Journal of Experimental Biology and Agricultural Sciences, August - 2018; Volume – 6(4) page 717 – 731

Journal of Experimental Biology and Agricultural Sciences

http://www.jebas.org

ISSN No. 2320 – 8694

EFFECT OF SILICON INTERACTION WITH NUTRIENTS IN RICE

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Received – April 17, 2018; Revision – June 17, 2018; Accepted – July 21, 2018
Available Online – August 10, 2018

DOI: http://dx.doi.org/10.18006/2018.6(4).717.731

KEYWORDS

EDX map

Silicon

Rice

Differential Si uptake

Macronutrients

Micronutrients

ABSTRACT

Present experiment was conducted by using 22 upland and lowland rice varieties grown in nutrient solution with four different concentrations of silicon (Control, 0.5, 1.0, 1.5 and 2.0 mM) for 30 days to observe the variable accumulation rate in Indica rice. Significant differences were observed in relative shoot growth than the relative root growth. The maximum relative shoot growth rates were observed in var. Swarna (203.45%), var. Pratikhya (202.70%), var. Ghanteswari (196.04%) and var. Jyortirmayee (154.55%) at 1.0 to 1.5 mM Si concentration after 20 days of treatment, higher concentration showed pessimistic effect. Relative shoot growth showed positive correlation with silicon accretion and highest accumulation in case of var. Swarna (3.96 mg/g) and var. Ghanteswari (3.79 mg/g) at 1.5 and 1.0 mM Si application, respectively. Among the 22 varieties used, six varieties were selected including 3-upland and 3-lowland on considering their differential Si-uptake potentials and analyzed for nutrient mobilization ability in combination with silicon. The EDX maps showed the different level of element deposition on leaf surface under Si influence in var. Ghanteswari. With the increase in external Si supply in each genotype, the amount of silicon deposited varies, influencing the other nutrient mobilization. Both major (P, K, Ca and Mg) and minor nutrients (Mn, Fe and Cu) showed affirmative correlation with increase in Si concentration. Thus, Si supplementation in rice is a cost effective, sustainable and environmental friendly nutrient management system for enhancing rice yield.

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Peer review under responsibility of Journal of Experimental Biology and Agricultural Sciences.

Production and Hosting by Horizon Publisher India [HPI] (http://www.horizonpublisherindia.in/).

1 Introduction

Rice is the essential staple crop and main source of carbohydrates intake chiefly in Asia and Africa. The highest paddy growing areas are found in Asia. According to FAOSTAT (2016), India occupies second position within the world after china in terms of rice production. But, the yield of rice achieved in India is 3.6 tonnes/ha, whereas in China it is 6.9 tonnes/ha (FAOSTAT, 2016). Existence of such wide yield gap in rice is attributed to several causes including climate change, low mechanization in fields, non-availability of labors at right time, nutrient deficient soils and multitudes of biotic and abiotic stresses (Lobell et al., 2011; DACFW, 2018). The aim of 'National Food Security Mission' is to increase rice production to 10 million tons by productivity enhancement through restoring soil fertility and productivity (DACFW, 2017). However, the improved agricultural practices adopted for exaggerating yield involves the multiple use of chemicals that had resulted into numerous sorts of pollutions causing serious health concern for all living organisms. Therefore, it is essential to think in the direction of sustainable agriculture for productivity enhancement. The utilization of organic sources of nutrients will play a crucial role in achieving sustainable and environment safe agriculture. An economic and efficient soil nutrient management scheme is an indispensable facet to be considered for crop production and liable for enhancing crop productivity. The entire nutrients essential for rice production had been proposed in its fertilization schedule, incorporating all major and minor nutrients. Various studies had indicated that Si accumulated in plants exerts several beneficial effects on plant growth. Silicon fertilizers are additionally recommended for rice cultivation as their accumulation improves cation exchange capacities, water holding capacity, increase plant disease recovery and thus affect yield and productivity. The most noticeable beneficial effect of Si is an enhanced resistance to both biotic and abiotic stresses (Ma & Yamaji, 2006). Silicon is especially important in typical Si-accumulating plants, such as rice (*Oryza sativa*) for healthy growth and high production (Epstein & Bloom, 2005). Rice accumulates Si in the shoots many times higher than N, P, and K (Ma & Takahashi, 2002; Nhan et al., 2012). However, the Si accumulation varies widely among different species. Japonica rice varieties generally showed higher rate of Si deposition than Indica rice varieties as reported earlier (Deren et al., 1992; Ma et al., 2007). Also, Indica rice varieties showed genotypic differences in Si accumulation rate (Swain et al., 2016). Ma et al. (2007) reported that the genotypic difference arise due to variation in abundance of Si transporters in rice roots.

Various studies reported that silicon based fertilization can improve growth and nutrient status of rice. Their application has consistently shown improved utilization of applied fertilizers. Silicon application in field had suggested the exploitation of appropriate mineral nutrition in plants. In 1990, Ma & Takahashi (2002) reported that Si beneficial effects are consistent with both low and high P levels. Rana & Masood (2002) reported that to understand metal interaction with plant, growth conditions and the presence of other ions must be studied. Silicon is well acknowledged to efficiently alleviate various chemical stresses caused by the toxicity of heavy metals like aluminum, cadmium, manganese, and iron. Williams & Vlamis (1957) reported that the Mn toxicity in barley, which can be alleviated by addition of a minute quantity of Si. Similar interaction has been reported in beans (Horst & Marschner, 1978), barley (Horiguchi & Morita, 1987), cowpea (Iwasaki et al., 2002) and cucumber (Shi et al., 2005). Hammond et al (1995) reported that Si can significantly mitigate effects of Al toxicity in barley. Liang et al. (2001) observed the mitigation results due to co-deposition of Al and Si, these results into formation of Si-Al complexes that are insoluble. Such detoxification methods are also adopted in plant system during Cd toxicity in plants in presence of Si (Chen et al., 2000; Liang et al., 2005). Silicon is to enhance nutrient accretions by augmenting root activity (Chen et al., 2011), increasing water uptake (Sonobe et al., 2010), and improving roots hydraulic conductance (Hattori et al., 2008). Ma et al. (2001) reported that Si is the only quasi-essential element, which showed no serious detrimental effects even if it accumulates excessively in plant tissues. However, it increases abrasiveness of foliage which acts as a key element in defense of grasses. Massey & Hartley (2006) reported that higher levels of silica in grasses causes increased abrasiveness of leaves resulting into deterring feeding by voles. As a result the growth rates of juvenile and mature female voles both reduces, which occur due to the decreased absorption of nitrogen from the foliage. Prasad et al. (2005) noted that silicon is a driving element in the evolution of dental adaptations seen in mammalian grazers. They found that vole feeding in grasses further increases the levels of silica in leaves, signifying a dynamic feedback mechanism in between grasses and their herbivores grazers. Therefore, the use of Si in agricultural had become a new trend as for the sustainable crop production strategies (Etesami & Byoung, 2018). The present study is to investigate the effect of differential Si accumulation among the upland and low land Indica rice varieties and their consequence on the nature of uptake and mobilization of other essential nutrients under different concentration of Si.

2 Materials and Methods

2.1 Genotype used

Twenty-two upland and lowland rice varieties were collected from Rice Research Station of Orissa University of Agriculture and Technology, Bhubaneswar, Odisha, India for the differential silicon uptake experiment. Among the twenty two selected verities, sixteen were upland varieties i.e. Annada, Annapurna, Badami, Ghanteswari, Jogesh, Jyotirmayee, Keshri, Khandagiri, Lalat, Lalitagiri, Pathara, Shankar, Subhdra, Udaygiri, Uphar, Varnaporva while the rest six were lowland varieties i.e. Jagbandhu, Manswini, Pratikhya, Rambha, Swarna, Swarna Sub-1.

2.2 Plant materials and growth conditions

Around 200 seeds of each variety were used for experimentation. They were firstly disinfected using mercury chloride (0.1%) for one minute followed by treatment of 0.5% (w/v) fungicide ‘Bavistin’ for 30 min with continuous shaking. The seed were rinsed several times and soaked in deionized water at room temperature for whole day. The experiment was conducted hydroponically using Yoshida’s nutrient solution (Yoshida et al., 1976). After 5-days of germination, 30 uniform seedlings were transferred to 10 cm plastic cups containing 50 ml of Yoshida’s nutrient solution (YNS, pH 5.0). The YNS was renewed every 4 days interval and pH was maintain till the completion of the experiment.

2.3 Silicon uptake treatments

Each plastic cup with 30 seedlings was fortified with four different concentrations of silicon (Si) i.e. 0.5, 1.0, 1.5 and 2.0mM along with control (0mM) in Yoshida solution. *Diatomaceous earth* (80-90% opal, $SiO_2 \cdot nH_2O$), an organic source of Si was used as the source of silicon supplementation. The plants grew in chambers with natural light and 50% humidity. On 5th day the seedlings were transferred to YNS and after establishment on 6th day, the Si supplementation was provided. After 10 and 20 days of treatment, three seedlings from each treatment of each variety were selected at random basis to measure the root and shoot growth. After 30 days of experiment, the samples were harvested and dried further to estimate the silicon content.

2.4 Plant growth study

Growth parameters of all twenty-two varieties in different concentration were recorded and analyzed. The root-shoot growths were measured 10 days after treatment (10 DAT) and 20days after treatment (20 DAT) application. The relative shoot and root growth compared to control conditions was calculated using following formula

$$\text{Relative growth (\%)} = \frac{\text{Length of shoot or root under treatment}}{\text{Length of shoot or root under Control}} \times 100$$

2.5 Estimation of silicon content

After 30 days grown in hydroponic culture, samples of different varieties of each treatment were collected and dried in oven at 70˚C for three days and powered for Si-estimation. For Si estimation, the samples were first digested and measured the Si content colorimetrically. Tissue analyses was done through digestion of powdered plant material (0.1g) by autoclaved-induced alkali digestion to release the various forms of silicon (mono and poly silicates) and quantified through colorimetric molybdenum blue method (Wei-min et al., 2005). About 1ml of digested sample was transferred further into a 50ml polyethylene tube with addition of 30ml of 20% acetic acid and 10ml ammonium molybdate (54 g/l, pH 7.0). Further, the solution was shaken thoroughly and kept for 5 min. and then addition 5ml of 20% tartaric acid and 1ml reducing solution. The reducing solution consisted of solution A (2g of Na_2SO_3 and 0.4 g 1-amino-2-naphthol-4-sulfonic acid in 25ml of doubled distilled water) and solution B (25 g of $NaHSO_3$ in 200 ml of doubled distilled water), and finally adjusted to 250ml with doubled distilled water. The volume was made up to 50ml with 20% acetic acid. The absorbance was taken at 650 nm under UV/Vis-Spectrophotometer (LAMBDA 365, Perkinelmer) after 30 min. The silicon was calculated with the help of standard calibration curve.

2.6 Atomic Absorption Spectroscopic (AAS) analysis

The dried ground plant material (0.5g) was digested with di-acid mixture of nitric acid and sulphuric acid used in 4:1 ratio. The samples were digested over a hot plate at 80℃ for 1 hour. After the acid digestion, the samples ash were cooled to room temperature and scrapped with spatula dissolved in 5ml double distilled water. The 5ml mixture was filtered using Whatman filter paper and an aliquot was diluted to be used in AAS analysis. The rice samples were analyzed for P, K, Ca, Mg, Fe, Mn and Cu concentration using atomic absorption spectrophotometer (iCE™ 3300 AAS. Thermofisher, Germany).

2.7 Energy Dispersive X-Ray Spectroscopy (EDX)

Scanning electron microscopy (SEM) coupled to EDX Spectroscopy was used for characterizing and mapping the relative elemental component in leaf surface of rice. The leaves were cut into small square piece (1 cm^2) and placed exposing the abaxial surface of leaf on stubs sprayed with thin layer (1.5–3.0 nm) of gold coating and system operation at voltage of 20 KV (Ensikat et al., 2010). SEM-EDX micrographs were obtained using Field Emission Gun-SEM (JEOL JSM-7600F FEG-SEM).

Table 1 Germination percentage and length of 5-day old vigorous seedling used in Si uptake screening experiment.

Varieties used	Germination percentage (%)	Seedling length (Mean±SD)	
		Shoot (cm)	Root (cm)
Annada	*80*	2.8±0.7	1.5±1.1
Annapurna	*95*	1.3±0.1	0.7±0.5
Badami	*100*	3.1±0.1	1.4±0.1
Ghanteswari	*100*	3.3±0.1	2.4±0.6
Jagbandhu	*95*	2.9±0.3	1.9±1.2
Jogesh	*95*	2.1±0.1	1.6±0.7
Jyotirmayee	*100*	1.4±.05	0.9±0.3
Keshri	*100*	3.9±.04	2.9±0.8
Khandagiri	*85*	1.6±.07	2.0±1.4
Lalat	*90*	2.8±0.3	1.5±0.4
Lalitagiri	*95*	1.5±0.1	0.9±0.3
Manswini	*90*	2.3±0.1	2.4±1.3
Pathara	*95*	2.3±0.1	2.4±0.7
Pratikhya	*100*	4.1±0.6	2.7±1.1
Rambha	*85*	2.2±1.4	1.1±0.9
Shankar	*90*	3.3±0.4	1.3±0.2
Subhdra	*95*	1.7±0.1	1.3±0.5
Swarna	*100*	3.1±0.4	2.5±0.7
Swarna Sub-1	*100*	3.1±0.3	3.1±0.4
Udaygiri	*95*	1.9±0.1	1.2±0.6
Uphar	*90*	3.4±0.6	3.6±0.9
Varnaporva	*100*	1.5±0.9	1.4±0.9

Data are means of three replicates ± SD

2.8 Statistical analysis

All analyses were carried out in triplicates. The experiment was set up in a randomized block design (RBD) and the experimental data obtained were subjected to statistical analysis by adopting Fisher's method for analysis of variance. The level of significance used in 'F' test and the Critical difference (CD) values calculated were at 0.05 significance level ($p<0.05$). All graphs were presented with relative standard deviation as error bars.

3 Result and Discussion

Rice is a classic example of Si-accumulating species as they can accumulate Si up to 10% of shoot dry weight. Tamai & Ma (2003) compared the whole gramineous species including wheat, triticale, sorghum, rye, maize and barley for their Si-uptake ability during a period of 24 hours under the same condition and observed that the Si uptake by rice roots was much higher than any other gramineous species. However, silicon content in the plant varies greatly among the plant species. Genotypic variations and its understanding are critical as it alters the quantity of Si accumulated. The alteration in Si accumulation indirectly affects the degree of resistance towards chemical stresses and adversely affects nutrient mobilization in crop species. Ma et al. (2007) reported that the presence of two Si transporter genes naming *Low silicon rice 1* and *2* (*Lsi1* and *Lsi2*) expressed highly in case of both japonica and indica species. They also highlighted the variation in their abundance as the core reason of genetic variation in rice species. Among the 22 studied genotypes, the germination ability, root and shoot growth were found to be varied in different concentration of Si (Table 1-3). Vigorously growing 5-days old germinated seedlings of each variety were used for the study. All of the indicia genotypes showed higher relative growth with increase in external Si supplementation, primarily in the concentration range of 1.0-1.5 mM, signifying the optimum Si concentration for the growth of Indica rice genotypes. Above 1.5mM Si, some of the tested genotypes exhibited depressing growth rate. Significant difference was recorded amongst the shoot growth rate of each cultivar with change in amount of Si added at 10days after treatment (Table 2). A prominent relative growth change was observed in shoot growth, but roots don't signify much variation. The maximum relative shoot growth was represented by the vars. Ghanteswari, Jyortirmayee, Pratikhya and Swarna after 10-days treatment. Other varieties like Annada, Annapurna, Keshri, Lalat, Lalitagiri, Manswini, Pathara, Rambha, Shankar, Swarna Sub-1, Uphar showed modest growth followed by Badami, Jagbandhu, Jogesh, Khandagiri, Shubdhra, Udaygiri and Varnaporva verities with lowest relative growth rate. But, after 20 days treatment of silicon (Table 3), both relative shoot growth and root growth rate compared to control were significantly different. Among all varieties used, variety Ghanteswari, Pratikhya and Swarna continued to show higher relative shoot growth rate, while other variety Badami, Jagbandhu, Jogesh, Khandagiri and Shubdhra sustained as low relative growth varieties. The remaining varieties were showing moderate growth rates under varying Si concentration. It was difficult to categorize all these varieties into high, moderate and

Table 2 Effect of different silicon treatments on relative growth percentage of shoot and root in comparison to control seedlings after 10-days of treatment.

	Relative Shoot Growth (%)					Relative Root Growth (%)				
	Different Concentration Of Silicon (mM)									
`	0.0	0.5	1.0	1.5	2.0	0.0	0.5	1.0	1.5	2.0
Annada	100	106.94	108.33	108.33	105.56	100	123.93	105.98	108.55	94.02
Annapurna	100	96.67	97.14	104.29	106.19	100	89.92	126.05	134.45	118.49
Badami	100	97.46	72.08	88.83	91.37	100	95.15	100.97	99.03	124.27
Ghanteswari	100	140.98	190.16	137.70	131.15	100	100.93	96.26	79.44	102.80
Jagbandhu	100	109.09	99.02	105.65	73.71	100	138.62	364.14	115.86	80.69
Jogesh	100	95.08	93.44	88.52	95.90	100	125.25	95.96	88.89	143.43
Jyotirmayee	100	146.77	203.23	169.35	158.06	100	96.67	74.67	90.67	110.00
Keshri	100	108.70	114.49	102.90	113.04	100	96.30	93.33	74.81	75.56
Khandagiri	100	102.19	96.61	92.43	106.97	100	90.00	127.50	119.17	103.33
Lalat	100	125.61	134.15	157.01	138.72	100	143.36	92.92	70.80	118.58
Lalitagiri	100	100.79	111.11	120.63	95.24	100	66.67	73.33	73.33	82.00
Manswini	100	109.59	121.92	104.11	101.37	100	109.91	123.42	101.80	112.61
Pathara	100	100.53	106.84	135.00	120.53	100	128.04	122.43	122.43	130.84
Pratikhya	100	185.45	214.08	199.53	205.16	100	141.13	141.13	536.29	128.23
Rambha	100	110.20	114.73	111.05	133.43	100	104.17	95.83	114.58	110.42
Shankar	100	103.03	104.55	107.58	103.03	100	97.75	95.51	88.76	105.62
Subhdra	100	90.46	85.26	70.81	72.25	100	94.90	132.65	75.51	118.37
Swarna	100	153.75	170.00	197.50	172.50	100	166.95	135.59	194.07	124.58
Swarna Sub1	100	116.60	110.12	117.41	124.70	100	118.54	115.89	125.83	99.34
Udaygiri	100	114.04	69.30	92.11	86.84	100	88.97	73.53	59.56	77.21
Uphar	100	108.74	110.68	89.32	104.85	100	101.14	99.43	76.00	93.71
Varnaporva	100	86.76	81.09	125.06	72.10	100	135.40	97.35	134.51	116.81
ANOVA results Not Significantly different (*) Significantly different (**)		Varieties** F-value=3.9 CD = 11.0 CV = 16.38	Treatments** F-value=9.2				Varieties* F-value= 0.6	Treatments* F-value=1.6		

[Note- Data was subjected to two ways ANOVA at *p*<0.05 and results are presented above with Critical Difference (CD) and Coefficient of Variation (CV).]

Table 3 Effect of different silicon treatments on mean relative growth percentage of shoot as well as root in comparison to control seedlings after 20-days of treatment.

Varieties used	Relative Shoot Growth (%)					Relative Root Growth (%)				
	Different Concentration Of Silicon (mM)									
	0.0	0.5	1.0	1.5	2.0	0.0	0.5	1.0	1.5	2.0
Annada	100	104.02	111.11	111.82	102.84	100	109.24	87.39	115.97	112.61
Annapurna	100	106.91	120.21	123.40	140.96	100	129.17	130.00	95.00	120.00
Badami	100	100.66	93.61	84.36	79.74	100	100.00	102.61	92.17	93.91
Ghanteswari	100	141.58	196.04	161.39	145.54	100	109.71	100.00	114.56	135.92
Jagbandhu	100	88.13	88.57	91.21	91.21	100	101.94	82.52	111.65	92.23
Jogesh	100	94.35	110.00	85.87	91.30	100	130.14	136.99	127.40	123.29
Jyotirmayee	100	118.18	154.55	126.45	117.63	100	109.23	90.77	98.46	76.15
Keshri	100	138.21	90.79	95.66	87.53	100	96.10	91.56	77.92	72.08
Khandagiri	100	93.55	99.46	84.05	85.84	100	91.89	132.43	71.17	84.68
Lalat	100	106.57	114.58	114.99	109.86	100	85.11	106.38	106.38	108.51
Lalitagiri	100	84.50	95.94	91.33	91.88	100	80.65	70.97	64.52	59.68
Manswini	100	100.55	106.56	98.36	115.03	100	63.38	71.13	80.99	105.63
Pathara	100	94.40	94.89	110.22	106.81	100	119.54	101.15	147.13	131.03
Pratikhya	100	155.86	202.70	170.27	151.35	100	164.00	146.67	160.00	156.00
Rambha	100	97.23	77.85	96.37	105.02	100	117.27	110.91	102.73	112.73
Shankar	100	102.43	109.19	109.46	104.86	100	92.86	71.43	76.43	62.14
Subhdra	100	104.28	117.12	97.52	92.57	100	123.53	112.75	92.16	85.29
Swarna	100	130.50	185.60	203.45	168.97	100	110.00	116.00	95.00	98.00
Swarna Sub1	100	100.57	115.91	117.61	137.50	100	125.00	109.26	137.96	104.63
Udaygiri	100	89.60	113.33	110.13	114.00	100	89.38	88.50	93.81	124.78
Uphar	100	105.87	114.18	107.09	102.44	100	100.91	104.55	103.64	120.91
Varnaporva	100	101.18	101.18	113.73	93.14	100	102.56	76.92	84.62	62.39
ANOVA results Not Significantly different (*) Significantly different (**)	Varieties** F-value=6.9 Treatments** F-value=3.9 CD = 9.6 CV = 15.67					Varieties* F-value= 0.6 Treatments** F-value=5.5 CD = 9.1 CV = 14.7				

[Note- Data was subjected to two ways ANOVA at $p<0.05$ and results are presented above with Critical Difference (CD) and Coefficient of Variation (CV).]

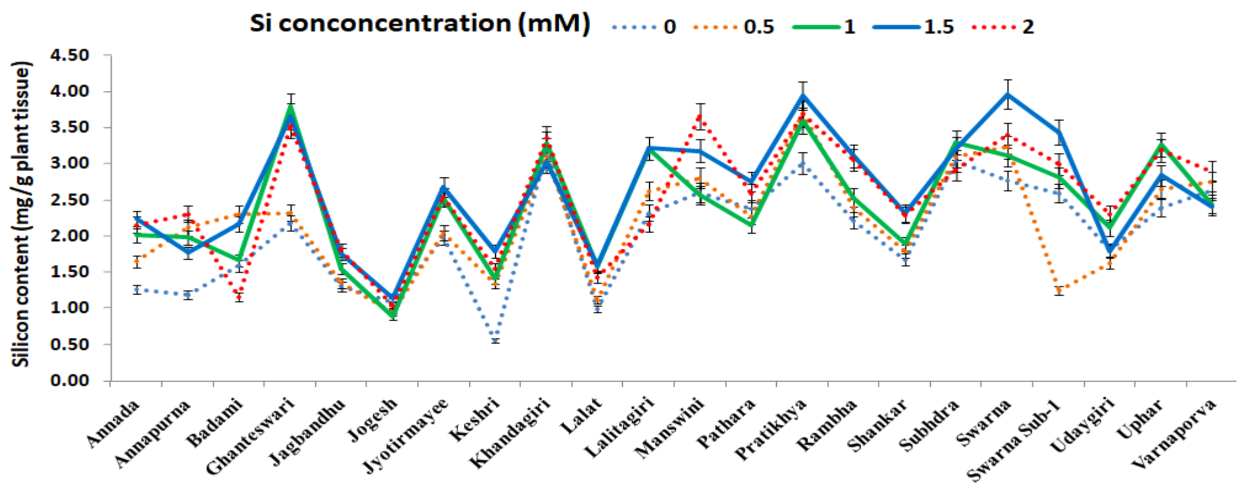

Figure 1 Impact of different silicon treatment on mean silicon accumulation of 22 varieties of indica rice represented as mg Si/g plant tissue. Data are means ± RSD (relative standard deviation, n=3) as error bars. Data showed significant differences between treatments $p < 0.05$ according to Fisher's protected LSD test (p-value= <.001, LSD=4.94).

low uptake groups based on only relative shoot growth rate. So, the amount of Si accumulated was also quantified to get a clear idea of the nature of uptake of these varieties as positive correlation was expected between relative growth rate and Si deposition. Each treatment harvested was estimated for its Si content by blue ammonium molybdate colorimetric method. Wei-min et al. (2005) proposed a reliable method for rapid determination of Si content in rice using a RIL population (cross Zhenshan 97B / Milyang 46) by adopting high-temperature alkaline fusion method for digestion. A linear and positive trend was obtained during silicon estimation study, which was similar to the trend obtained in relative shoot growth rate study. The highest Si deposition was obtained in 1.0-1.5mM Si treatments which are parallel to the pattern of maximum relative growth rate (Figure1). The present study specifies and confirms a direct positive correlation between the silicon deposition and relative growth in rice. But, in few varieties such as Annapurna, Khandagiri, Manaswini, Udaygiri, and Varnaporva even showed higher silicon accumulation till 2 mM external Si supply. Hence, by considering both relative shoot growth rate and amount of silicon accumulated by the each variety, they are categorized as into three groups and designated as- 'high-accumulators', 'moderate-accumulators' and 'low-accumulators' of silicon (Table 4).

Nutrient management in rice is an essential criterion for improving the production and productivity of crop. Tamai & Ma (2008) reported the effects of silicon on rice growth and production under field conditions by developing a low silicon mutant. They observed that at a later growth stage of rice, the wild type showed slightly higher plant height and shoot dry weight than the mutant one. They estimated that the silicon deposited in the shoots during harvest of both the wild type and mutant and reported as 4.14% and 0.22% of the dry weight basis, respectively. They suggested that's why the plant height and shoot dry weight was increased in mutant. They also explained that beneficial effects of silicon are underestimated in most studies as the effect of Si on plant growth are more pronounced under stress conditions and is much emphasized, while its effects

Table 4 Based on the data of mean relative shoot growth percentage (Table 2 & 3) and mean silicon accumulation (Figure-1) in 22 rice varieties, they are categorized into three silicon differential accumulators group. Two varieties (one upland and one lowland) were selected from each group for further nutrient load analysis.

Si-differential accumulators	Varieties
High Si-accumulators	var. Ghanteswari, var. Swarna, var. Swarna Sub-1
Moderate Si-accumulators	var. Annada, var. Annapurna, var. Jyotirmayee, var. Keshri, var. Lalat, var. Lalitagiri, var. Manswini, var. Pathara, var. Pratikhya, var. Udaygiri, var. Uphar
Low Si-accumulators	var. Badami, var. Jagbandhu, var. Jogesh, var. Khandagiri, var. Rambha, var. Shankar, var. Shubdhra, var. Varnaporva

were left unnoticeable under non-stressed conditions. Cuong et al. (2017) observed that optimum dose of silicon fertilizer (329 Kg/hm^2 SiO_2) can maximize the grain yield 3716 Kg/hm^2 and enhanced the nutrient uptake of N, P, K and Si significantly. Si build up in leaf surface results into increased shoot-root biomass, reduced transpiration, increased chlorophyll synthesis, maintain homeostasis and osmoregulation inside cells ultimately influencing photosynthesis and plant growth and production. Therefore, essentially to understand the effect and consequence of genotypic differential Si-accumulation on the mobilization of other nutrient ions in rice, 22 varieties were categorized into three groups as high, medium and low accumulators. One variety of each group, (Table 4) one belonging to upland and other from lowland Indica rice were selected as representative of group and further experimented for studying the interaction of silicon and mobilization of macronutrients (P, K, Ca and Mg) and micronutrients (Fe, Mn and Cu). The three upland varieties viz., Ghanteswari (High-accumulator), Jyotirymee (Moderate-accumulator), Badami (Low-accumulator) and three lowland varieties viz., Swarna (High-accumulator), Pratikhya (Moderate-accumulator) and Jagbandhu (Low-accumulator) were used for Si and other nutrient interaction study. The nutrient mobilization trend of micro- and macronutrients were studied in 'basic nutrient solution (Yoshida, pH=5)' under varying silicon concentration in comparison to control. The other nutrients were quantified through the Atomic absorption spectroscopy (AAS) technique widely used for determining traces of metals in any decomposed or hydrolyzed solid sample. The hydrolyzed solid sample is converted in aqueous aliquot and diluted to aspirate the metal analyte into an air-acetylene flame, causing evaporation of the solvent and vaporization of the free metal atoms.

The first macronutrient focused was Phosphorous (P), which is an important nutrient necessary for plant growth and can limit rice productivity due to its low availability in soil. Phosphorous amount was estimated through atomic absorption spectrophotometer (AAS) in all the 30 samples of six varieties with different Si concentration (i.e. five treatments of each variety). Based on quantification results (Figure-2a), it was observed that phosphorous has optimistic association with the increase in Si concentration. The high and moderate accumulator's showed increase in P uptake among the tested varieties, Ghanteswari and Pratikhya showed improved uptake till 1.0mM concentration of Si and subsequently declined. Whereas, variety Swarna and Jyotirmayee showed continuous higher uptake even up to 2.0mM external application silicon. However, the low accumulator varieties such as 'Badami' and 'Jagbandhu' did not show any regular trend of P deposition. The present study showed that there was a genotypic differential for Si- accumulation in rice varieties used. Ma & Takahashi (1990) reported that Si fertilizer can improve plant P utilization by escalating both P content of rice and enhancing phosphate fertilizer efficiency, but the study

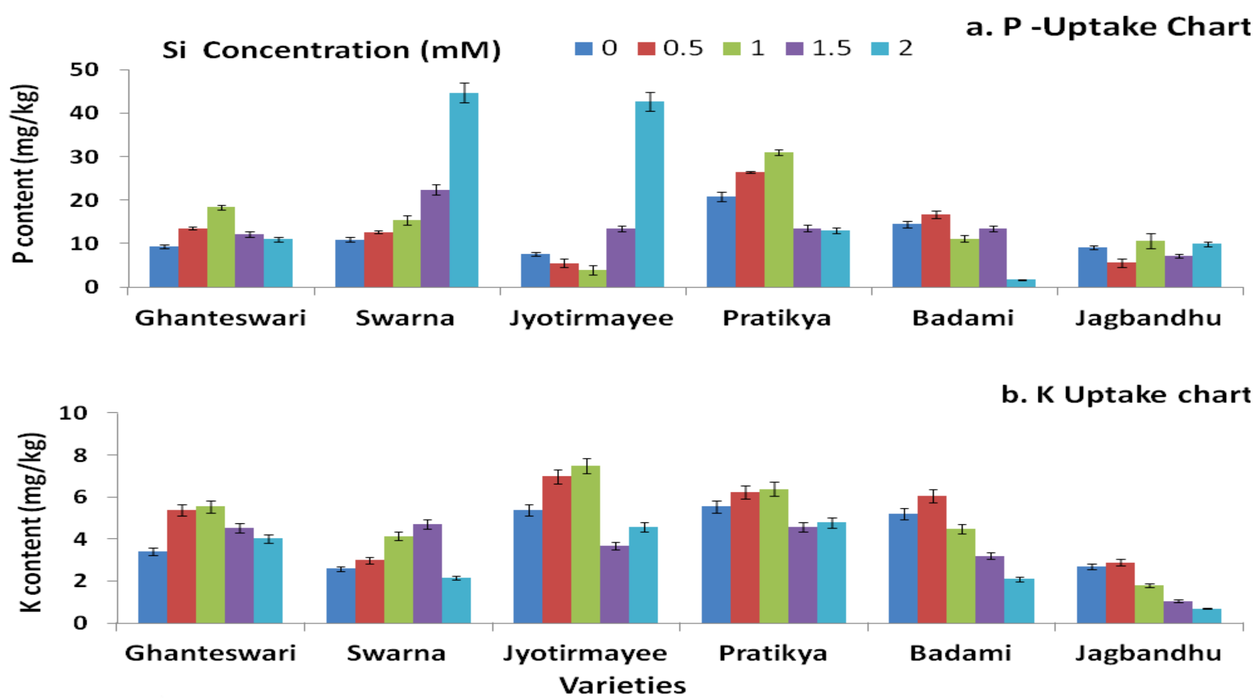

Figure 2 Effect of different silicon treatment on nutrient load of macronutrient Phosphorous (P) and Potassium (K) (expressed in mg/kg) in 6 selected Si-differential accumulator varieties. Data are means ± RSD (relative standard deviation, n=3) as error bars. Results for P were not significantly different and for K results were significantly different between treatments at $p < 0.05$ (=0.0034) according to Fisher's protected LSD test.

was conducted in different P levels in nutrient media. Agostinho et al. (2017) reported that there was significant variation of rice grain yield and Si accumulation on application of varying P-concentration through pot culture experiments. However, they proposed that P and Si in the soil can creates a synergistic effect on soil having Al, Mn, and As ($P < 0.01$), but not in growth of plant and P uptake.

The present study also emphases on the effect of genotypic differential uptake of silicon in presence of potassium (K), which is essential for maintenance of osmotic pressure and homeostatic of plant cell. In case of K, all the six varieties showed augmented accumulation of K with the increase in Si concentration (Figure 2b). The result also suggests that increment in K uptake was supported only up to optimum level of Si (1.0 – 1.5 mM) in plant tissue, after which it declined**.** Hence, it is opinion that Si show affirmative correlation with enhancement in the K uptake in rice. Chen et al. (2016) applied Si to K-deficient (0.5 Mm) sorghum seedlings for 15 days and found that K^+ content in xylem sap was significantly increased and osmotic potential of xylem sap was decreased. They explained that the major reason for Si-induced improvement was enhanced K^+ concentration in xylem sap, which increases the osmotic gradient and hence xylem hydraulic conductance. However, they reported no effect on sorghum growth by Si application (1 mM) under control conditions. Also, the growth and water status were reduced by K-deficient stress, but Si suppementation significantly alleviated these decreases in parameters.

In another experiment, the addition of Ca with silicon showed a variation with regard to upland and lowland varieties (Figure 3a). In case of Ca interaction with silicon, the upland varieties Ghanteswari, Jyotirmayee and Badami showed positive link with silicon accumulation. The increment in Ca uptake can be seen up to 1.0mM Si in all the genotypes, while in variety Jyotirmayee further enhanced uptake up to 2.0mM silicon application was reported. But, the lowland varieties like Swarna, Pratikhya, and Jagbandhu showed a disturbed Ca uptake trend with increase in Si concentration. Calcium uptake trend seems to be negatively correlated with Si uptake. The impact of silicon on growth and nutrient status of rice plants grown under different concentrations of zinc regimes has been reported (Mehrabanjoubani, 2014). They reported that silicon application increased with Ca^{2+}, K^+, P and B contents in plants under 50 µg/l Zn. Marxen et al. (2016) reported that the total uptake of P was increased and decreased of Ca due to the high Si application in rice. The present study showed the contradictory trend among the lowland and upland varieties and suggesting the variation in the need of nutrient in the rice crop based on their growth conditions. Quiet similar interaction pattern (Figure 3b) was observed in case of Magnesium (Mg), i.e. the upland varieties showed increased Mg uptake under low levels of silicon application up to 1.0mM. Above 1.0mM silicon application, the lowland varieties showed continuous decrease of Mg deposition. Therefore, it is concluded that higher silicon deposition in rice has a negative effect on Mg uptake. Further, Korkmaz et al. (2018) studied the effect of different concentration of Si (0, 0.5, 1.0, 2.0 mM) on tomato plant

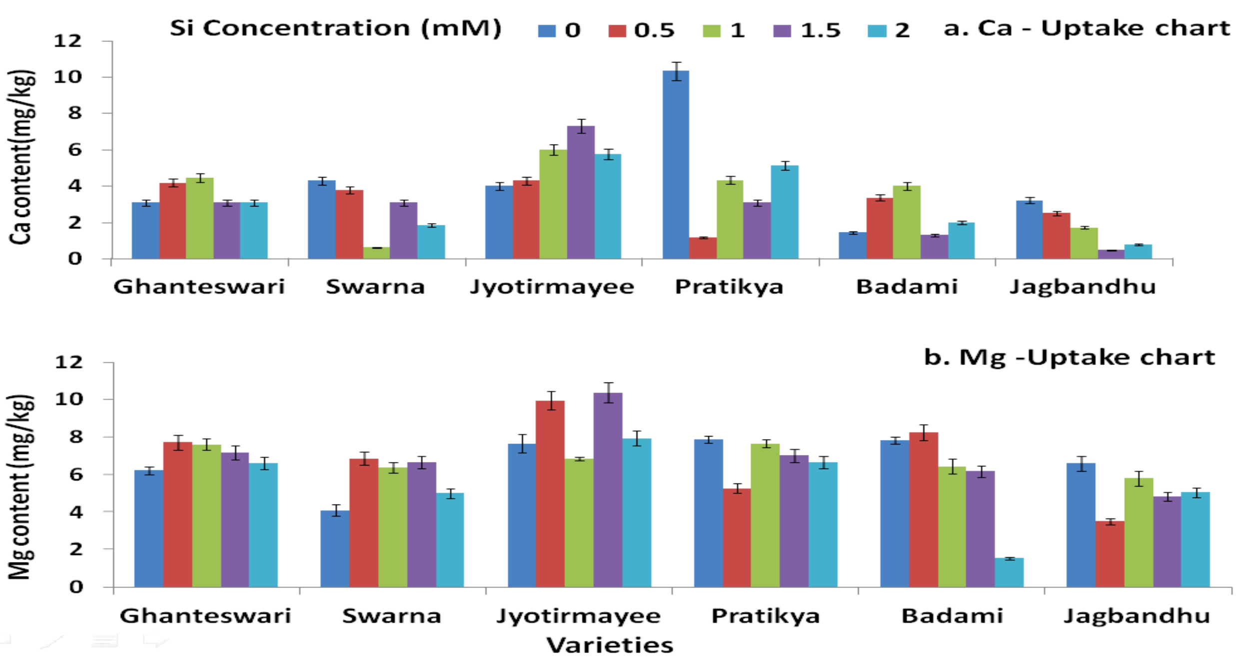

Figure 3 Effect of different silicon treatment on nutrient load of macronutrient calcium (Ca) and Magnesium (Mg) (expressed in mg/kg) in 6 selected Si-differential accumulator varieties. Data are means ± RSD (relative standard deviation, n=3) as error bars. Results for both Ca and Mg were not significant different between treatments, but were significantly different between varieties at $p < 0.05$ according to Fisher's protected LSD test.

under salt stress and reported no significant (p>0.05) change in Ca and Mg concentration in leaves. However, Horuz & Korkmaz (2014) previously reported that Si fertilization improved the ratios of K/Na, Ca/Na, Mg/Na and P/Na in rice under salt stress and also that it decreased the damage caused salt.

Micronutrients are also essentially important in nutrient management system as involved in many biological processes as catalyst. They are not generally incorporated in fertilization programs, but crops even have prescribed recommendation for micronutrients. Silicon was not considered as essential nutrient in plants, but performs several beneficial activities in its accumulating species. The present study was to know the interaction of silicon with microcronutrient like manganese (Mn), iron (Fe) and copper (Cu). The result showed that silicon accumulation was optimum and positive association with all the micronutrients by encouraging there utilization (Figure 4a). However, in case of Fe the trend differs with different Si accumulating group of rice (Figure 4b). The high and moderate Si accumulators showed better and enhanced utilization of Fe with increasing silicon deposition. Furthermore, the upland varieties showed increment up to optimum Si (1.0 – 1.5 mM) deposition, while the lowland varieties Swarna and Pratikya showed continuous increased uptake with silicon supplementation. The low accumulator of both

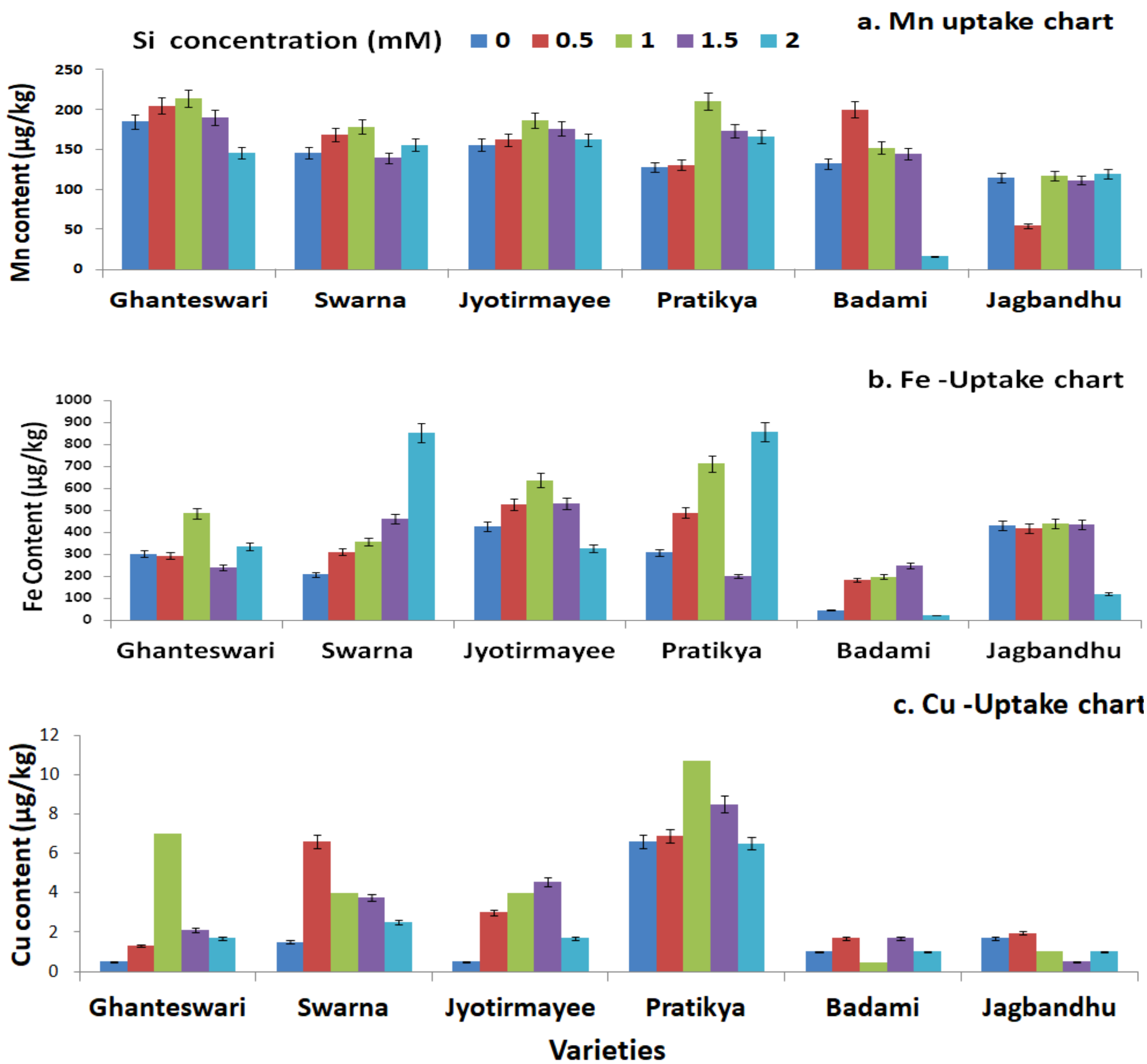

Figure 4 Effect of different silicon treatment on nutrient mobilization of micronutrient manganese (Mn), iron (Fe) and copper (Cu) (expressed in µg/kg) in 6 selected Si-differential accumulator varieties. Data are means ± RSD (relative standard deviation, n=3) as error bars. Results for all Mn, Fe and Cu were significantly different between varieties at $p < 0.05$ according to Fisher's protected LSD test.

upland (Badami) and lowland (Jagabandhu) varieties were not able to utilize Fe better from the nutrient media. Finally, the Cu interaction with silicon was observed and presented in Figure-4c and found close similarity with Fe. It was also reported that high and moderate Si accumulator varieties showed better mobilization of Cu from nutrient solution. But, the low or poor silicon accumulator fails to deposit silicon in their cells and thus show very low rate of uptake of nutrients from media. Overall all micronutrients i.e. Mn, Fe and Cu have enriched in plant tissue and establish a favorable link with Si. Majority of silicon and micronutrient interaction studies in rice were concentrated on the silicon induced alleviation effects of micronutrient and heavy metal toxicity (Mn, Fe, Cu, Al, Zn, Cd, Pb, As, etc.). All these studies reported the decrease in level of toxic micronutrient ions in their rich supply environment. Hernandez-Apaolaza (2014) studied the physiological role of Si in micronutrients deficiency in cucumber under hydroponic system. He explained that the Si supply mitigates the symptoms of Fe deficiency under Fe-deficient condition, but this effect was not clear under Zn- or Mn-deficient conditions. He suggested that the Si actually interact with the metal distribution in the plant which results into diminishing the damage. Carrasco-Gil et al. (2018) reported that under Fe sufficiency conditions, Si supply increased in Fe root plaque formation. So, Fe acquisition strategies got activated and subsequently the rate of Fe translocation to the aerial parts. They also reported that under Fe deficiency, Si-accumulator plants absorbed Fe from the plaque more rapidly than Si –accumulator plants. The present study represented the EDX maps of the nutrients present on the leaf surface (Figure-5). EDX map was developed to identify the change in accumulation of different macronutrient and micronutrient in leaf under silicon influence. It can't provide the clear idea of the rate of accumulation, still will be useful in providing the relative evidence of nutrient deposition. It was observed that the variety Ghanteswari grown in nutrient solution with or without Si showed differential accumulation of mineral with significant variation (Figure 6 & 7). The elemental spectrum of leaf surface was also presented in Figure-5, depicting the quantity and presence of other nutrients. With increase in the external application of Si, the amount of silicon deposition was more in leaf tissues and also influencing the other enhanced uptake of nutrients like K and Ca (Figure-5). The plant grown without silicon showed concentration of carbon backbone as 42.53% and silicon concentration as 0.41%. In plant grown with the addition of 1.0 mM silicon supplementation, the concentration of carbon backbone was found to be 38.69% and silicon concentration as 11.8%. Thus, we could conclude that the silicon contribute to the structural formation of leaf tissue providing strength, protection against penetration from pest and grazing animals.

In conclusion, considering the various studies conducted the present study concluded that 'silicon act differently based on the growth conditions of rice' (Figure 8). Under metal toxicity (Al, Cd, As, Mn, Fe, Zn, etc.) conditions in rice, silicon causes mitigation of harmful toxicity effects of metals. It basically alleviates the uptake of harmful metal ions and thus reduces the oxidative stress caused by them. Sometimes essential macronutrients such as P and N are applied in excess to the rice, resulting into negative consequences on growth and productivity. Silicon can also provide advantageous outcomes under such stress situations. It allow lower uptake of excess P and N and improve their utilization all along the whole rice plant. Thus reducing the injurious effects caused by excess fertilizers applications. All of these effects of silicon application under stress are enhanced in case of 'high Si-accumulator varieties', whereas lowered if it's a 'low Si-accumulator'. While under unstressed conditions the effects of silicon supplementation is not much pronounced, however showed positive results in rice nutrient mobilization. The nutrient such as P and K showed higher uptake and better mobilization inside rice plant with silicon supplementation. Other essential macronutrients such as Ca and Mg also showed higher uptake, but results varies with the nature of rice. In case of the lowland varieties, Ca accumulation is lower with enhanced Si uptake. Opposite the above results, in upland varieties higher accumulation of Ca occurs with improved Si uptake. Micronutrient also reported to demonstrate better utilization and uptake under higher silicon accumulation. But, all these characters are highly influence by the genotypic differential silicon uptake nature of rice. More than the stress conditions, in unstressed state the genotypic differential Si-uptake nature are essential to considered. The differential nature of rice decides the modification in rate of higher nutrient mobilization properties. As the property depends on rate of silicon accumulation, the higher the Si-uptake potential better is the nutrient mobilization property of a variety. Low silicon accumulating varieties showed unpredicted results in most of the cases even in presence of high silicon application. This study focused on identifying the relationship of genotypic difference in Si-uptake and its interaction with other nutrient in rice. Therefore, the nutrient absorption capacity of plant can be improved further and the nutrient deficient soil can also be effectively used for enhancing the productivity of rice.

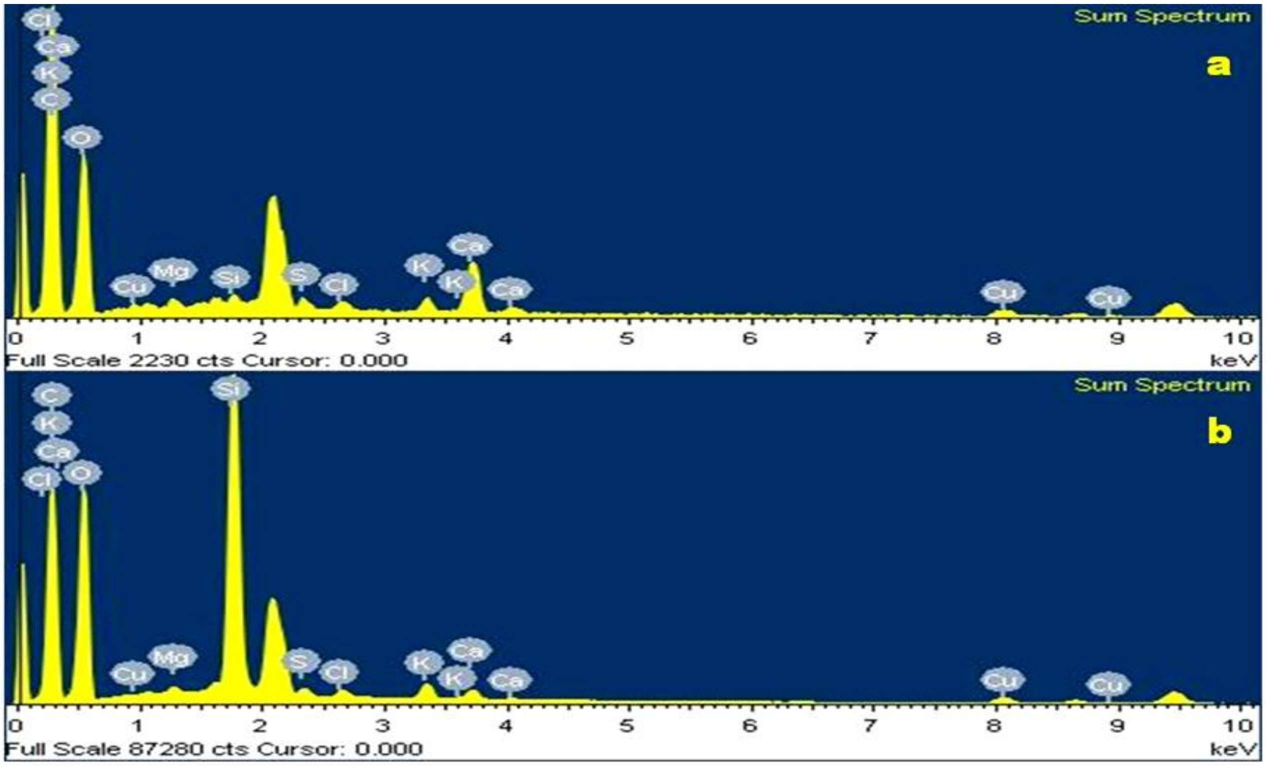

Figure 5. EDX Spectrum obtaied during EDX analysis of abaxial leaf surface of va. Ghanteshwari grown in **a)** –**Si** nutrient solution and **b)** +**Si** (1.0mM) nutrient solution at 20KV voltage detecting the distribution of different elements n leaf surface.

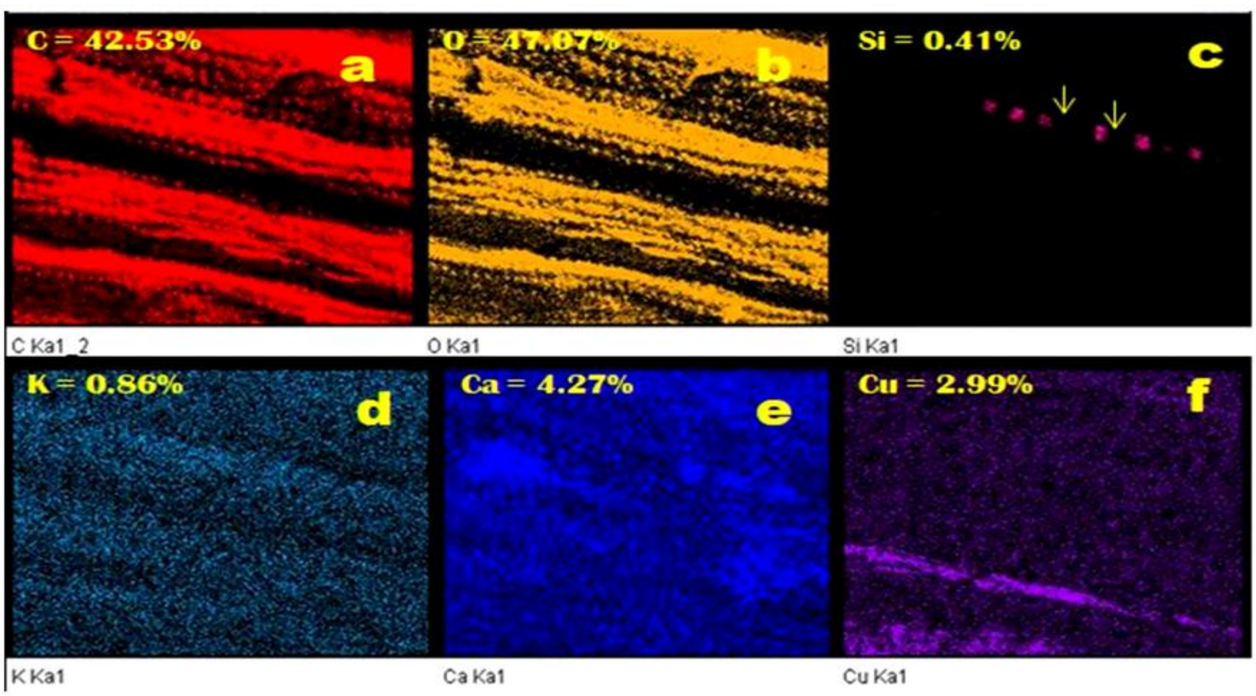

Figure 6. EDX element mapping of var.Ghanteswari leaf grown on nutrient solution with no added Si. **a)** Carbon-C, **b)** Oxygen-O **c)** Silicon-Si, **d)** Potassium-K, **e)** Calcium-Ca, and **f)** Copper-Cu.

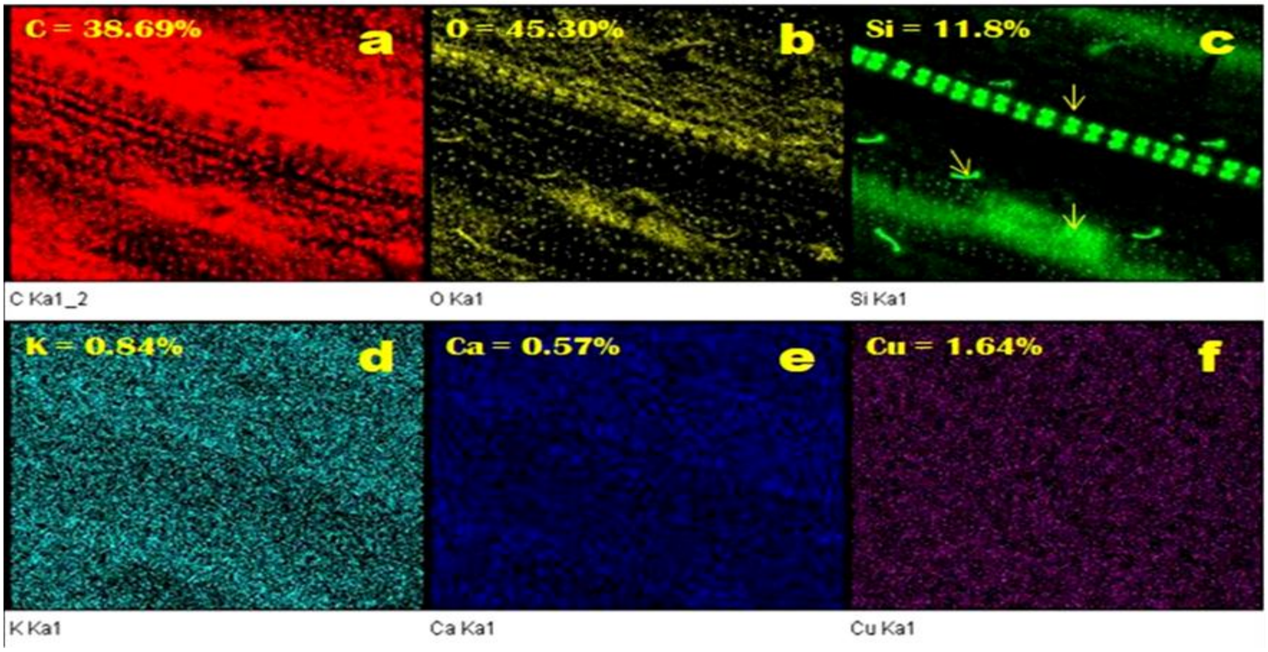

Figure 7. EDX elemental mapping of var.Ghanteswari leaf grown on nutrient solution with 1.0mM added Si. **a)** Carbon-C, **b)** Oxygen-O, **c)** Silicon-Si. **d)** Potassium-K, **e)** Calcium-Ca, and **f)** Copper-Cu.

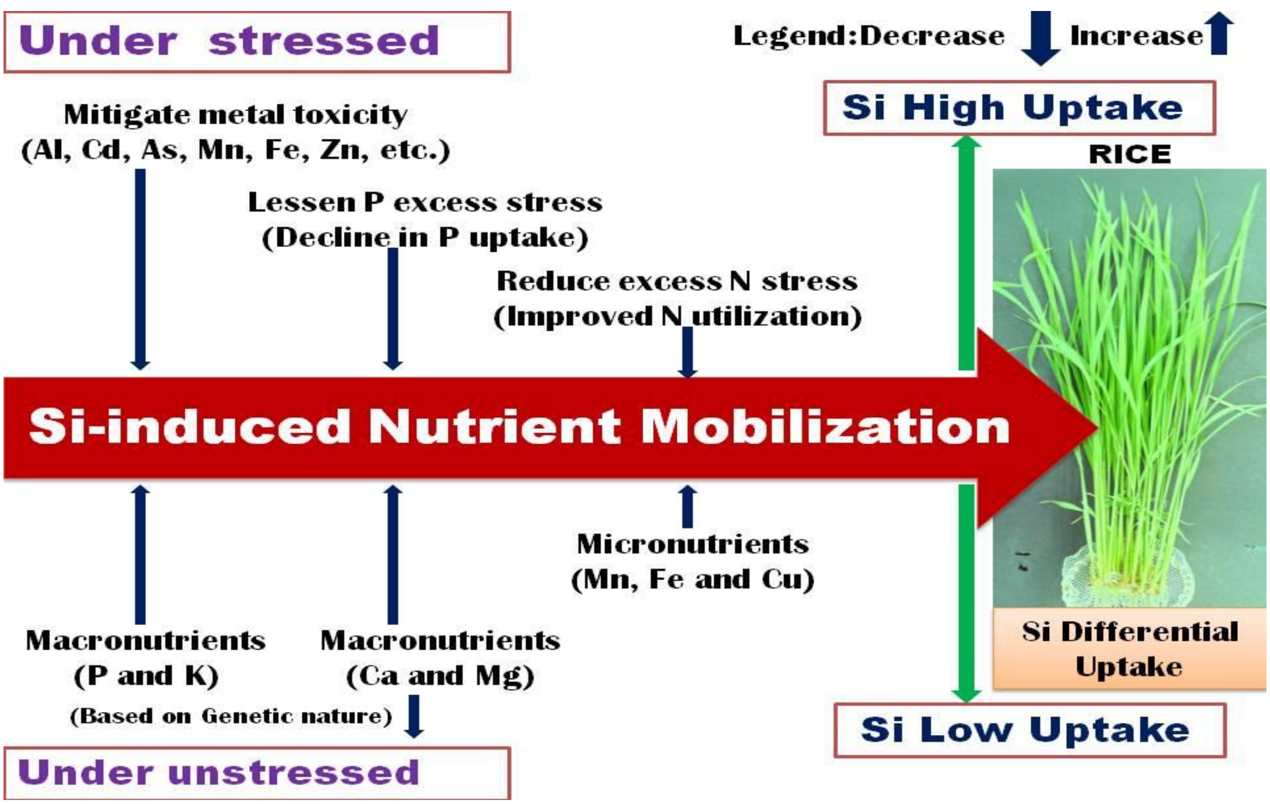

Figure 8. A schematic diagram: depicting the impact of silicon on nutrient mobilization and its relationship

Acknowledgments

The authors wish to acknowledge to the Department of Science & Technology, Govt. of India for providing the instrumental facilities under the FIST grant. This research was financial supported by DST-Inspire. We also thank the SAIF, IIT Bombay for providing SEM-EDX analysis.

Conflict of Interest

The authors declare that there is no conflict of interest regarding the publication of this research paper.

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