crop was harvested after a growth period of 82 days; the picture (Online Resource 3) shows rice in the experimental field at maturity stage.

Soil solution sampling and analyses

We sampled soil solution using permanently installed suction cups (Rhizon SMS, Rhizosphere research products, Wageningen, the Netherlands). Five days after seeding (DAS) in the first experimental cropping season, three suction cups were installed in each core plot, one in the center and two in opposite corners. The suction cups were inserted horizontally into the topsoil to a depth of 15 cm. They consisted of a 5-cm-long porous part made of polyethersulfone with an outside diameter of 2.5 mm and a pore size of 0.12–0.18 μm and were connected to a 60-cm PE/PVC tube with female/male luer lock system at the end of the tube. During both cropping seasons, every 10 days, a syringe was connected to the end of the tube above the soil surface to remove 5 ml of soil solution. The three samples taken per plot were combined into a vial that contained 300 μl of nitric acid (65%) to prevent co-precipitation of dSi and iron (Fe) oxides and sorption of dSi onto Fe oxide surfaces (Sauer et al. 2006). Silicon was determined by inductively coupled plasma optical emission spectrometry (ICP-OES; Ultima 2, Horiba Jobin-Yvon, Longjumeau, France).

Plant sampling and analyses

At tillering stage, i.e., 28 DAS and 34 DAS in the first and second cropping seasons, respectively, rice leaves were sampled in all core plots to determine the plants’ nutritional status at a critical growth stage. Sampling was

conducted by cutting the youngest fully developed leaf from several random plants until a sample size of 50 g fresh mass was obtained (Dobermann and Fairhurst 2000). Samples were dried in an oven at 65 °C for 48 h, shipped to Germany, and ground for analyses.

At maturity stage, i.e., 85 DAS and 78 DAS in the first and second cropping seasons, respectively, straw production and grain yield were measured. Three samples were taken per core plot by randomly choosing a 4 m² area, respectively (altogether 12 m²), and harvesting all enclosed plants by cutting them directly above the soil surface. Grains with hulls were separated from the straw, and both fractions were weighed. Subsamples (150 g of straw and 100 g of grains for each sample) were air-dried until constant weight was reached, weighed, and shipped to Germany. The three samples of the same plant fraction and plot were pooled, and samples were ground for analysis.

X-ray fluorescence (XRF) spectroscopy was applied to determine concentrations of Si, phosphorous (P), potassium (K), Ca, and Mg in plant material. Ground samples were dried at 85 °C for 48 h, and 3 g of each sample was mixed with 650 mg of wax; then, 32-mm pellets were prepared by pressing with a force of 12 Mg. The XRF measurements were taken using a wavelength-dispersive XRF spectrometer (S4 Pioneer, Bruker AXS, Karlsruhe, Germany), equipped with a 4-kW-Rh X-ray tube (75- μ m Be window), 60-kV generator, and an eight-position crystal changer. The spectrometer operating conditions were vacuum, 23-mm collimator mask, and 0.46° collimator in conjunction with the analyzing crystal PET, and 30 kV at current of 80 mA. Calibration was performed using plant standard materials for all elements. In the case of Si, elemental concentrations in the samples were above the calibration range. Therefore, a standard addition method was applied as described in Marxen et al. (2016): A dried and ground grass sample was mixed with different amounts of SiO₂ (2–14% in steps of 2%), pressed to pellets, and measured under above-described conditions.

Concentrations of carbon (C) and nitrogen (N) were determined using a dry combustion analyzer (Vario EL cube, Elementar, Hanau, Germany).

Pest and disease assessment

During the first cropping season, neither leaf folders nor the rice blast disease appeared in the field; thus, both were only assessed during the second cropping season.

Leaf folders were assessed according to the standard protocol used by the Southern Regional Plant Protection Center (SRPPC) in Tien Giang Province, Southern Vietnam. A wooden frame of 0.2 m² area was placed in each core plot at tillering stage (41 DAS in the second cropping season) at five random locations (altogether 1 m² area per

plot), counting the number of leaf folder larvae feeding on rice plants and the number of damaged leaves within the enclosed area.

Leaf and neck blast were assessed at tillering stage (34 DAS in the second cropping season) and ripening stage (74 DAS in the second cropping season), respectively. Severity of leaf blast was estimated by randomly assessing 3 leaves of 25 different tillers (altogether 75 leaves) per core plot. Severity of neck blast was estimated by randomly selecting 50 panicles of different plants per core plot. Severities of leaf and neck blast were evaluated according to the scoring system developed by the International Rice Research Institute (IRRI 1996).

Statistics

Data were analyzed using SigmaPlot software version 12 (Systat Software Inc.). All data were tested for normality distribution and equal variances to choose the appropriate analysis. Seasonal means of [dSi] were calculated for each individual plot. These data were used to test treatment effects on [dSi] by applying the Kruskal–Wallis ANOVA on ranks followed by Dunn’s HSD test for pairwise comparisons between the treatments. In order to test for differences in [dSi] between the two cropping seasons, we applied paired t tests on data for each of the treatments. To analyze treatment effects on nutrient concentrations in leaves and straw, production of straw and grains, total Si uptake, and the severity of leaf and neck blast, the Kruskal–Wallis ANOVA on ranks was used; when the differences were significant, Dunn’s test for pairwise comparisons between the treatments was applied. To compare the production of straw and grains and total Si uptake between the two cropping seasons, we used the Mann–Whitney rank-sum test instead of the t test because the data were not normally distributed. To analyze the data on leaf folders and damaged leaves, one-way ANOVA followed by Tukey’s HSD test was used. Results were considered to be significant for $P \leq 0.05$.

Results

Dynamics of dSi in the soil solution

Means of [dSi] values in the first cropping season were 4.3, 4.6, 5.7, and 6.6 mg Si l⁻¹ in the control and the 0.1, 0.4, and 1.5 Mg Si ha⁻¹ treatment, respectively. Concentrations significantly differed between the control and the 1.5 Mg Si ha⁻¹ treatment and between the 0.1 Mg Si ha⁻¹ and the 1.5 Mg Si ha⁻¹ treatment (Fig. 1a). In the second cropping season, we found significantly higher [dSi] than in the first cropping season for all treatments (Fig. 1a). The temporal

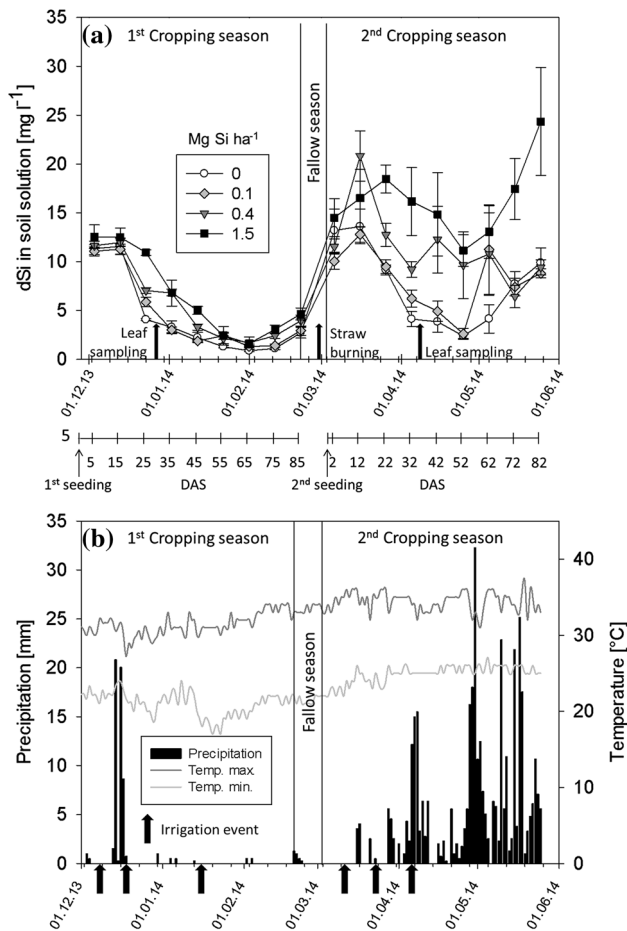


Fig. 1 **a** Dissolved Si concentration [dSi] in soil solution (\pm SE); additional x-axes show individual sampling dates in days after seeding (DAS) and **b** daily precipitation and temperature during the first and second cropping seasons (data taken from www.accuweather.com in August 2015); times of irrigation are plotted in the graph; amounts of irrigation were not measured

course of [dSi] followed the same pattern in both cropping seasons and in all treatments. In the second cropping season, means of [dSi] were 7.6, 8.3, 11.4, and 16.3 mg Si l⁻¹ in the control and the 0.1, 0.4, and 1.5 Mg Si ha⁻¹ treatment, respectively. Concentrations only differed significantly between the control and the 1.5 Mg Si ha⁻¹ treatment.

Si uptake and biomass production

In both cropping seasons, Si concentrations in rice leaves at tillering stage were positively related to Si application rates. However, when compared to the control, the increases were only significant for the 1.5 Mg Si ha⁻¹ treatment (Fig. 2). In the first cropping season, also Si concentrations in rice straw at maturity stage were positively related to Si application rates; however, the increase was only significant for the 1.5 Mg Si ha⁻¹ treatment, too.

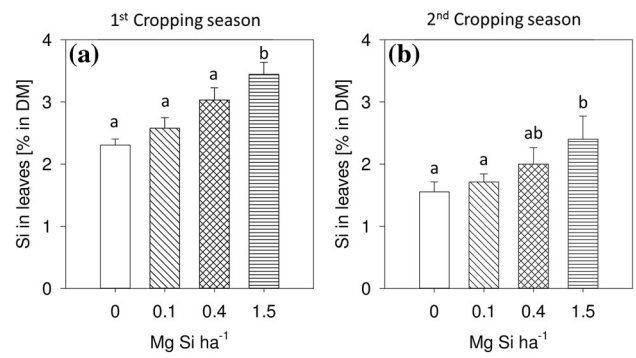


Fig. 2 Si concentration in rice leaves at tillering stage **a** in the first cropping season at 28 DAS and **b** in the second cropping season at 34 DAS; error bars represent SE; letters give significant differences ($P \leq 0.05$)

In the second cropping season, Si concentrations in straw at maturity stage did not differ between the four treatments (Fig. 3).

In the first cropping season, straw production and grain yield did not differ between the treatments. In the second cropping season, they both increased slightly but not significantly with increasing Si application, except for straw production in the control treatment (Table 2). In the second cropping season, straw production was significantly higher and grain yield was significantly lower than in the first cropping season. Total Si uptake by the plants increased slightly with increasing Si application in both cropping seasons, except for the control treatment in the second cropping season with a very high standard error (SE); when compared to the controls, the increase was only significant for the highest Si application rate in the first cropping season (Table 2).

Concentrations of essential nutrients in plant tissue

In both cropping seasons, Si application did not affect the concentrations of C, N, P, K, Ca, and Mg in rice leaves (Table 3). In the first cropping season, concentrations of C

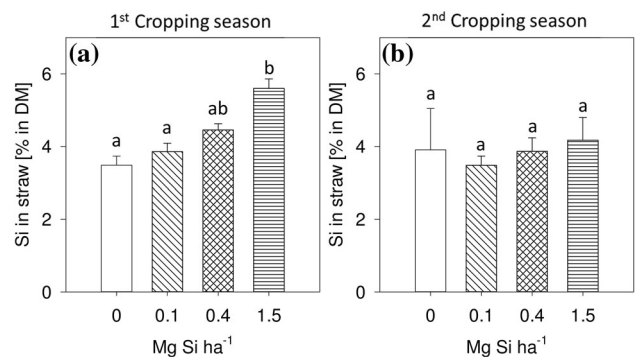


Fig. 3 Si concentration in rice straw at maturity stage **a** in the first cropping season and **b** in the second cropping season; error bars represent SE; letters give significant differences ($P \leq 0.05$)

Table 2 Straw biomass production, grain yield, and total Si uptake by aboveground biomass during the first and second cropping seasons

Treatment	Straw (Mg DM ha ⁻¹)	Grains with hulls (Mg DM ha ⁻¹)	Si uptake (kg ha ⁻¹)
First cropping season (dry season)			
0 Mg Si ha ⁻¹	9.6 (0.4) ^a	6.7 (0.3) ^a	398 (15) ^a
0.1 Mg Si ha ⁻¹	9.3 (0.3) ^a	6.7 (0.4) ^a	420 (11) ^a
0.4 Mg Si ha ⁻¹	9.3 (0.3) ^a	7.1 (0.3) ^a	484 (20) ^{a,b}
1.5 Mg Si ha ⁻¹	9.3 (0.4) ^a	6.6 (0.1) ^a	592 (34) ^b
Second cropping season (wet season)			
0 Mg Si ha ⁻¹	11.2 (0.6) ^a	4.8 (0.2) ^a	485 (69) ^a
0.1 Mg Si ha ⁻¹	11.0 (0.3) ^a	5.0 (0.1) ^a	427 (05) ^a
0.4 Mg Si ha ⁻¹	12.2 (0.3) ^a	5.1 (0.1) ^a	518 (17) ^a
1.5 Mg Si ha ⁻¹	12.9 (0.8) ^a	5.3 (0.2) ^a	595 (66) ^a

SE in brackets; letters give significant differences within one season ($P \leq 0.05$)**Table 3** Nutrient concentrations (g kg⁻¹) in rice leaves at tillering stage in the first cropping season at 28 DAS and in the second cropping season at 34 DAS

Treatment	C	N	P	K	Ca	Mg
First cropping season (dry season)						
0 Mg Si ha ⁻¹	409 (5)	41.4 (1.2)	3.92 (0.07)	27.1 (1.0)	4.49 (0.21)	2.46 (0.09)
0.1 Mg Si ha ⁻¹	409 (2)	41.6 (1.0)	3.95 (0.05)	26.7 (1.0)	4.38 (0.22)	2.37 (0.08)
0.4 Mg Si ha ⁻¹	401 (3)	41.6 (0.7)	4.08 (0.10)	27.4 (0.8)	4.27 (0.20)	2.24 (0.07)
1.5 Mg Si ha ⁻¹	398 (1)	40.9 (0.7)	3.99 (0.10)	26.7 (0.7)	3.86 (0.08)	2.11 (0.07)
Second cropping season (wet season)						
0 Mg Si ha ⁻¹	432 (1)	27.0 (1.0)	2.95 (0.13)	22.0 (1.1)	3.82 (0.28)	2.29 (0.17)
0.1 Mg Si ha ⁻¹	429 (2)	27.0 (0.7)	3.04 (0.14)	23.9 (1.3)	4.21 (0.36)	2.46 (0.13)
0.4 Mg Si ha ⁻¹	427 (3)	26.5 (0.5)	2.88 (0.13)	23.7 (1.1)	4.16 (0.43)	2.29 (0.16)
1.5 Mg Si ha ⁻¹	423 (4)	27.8 (0.5)	2.85 (0.09)	23.7 (2.1)	3.76 (0.48)	2.20 (0.16)

SE in brackets; there were no significant differences between the treatments within one cropping season for any nutrient

and Mg in rice straw slightly decreased with increasing Si application; when compared to the control; the decrease was only significant for the highest Si application rate (Table 4). Concentrations of N, P, K, and Ca in straw did not differ between the treatments. In the second cropping season, Si application did not affect the concentrations of C, N, P, K, Ca, and Mg in straw (Table 4).

Resistance of rice against leaf folders, leaf blast, and neck blast

The number of leaf folder larvae at 42 DAS in the second cropping season was significantly higher in the control than in the treatments with Si application (Fig. 4a). Likewise, the number of damaged leaves was slightly but not

Table 4 Nutrient concentrations (g kg⁻¹) in rice straw at maturity stage in the first and second cropping seasons

Treatment	C	N	P	K	Ca	Mg
First cropping season (dry season)						
0 Mg Si ha ⁻¹	387 (4) ^a	5.81 (0.37) ^a	1.34 (0.11) ^a	19.7 (0.2) ^a	3.53 (0.15) ^a	2.06 (0.12) ^a
0.1 Mg Si ha ⁻¹	383 (2) ^a	6.01 (0.19) ^a	1.43 (0.03) ^a	20.0 (0.3) ^a	3.75 (0.21) ^a	2.00 (0.02) ^a
0.4 Mg Si ha ⁻¹	375 (2) ^{a,b}	5.80 (0.17) ^a	1.33 (0.05) ^a	19.5 (0.6) ^a	3.39 (0.14) ^a	1.81 (0.05) ^{a,b}
1.5 Mg Si ha ⁻¹	363 (1) ^b	6.13 (0.19) ^a	1.39 (0.07) ^a	19.2 (0.2) ^a	3.14 (0.08) ^a	1.54 (0.05) ^b
Second cropping season (wet season)						
0 Mg Si ha ⁻¹	387 (8) ^a	7.41 (0.14) ^a	1.69 (0.08) ^a	18.3 (0.3) ^a	3.00 (0.20) ^a	1.65 (0.09) ^a
0.1 Mg Si ha ⁻¹	397 (2) ^a	8.05 (0.10) ^a	1.74 (0.06) ^a	17.8 (0.8) ^a	2.89 (0.13) ^a	1.70 (0.08) ^a
0.4 Mg Si ha ⁻¹	393 (2) ^a	8.05 (0.22) ^a	1.66 (0.02) ^a	18.8 (0.7) ^a	2.77 (0.09) ^a	1.55 (0.05) ^a
1.5 Mg Si ha ⁻¹	389 (4) ^a	7.89 (0.37) ^a	1.57 (0.08) ^a	19.5 (0.3) ^a	2.77 (0.19) ^a	1.61 (0.07) ^a

SE in brackets; letters give significant differences within one cropping season ($P \leq 0.05$)

significantly higher in the control (Fig. 4b). The maximum observed damage was 20% of leaf area.

Increased Si application decreased leaf blast severity at 34 DAS and neck blast severity at 75 DAS; when compared to the control, the increases were significant for the 0.4 and 1.5 Mg Si ha⁻¹ treatments (Fig. 5b).

Discussion

General observations on controls of dissolved Si in soil

The temporal courses of [dSi] reflect relationships between the rates of both dSi inputs and exports. Besides the application of silica gel, inputs of dSi occur by irrigation (while rain water typically shows very small or no [dSi] according to Klotzbücher et al. 2015b) and by release of Si from the soil matrix and plant residues. Exports occur by losses of water (percolation and drainage) as well as by plant Si uptake and harvest.

One clear pattern observed in both cropping seasons was that [dSi] decreased in all treatments during the period with highest biomass gain (Fig. 1a). Similar results were shown for paddy soils in California (Seyfferth et al. 2013) and Japan (Mihara et al. 2016). We refer this finding mainly to high rates of Si uptake by rice plants, which obviously excelled the rates of Si inputs by dissolution of silica gel and minerals in the strongly weathered soil. The pattern is not ubiquitous: Klotzbücher et al. (2015b) reported for Philippine paddy soils characterized by high amounts of weatherable silicate minerals that [dSi] remained constantly high during the cropping season.

Silicate mineral dissolution rates are controlled by [dSi] (e.g., Frayse et al. 2006); hence, we assume that in our field plots, rice plants can improve the release of plant-available Si from soil by strongly decreasing [dSi] during the period when they need Si for growth. In the last third of the cropping season, [dSi] increased in all treatments presumably as plant-Si-uptake rates decreased. In this period,

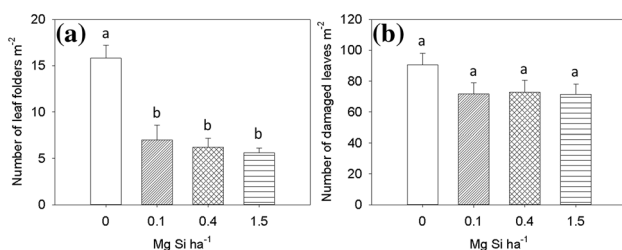


Fig. 4 Number of **a** leaf folders and **b** damaged leaves per m² in the second cropping seasons at 41 DAS; error bars represent SE; letters give significant differences ($P \leq 0.05$)

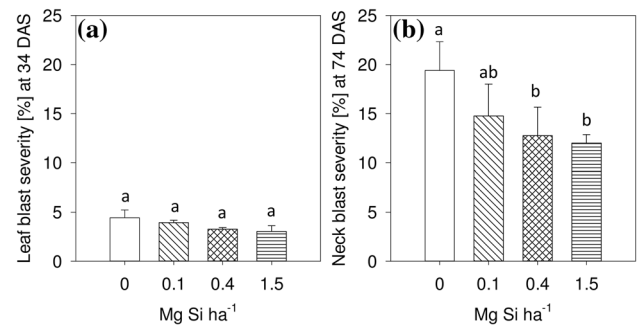


Fig. 5 Severity of **a** leaf blast at 34 DAS and **b** neck blast at 74 DAS according to the scoring system of the International Rice Research Institute (IRRI 1996) in the second cropping season; error bars represent SE; letters give significant differences ($P \leq 0.05$)

the plants primarily fill grains, which typically contain very low concentrations of Si (Klotzbücher et al. 2015b).

During the fallow season, [dSi] increased considerably (Fig. 1a). Reasons for this finding should include the lack of Si export by plant uptake and input of rice straw ash into topsoil (i.e., after the first cropping season, rice straw was burned on the respective plot). Also, a recent laboratory study by Nguyen et al. (2014) demonstrates high rates of Si release from burned rice straw. Likewise, untreated rice straw was shown to be a significant source of plant-available Si in a field study by Seyfferth et al. (2013) and in several laboratory studies (Hossain et al. 2001; Watanabe et al. 2013; Marxen et al. 2016). Our study thus supports these findings and emphasizes the importance of recycling crop residues for the Si supply to rice plants, especially in regions with strongly weathered soils where low levels of plant-available Si might limit plant growth.

Relations between Si in soil solution and Si uptake by plants

Our results show that silica gel released Si into the soil solution because mean [dSi] (Fig. 1a) increased with increasing amounts of silica gel during the first cropping season. Also in the second cropping season, [dSi] differed between the treatments. This finding firstly suggests that the applied silica gel did not dissolve completely during the first cropping season but still released Si into soil solution during the second cropping season. Also Si release from rice straw ash may have differed between the treatments during the second cropping season as the concentrations of Si in the straw, which was produced during the first cropping season, increased upon silica gel application. Silicon potentially recycled with straw during the second cropping season accounted for 69% (control treatment) to 87% (1.5 t Si ha⁻¹ treatment) of total Si taken up by the aboveground biomass.

The Si dynamics clearly differed between the two cropping seasons. In the first cropping season, we observed a positive relationship between [dSi] and Si concentrations in rice straw. Such a relationship has also been observed in laboratory experiments (Fleck 2013; Gocke et al. 2013; Marxen et al. 2016). However, in the second cropping season, Si concentrations in rice straw at maturity stage did not differ between the treatments (Fig. 3b) despite the large differences in [dSi], which were even more pronounced than during the first cropping season (Fig. 1a). Furthermore, total Si uptake did not differ between the cropping seasons even if average [dSi] values were generally higher in the second than in the first cropping season. Taken together, these observations suggest that [dSi] in soil solution was not related to Si concentration in rice straw at harvest stage or to total Si uptake by the plants. They contradict observations from the above-mentioned laboratory studies. This result was surprising, and we can only speculate about the factors explaining it. A main difference between the seasons was climate, and in particular precipitation, which was 389 mm during the second versus 59 mm during the first season. As a result of high precipitation, Si transport from roots to shoots, in part driven by the transpiration stream (Yamaj et al. 2015), might have been lower in the second than in the first cropping season due to higher air humidity. Also leaching of ions from the standing biomass, induced by the heavy rainfall, might have played a role. This is in part supported by the slightly lower K, Ca, and Mg concentrations in straw in the second cropping season. The hypothesized climate effects on Si in rice plants, however, need to be examined directly in subsequent studies.

Si fertilization effects on plant growth and pest occurrence

Effects of silica gel application on biomass production were not observed, indicating that Si was not growth limiting despite Si concentrations below or close to the threshold of 5%, which is regarded as critical value for rice plant Si status (Dobermann and Fairhurst 2000). None of the other measured macronutrients was below the critical values given for rice (Dobermann and Fairhurst 2000). There was also no indication from growth habit or leave discoloration for any other nutrient deficiency or toxicity. The grain yield of 6.1–6.7 Mg ha⁻¹ in the first cropping season was good for farmer's field in the area.

With increasing Si application rates, the occurrence of leaf folders and of leaf and neck blast decreased slightly (not always significantly). This is in accordance with the slight (also not always significant) effects of increasing silica gel application on Si concentration in leaves at tillering stage. Thus, our study basically confirms the

literature showing that increased Si uptake by rice plants mitigates damage by leaf folders (Han et al. 2015) and rice blast by increasing the resistance of rice plants against these stress factors (Datnoff et al. 1997; Seebold et al. 2000, 2004). However, in our study, the positive effects of Si on stress resistance did not induce measureable effects on plant growth. An explanation for this finding is that the occurrence of leaf folders and rice blast (and possibly also other pests and diseases) was in general low during our study period. The benefits of Si for rice plants might be more important when stress levels are higher; then, positive Si fertilization may also enhance plant growth.

Previous work showed that differences in Si availability can affect the uptake of other nutrients by plants as reviewed by Guntzer et al. (2012). In our study, Si fertilization showed no significant effect on concentrations of N, P, K, and Ca in rice plants. However, Mg concentration in straw at harvest stage decreased slightly with increasing Si concentration (Table 4). To our knowledge, no mechanism on Si–Mg interaction exists. Thus, the decrease in Mg concentration might be an effect of the reduced plant mass that does not consist of Si (similar straw biomass in the treatments but increasing Si concentration with increasing Si application).

Also C concentrations in rice straw at harvest stage decreased with increasing Si concentration (Table 4). This is in accordance with the result of our field experiment in Northern Vietnam (Marxen et al. 2016) and might be explained by a decreased formation of structural C compounds (lignin or cellulose) due to increased formation of phytoliths as proposed by Raven (1983). Both plant constituents seem to perform similar functions, including cell strength and tissue support, mechanical defense against pest and fungal diseases, and alleviation of abiotic stresses (Cooke and Leishman 2011). Hence, phytolith formation following Si uptake could be an energetically cheaper alternative to the synthesis of structural C compounds (Raven 1983).

Summary and conclusions

Our assessment demonstrates positive effects of Si application on resistance of rice plants against biotic stress. However, in contrast to previous reports, Si application did not affect rice growth. The finding might be explained by generally low stress levels during our study period. Hence, it invites further field research on the links between level of stress and Si nutrition effects on plant growth.

Concentrations of dissolved Si in paddy topsoil with inherently low plant-available Si are subject to strong temporal changes. Our results confirm previous studies showing that Si uptake by plants is a major factor causing

decreasing concentrations of dissolved Si, while recycling of phytoliths with rice straw ash increases dissolved Si in the soil solution during the fallow season. Our study furthermore demonstrates that, under field conditions, the relationships between dissolved Si in soil solution and Si uptake by plants are not always as straightforward as reported previously in laboratory experiments. More particularly, Si concentrations in straw at maturity stage were not related to differences in dissolved Si in soil solution in one of the two cropping seasons under study, suggesting that Si uptake was not related to potential amounts of Si available to the plants. The finding points to knowledge gaps about major factors of Si uptake by rice plants, which currently limit our ability to efficiently ‘manage’ Si nutrition of rice. We speculated that differences in climate between the cropping seasons might be an explanation for our findings. However, climate effects on Si uptake by plants have not yet been studied directly and thus should be in the focus of future research.

Acknowledgements This work has been financed by the LEGATO project (Funding Codes: 01LL0917A and 01LL0917N) of the Federal Ministry for Education and Research (BMBF). We thank the farmer, Mai Van Tu, for allowing us to establish the experiment on his paddy field. Tran Van Chau and his colleagues from the SRPPC helped us with the field work. Alexandra Boritzki, Aleksey Prays, Susanne Horka, Andreas Rämmler, Jutta Fröhlich, and Bernd Apelt did the laboratory work.

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