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RESEARCH ARTICLE

Changes in Growth, Yield, Juice Quality and Biochemical Attributes of Sugarcane in Response to Orthosilicic Acid Granules

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Abstract A field experiment was conducted to study the effect of silica granules in the form of orthosilicic acid on growth, yield and juice quality attributes of sugarcane variety CoLk 94184 at IISR experimental farm, Lucknow. The treatments comprised of control (T1), orthosilicic granules @20 (T2), 40 (T3) and 80 kg/ha (T4) and calcium silicate @2 t/ha (T5). Silica application showed higher shoot population, specific leaf weight and total dry matter accumulation relative to control. Cane girth, cane height and cane yield were increased due to silica application. Highest increase in these traits was obtained in T3 treatment. Juice quality attributes, viz. °Brix, sucrose percent juice, juice purity, juice extraction, CCS percent juice, *S*/*R* ratio and SPS activity, were higher in silica-treated plants relative to control. CCS increase was recorded about 15.2–31.8 % over control with highest increase (31.8 %) in T4 treatment. Soluble silica content was significantly higher in leaf and root tissues of treated plants. Findings suggest that application of orthosilicic acid may be beneficial in improving cane yield and juice quality of sugar cane.

Keywords Silica granule · Orthosilicic acid · Sugarcane · Juice quality · Cane yield

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Abbreviations

OSA	Orthosilicic acid
Si	Silicon
CCS	Commercial cane sugar
SAI	Soluble acid invertase
SPS	Sucrose phosphate synthase
HEPES	4-(2-Hydroxyethyl)-1-piperazine-*N*′-2-ethanesulfonic acid
UDPG	Uridine diphosphate glucose
NaOH	Sodium hydroxide
PMSF	Phenylmethylsulfonyl fluoride

Introduction

Silicon is the second most abundant element in the Earth's crust (about 28 % by mass) and usually found in the form of complex silicate minerals and less often as silicon dioxide. It is recently been considered as an essential nutrient for plants (Epstein 2009). Many plant species, especially monocotyledonous angiosperms including rice, sugarcane and some members of the Cyperaceae, have reported beneficial effects of silica on the growth (Epstein 2009; Liang et al. 2007). Plants absorb silicon as monosilicic acid (H_4SiO_4), being proportional to the Si concentration in soil solution (Jones and Handreck 1967; Fox et al. 1967) in monocotyledons. The benefits of Si for sugarcane were realized early in Hawaii in 1965 when field experiments with Si-rich compounds provided highly significant yield increase. It has been reported that under certain conditions sugarcane may absorb more silicon than any other nutrient from the soil. In Puerto Rico, the above ground parts of a 12-month crop contained 379 kg ha^{-1} of

Si, compared with 362 kg ha^{-1} of K and 140 kg ha^{-1} of N (Samuels and Alexander 1969). Silicon (Si) is effective in alleviating both biotic and abiotic stress in most of the crops (Yoshida 1975). Datnoff et al. (2002) suggested that application of Si suppressed brown spot, stem rot, sheath brown rot in rice, and *Fusarium* wilt and corynespora leaf spot on cucumber. Similarly, Cherif et al. (1994) reported increasing resistant to the fungal disease caused by *Pythuim ultimum* and *P. aphanidermatum* in cucumber roots by Si application. Besides its role in improving the resistant of sugarcane against pest and disease, Si has also been reported as an important enzyme regulator in sugar synthesis, storage and retention in sugarcane (Meyer and Keeping 2000).

Sugarcane is a typical Si-accumulating plant which is known to absorb a large amount of silica from the soil. Silicon plays an important role in inducing resistance to various biotic and abiotic stresses in plants, helps in controlling Al, Mn and Fe toxicities, increases P availability, reduces lodging and improves rate of photosynthesis by effective use of sunlight and water. The present investigation was aimed to study the effects of orthosilicic acid granules (0.8 % silica) (OSA) on sugarcane growth, yield and juice quality attributes.

Materials and Methods

Three-bud setts of sugarcane variety CoLk 94184 were planted in the month of October 2013 at IISR farm in the randomized block design (RBD) with three replications. Treatments comprised of control (T1), orthosilicic acid granules @20 (T2), 40 (T3) and 80 kg/ha (T4), and calcium silicate @2 t/ha (T5). Soil of experimental field had pH 7.0, EC 0.16 and OC 0.49 %. Shoot population, plant height, leaf area, specific leaf weight (SLW) and dry matter were determined at grand growth stage, and at ripening, SPS activity and SAI activity were analyzed in cane stalk by following the method of Jain et al. (2013) and Hatch and Glasziou (1963), respectively. At harvesting, cane yield and juice quality attributes were measured.

For dry matter partitioning, plants from each plot were sampled in three replications and oven dried at 80 °C till a constant weight obtained and dry weight was expressed in kg dry weight.

At harvesting, cane yield attributes, viz. cane height, girth and weight, were determined using 5 canes per samples in 3 replications. Number of millable canes and cane yield was recorded by counting and weighing of canes on per plot basis.

Juice quality parameters, viz. °Brix, sucrose percent juice, juice purity, juice extraction percent and CCS percent juice, reducing sugars and *S*/*R* ratio, were determined after cane crushing in the month of December 2014. For sucrose % juice, the fresh cane juice was clarified by adding lead acetate and filtered through Whatman filter paper No 1. Sucrose percent juice was determined in clear juice by automatic saccharimeter (Rudolph Autopol). The amount of reducing sugars in cane juice was determined by Nelson's arsenomolybdate reagent (Nelson 1944). Ratio of sucrose to reducing sugar (*S*/*R* ratio) was calculated and expressed as *S*/*R* ratio. CCS percent juice was determined using formula:

$$\text{CCS\% juice} = (1.022\ \text{Pol\%}\ \ \text{juice} - 0.292\ \text{Brix})$$

% Extraction juice was determined by using the formula:

$$\text{Juice extraction percent} = \frac{\text{Cane}\ \ \text{weight} - \text{juice weight}}{\text{cane weight}} \times 100$$

All the data were determined in three replications.

Enzyme Extraction

For SPS and SAI estimation, middle part of stalk tissues was frozen in liquid nitrogen, ground and solubilized in 100 mM HEPES–NaOH buffer (pH 7.5), containing 10 mM Na–EDTA, 10 mM $MgCl_2$ and 1 mM PMSF. The homogenate was centrifuged at 15,000 rpm for 15 min, and supernatant was collected as enzyme extract. SPS activity was determined in a reaction mixture (1.0 ml) containing 50 mM HEPES–NaOH buffer, pH 7.5, 5 mM $MgCl_2$, 10 mM UDPG, 30 mM glucose-6-phosphate (to maintain equilibrium of glucose in a reaction mixture), and 10 mM fructose-6-phosphate, enzyme extract (0.4 ml) was incubated at 37 °C for 30 min, and 0.1 ml 1 N NaOH was added to stop the reaction (Jain et al. 2013). Un-reacted fructose-6-phosphate was destroyed by heating the reaction mixture tubes in boiling water bath. After cooling, 0.5 ml of 0.1 % (w/v) resorcinol in acetic acid and 3.5 ml HCl (5:1) were added in a suitable aliquot of reaction mixture and incubated at 80 °C for 10 min. The mixtures were then cooled to room temperature, and absorbance was measured at 520 nm wavelength (Roe 1934). Results were expressed as µg sucrose formed per mg protein.

SAI activity was determined in a reaction mixture containing 50 mM Na-citrate, pH 5.0 and 125 mM sucrose in 1 ml final volume. After about 30 min, reaction was stopped by adding 0.1 ml ethanol and 0.1 ml saturated sodium sulfate solution and boiled for 3 min. The amount of hexose sugars produced was determined by Nelson's arsenomolybdate reagent (Nelson 1944). Enzyme activity was expressed in µg reducing sugars formed per mg protein. Protein concentration was determined in enzyme extract by the method of Lowry et al. (1951) using bovine serum albumin as a standard protein.

Soluble Silica Estimation

For soluble silica determination, dry leaf and root samples were predigested in tri-acid mixture containing HNO_3:H_2SO_4:HCl (5:1:2 ratios); 2 ml soluble filtrate was mixed with 1 ml 0.1 N HCl and 2 ml 10 % ammonium molybdate solution. After about 5 min, 1.5 ml 10 % oxalic acid and 2 ml amino-naphthol sulfonic acid were mixed well and measured absorbance after about 10 min at 815 nm using UV–Vis spectrophotometer (Systronics). Results were expressed in mg silica per 100 mg dry weight.

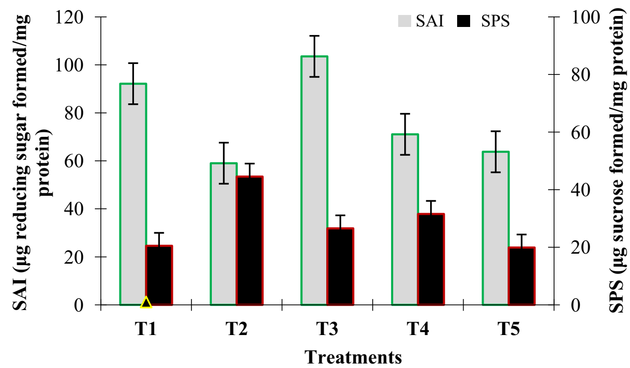

Fig. 1 Effect of orthosilicic acid granule on SPS and SAI activities. *Vertical bars* represent ± SE ($n = 3$)

Results and Discussion

Application of orthosilicic acid granules increased shoot population as compared to untreated control with maximum in T3 treatment (@40 kg/ha) (Table 1). Plant height ranged between 236 and 249 cm in different treatments with maximum in T3 treatment. Yavarzadeh et al. (2008) also reported increase in plant height which may be due to deposition of silica on the plant tissues causing erectness of leaves and stem (Table 1). Leaf area ranged between 396 and 525 cm^2 in different treatments; however, orthosilicic acid application did not show any improvement (Table 1).

Table 1 Effect of orthosilicic acid granules on growth parameters at grand growth phase

Treatment	Plant height (cm)	Leaf area (cm^2)	Shoot population/plot	SLW (gm dry wt/cm^2)
T1	247.0 ± 3.8	524.75 ± 1.48	823 ± 1.73	$9.85 \pm 0.23 \times 10^{-3}$
T2	243.7 ± 1.9	400.50 ± 1.97	918 ± 1.15	$10.9 \pm 0.58 \times 10^{-3}$
T3	233.5 ± 2.3	440.25 ± 1.88	1184 ± 2.31	$10.8 \pm 0.4 \times 10^{-3}$
T4	248.9 ± 2.5	462.20 ± 0.12	927 ± 1.15	$11.4 \pm 0.78 \times 10^{-3}$
T5	236.6 ± 3.6	396.25 ± 0.72	815 ± 1.15	$12.8 \pm 1.2 \times 10^{-3}$

Data are mean of three replicates ± SE ($n = 3$)

Table 2 Effect of orthosilicic acid granules on dry matter accumulation in different plant parts of sugarcane

Treatment	Dry wt. (kg)				
	Stalk	Leaf lamina	Leaf sheath	Root	Whole clump
T1	0.555 ± 0.02	0.126 ± 0.01	0.103 ± 0.004	0.010 ± 0.001	0.894 ± 0.035
T2	0.693 ± 0.01	0.161 ± 0.002	0.105 ± 0.01	0.012 ± 0.001	0.971 ± 0.023
T3	0.852 ± 0.01	0.163 ± 0.005	0.150 ± 0.02	0.017 ± 0.003	1.182 ± 0.038
T4	0.804 ± 0.01	0.135 ± 0.01	0.108 ± 0.001	0.011 ± 0.002	1.058 ± 0.023
T5	0.513 ± 0.02	0.095 ± 0.003	0.058 ± 0.01	0.007 ± 0.001	0.673 ± 0.034

Data are mean of three replicates ± SE ($n = 3$)

Table 3 Effect of orthosilicic acid granule on yield attributes

Treatment	Cane length (cm)	Cane girth (cm)	NMC/plot	Cane yield (kg/plot)
T1	333.4 ± 1.85	3.4 ± 0.12	630 ± 1.45	480 ± 1.15
T2	336.17 ± 1.5	3.5 ± 0.17	706 ± 1.53	528 ± 0.58
T3	354.2 ± 1.22	3.6 ± 0.17	787 ± 0.33	605 ± 0.67
T4	322.0 ± 2.31	3.3 ± 0.12	700 ± 0.88	518 ± 1.76
T5	294.0 ± 1.15	2.9 ± 0.06	620 ± 1.15	458 ± 1.15

Data are mean of three replicates ± SE ($n = 3$)

Table 4 Effect of orthosilicic acid granule on juice quality attributes

Treatment	°Brix	Sucrose % juice	Juice purity (%)	Extraction %	Reducing sugars	*S*/*R* ratio	CCS %
T1	13.31 ± 0.25	10.17 ± 0.02	76.37 ± 0.16	47.62 ± 1.37	0.464 ± 0.03	21.91 ± 1.26	6.51 ± 0.09
T2	14.52 ± 0.25	11.49 ± 0.01	79.16 ± 0.03	48.70 ± 2.19	0.474 ± 0.02	24.23 ± 1.15	7.50 ± 0.08
T3	15.59 ± 1.68	12.75 ± 1.82	80.76 ± 3.72	49.97 ± 0.72	0.483 ± 0.02	26.42 ± 3.83	8.48 ± 2.13
T4	16.03 ± 0.6	12.98 ± 0.04	80.92 ± 0.24	50.77 ± 2.14	0.415 ± 0.03	31.23 ± 2.66	8.58 ± 0.15
T5	15.58 ± 1.83	12.60 ± 1.67	79.99 ± 2.88	39.50 ± 1.37	0.459 ± 0.02	27.46 ± 4.99	8.32 ± 1.76

Data are mean of three replicates ± SE ($n = 3$)

Table 5 Soluble silica content in leaf and root tissues of sugarcane

Treatment	Soluble silica (mg per 100 mg dry weight)	
	Leaf	Root
T1	1.28 ± 0.01	0.36 ± 0.01
T2	4.28 ± 0.04	1.11 ± 0.01
T3	3.51 ± 0.01	0.86 ± 0.02
T4	3.07 ± 0.02	0.78 ± 0.02
T5	2.51 ± 0.02	1.50 ± 0.01

Data are mean of three replicates ± SE ($n = 3$)

Orthosilicic acid granules showed an increase in SLW as compared to control. This may be due to the maintenance of high photosynthetic activity and efficient utilization of light and translocation of assimilated products to sink due to Si application as reported by Rani et al. (1997). Similar results were observed earlier in maize due to Si application (Rohanipoor et al. 2013).

Silica increases the hardness of the crop and helps in accumulation of dry matter. In the present study, silica application improved dry matter accumulation in different plant parts; highest dry weight of stalk, leaf lamina, leaf sheath and root tissues was recorded in T3 (Table 2). Jawahar and Vaiyapuri (2010) also reported higher dry matter accumulation in rice by applying Si in the growing medium.

At harvest, cane girth, cane height and cane yield were recorded highest in T3 followed by T2 (@20 kg/ha) (Table 3). Beneficial effect of Si in improving the growth and yield has been reported earlier in rice (Jawahar et al. 2015), sugarcane (Elawad et al. 1982) and several dicots (Jones and Handreck 1967).

Orthosilicic application showed higher activity of SPS enzyme at ripening phase as compared to control, the highest being recorded in T2 treatment (Fig. 1). In contrast to SPS, SAI activity decreased due to silica application. Silica in the form of sodium metasilicate (SMS) has been found to be strong inhibitor of acid invertase (Rosario and Santisopasri 1977, Alexander 1973, Batta et al. 1991) which helps in improving sucrose content in cane stalk and controlling juice quality attributes at cane maturity.

°Brix, sucrose content, juice purity, juice extraction percent, *S*/*R* ratio and CCS percent were increased due to silica application; these parameters were recorded highest in T4 (@80 kg/ha) treatment (Table 4). CCS increase ranged between 15.2 and 31.8 percent over control due to silica treatment, the highest (31.8 %) being recorded in T4 treatment.

Soluble silica content was significantly higher in orthosilicic acid-treated plants as compared to control, and it ranged between 1.28 and 4.28 % in leaf and 0.36–1.5 % in roots (Table 5). Highest Si content in leaves and roots was found in treatments T2 and T5, respectively. Higher content of silica in leaf tissues of orthosilicic acid treated plants (T2, T3 and T4) as compared to calcium silicate (T5) indicated that orthosilicic (OSA) form of silica is highly soluble and readily transported to leaf site, while calcium silicate (T5) is relatively less soluble showing highest accumulation in root tissues.

The present findings revealed that basal application of orthosilicic acid granules with recommended dose of NPK fertilizer at the time of planting may be useful for improving cane yield and juice quality attributes of sugarcane.

Compliance with Ethical Standards

Conflict of interest The authors declare that they have no conflict of interest.

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