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Effects of OryMax^{SL} and Siliysol^{MS} on Growth and Yield of MTL560 Rice

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Abstract: Silicon is used in diverse plant functions improved resilience against biotic and abiotic stress which contribute to maintaining plant yield. Two commercial agro-silicon products, OryMax^{SL} and Siliysol^{MS}, were sprayed at different doses on MLT560 rice cultivar planted in pots to examine their effects on growth and yield. The silicon products showed no enhancement on photosynthetic pigments and internode length, but they significantly enhanced rice yield up to 15% higher than the control. The treatments were effective in increasing internode diameter, internode wall thickness, filled grain ratio, silicon accumulation in rice shoot and husk. Although silicon levels on the product labels are unequal, their actual silicon concentrations were determined insignificant, 25.43±0.89 and 22.83±2.15 in percentage on the weight basis, respectively for the OryMax^{SL} and Siliysol^{MS}. Silicon application from these formulations has positive benefits to rice yield and growth. And function response requires further investigation.

Key words: Growth · Internode · Rice · Silicon · Yield

INTRODUCTION

Silicon is the second most abundant element in the earth crust after oxygen [1] and accounts for 50-70% of soil mass [2]. It deposits in soil as two main forms of silicates and aluminosilicates [1]. In the soil solution, silicon is present in the form of silicic acid (H₄SiO₄) with the concentrations ranging from 0.1 to 0.6 mM [3]. However, its proportional composition in plants may vary between 0.1% and 10% on the dry weight depending on plant species [4]. [1] summarized some important functions of silicon in plant biology such as resistance to biotic stresses (bacteria, fungi, herbivores) and abitotic stresses (salinity, drought, low temperatures, heavy metal and aluminum toxicity), improvement of other nutrient imbalance, prevention of lodging. It also plays a critical role in both physical and chemical plant defence [5].

In many plant species, silicon is found in variably constitutive forms such as hydrated amorphous silica, silica-cuticle double layers, or silica bodies [6]. Rice can accumulate silicon up to the level of 10% of shoot dry weight which is higher than many macronutrients

including nitrogen, potassium or phosphate [7]. Recently, several genes (Lsi) responsible for silicon uptake and transport have been identified in rice [6, 8]. The rice plant absorbs silicon actively in the form of silicic acid through the root hairs [9, 10, 11] and silicon accumulation in rice is genotype dependent [12]. Up to now there is no evidence of the involvement of silicon in any biochemical reaction in plants [10, 13] but silicon supplement through fertilizers has been reported to improve yields significantly in many crops [14] including rice [15-19]. Silicon depletion in soils of rice culture could be a possible limiting factor to decline or stagnate the rice yield [20-22]. Research focusing on silicon benefits to rice performance has been done intensively in Japan where rice is grown in smaller scale than in other countries in Asia. Limited study has been done on the roles of silicon in Vietnam on rice production, particularly in the main rice region of the country, the Mekong Delta. There are also not many agro-silicon products available on the domestic market. Therefore, this study aims to examine the contributory effects of two agro-silicon products, OryMax^{SL} and SilySol^{MS}, on rice growth and development before they can be widely applied on the rice field.

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MATERIALS AND METHODS

MTL560 rice cultivar was grown in pots with the surface area of 0.08 m² contained 5 kg of soil. In each pot, 3 seedlings after 3 days of germination were placed in equilateral triangle form. Plants were exposed to natural climate conditions from the middle of September until harvest in December 2010 at location of 10°7'21''N and 105°34'48''E. Chemical fertilization equivalent to a total per hectare of 90 kg N, 60 kg P₂O₅ and 30 kg K₂O was applied at 8 (accounting for ½P₂O₅, 1/3 N, ½ K₂O), 20 (accounting for ½P₂O₅ and 1/3 N) and 40 (the rest) days after sowing (DAS).

The two commercial Agro-silicon products (Agro Genesis Pte Ltd, Singapore) used in the experiment were OryMax^{SL} and SilySol^{MS} with silicon contents as SiO₂ of 10% and 25%, respectively. The experiment was carried out with 5 treatments and 3 replicates (3 pots per treatment). Silicon was sprayed until all leaves of plants in each pot were completely wet. Each silicon product was used at two doses: 6 and 12 mL L⁻¹ for OryMax^{SL} and 4 and 6 g L⁻¹ for SilySol^{MS}. Silicon was applied at 20, 40 and 60 DAS. Control plants were sprayed with distilled water.

Plant height was measured at 20, 40 and 60 DAS. Internode length, largest outside diameter and the thickness of 4 internodes from top were measured at harvest. Rice yield was calculated after converting the seed moisture content to standard level of 14%.

Chlorophyll a, b and total carotenoids in rice leaves were analyzed at 45 DAS by the method of [23]. Fresh leaf per plant (0.2 g) were ground with liquid nitrogen immediately after removing from the plant. Photosynthetic pigments were extracted by mixing well with 10 mL of 80% acetone. The sample was left at room temperature under low light condition for 30 minutes and then 0.5 mL of the solution was diluted with 4.5 mL of 80% acetone. The diluted solution was measured for absorbance at 663.2, 648.8 and 470 nm.

Silicon levels in rice stem and husk were determined by adapting the methods of [24, 25]. Samples were placed in an oven at 70°C until constant weight and about 0.2 g was used for analysis. The dried sample was mixed with 3 mL of 60% HNO₃, 3 mL of 30% H₂O₂ and 2 mL of 46% HF. The mixture was digested in an ultrasonic cleaner (Sonorex Super 10P, BANDELIN Electronic, Germany) for 10 minutes. Afterwards, the digested sample was diluted to 100 mL with 4% boric acid. The diluted solution (50 μ L) was used for silicon determination. Distilled water of 1300 μ L was added to the sample followed by addition

of 750 μ L of 0.26 N HCl, 100 μ L of 10% (NH₄)₆ Mo₇O₂₄, 200 μ L of 20% tartaric acid and 100 μ L reducing agent which was prepared by dissolving 1 g of Na₂SO₃, 0.5 g of 1-amino-2-naphthol-4-sulfonic acid and 30 g of NaHSO₃ in 200 mL of distilled water. The sample was vortexed during 15 seconds, left at room temperature for 30 minutes. The formation of blue complex was measured for absorbance at 600 nm by a spectrophotometer (HE λ IOS α , Thermo Spectronic, England).

Silicon standard solution (Sigma-Aldrich, USA) containing 1013 ppm of Si in 2% NaOH was diluted to a concentration of 20 ppm with water and employed for constructing silicon standard curve. The amounts of silicon solution of 20 ppm for building the standard curve were 0, 50, 150, 200, 300 and 350 μ L. The addition of distilled water to each standard point was 1300, 1250, 1150, 1100, 1000 and 950 μ L, respectively. For blank sample, 50 μ L of digested mixture was used. For each treatment, the measurement was carried out with 3 replicates. For data analysis, SPSS 10.0 was used.

RESULTS

The analyzed data presented in Table 1 demonstrated that the biosynthesis of photosynthetic pigments in rice plants at 45 DAS were not stimulated by silicon treatments. The observed data (not presented) showed that rice plants at harvest in all treatments with or without silicon application were not significantly different in plant height, internode length, flag leaf size, number of green-remaining leaves. The number of plants per pot at harvest were also similar with all applied treatments. However, silicon application enlarged the internode diameter of rice stem at any providing doses (Table 2). In 4 internodes examined, the largest outside diameter of rice internode from the control was always smaller than those in the remaining treatments but the difference neither occurred between two silicon products nor concentrations of each product excepting for the 2nd internode. All spraying doses improved rice internodes diameter significantly with the minimal enlargement of 8% for the 1st internode when 4 g L⁻¹ SilySol^{MS} was applied. The highest increase in internode outside diameter was obtained for the 2nd internode at the spraying rate of 12 mL L⁻¹ OryMax^{SL}. In this case, silicon supply has widened the internode diameter up to 20% in comparison to the control. When spraying MTL56 rice cultivar with OryMax^{SL} and SilySol^{MS}, the variation in internode outside diameter between treatments occurred mainly at the 2nd internode.

Table 1: Effect of spraying silicon containing products on phothosynthetic pigments in rice leaves

Treatment	Photosynthetic pigments ($\mu g g^{-1} FW$) in rice leaves at 45 DAS			
	Chlorophyll a	Chlorophyll b	Total carotenoids	
Control	524.9	168.9	4257	
6 mL L ⁻¹ OryMax	597.8	195.2	6561	
12 mL L ⁻¹ OryMax	748.3	247.9	6243	
4 g L ⁻¹ SilySol	677.1	221.9	5596	
6 g L ^{−1} SilySol	759.1	260.3	4916	
P _{0.05}	ns	ns	ns	
CV (%)	15.3	19.0	19.1	

ns: not significant difference; CV: coefficient of variance

Table 2: Silicon application enhances rice internode outside diameter

	The largest outside			
Treatment	1 st	2 nd	3 rd	4 th
Control	3.05b	4.19d	4.91b	5.51b
6 mL L ⁻¹ OryMax	3.33a	4.85ab	5.67a	6.31a
12 mL L ⁻¹ OryMax	3.46a	5.04a	5.69a	6.47a
4 g L ⁻¹ SilySol	3.31a	4.55c	5.37a	6.27a
6 g L ⁻¹ SilySol	3.30a	4.67bc	5.59a	6.21a
P	*	**	**	**
CV (%)	3.5	2.7	3.7	3.7

^{*, **:} significant difference at 1% and 5% level, respectively. CV: coefficient of variance. Values presented are the means of n = 9, 3 for each replicate. In a column, the numbers followed by the same letter(s) are not significant difference by DMRT

Table 3: Silicon application contributes to increase the wall thickness of rice internodes

	The thickness of in	nternode wall (µm) from top at harve	st	
Treatment	1 st	$2^{ m nd}$	3 rd	4 th
Control	250.7c	508.7b	722.0b	852.0b
6 mL L ⁻¹ OryMax	272.7b	624.7a	821.3a	950.0a
12 mL L ⁻¹ OryMax	298.0a	660.7a	854.6a	962.0a
4 g L ⁻¹ SilySol	274.7b	644.6a	840.7a	946.0a
6 g L ⁻¹ SilySol	271.3b	614.7a	824.7a	944.7a
P	**	**	**	**
CV (%)	4.2	5.4	3.1	3.2

^{**:} significant difference at 5% level. CV: coefficient of variance. Values presented are the means of n = 9, 3 for each replicate. In a column, the numbers followed by the same letter are not significant difference by DMRT

Silicon spraying not only contributed to increase stem diameter but also stem thickness. The values presented in Table 3 showed that silicon treated plants had thicker internodes walls in comparison to those of the control. As above mentioned, plant heights were similar to all treatments but bigger stem diameter and thicker internode walls could contribute to improve stem hardness which would be an advantage for MTL56 rice cultivar against lodging.

Silicon application improved many rice yield components except for the number of panicles per pot and the weight of 1,000 grains (Table 4). Spraying silicon products increased the number of filled grains, at least 13 filled grains per panicle more than the control. As a result, the filled grain ratio has been improved significantly in all silicon added treatments. All enhanced parameters of rice yield accumulated to augment the rice

production per pot with the highest record of approximately 15% for the $OryMax^{SL}$ of 12 mL L^{-1} . The observed data exposed a possible suspect that applying higher doses of these products were not simultaneous with the rice yield. Spraying silicon form in solution of the $OryMax^{SL}$ to MTL56 rice cultivar seemly achieved better components on rice yield than microsuspended form of silicon in the $SilySol^{MS}$.

Silicon supplement enhanced silicon accumulation in rice stem and husk of MTL56 rice cultivar (Table 5). At all the applying doses, silicon spraying has increased the silicon levels in rice stem and husk significantly. However, OryMax^{SL} and SilySol^{MS} application augmented the silicon accumulation in stem more prominent than in rice husk, particularly for OryMax^{SL} of 12 mL L⁻¹ which boosted the silicon level almost twice more than that of the control stems. Both silicon products improved silicon

Table 4: Effects of silicon application on rice yield components

	Rice yield components				
Treatment	No. of panicles pot ^{-1#}	No. of filled grains panicle-1#	Filled grain ratio (%)#	Weight of 1000 grains (g)	Yield pot ⁻¹ (g)
Control	25.0	125c	91.3c	25.2	61.5c
6 mL L ⁻¹ OryMax	25.3	148ab	93.2b	25.4	66.6abc
12 mL L ⁻¹ OryMax	26.7	153a	94.6a	25.2	70.7a
4 g L ⁻¹ SilySol	26.7	150ab	93.1b	25.3	67.9ab
6 g L ⁻¹ SilySol	25.0	138a	93.6ab	25.3	63.7bc
P	ns	**	**	ns	*
CV (%)	4.26	5.06	0.68	0.40	4.7

ns: not significant difference; *, **: significant difference at 1% and 5% level, respectively. CV: coefficient of variance. $^{\#}$ Values presented are the means of n = 9, 3 for each replicate. In a column, the numbers followed by the same letter(s) are not significant difference by DMRT

Table 5: Effect of silicon application on silicon content in rice stem and husk

	Silicon content on dry weight (%)	
Treatment	In stem	In husk
Control	7.3c	10.7b
6 mL/L OryMax	11.6b	12.4a
12 mL/L OryMax	13.7a	13.1a
4 g/L SilySol	11.3b	13.8a
6 g/L SilySol	11.5b	13.4a
P	**	**
CV (%)	8.2	7.7

*,**: significant difference at 1% and 5% level, respectively. CV: coefficient of variance. In a column, the numbers followed by the same letter are not significant difference by DMRT

accumulation in rice husk clearly, at least 15% in comparison to the free silicon spraying treatment. Basing on the analyzed data presented in Table 5, it was suspected that silicon in solution of the OryMax^{SL} was probably more transportable to the other parts of rice plant than the microsuspension of silicon in the SilySol^{MS}.

DISCUSSIONS

Before this experiment was carried out, two other trials have been already done on the field for the same cultivar at different locations in the region but there was no significant difference in yield if silicon amounts of these products suggested by the manufacturer were applied. Since, in this experiment we applied higher doses of the products to recognize whether not silicon application from these products has a perspective enhancement on rice yield before they would be widely used.

The supplier has provided only the estimated levels of silicon on the product labels but with the big difference in silicon contents between them which caused difficulty in explanation for the experimental results. The analyzed data showed that both products contained similarly

silicon levels, 25.43 ± 0.89 and 22.83 ± 2.15 in percentage on the weight basis, respectively for the OryMax^{SL} and the SilySol^{MS}.

The data presented in Table 1 approved that OryMax^{SL} and Siliysol^{MS} spraying have no contribution to the synthesis of rice photosynthetic pigments although the doses of silicon application have been used at least double the recommended dose. However, the absolute values of chlorophyll a, chlorophyll b and total carotenoids from the silicon sprayed treatments were always higher than those of the control. The high value of coefficient of variance might be responsible for non-significant difference between the treatments.

At the beginning of experiment, 3 plants were planted in a pot and at the end of life cycle about 25 productive panicles per pot were harvested. That means the effective tillage ratio of MTL56 rice cultivar was more than 8 for each individual plant. There were no difference in plant height, number of panicles per pot and weight of 1,000 grains between silicon application levels in this trial or in another finding albeit calcium silicate supplement at very large amount of 500 kg ha⁻¹ [26]. Silicon fertilizer contributed only to other rice yield parameters such as number of filled grains per panicle and filled grain ratio (Table 4). Although silicon spraying caused no effect on suppressing the plant height as well as internode length, it improved the outside diameter of internodes and the thickness of internode walls which might be among effective factors contributing to the hardness of rice stem that could prevent lodging. The larger outside diameter and the thicker walls of the internodes found in the silicon sprayed rice plants could be due to formation of thicker cell walls below the cuticle layer [27]. Our results showed that silicon application increased rice yield without promoting the biosynthesis of photosynthetic pigments, although other experiments reported that silicon supplements maintained and enhanced photosynthetic

activity in rice, being one of the possible reasons for accumulating greater dry matter [28, 29]. The reduction of unfilled grain ratio in OryMax^{SL} and SilySol^{MS} treatments resulting in an increase of rice yield could be one of the contributions of silicon in increasing spikelet fertility [30]. Other experimental outcomes also stated that silicon addition caused no effect on the number of panicles per square unit or weight of 1,000 seeds [31].

Silicon application in the form of calcium silicate has imporved the grain and straw yield of rice, the ratio of filled grains and its content in straw [26, 32]. Therefore, rice farmers should think of exploiting the rice residues after harvesting as a potential and practical silicon source for rice because the silicon from rice husk ash has been demonstrated to improve rice growth and yield [33, 34]. The ability of taking silicon from soil is an extremely important factor for the rice yield since molecular evidence shows that the rice plant lacking of *Lsi* genes has 10 times lower in yield than the wild type [8]. With 15% yield increase, our result is in agreement with the former investigation of silicon application on rice yield ranging from 10 to 30% [21]. However, the contribution of silicon to rice yield should be investigated in further details because the significant increase of silicon in rice husk could result in an enhancement of rice yield.

In rice seedlings, silicon supplement in the form of sodium silicate responsed to an increase of its content in both roots and leaves concomitantly with the increasing doses of silicon application [35]. In this study, it might be predicted that applied higher doses of these two agro-silicon products than those presented in Table 4 would response for an unclear enhancement on rice yield. By those levels of silicon spraying, MTL560 seemed to satisfy with silicon utility on its yield components. Supplying silicon at an appropriate level could speed up silicon accumulation nearly twice in rice shoot (Table 5) thus it contributed to stem morphology or other agronomical traits should be further studied. Silicon is just considered as beneficial element for many plants [10] but its accumulation in their bodies concomitantly with the silicon availability in surrounding environment has been observed not only in rice but also wheat [36, 37]. With recently fast increase in literature on the silicon functions in plants, there is no doubt in stating that silicon is an emerging-focused element in plant science.

CONCLUSION

Spraying rice with silicon via two agro-chemical products, OryMax^{SL} and SilySol^{MS}, as leaf fertilizer can enlarge the internode diameter, increase the internode wall

thickness, reduce the unfilled grain ratio, enhance the filled grains per panicle which results in increasing the rice yield significantly.

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