

Response of Sugarcane and Sugarcane Stalk Borers *Sesamia* spp. (Lepidoptera: Noctuidae) to Calcium Silicate Fertilization

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Abstract

Sugarcane is grown extensively throughout the world including more than 100,000 ha in Khuzestan province, Iran. The pink stalk borers *Sesamia* are key pests of sugarcane in this region, while other stalk borers will occur in sugarcane worldwide. Application of silicon as a soil amendment has provided plant mitigation to both biotic and abiotic stresses. Silicon has been shown to enhance resistance of sugarcane against stalk borers. Field trials were conducted to determine the effects of calcium silicate against infestations of stalk borers *Sesamia* spp. and on yield quality. Experiments were conducted with three sugarcane varieties CP69-1062, IRC99-01, and SP70-1143 and two rates of calcium silicate (400 and 800 kg/ha). Percentage of stalk damaged, percentage of bored internodes, length of borer tunnel (mm), number of larvae+pupae per 100 stalks, number of exit holes, and cane yield quality were determined. We demonstrate significant reduction on borer population and damage under silicon treatment, but greater reduction in the percentages of stalk damage, bored internodes, moth exit holes, and length of borer tunnel and number of larvae and pupae per 100 stalks were observed in the susceptible variety CP69-1062. Silicon treatment positively affected cane and sugarcane juice quality of for the variety CP69-1062, but not for SP70-1143. We conclude that the benefits of silicon to sugarcane quality and sugarcane resistance to stalk borers are dependent on the sugarcane variety.

Introduction

Sugarcane is a strategically important crop that has economic and social impact in many countries, but it is vulnerable to many biotic stressors and among them, lepidopteran stalk borers are the most destructive in many sugar-producing countries, including Brazil (Volpe *et al* 2014), South Africa (Keeping 2006), Indonesia (Goebel *et al* 2014), Reunion Island (Nibouche & Tibère 2008), the USA (Showler & Reagan 2012), Mexico (Vejar-Cota *et al* 2008), Iran (Jamshidnia *et al* 2010), Colombia (Vargas *et al* 2013), India (Srivastava *et al* 2012), and Papua New Guinea (Kuniata & Sweet 1994).

Two species of stalk borers are important in the province of Khuzestan, the major sugarcane producer in Iran

(Sadeghzadeh-Hemayati *et al* 2011), *Sesamia cretica* Lederer and *Sesamia nonagrioides* Lefebvre (Lepidoptera: Noctuidae). Both are capable of causing economic losses to commercial varieties and affect the sugar industry by both direct and indirect crop damages (Leslie 2004, Askarianzadeh *et al* 2008, Showler & Reagan 2012). Management of stalk-boring lepidopterans in sugarcane is multi-tactic, and several control options are used around the world, including biological control (Kuniata & Sweet 1994, Nikpay *et al* 2014), cultural practices (Reay-Jones *et al* 2005, Beuzelin *et al* 2011, Sandhu *et al* 2011), varietal resistance (Keeping 2006), and insecticide treatment (Legaspi *et al* 1999). One relatively new approach to manage stalk borers is the application of silicon as a nutritional amendment. This approach is classified as a

nutritional integrated pest management (IPM), as it involves improving crop resistance by improving crop health (Reynolds *et al* 2009, Keeping *et al* 2013). Silicon is an important nutrient element that promotes the growth and development of many agricultural and horticultural crops (Ma 2004, Ma & Yamaji 2006). Silicon is absorbed by plants in the form of monosilicic acid [$\text{Si}(\text{OH})_4$], the most common form of silicon in the soil solution at a pH below 9 (Jones & Handreck 1967). After the uptake and being transported from roots to vegetative shoots, silicic acid becomes concentrated due to water loss or physiological processes, and finally is deposited as silica gel (Ma & Yamaji 2006).

Silicon has both direct and indirect effects on insect pests (Reynolds *et al* 2009). Direct effects are related to reduction in insect growth, reproduction, and damage in host plants treated with different sources of silicon (Keeping *et al* 2009, Korndörfer *et al* 2011, White & White 2013, Nikpay & Soleyman Nejadian 2014). Indirect effects are related to insect development delay with a postponement in crop penetration, resulting in a greater exposure of the pest to natural enemies, adverse climatic conditions, and insecticidal sprays (Reynolds *et al* 2009). Recent research has indicated that silicon also enhances plant defenses at the tritrophic level. The plant-insect pest-parasitoid/predator interaction is altered through herbivore-induced plant volatile (HIPVs) emissions that silicon applications alter. This effect can attract more parasitoids to plants treated with silicon (Kvedaras *et al* 2010).

In the present study, the possible use of calcium silicate against the pink stalk borers *Sesamia* spp. was evaluated. The objectives of the study were to determine the effects of calcium silicate fertilizer treatments on sugarcane stalk borer damage and cane quality characteristics of three commercial varieties under field conditions.

Material and Methods

Experimental site details and cultivation of varieties

Trials were carried out during 2012–2013 at Salman Farsi Agro-Industry Farms (48°35'E, 31°8'S) Ahwaz, Iran. Evaluations were established in fields that had a history of moderate to high levels of stalk borer infestation. The soil was a loam (41.6% silt, 31.6% sand, and 26.8% clay, 161 mg/kg Ca, 48 mg/kg Mg, and 124 mg/kg K), with a pH of 7.7 (pH water), and EC of 4.9 ds/m.

Three sugarcane varieties, CP69-1062 (Canal Point, USA), IRC99-01 (cross made in Cuba and selected in Iran), and SP70-1143 (São Paulo, Brazil), were used in the experiments. These three varieties express different levels of resistance to stalk borers under field conditions (Askarianzadeh *et al* 2008, Nikpay *et al* 2014). All varieties were cultivated under

standard tillage, complete plow-out of previous crop remaining, following by ridging at 1.8-m row spacing through the field. Before planting of sugarcane varieties, phosphorous fertilizer (super phosphate triple/300 kg per hectare) was added with a pneumatic fertilizer machine based on standard procedures of sugarcane nutrient treatments in Iran. Each sugarcane variety was planted as billets (50–70 cm and free from stalk borer infestation), and following planting of seed cane sets, all furrows were treated with Atrazine and Sencor herbicides (3+2 kg per hectare) based on local recommendations as early post emergence application for suppression of annual weeds.

Experimental design and calcium silicate treatments

The trial was set up as a randomized complete block with a factorial design (sugarcane varieties as the main factor and Si rates as the secondary factor) with four replicates. Each experimental plot (block) consisted of four rows, 8 m long, and 1.8-m inter-row spaces (43.2 m²) in different areas of the field (in plant cane). This plot configuration was used for our experiments because plots for trials in sugarcane are recommended to be at least 20 m² (Laycock 2004). Each plot within each variety treatment with calcium silicate was separated by two buffer rows. Each variety received three treatments: calcium silicate (Ca_2SiO_4) (soluble $\text{SiO}_2 \geq 20\%$, Dalian Siliconfat Co., Ltd., Dalian, China; imported by Ghaem Agricultural and Chemical Company, Tehran, Iran) at 0, 400, and 800 kg per hectare. The rates were chosen based on previous research on pest control in rice and sugarcane (Hou & Han 2010, Korndörfer *et al* 2011), both of which are major agricultural crops with high uptake of silicon (Ma 2004). Calcium silicate samples were weighed with a digital balance (Sartorius BP1200, Gottingen, Germany) for each treatment row and stored into plastic bags. Silica was sprinkled by hand into furrows, and then carefully mixed in the soil with a hand hoe to a depth of 35 cm. Calcium silicate treatments were applied before herbicide applications and first watering of planted canes. All sugarcane varieties were planted in late August 2012.

At the harvest time of each variety, 20 stalks were randomly collected from the central rows of each experimental plot (Reay-Jones *et al* 2005) for *Sesamia* spp. damage assessments. The leaves of all stalks were completely removed (at the natural breaking point after the last fully expanded internode) before weighing stalk samples. The number of internodes per stalk, number of internodes bored per stalk, number of moth exit holes, percentage of stalks damaged (number of stalks bored per plot/total number of stalks sampled per plot×100), percentage of internodes bored (number of internodes bored per plot/total number of internodes sampled per plot×100), length of each borer tunnel (mm), number of live borers per stalks (expressed as *Sesamia* larvae or

pupae per stalk, $S/100$), height of plant (at the natural breaking point), and weight of stalks from each plot were determined and recorded. For assessing the effects of calcium silicate treatments on sugar quality, a randomized block design (Si treatment rates) was used. Twenty whole stalks (in each plot and taken in addition from previous sampling) were randomly harvested from each plot. This procedure was performed prior to harvest in 2013. These stalks were topped by hand at the natural breaking point (after the last fully expanded internode). Each bundle of 20 stalks was fed through a chipper disintegrator, and subsamples were analyzed for cane juice quality (polarity, brix, purity, and percentage of refined sugar). The polarity (%Pol) and brix of cane juice (%Brix) were obtained using a polarimeter (Optical Activity Ltd., England) and a refractometer (Index Instruments, England).

Data analysis

All data were analyzed for normality and homogeneity of variance (Bartlett's test), and appropriate transformations [\arcsin , $\log(x)$ or $\log(x+1)$] were applied where these conditions were not met, before analysis of variance was performed. However, untransformed data are presented in the tables. All analyses were performed with SPSS software (SPSS version 16, SPSS International, Chicago, USA), using variety as the main factor and calcium silicate treatment as a secondary factor. Tukey HSD test was used for mean comparisons between treatments at 0.05 significance levels.

Results

The interactions of sugarcane varieties and calcium silicate treatment were significant for all evaluated parameters regarding stalk borers damage (Table 1). There were also

significant differences among the three tested varieties to borer damage, especially for the stalk damage (%), length of borer tunnel (mm), and number of exit holes (Table 2). The variety SP70-1143 was the least damaged and the most resistant variety tested, while the variety CP69-1062 was the most susceptible to *Sesamia* infestation (Table 2). The effect of calcium silicate treatment on borer damage was affected by the treatment rate (Table 3), with 800 kg/ha being most effective for reducing stalk borers damage (Table 3). The high rate of calcium silicate (800 kg/ha) reduced the percentage of internodes bored to 1.3% for CP69-1062 and SP70-1143. For the variety IRC99-01, the percentage of bored internodes was approximately 2.4%, which did not differ significantly compared with the other two tested varieties at this rate (Table 1).

Significantly reduced tunnel length bored was also observed in the variety CP69-1062 between the two calcium silicate rates and in the variety IRC99-01 and SP70-1143 between control and the highest (800 kg/ha) calcium silicate rate for tunnel length bored (Table 1).

With respect to the length of borer tunnel per stalk, the larvae were able to produce longer tunnels in the control group of CP69-1062 variety. This represents a greater feeding tendency by the pest to this variety. The concentrations of 400 and 800 kg/ha of calcium silicate significantly reduced the performance of the larvae into the stalks (Table 1).

The number of borers (larvae+pupae) per 100 stalks ($S/100$) was significantly affected by calcium silicate treatment in the variety CP69-1062, but differences for varieties IRC99-01 and SP70-1143 were only observed at the highest rate (800 kg/ha). Overall, mean $S/100$ was lower in silicon-treated plants than in the control (0 kg/ha).

The number of *Sesamia* spp. larvae and pupae per 100 stalks on the three sugarcane varieties showed significant

Table 1 Interaction of different sugarcane varieties and silicon treatments on the percentage of stalk borers damage (SD), bored internodes (IB), length of borer tunnel (LB), mean number of larvae and pupae of *Sesamia* per 100 stalks ($S/100$), and exit holes (EH).

Varieties	Rate (kg/ha)	SD (%)	IB (%)	LB (mm)	($S/100$)	EH
CP69-1062	0	53.7±1.25a	10.1±0.55a	143.0±1.84a	45.0±2.04a	1.8±0.19a
	400	36.2±1.25b	3.8±0.32c	74.7±2.52bc	32.5±1.44bc	0.52±0.08bcd
	800	21.2±2.39e	1.3±0.18e	24.3±1.89d	21.2±2.39d	0.11±0.03 cd
IRC99-01	0	38.7±1.25b	5.3±0.26b	92.0±3.10b	37.5±1.44ab	0.87±0.13b
	400	32.5±1.44bc	3.6±0.20 cd	67.5±2.66bcd	33.7±2.50bc	0.52±0.09bcd
	800	23.7±2.39de	2.4±0.22de	40.8±2.28 cd	22.5±1.44d	0.25±0.06 cd
SP70-1143	0	31.2±1.25bcd	3.8±0.27c	61.7±4.95bcd	31.2±1.25bc	0.56±0.10bc
	400	26.2±1.25cde	2.8±0.16 cd	52.1±2.48 cd	26.2±1.25 cd	0.36±0.07 cd
	800	18.7±1.25e	1.3±0.10e	22.1±1.64d	18.7±1.20d	0.10±0.03d
$F_{4,35}$		11.93 ^{<0.0001}	36.70 ^{<0.0001}	60.89 ^{<0.0001}	4.15 ^{0.010}	12.63 ^{<0.0001}

Means followed by the same letter in each column are not significantly different using Turkey's test at $p>0.05$.

Table 2 Effects of different sugarcane varieties on the percentage of stalk damaged (SD), bored internodes (IB), length of borer tunnel (LB), mean number of larvae and pupae of *Sesamia* per 100 stalks (S/100), and exit holes (EH).

Mean±SE					
Varieties	SD (%)	IB (%)	LB (mm)	(S/100)	EH
CP69-1062	37.0±4.10a	5.0±1.13a	98.1±5.14a	32.9±3.10a	0.81±0.08a
IRC99-01	31.6±2.07ab	3.7±0.37a	70.8±2.86b	31.2±2.05a	0.55±0.05b
SP70-1143	25.4±1.68b	2.6±0.37a	48.6±3.00c	25.4±1.68a	0.34±0.04 b
$F_{2, 33}$ p value	4.26 ^{0.022}	2.93 ^{0.067}	35.3 ^{<0.0001}	2.78 ^{0.076}	12.91 ^{<0.0001}

Means followed by the same letter within each column are not significantly different by Turkey's test at $p>0.05$

differences across calcium silicate concentrations (Table 1). There were no significant differences across all three tested varieties at a high rate of calcium silicate (Table 1). Furthermore, the number of exit holes was higher in the susceptible variety CP69-1062. The number of exit holes was lower when varieties were amended with 800 kg/ha calcium silicate (Table 1).

Results of the current study indicated that 800 kg/ha of calcium silicate was sufficient to increase the percentages of polarity [Pol (%)], brix [Brix (%)], and refined sugar (%), purity, and mean weight of stalks of the variety CP69-1062 (Table 4). In the case of the variety IRC99-01, 800 kg/ha of calcium silicate significantly increased the purity, percent of refined sugar, and weight of stalk as compared to the control (Table 4). There were no significant differences in quality characteristics between control and calcium silicate-treated plants belonging to the variety SP70-1143 (Table 4).

Discussion

Sugarcane is an important silicon-accumulating crop, and it has been demonstrated that silicon has a significant effect on the sugarcane plant growth and development as well as beneficial effects in ameliorating a wide range of biotic and abiotic stresses (Ma 2004).

Also, the accumulation of silicon in sugarcane has proven to be an important element of resistance against various insect and mite pests (Reynolds *et al* 2009). Silicon accumulated in tissues of stalk and leaves provides a physical stiffness and barrier layer against chewing and sucking pests (Korndörfer *et al* 2011, Keeping *et al* 2013, White & White 2013, Nikpay & Soleyman Nejadian 2014).

The calcium silicate fertilizer applied in this research affected the level of damage caused by pink stalk borers in all three sugarcane varieties tested. The results of our study extend the effects of calcium silicate as a nutritional means of increasing sugarcane resistance to an additional genus of stalk borer *Sesamia*. Reduction in stalk damage due to calcium silicate treatments expressed as in percent of bored stalks by *Sesamia* spp. was recorded in all three varieties. This reduction was significant in the variety CP69-1062 for all calcium silicate treatments, while significant differences between calcium silicate treatments in IRC99 and SP70 were observed between the control and the highest (800 kg/ha) calcium silicate rate for the percentage of damage stalks.

A significant reduction in the percent of stalks damaged, percent of bored internodes, and tunnel length bored per stalk in susceptible varieties N27 and N35 was reported by Keeping *et al* (2013) when using different sources of calcium silicate under field conditions. Application of silicon can

Table 3 Effects of different silicon treatments on the percentage of stalk damaged (SD), bored internodes (IB), length of borer tunnel (LB), mean number of larvae and pupae of *Sesamia* per 100 stalks (S/100), and exit holes (EH).

Concentration (kg/ha)	Mean±SE				
	SD (%)	IB (%)	LB (mm)	(S/100)	EH
0	41.2±2.89a	6.4±0.83a	106.5±3.84a	37.9±1.89a	1.08±0.09a
400	31.6±1.42b	3.4±0.17b	66.0±1.81b	30.8±1.20b	0.47±0.04b
800	21.2±1.25c	1.6±0.22c	29.8±1.66c	20.8±1.03c	0.15±0.02c
$F_{2, 33}$ p value	25.0 ^{<0.0001}	22.8 ^{<0.0001}	137.3 ^{<0.0001}	36.1 ^{<0.0001}	56.8 ^{<0.0001}

Means followed by the same letter within each column are not significantly different by Turkey's test at $p>0.05$

Table 4 Response of sugarcane varieties to calcium silicate application.

Concentration (kg/ha)	Pol (%)	Brix (%)	Purity (%)	Refined sugar (%)	Weight of one stalk (g)
Variety CP69-1062					
0	15.3±0.17b	17.9±0.18b	85.0±0.25b	9.2±0.11b	598.7±4.26c
400	15.5±0.13b	18.1±0.13b	85.7±0.13b	9.4±0.08b	630.0±3.53b
800	16.4±0.08a	18.9±0.11a	86.9±0.34a	10.1±0.06a	665.0±11.72a
$F_{2,11}$ ^p value	20.60 ^{0.001}	11.22 ^{0.004}	14.77 ^{0.001}	24.89 ^{0.001}	19.58 ^{0.001}
Variety IRC99-01					
0	17.4±0.29a	19.9±0.36a	87.0±0.12b	10.5±0.06b	1006.2±3.14b
400	17.1±0.09a	19.6±0.11a	87.2±0.08ab	10.5±0.03ab	1015.0±5.40b
800	17.9±0.33a	20.5±0.33a	87.6±0.20a	11.1±0.22a	1046.2±10.48a
$F_{2,11}$ ^p value	2.52 ^{0.134}	2.06 ^{0.183}	4.24 ^{0.050}	5.43 ^{0.028}	8.90 ^{0.007}
Variety SP70-1143					
0	16.8±0.17a	19.0±0.11a	88.2±0.39a	10.1±0.13a	730.0±7.90a
400	17.1±0.08a	19.2±0.14a	88.5±0.17a	10.4±0.02a	731.2±3.75a
800	17.1±0.16a	19.2±0.10a	89.0±0.42a	10.5±0.09a	738.7±4.26a
$F_{2,11}$ ^p value	1.22 ^{0.339}	0.99 ^{0.408}	1.25 ^{0.330}	2.95 ^{0.103}	0.709 ^{0.518}

Means followed by the same letter in each column are not significantly different within the same variety using Turkey's test at $p>0.05$

increase resistance against stalk borers due to increased silica concentration in leaf and stalk tissues resulting in more rigid tissues and, consequently, reducing larvae penetration and borer feeding (Keeping *et al* 2009, Reynolds *et al* 2009, White & White 2013).

In a recent study, White & White (2013) found that cumulative length of borer tunneling was reduced by 27% in the variety HoCP96-540 with application of silicon. Our results are in accordance with Keeping *et al* (2013) and White & White (2013), which indicate that silicon can reduce the percent of stalk damage and length of borer tunneling. Likewise, Keeping *et al* (2009) found that application of silicon (calcium silicate) could enhance resistance of susceptible varieties to levels similar to those of resistant varieties without silicon. Our data are in accordance with Keeping *et al* (2009, 2013). The number of moth borer exit holes was significantly reduced in CP69-1062 and SP70-1143 and in the variety IRC99-01. Silicon application generally decreased the number of exit holes, although this reduction was not significant in our study.

Application of silicon in sugarcane can also increase varietal resistance to other pests. Under laboratory conditions, Korndörfer *et al* (2011) found that silicon fertilization reduced the number of sugarcane spittle bug, *Mahanarva fimbriolata* Stål (Hemiptera: Cercopidae) nymphs. They noted that silicon treatment (potassium silicate) reduced longevity of female and male spittlebug on the variety SP80-1816. Adults fed on plants with a high concentration of silicon lived 2 days less compared to those fed on untreated plants.

We conclude that the reduced damage to our three sugarcane-treated varieties may likely have resulted from blocking/delaying the insect penetration or the insufficient

digestibility of silicon-treated sugarcane stalk tissues. The silicon in plant tissue cell walls can affect stalk borer larval feeding (and other chewing and sucking insect pests such as locusts and aphids) at both nutritional and physical levels, affecting the acquisition of required levels of nutrients and water by insects (Panda & Khush 1995, Hou & Han 2010). According to the literature, susceptible sugarcane varieties will likely benefit more than resistant varieties with silicon application (Keeping *et al* 2009, Reynolds *et al* 2009). Indeed, our data indicated that the greatest reduction of damaged stalks, bored internodes, length of larval tunnel, and number of moth borers per 100 stalks due to application of calcium silicate was on CP69-1062, a variety that was the most susceptible among the tested varieties. Therefore, the application of silicon in sugarcane cultivation systems, especially where susceptible varieties are grown and where soils are deficient/low in silicon, may decrease infestation and damage by stalk borers under field condition. This strategy has several advantages such as ease of use, no reported resistance of insect pests to silicon, and adoption of application technology by growers, once silicon becomes a popular tactic in sugarcane best management practice and IPM (Keeping *et al* 2009, Reynolds *et al* 2009, Savant *et al* 1999). Despite our findings that suggest silicon can be used effectively in integrated management of sugarcane stalk borers and yield quality, additional studies are needed to determine the level of silicon application on different soil types and on other commercial varieties.

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