

Unusual Accumulation of Silver in the Aleurone Layer of an Indian Rice (Oryza sativa) Landrace and Sustainable Extraction of the Metal

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Supporting Information

ABSTRACT: Uptake of noble metals by cereal plants is not reported in literature. Our study of 505 native rice landraces showed that nine of them accumulate silver at a high concentration when grown in the same soil. Among these, a medicinal rice landrace from West Bengal, *Garib-sal* was found to accumulate silver at an especially high concentration in the grains. Cultivation of *Garib-sal* rice in three successive years in Basudha farm in the rice growing period of June–October confirmed that for the same concentration of silver in the soil (~0.15 mg/kg), *Garib-sal* accumulates it in the grains to the extent of ~15 mg/kg. Laboratory experiments also demonstrated that silver uptake by *Garib-sal* is significantly greater than for other varieties grown on the same soil, and that the metal accumulates mostly in the grains, secondary ion mass



spectrometry was performed. The images reveal that the silver is concentrated in the aleuronic layer of the rice bran. Its concentration decreases in the subaleurone and becomes negligible in the endosperm. Accumulation of silver does not alter the grain morphology and chemical characteristics. The metal may be extracted from the bran after milling of the rice, thereby causing no loss of the foodstuff.

KEYWORDS: Rice, Bioaccumulation, Analytical chemistry, Mass spectrometry, Noble metals, Agriculture, Ethnobiology

INTRODUCTION

Rice (Oryza sativa) is one of the three most important cereals, being the staple food for more than half of the world's population.¹ A part of recent rice research is engaged in developing or genetically modifying strains of rice, in order to provide additional nutrients essential for human health.²⁻⁴ Though the phenotypic characters of different rice varieties seem to be largely genetically determined,⁵ micronutrient contents of rice may vary with the characteristics of the farm soil,⁶ and also with the water used for irrigation.⁷ In regions where groundwater is heavily contaminated with arsenic, rice grain can accumulate arsenic,⁸ which then enters the food chain affecting the consumer population. $^{9-11}$ A few other heavy metals, e.g., zinc and iron, are also reported to accumulate in rice grains.¹² In most cases, heavy metals are environmental stress factors for plants, and are bioaccumulated principally in the root epithelium and leaves.^{13,14} Silver is not known to bioaccumulate in the reproductive tissue of any cereal.¹⁵ Its typical concentration in soil does not exceed ~0.10 mg/kg, and in agricultural crops the value is <1 mg/kg of dry weight in vegetative tissues.

In the present study, we have analyzed the concentration of 12 heavy metals in the grains of 505 rice landraces from 1120 landraces grown every year on the conservation farm of Basudha (http://www.cintdis.org/basudha), as part of *in situ*

rice genetic diversity conservation effort. Regardless of the silver content in the farm soil, none of the 505 varieties reported herein showed high silver content in the grains, except nine varieties. Among these nine, Garib-sal showed an exceptionally high concentration of silver at 15.61 mg/kg in its grain.¹⁷ This landrace from West Bengal was traditionally used by indigenous people for medicinal purpose, to cure gastro-intestinal (GI) infection.¹⁷ Laboratory experiments confirmed high concentration of silver in the grains of Garibsal rice, compared to no silver uptake in the control, BPT 5204. To investigate the exact location of silver deposition in the grain, we performed secondary ion mass spectrometric imaging of the rice. An elemental map shows that silver concentration is highest in the aleuronic layer of rice grain, but negligible in the endosperm. Thus, metallic silver can be recovered from the bran coat after its separation by milling and polishing of rice grains. Noble metals have traditionally been extracted by the alkaline cyanide method.¹⁸ As cyanide is highly toxic, it can cause severe environmental hazards. Therefore, there is a need for green and sustainable methods for the extraction of noble metals.

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METHODS AND EXPERIMENTS

Materials. All the rice varieties were grown by Centre of Interdisciplinary Studies, Kolkata in its Basudha farm (http://www.cintdis.org/basudha) located in Bissam Cuttack block, Rayagada district of southern Odisha (19° 42′ 32.0″N, 83° 28′ 8.4″E). A few varieties, including a population of *Garib-sal* (coded G02-2) were grown on a farmer's land, ca. 14 km away from Basudha in Ankulpadar village, Rayagada district of Odisha (19° 26′ 27.1″N, 83° 33′ 7.3″E). Samples of the original population of *Garib-sal* grown on Basudha farm (coded G02-1) and G02-2, along with BPT-5204 (control, procured from Rajendranagar market, Hyderabad, India) were grown in our lab at IIT Madras. Nitric acid (assay 69–71%) and hydrogen peroxide (30% w/v) were purchased from Fisher Scientific. All the other materials needed for the analytical procedures are mentioned in the following sections.

Methods. Cultivation of Rice in the Donor Farm. Garib-sal rice were grown on both Basudha farm and the donor farmer's field, following traditional organic methods (with organic manures, and leaf mulch, without external input of synthetic agrochemicals).

Cultivation of Rice in Laboratory. Two samples of Garib-sal (coded G02-1 and G02-2, procured from two different farm fields, of two different soil characteristics), along with a common rice variety (BPT 5204, a breed introduced by Acharya N. G., Ranga Agricultural University), were grown in the laboratory in the Rabi (summer) season, following organic cultivation method. Each of the three rice samples were grown in 6 pots as replicates, with 4 seedlings grown in each pot. Three replications in each group were kept in normal soil without addition of salts, and the other three were injected with AgNO₃ solution. Silver was added in three different steps, once in 30 days (1 mg), 50 days (0.5 mg) and 75 days (0.5 mg) after the day of planting. The total amount of silver addition was 2 mg in 7 kg of soil (to replicate the silver concentration of farm soil). All the plants were watered every day for 120 days until the grains reached maturity. All grains were harvested after 150 days of plantation and dried in ambient condition and tested for metal ion uptake. All the measurements were performed in triplicate and the average values are reported.

Metal Detection and Analysis. Samples of the rice grains and different parts of the plants were digested using Anton Paar Multiwave 3000 microwave digester. For this, 500 mg of the dry weight of the sample was placed in a Teflon tube and 5 mL of HNO₃ followed by addition of 2 mL of H_2O_2 and 1 mL of H_2O carefully, and the mixture was kept inside the microwave digester. The program was set as 10 min of ramping time from 0 to 800 W and it was held for 15 min at 800 W. The vessels were allowed to cool for 1 h. Clear solutions indicated that the digestion was complete. This solution was further diluted ten times for the analysis of different metal ions using inductively coupled plasma mass spectrometry (ICPMS).

Resin Embedding and Microtomy. Embedding was done using standard protocols with Epofix resin, an epoxy resin (procured from Struers, Denmark, Cat. No: 40200029). The grains for secondary ion mass spectrometry (SIMS) analysis were decorticated, but no polishing was done so that the rice bran was intact. Some whole gains (without decortication) were also embedded for optical imaging. As all biological samples contain some amount of water, the grains were kept in a vacuum (10^{-2} mbar) for 10 days before embedding them in the resin. The resin was kept separately in a vacuum (10⁻ mbar) for 1 day, before use, for degassing. Whole grains were embedded in the resin using Eppendorf tubes of 10 mm diameter. The samples were kept in a vacuum impregnation unit (10^{-2} mbar) (CytoVac, Struers, Denmark) for 30 min to remove any trapped air bubbles and were then cured in air for 16 h at room temperature. When the resin was totally dried, the block was taken out and cut into sections of 5 mm thickness using a rotary cutter (Discoplan TS, Struers, Denmark). Microtomy (Leica EM UC6) was performed using the 5 mm cut sections of embedded rice grain to make a flat surface for SIMS analysis. The sections were then fixed on silica substrates with an adhesive (Fevi kwik, Pidilite) procured from the local market. To study the cut surface of the grain, SEM imaging was done using freshly

microtomed samples. Samples went through a precleaning step before SIMS imaging.

Instrumentation. The concentration of silver ion in the solution was measured using a PerkinElmer ICPMS NexION 300X. ICPMS measurements were done after calibrating the instrument. Silver was calibrated with silver atomic absorption spectroscopy (AAS) standard solution (1000 mg/L) purchased from Fisher Scientific (product no: AAAGH). The other elements like Cr, Mn, Fe, Ni, Co, Cu, Cd, As, Pb, Sb and Zn were calibrated by PerkinElmer Instrument Calibration Standard 2 (PE#N9301721). For all cases, five calibration points were used; 0 (called as blank) 10, 100, 1000 and 10000 ppb, respectively. In all the cases, five concentrations of the calibrant were analyzed first until a calibration curve was achieved with $R^2 = 0.9999$. After that, the two samples of known concentration (>100 and <1000 ppb) were checked at least three times to get reproducible data with deviation <0.5 ppb (for samples concentration higher than 1000 ppb). Detection limit for ICPMS is 0.01 ppb for these ions. Scanning electron microscopy (SEM) equipped with EDAX (FEI Quanta 200, Czechoslovakia) was used to record the surface morphology, elemental composition and elemental mapping of the samples. XPS measurements were done with Omicron ESCA Probe spectrometer with monochromatic Mg K α X-rays ($h\nu$ = 1253.6 eV). Most of the spectra were deconvoluted to their component peaks using the software CasaXPS. The energy resolution of the spectrometer was set at 0.1 eV at a pass energy of 20 eV. Binding energy was corrected with respect to C 1s at 284.5 eV. For XPS analysis, samples were ground and pellets were prepared using a pelletizer, they were further dried in a desiccator for 3 days so that a vacuum better than 1.1×10^{-8} Torr was attained throughout the measurement.

All SIMS analyses were performed using CAMECA NanoSIMS 50. The rice grain surface was scanned with focused secondary ion beam of 16 keV Cs⁺ and the analysis was done with a double-focusing mass spectrometer. We employed high resolution NanoSIMS 50 for recording the Ag deposition on different parts of the rice grain. Before each image acquisition, precleaning was performed for 15–20 min at 16 keV Cs⁺, 1–2 nA over 60 × 60 μ m. All image data were recorded with 16 keV Cs⁺ primary ions and negative secondary ions, with high mass resolution ($M/\Delta M \sim 5000$).

RESULTS AND DISCUSSION

The presence of essential and heavy metal ions were analyzed in 505 rice varieties grown in the Basudha farm (Table S1). Out of 505 rice varieties reported herein, nine varieties showed silver concentration more than 2 mg/g in the grains as shown in Table 1; the concentration of silver in the soil was ~0.20 mg/kg. Among these nine, *Garib-sal* showed an exceptionally high concentration of silver at 15.61 mg/kg in its grain.

To test the possible effect of silver concentration in soil on the silver uptake in the rice grain, the same *Garib-sal* rice was grown on another farm with lower silver concentration in the soil at <0.01 mg/kg, which showed a concentration of ~0.20 mg/kg in the grain. In our analysis, rice samples of *Garib-sal*

Table 1. Rice Landraces	with	High	(>2	mg/kg)	Silver
Content in Grains					

Sl. no.	accession code	landrace	Ag (mg/kg)
1	D03	Deputy-sal	2.40
2	G02	Garib-sal	15.61
3	К03	Kaya	2.05
4	K45	Kankhira	2.26
5	N03	Noichi	2.73
6	Q11	Kharah	5.60
7	S11	Subasita	4.70
8	V06	Bhuri shulah	2.50
9	Z06	Jhinga	3.70

Table 2. Mean Silver Concentration (mg/kg) in the Soil and Various Parts of *Garib-sal* (G02) and BPT^{*a*} Rice Plants Grown in the Laboratory

	concentratio	on in Ag deficie	nt soil (C_x) concentration in Ag enriched soil (C_y)			ed soil (C _y)	times increase in Ag concentration $(C_y - C_x)/C_x$		
cultivars	G02-1	G02-2	BPT	G02-1	G02-2	BPT	G02-1	G02-2	BPT
soil		0.033			0.33			10.00	
grain	0.04	0.07	0.04	2.19	2.77	0.13	53.75	38.57	2.25
husk	0.05	0.09	0.11	2.62	2.83	0.20	51.40	30.44	0.82
lower stem	0.08	0.07	0.04	0.67	0.73	0.52	7.38	9.43	12.00
upper stem	0.08	0.08	0.07	0.54	0.67	0.54	5.75	7.38	6.71
leaves	0.04	0.05	0.05	0.24	0.37	0.28	5.00	6.40	4.60
roots	0.10	0.09	0.11	2.35	2.60	0.57	22.50	27.89	4.18
^a BPT refers to BPT 5204, a modern cultivar.									

showed markedly enhanced silver concentration than that of 7 popular marketed varieties grown in the same soil with Ag concentration of ~0.15 mg/kg (Table S2). The list of 7 varieties compared here includes a popular modern high-yield variety MTU 7029 (syn. Swarna), released in 1982. The Ag uptake data, corresponding to high and low silver-containing soils, suggest that the *Garib-sal* variety has an exceptionally high capacity to bioaccumulate silver, in the range of 100 times that of ordinary rice cultivars. Most of the other cultivars refuse to take up silver altogether, when cultivated in the same soil.

To examine the effect of soil Ag concentration on the silver uptake by Garib-sal (accession code G02) rice, samples of two populations of the same landrace (coded G02-1 and G02-2, harvested from two different farms) were cultivated in our laboratory in the Rabi (summer) season of 2014. The seedlings were grown in experimental pots while intermittent doses of AgNO₃ were injected into the soil until flowering of the rice plant, so that the final silver concentration in the soil was ~0.35 mg/kg (3.2 mg of AgNO₃ added to 7.0 \pm 0.2 kg of soil), alongside a control with no silver injection (see Methods for details). The results were compared with a modern rice variety (BPT 5204), under identical conditions (with and without silver addition to soil). Our measurements of the silver concentrations in various plant parts and the soil are given in Table 2, which shows that the accumulation of Ag in all rice plants increased significantly when the Ag content in the soil increased 10-fold. However, the increase in Ag uptake from the Ag-enriched soil by the check cultivar, BPT 5204 was only 2.25fold, whereas the accumulation of silver in G02 rice grains was \sim 39–54 times higher when grown in Ag-enriched soil than in Ag-deficient soil. Thus, higher silver content in the soil significantly enhanced the plant's uptake of the metal. Although the relative increase in silver accumulation was different in different tissues; the highest increase was in the grains, and the lowest in the leaves.

Differences in bioamplification in the rice plant are likely to be related to the differences in the ambient conditions such as temperature and soil characteristics.¹⁹ For instance, typical temperature range and annual rainfall in the farm were 28–34 °C and ~805 mm, respectively (during the cultivation period), in contrast to 35–41 °C and ~258 mm of water irrigated in the laboratory. The same rice was also grown in three different seasons in the same farm soil, with silver concentration around ~0.15 mg/kg in the farm, and was found to have accumulated nearly equal concentration of silver (Table S3). This suggests the possibility to vary the uptake profile by controlling environmental factors. Enhanced silver concentration does not seem to alter the composition of rice in terms of contents of other metals (such as Fe, Mn, Cr, Cu and Ni), except for Zn (data presented in Table S4), whose uptake seems to be especially high in *Garib-sal*.¹⁷ We examined the enhanced silver accumulation in different parts of the G02 rice grain. Silver deposition was largely in the bran of the rice grain. In the polished rice (with the bran removed), Ag concentration was less than half of the initial value (Table S5).

Figure 1A shows photographs of the *Garib-sal* rice panicles, whose length and grain density appear to be the same when



Figure 1. Photographs of rice grains. Panicles of G02 rice grown in the lab in (A) normal soil; (B) silver enriched soil (sb: 50 mm); (a1 and a2) photograph of a few rice grains corresponding to A and B (sb: 8 mm); (b1 and b2) magnified photograph of decorticated grains corresponding to A and B (sb: 20 mm). See text for details.

they are grown in normal and silver enriched soils (Figure 1A,B, respectively). Figure 1a1,2 show enlarged pictures of the raw rice (with hull); and Figure 1b1,2 are the same for decorticated rice, corresponding to Figure 1A,B. The scale bars (sb) are shown in the figure. However, standard spectroscopic and microscopic characterization did not reveal any significant morphological difference between the rice samples grown in Ag-enriched and Ag-deficient soils. X-ray photoelectron spectra showed similar features although minor changes in intensities are seen in the C 1s and O 1s regions (Figure S6). The sensitivity of XPS is of the order of parts per thousand, whereas the concentration of silver in the rice grain is 15 mg/kg (i.e., 15 ppm); we were therefore unable to get any signal of Ag using XPS. No difference was detected in the binding energies of the features. SEM examination of the grains (Figure S7) did not show changes in the grain microstructure. Elemental analysis with EDS was not sensitive enough to view the silver accumulation as the concentration was below the detection



Figure 2. NanoSIMS imaging of a G02 rice grain. (A) Collection of nanoSIMS images from an embedded rice grain showing the distribution of ¹⁰⁷Ag at different positions of the grain. Grain border is marked and distinguished from the embedding resin. There is a gap between the resin and the grain upon curing, which is marked. Relatively higher concentration of Ag is found at the periphery of the grain and the concentration decreases as it moves toward the endosperm. (Ai) Higher magnification SEM image on which nanoSIMS was performed and a graphical representation is marked in Figure 3A. (Aii) SEM image of the horizontally cut rice grain embedded in epoxy resin, the image in Figure 3Ai was taken from the portion marked in the panels B and C. Enlarged nanoSIMS image of positions 7 and 1 of Figure 3A using ¹⁰⁷Ag. The intensity color profile is shown on the right. (D) Schematic representation of a rice plant showing the concentration of Ag accumulation at different parts. The Ag concentration color profile is shown on the right of the figure.

limit of EDS. The only element clearly visible in images other than C, O, N, K and Mg was Si (shown in SI Figure S7).

To detect and image the silver accumulation on different parts of the rice grain, we employed SIMS analysis using NanoSIMS 50 (see Methods for details). As illustrated in Figure S8, several peaks of interference appeared at masses 107 and 109 amu of Ag. However, as the Ag peak at 107 amu showed less mass interference than at 109 amu, we preferred to follow the ¹⁰⁷Ag isotope (marked with a red line shown in SI Figure S8). The ratio between the ¹⁰⁷Ag and ¹⁰⁹Ag, i.e., the terrestrial ratio, was found to be ~52/48.

A longitudinally cut decorticated *Garib-sal* rice grain, embedded in an epoxy resin, was imaged using NanoSIMS. An HRSEM image of a portion of the grain is shown in Figure 2Ai. This position is graphically represented in Figure 2A. The red border separates the embedded resin and the rice grain. The image shows the aleurone layer in more detail along with the subaleurone layer and endosperm, as marked in Figure 2Ai. Various regions of the grain can be understood from Figure S9, which shows a scheme of a longitudinally cut rice specimen (left) along with a high resolution photograph of the whole grain (right). The full SEM image of embedded rice grain is shown in Figure 2Ai.

NanoSIMS images of different parts of the rice grain (i.e., at the periphery, subaleurone and at the endosperm) reveal a heterogeneous distribution of ¹⁰⁷Ag. The periphery of the grain accumulates more Ag, clustered in the aleurone layer (marked as region 1, 2 and 3), with its intensity gradually decreasing toward the subaleurone layer (marked as region 4 and 5). A negligible amount of Ag was seen to have accumulated in the interior of the endosperm (marked as region 6 and 7). Enlarged images of positions 7 and 1 in Figure 2A are presented in Figure 2B,C, respectively. Figure 2B shows that negligible amount of Ag is present in the endosperm. Figure S10 in the SI shows the distribution of ¹²C, ¹²C¹⁴N, ³²S and ¹⁰⁷Ag at position 7. Figure 2C shows high accumulation of ¹⁰⁷Ag in the aleurone layer, with nonuniform distribution. The intensity decreases from the outer to the inner layers of the grain, and is minimal in the endosperm. An intensity profile is shown on the right of Figure 2C. A schematic diagram of the accumulation of Ag in various parts of a rice plant is shown in Figure 2D.

The distribution of 107 Ag (marked #a) along with overlaid images of $^{12}C^{14}N^{32}S^{107}$ Ag (marked as #b and #c) for various regions are shown in Figure 3. In images #b and #c, the elements ^{12}C , ^{14}N and ^{32}S are represented in different colors, whereas 107 Ag is uniformly red. The peripheral positions 1, 2, and 3 (as marked in Figure 2A) were enriched with silver, especially in the aleuronic layer as shown in Figure 3. The concentration of Ag decreases subcutaneously in subaleurone as seen in regions 4 and 5 and it is negligible in the endosperm



Figure 3. Silver distribution of Ag at different positions of the G02 rice grain. The distribution of 107 Ag and the superimposition of 12 C, 14 C¹⁴N, 32 S, 107 Ag marked at positions 1–7 (as in panel A). (a) Deposition of 107 Ag. (b) Superimposed images of 12 C¹⁴N 32 S¹⁰⁷Ag (blue-blue-green-red), and (c) 12 C¹⁴N 32 S¹⁰⁷Ag (green-green-blue-red).

(regions 6 and 7). Figure S11 shows the distribution of these elements in other regions.

To demonstrate viable extraction of silver, 64 g of *Garib-sal* rice was taken and digested using a 5:3 HNO₃ and H_2O_2 mixture in a microwave digestor (for details, see S12), which made a clear solution. The clear yellow solution indicated that the digestion was complete (Figure 4a). The yellow color arose due to dissolved NO₂, which disappeared when the solution was evaporated and diluted (Figure 4b). ICPMS data ensured that the sample contained 14.60 mg/kg of silver and no lead and mercury were present in the sample. Figure 4c shows the formation of a white precipitate after addition of dil. HCl. After 1 h, the whole solution got turbid as shown in Figure 4d. As Ag is in group 1 (in the analytical group of cations) and in the

absence of the other interfering ions (Pb and Hg), the only possibility for the white precipitate is AgCl. The turbid solution was centrifuged and washed multiple times and was dried for characterization. The dry weight of the precipitate was ~ 1.10 mg (some amount was lost during washing and centrifugation, quantitative calculation is given in \$12). The precipitate was pelletized using an infrared pelletizer and EDS spectra were collected, from multiple points of the pellet. Figure 4e shows the presence of Ag and Cl in the EDS spectrum. Small amounts of Si and O were also seen at some places, which are due to the ITO plate exposed in some regions as the pellet was thin. A control sample of AgCl was also prepared by addition of dil. HCl to aqueous AgNO3 (purchased from Sigma-Aldrich), and corresponding data are shown in Figure 4f. The elemental ratio (at. %) for Ag:Cl was \sim 1:1 shown in the insets of Figure 4e,f. These data confirm the presence of silver in G2 rice sample and also show that it can be extracted by a simple procedure.

Solid state extraction of gold by cyclodextrin (CD), a cyclic carbohydrate, has been reported.²⁰ Similar binding of heavy metals is expected for carbohydrates in general, not limited to CD. Our previous studies have shown that glucose and carbohydrate can cause chemical corrosion of silver.^{21,22} We have also found that uranium species in water, existing as $UO_2^{2^+}$, binds with starch and gets accumulated in rice upon cooking.²³ These studies suggest that uptake of silver and its selective binding with carbohydrate are possible, although the molecular mechanisms of uptake, transport and accumulation require further investigation.

CONCLUSION

We report a novel rice landrace *Garib-sal* that accumulates about 100-fold greater amount of silver in the grain from silverenriched ($\sim 0.15 \text{ mg/kg}$) soils than any standard rice variety. The grain was imaged using secondary ion mass spectrometry, and the results shows that silver was accumulated mostly in the aleuronic layer of the rice bran. The concentration decreases subcutaneously in subaleurone and was found negligible in the endosperm. We suggest growing this landrace as a possible means of agricultural extraction of the noble metal from the rice bran after polishing the grain. A majority of marketed rice in



Figure 4. Extraction of silver as silver chloride. Photographs showing the (a) digested extract of G2 rice, (b) after evaporation and further dilution with DI water (5 times), (c and d) 1 min and 1 h after addition of dil. HCl, respectively. (e) EDS spectrum of the white precipitate and (f) EDS spectrum of standard AgCl. The elemental ratio are shown in the insets of the panels e and f. Precipitation of AgCl is noticed in panel c and it is complete in panel d.

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South Asia is sold as polished grain, from which the bran is removed. The removed bran from the polished *Garib-sal* rice may be used for the extraction of silver. Furthermore, we endorse the genius of traditional indigenous medicinal knowledge of the use of this particular silver rice in the treatment of GI infections. Because silver ions are known to be detrimental to pathogenic microbes,²⁴ the Ag-fortified rice is likely to be a potent curative of GI microbial infections. *Garibsal* rice is the only cereal to contain silver in its grains at concentrations greater than 10 mg/kg. This study is the first investigation of the deposition of silver in rice grain.

ASSOCIATED CONTENT

S Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acssuschemeng.7b02058.

Additional details describing concentration of various metals in different rice varieties, concentration of silver in marketed rice varieties, reproducibility of the uptake of silver grown on the same field in three different seasons, concentration of different metals in G02 variety grown in different soils, analysis of G02 rice variety for silver concentration in different parts of the grain, spectroscopic analysis of G02 rice, SEM/EDS spectrum of transverse section of husked rice, calibration of Ag peaks in NanoSIMS, labeled cross-sectional diagram of rice, NanoSIMS imaging of endosperm, NanoSIMS imaging of aleurone and subaleurone layer and digestion procedure (PDF)

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Notes

The authors declare no competing financial interest.

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