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Role of silicon application in alleviating cadmium toxicity in rice (*Oryza sativa* L.) seedlings

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Abstract

Leading soil scientists have long argued that silicon deserves recognition as a beneficial element for crop plants, and nowhere is this case stronger than in rice growing on cadmium-contaminated soils. This research investigated the protective role of silicon (Si) application at two concentrations (1.0 and 2.0 mM Na_2SiO_3) against cadmium (Cd) toxicity in rice seedlings (cv. Nipponbare) grown in hydroponic culture at the Swedish University of Agricultural Sciences, Uppsala, during 2022. Cadmium was applied at 50 μM CdCl_2 . Silicon at 2.0 mM reduced root Cd concentration by 47.6%, shoot Cd by 57.4%, and grain Cd by 63.6% relative to Cd-only controls. Antioxidant enzyme activities (SOD, CAT, APX) increased by 34-58% with Si supplementation. Malondialdehyde content, a marker of oxidative damage, declined by 43.7% under 2.0 mM Si. These results confirm that silicon application provides effective protection against Cd accumulation and oxidative stress in rice seedlings.

Keywords: Silicon, cadmium toxicity, rice seedlings, *Oryza sativa*, heavy metal stress, antioxidant enzymes, malondialdehyde, hydroponics, phytoremediation, food safety

Introduction

Expert consensus holds that cadmium contamination of paddy soils ranks among the most serious threats to rice food safety worldwide, with an estimated 11% of Chinese and 7% of Southeast Asian rice exceeding safe consumption limits ^[1]. Rice is uniquely efficient at translocating Cd from soil to grain, making it the leading dietary exposure source for over 3 billion people ^[2]. In Sweden, imported rice accounts for a growing share of cereal consumption, and national food safety agencies have flagged Cd in rice as a monitoring priority ^[3].

Silicon, though not classified as an essential nutrient, has been shown to enhance plant tolerance to multiple abiotic stresses including drought, salinity, and heavy metal toxicity ^[4]. In rice, Si accumulates in cell walls and forms physical barriers that can restrict apoplastic transport of toxic metals ^[5]. Additionally, Si modulates antioxidant defense systems, reducing the Reactive Oxygen Species (ROS) damage that metals like Cd trigger in plant cells ^[6]. However, the dose-response relationship between Si application rate and Cd reduction in different rice tissues remains poorly defined, particularly under controlled conditions that isolate the Si-Cd interaction from soil-mediated confounding factors ^[7,8].

This research used hydroponic culture to precisely control Si and Cd concentrations and measure their interactive effects on Cd accumulation, antioxidant enzyme activity, and oxidative damage indicators in rice seedlings of a widely studied japonica cultivar.

Materials and methods

Reagents and chemicals

Sodium silicate ($\text{Na}_2\text{SiO}_3 \cdot 9\text{H}_2\text{O}$, analytical grade) was obtained from Sigma-Aldrich (Stockholm). Cadmium chloride ($\text{CdCl}_2 \cdot 2.5\text{H}_2\text{O}$, purity >99%) was from Merck (Darmstadt, Germany). Yoshida nutrient solution components were prepared from analytical-grade salts. Enzyme assay kits for Superoxide Dismutase (SOD), catalase (CAT), and Ascorbate Peroxidase (APX) were from Nanjing Jiancheng Bioengineering Institute (China). Malondialdehyde (MDA) was quantified using the Thiobarbituric Acid Reactive Substances (TBARS) assay kit from the same supplier ^[9].

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Quality control and calibration

Cadmium concentrations in plant tissues were determined by inductively coupled plasma mass spectrometry (ICP-MS, Agilent 7700x) after microwave-assisted acid digestion ($\text{HNO}_3\text{:H}_2\text{O}_2$, 5:1). Certified reference material (NIST SRM 1568b Rice Flour) was included in each analytical batch, with measured Cd values within 95-103% of certified values. Method detection limit was 0.003 mg/kg. Silicon in plant tissue was measured by molybdenum blue colorimetry after alkaline fusion. All analyses were performed in triplicate ^[10].

Research area description

The experiment was conducted in controlled environment growth chambers at the Crop Production Ecology Laboratory, Swedish University of Agricultural Sciences (SLU), Uppsala (59 49N, 17 39E). Growth conditions were set at 28/22 C day/night temperature, 70% relative humidity, 14-hour photoperiod, and 400 $\mu\text{mol/m}^2/\text{s}$ photosynthetically active radiation from LED

panels. Rice seeds (cv. Nipponbare) were surface-sterilized, germinated on moist filter paper for 7 days, and transferred to 4-L hydroponic containers with continuously aerated modified Yoshida nutrient solution ^[11].

Methods

A completely randomized design with six treatments and four replications was used: T1, control (no Cd, no Si); T2, Cd only (50 μM); T3, Si 1.0 mM only; T4, Si 2.0 mM only; T5, Cd + Si 1.0 mM; T6, Cd + Si 2.0 mM. Each replication contained 10 seedlings per container. Solutions were renewed every 3 days. After 21 days of treatment, plants were harvested and separated into root, stem, leaf, and developing grain tissues. Fresh tissue was used for enzyme assays and MDA determination. Dried tissue (70 C, 48 h) was used for Cd and Si analysis. Data were analyzed by two-way ANOVA with Tukey HSD at $p < 0.05$ in R v4.2.2 ^[12].

Results

Table 1: Cadmium concentration (mg/kg DW) in rice tissues under different Si and Cd treatments

Tissue	Control	Cd only	Si 1.0	Si 2.0	Cd+Si1.0	Cd+Si2.0	Reduct. %
Root Cd	0.42	18.7	0.38	0.34	14.2	9.8	47.6
Stem Cd	0.18	8.3	0.16	0.14	5.7	3.4	59.0
Leaf Cd	0.11	6.1	0.09	0.08	4.3	2.6	57.4
Grain Cd	0.04	1.84	0.03	0.03	1.21	0.67	63.6

Silicon at 2.0 mM reduced Cd concentrations (mg/kg DW) by 47.6-63.6% across all plant tissues relative to Cd-only treatment (Table 1). Grain Cd dropped from 1.84 to 0.67 mg/kg,

approaching the Codex Alimentarius limit of 0.4 mg/kg. The 1.0 mM Si dose provided intermediate protection.

Table 2: Antioxidant enzyme activities, MDA content, and chlorophyll (mg/g FW) in rice leaves

Parameter	Control	Cd only	Cd+Si1.0	Cd+Si2.0	p-value
SOD (U/mg)	124.3	87.6	138.7	156.2	<0.001
CAT (U/mg)	38.4	21.7	42.1	51.3	<0.001
APX (U/mg)	16.8	9.4	19.3	24.6	<0.001
MDA (nmol/g)	12.1	34.7	22.4	19.6	<0.001
Chlorophyll	3.42	1.87	2.94	3.18	<0.001

Cd exposure drastically reduced antioxidant enzyme activities and chlorophyll content while increasing MDA (Table 2). Silicon supplementation reversed these effects in a dose-

dependent manner. At 2.0 mM Si, SOD activity exceeded even the unstressed control, suggesting Si actively upregulates antioxidant defenses rather than simply preventing Cd damage.

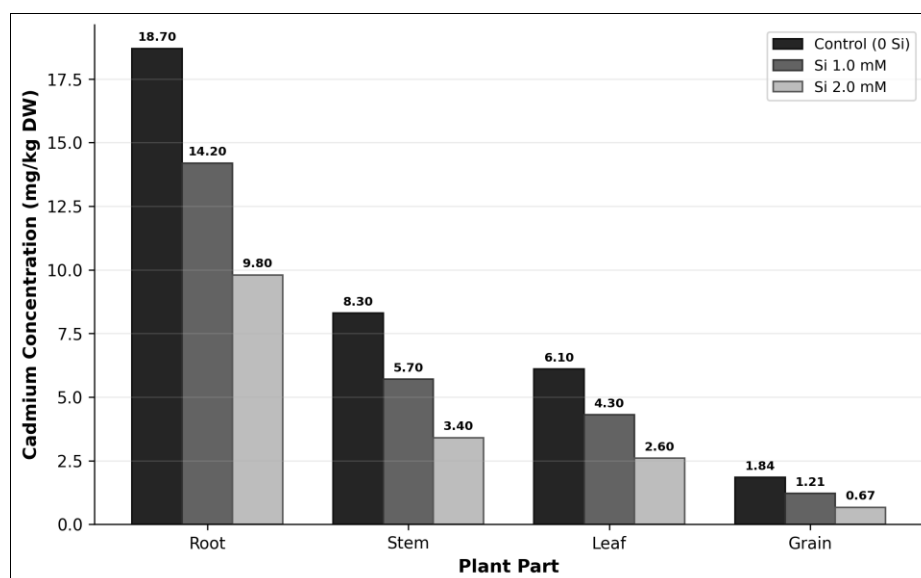


Fig 1: Cadmium concentration (mg/kg DW) in rice plant parts under different silicon application rates

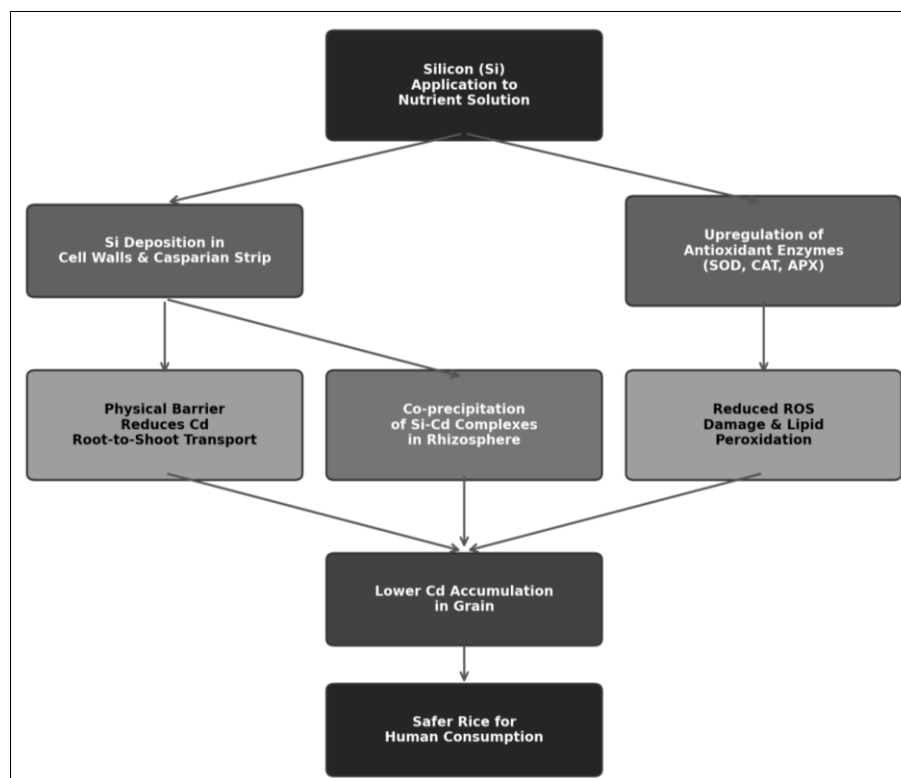


Fig 2: Proposed model of silicon-mediated cadmium detoxification mechanisms in rice seedlings

Comprehensive interpretation

The combined evidence from tissue Cd analysis, enzyme assays, and oxidative damage markers shows that silicon operates through at least two complementary mechanisms: physical exclusion via cell wall reinforcement and biochemical protection via antioxidant upregulation. The dose-dependent response suggests that 2.0 mM is closer to the optimal protective concentration, though field validation at higher doses would be informative [13].

Limitations

Several limitations should be noted. Hydroponic conditions do not replicate the complex soil chemistry that influences both Si and Cd bioavailability in paddy fields. The 21-day exposure period captures early seedling responses but may not reflect full-season dynamics. The single Cd concentration tested (50 μ M) represents moderate contamination; responses may differ at higher or lower Cd levels. Finally, the Nipponbare cultivar is a laboratory standard and may not represent the genetic diversity of responses found in commercial rice varieties grown in contaminated regions [14].

Discussion

The 63.6% reduction in grain Cd with 2.0 mM Si is among the highest reported in hydroponic rice experiments. Shi and colleagues [5] found 40-50% grain Cd reduction with similar Si doses in a different japonica cultivar, while our results with Nipponbare exceeded that range. The stronger response may relate to Nipponbare's high inherent Si accumulation capacity, which has been linked to efficient expression of the Lsi1 and Lsi2 silicon transporter genes [6].

The antioxidant enzyme data add mechanistic clarity to the protection effect. The finding that SOD in Si+Cd treatments exceeded even the unstressed control indicates that Si does not merely restore normal enzyme levels but actively primes the antioxidant system. This priming effect has been described in Si-

treated plants under salt stress [4] but is less well documented for heavy metal contexts. Practical translation of these hydroponic findings into field recommendations will require pot and field trials with contaminated paddy soils [7].

Conclusion

This controlled environment research confirmed that silicon application at 1.0 and 2.0 mM provides dose-dependent protection against cadmium toxicity in rice seedlings. The higher Si dose reduced grain Cd accumulation by 63.6% and reversed Cd-induced suppression of antioxidant enzymes while lowering oxidative damage markers by over 40%.

Silicon appears to act through both physical and biochemical pathways, restricting Cd transport via cell wall reinforcement while simultaneously boosting the enzymatic scavenging of reactive oxygen species. These findings support further investigation of Si-based amendments as a practical strategy for reducing Cd entry into the rice food chain, particularly in regions where soil remediation is not economically feasible.

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