RESEARCH ARTICLE
doi: 10.2306/scienceasia1513-1874.2022.045

ScienceAsia 48 (2022): **326–334**

Potential of biofertilizers and natural soil amendments to mitigate heavy metal contents of soil in lowland rice (*Oryza sativa* L.) farming

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Received 20 Apr 2021, Accepted 16 Oct 2021
Available online 28 Feb 2022

ABSTRACT: Chemical fertilizers have been extensively used in Sri Lanka, and they have contributed to the pollution of groundwater and soil. Therefore, a field experiment was conducted to reduce the potential risk of toxic metal contamination of rice farming through sustainable nutrient management, with the scope of replacing the conventional chemical fertilizers. The field experiment comprised the addition of organic amendments, consortium of biofertilizers, and chemical fertilizers. Two improved rice varieties and three traditional rice varieties were grown. The plant-available and total Cd, As, Pb, and Hg in soil, rice roots, and grains in each system were analyzed. The results revealed that organic soil management is an effective soil remediation technique for rice soil to neutralize the toxic heavy metals (e.g. Pb, Hg, Cd) and metalloids (As) and lead to a harvest with minimum heavy metal contamination ($p < 0.05$). Natural soil amendments such as compost, biochar, and biofertilizers, which reduce the heavy metal concentrations of soil and rice grains, could be recommended for soil application, instead of chemical fertilizers, for both the traditional and the improved rice varieties.

KEYWORDS: rice, heavy metal remediation, nutrient management, biofertilizer, compost, biochar

INTRODUCTION

Pollution of water and contamination of soil by heavy metals have caused serious problems threatening human health, environment, food safety, and sustainable development of agriculture. Among these metals, several metals such as iron (Fe), zinc (Zn), copper (Cu), nickel (Ni), cobalt (Co), manganese (Mn), and molybdenum (Mo) are essential and must be present in biological systems within certain concentrations [1]; nevertheless, they will become toxic at higher concentrations. However, there is no beneficial function of other metals such as lead (Pb), mercury (Hg), cadmium (Cd), and metalloid arsenic (As) found in plants and animals, and are very toxic even if at low concentrations [2].

It is identified that the agricultural soils have been highly contaminated with potential toxic metals, which could pass down into living systems and human body via food chains due to intense usage of variety of agricultural chemicals and fertilizers [3]. Since rice is grown in submerged conditions in Sri Lanka, there is an increased tendency for the rice to be exposed to heavy metals from the soil and water sources [4]. The extensive use of fertilizers in paddy cultivation has resulted in the rice contaminated with heavy metals and consumed in Sri Lanka [5]. Rice plant generally shows a tolerance to heavy metal toxicity due to hyper-accumulating nature of rice [4,5]. Human health problems caused by the heavy metal toxicity were reported in Sri Lanka, mainly chronic kidney disease of unknown etiology (CKDu) [5]. Therefore, there is an urgent need of ensuring a sustainable nutrient management in rice farming. Organic agricultural practices primarily use ecological processes rather than external inputs to facilitate crop growth and livestock production [6,7]. Organic fertilizers include compost, farm yard manure, slurry, worm castings, urine, peat, green manure, dried blood, bone meal, fish meal, and feather meal. It was reported that the concentration of heavy metals is very low in the widely used standard organic manures in Sri Lanka [7]. Therefore, incorporation of standard organic manures to soils un-contaminated with heavy metals would be one strategy to reduce the heavy metal loading and bio-availability through fixation and bioremediation. Compost is mainly enriched with humus, and its neutral pH facilitates the heavy metal immobilization from the contaminated soil [6, 8].

The added organic amendments into soil increases the heavy metal absorption, hence, decreases the heavy metal concentration in soil solution. Biochar, made from pine and oak wood and bark at 400–450 °C, has been found to adsorb a variety of heavy metals, including Pb, As, and Cd due to its large surface area and high pore volume [9]. Furthermore, some studies revealed that the biochar generated from carbonized rice hulls, wheat straw [10], sewage sludge [11], and water hyacinth [12] can immobilize Cd and other heavy metals such as Cu, Zn, Cr, and Ni in paddy soils.

Microorganisms have proven to be effective in detoxifying soil pollutants including heavy metals and metalloids (Pb, Hg, Cd and As). The contribution of the soil microbes in humification process during organic matter decomposition is also important. The humic substances play an important role in mitigating the adverse effects of organic and inorganic pollutants in the soil. Additionally, a group of soil fungi, arbuscular mycorrhizal fungi (AMF) possess useful symbiotic association with plant roots. Arbuscular mycorrhizal fungi can take up heavy metals through the fungal hyphae and transfer them to the host plant roots. Some AMF colonized plant roots can enhance heavy metal uptakes and root-to-shoot transports; while in other cases, AMF immobilizes heavy metal within the soil [13].

Therefore, the present study was planned to assess the effects of different nutrient management systems, such as the use of compost, biochar, and biofertilizer, on heavy metal contents of soil and rice grains. In addition, the scope of the study also aimed to identify a sustainable rice farming system by replacing conventional chemical fertilizers [10].

MATERIALS AND METHODS

Experimental site and design

A field experiment was carried out at Ranpathwela in Anuradhapura, North Central Province, Sri Lanka (8°23′30.8″ N, 80°39′03.2″ E) during *Yala* season in 2016. The mean annual precipitation was 1750 mm, and the mean annual temperature was 30–35 °C. The soil type of the area where experimental field was located was reddish brown earths Alfisols [14]. The study was planned as a two-factor factorial experiment with three replications and designed as the randomized complete block design. The first factor of the experimental design was the five varieties of rice: two improved rice varieties, BG 300 (BGSP1) and BG 304 (BGSP2); and three traditional rice varieties, Suwandel (TRSP1), Madathawalu (TRSP2), and Kaluheenati (TRSP3). The second factor consisted of a control and seven treatments: CON, control (no application of soil amendments and biofertilizers); AMF, AMF inoculants (2 Mg/ha); RAMF, Epaawela rock phosphate (ERP) (153.3 kg/ha) with AMF inoculants (2 Mg/ha); MC, mixed microbial culture [mixed culture of *Azospirillum* sp., *Pseudomonas* sp., and *Bacillus* sp. (5 l/ha)]; RMC, ERP (153.3 kg/ha) with mixed microbial culture (5 l/ha); BC, biochar (6 Mg/ha); CP, standard compost (10 Mg/ha); and IF, inorganic chemical fertilizer (125 N kg/ha, 62.5 P_2O_5 kg/ha, and 50 K_2O kg/ha).

Preparation of biofertilizers and organic amendments

Preparation of the native AMF inoculum: trap culture establishment

Soil samples with the fine root fragments of herbs were collected from the upper layer (0–15 cm) of the soil, where traditional rice was grown by exercising organic management practices in Anuradhapura District. Those fine root fragments were used as an indigenous AMF inoculum. Trap cultures were established using maize (*Zea mays* L.) to produce AMF inoculum. Finally, rhizosphere soil containing an average of 200 AMF spores per 100 g and AMF colonized root fragments (approximately 60–75% potential colonization) was used as the source of inoculum [13].

Isolation of Azospirillum sp.

Azospirillum sp. was isolated from the rhizosphere soil of a rice field in Anuradhapura Province, Sri Lanka. Nfb semi-solid medium in screw-capped tubes was inoculated with 0.1 ml of soil suspension and incubated at 37 °C for 72 h. Cultures in the slants were streaked on the plates of malate agar medium containing 0.1% NH_4Cl to get pure colonies and stored at −20 °C [15]. The density of isolated *Azospirillum* sp. was adjusted to approximately 10^8 CFU/ml in nutrient broth (NB) medium and used for *Azospirillum* inoculant production.

Isolation of Pseudomonas fluorescens

Pseudomonas fluorescens strains were isolated from the rhizosphere soil, from a depth of 5–20 cm at two independent rice fields in Anuradhapura and the Eppawala rock phosphate deposit. A soil suspension was prepared [16] and serial dilutions (10^{-1} to 10^{-10}) were made, and 0.1 ml aliquots of each suspension were spread onto King's B medium (KB) agar and incubated at 28 °C for 2 days. Fluorescent *Pseudomonas* strains were identified using a UV illuminator at 366 nm and sub-cultured on KB agar plates; and pure cultures of *Pseudomonas* sp. were obtained. Colonies of selected *Pseudomonas* sp. were then transferred to the NB and kept on the shaker for 72 h at 100 rpm and 30 °C to reach an approximate 10^8 CFU/ml [16].

Isolation of potassium solubilizing Bacillus sp.

Five grams of soil sample were taken from an organically grown lowland rice field and diluted to 10^{-8} suspension using sterile distilled water. Then, 0.l ml of soil suspension was spread over a Petri dish containing Aleksandrov medium and incubated at 30 °C for 72 h [17]. Screened strains of *Bacillus* approximately 10^8 CFU/ml were transferred to culture in a NB medium and stored at −20 °C.

Compost for organic soil amendment

Compost was produced under practical field conditions by windrow type composting with locally available resources: green cuttings (30 kg), rice straw (60 kg), leaf crop residues and leaves of *Leucaena leucocephala* and *Gliricidia sepium* (40 kg), cow manure (40 kg), and loam (15 kg). The temperature was regularly checked and kept at 60–65 °C at the center of the pile. During the 90 days of composting, the pile was turned three times; and at each turning, the materials were mixed thoroughly and moistened with water [18].

Preparation of biochar

Wood chips, air-dried rice straws, and rice husks (3:2:2 ratio) were used to produce biochar using two-barrel method [19]. The biochar was air-dried for 6 days, ground to pass through a 2 mm sieve, and mixed uniformly.

Establishment of treatments

The experiment was designed as a randomized complete block design with three replications. It comprised 24 plots of 10 m × 3 m. Plots were arranged as rows and separated by 45 cm double mud bunds. Ridges were made around the plot area about 45 cm in height and width. Each plot was subdivided equally into 5 subplots for 5 different rice varieties. AMF inocula (2 Mg/ha) were applied on the surface of the soil in respective treatments (AMF and RAMF), one day before transplanting and reapplied periodically in 1.5 month intervals. Rock phosphate was applied at a rate of 153.3 kg/ha per treatment as basal applications (RAMF and RMC).

The prepared biofertilizer in a NB was applied by dipping the roots of the seedlings into the slurry, 1 h before transplanting in the field as per treatment (MC and RMC). Biochar (6 Mg/ha) was spread, thoroughly mixed with the soil, and then ploughed to a 20 cm depth, one week before transplanting of rice seedlings in biochar amended plots (BC). Half of the recommended compost amount (5 Mg/ha) was spread on the soil as basal dressing at one week before transplanting; and two split doses, each of 2.5 Mg/ha, were added to the soil after as per treatment (CP). Inorganic chemical fertilizers (125 N kg/ha, 62.5 P_2O_5 kg/ha and 50 K_2O kg/ha) were applied to the IF treatment plots (IF). Entire dose of phosphorus and potassium and half of nitrogen (62.5 N kg/ha) were applied as the basal fertilizer, 1 h before transplanting rice. The remaining dose of nitrogen was top dressed equally at active tillering and panicle initiation stages.

Soil and plant sampling

Rhizosphere soil and bulk soil samples of 500 g each were collected from the different rice field plots at a depth of 0–15 cm 2 days prior to the harvest. Composite soil sample was made of subsamples that were collected from a 2 m × 2 m sampling grid, excluding the edges of the plot. The soil samples were air-dried, gently crushed, passed through a 2 mm sieve, and stored in sealed polythene bags.

Three to four rice plants at the early tillering stage (3 weeks of growth) were collected by pulling out plants from each subplot in an area of 1 m^2 and kept for the assessment of the establishment of AMF. The whole rice plant including the roots and panicles with grains was collected randomly just before harvest and then washed with clean water to remove soil particles. The whole plant samples were washed again with deionized water, and dried in the oven at 65 °C for about 2 days until a constant weight obtained. The dried samples were weighed and prepared for analysis.

Heavy metal analysis

Analysis of available concentrations of heavy metals and metalloids in soil samples

Soil samples were digested by wet acid digesting method adding HNO_3 along with HCl in the ratio of 3:1 (v/v) and heated on a hot plate for 2 h [20]. The digested samples were quantified for soil available concentrations of K, Na, Fe, Zn, Al, Cd, Pb, As, and Hg using Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-OES) (Thermo Scientific ICAP7400 DUO).

Analysis of total concentrations of heavy metals and metalloids in rice root and grain samples

Ground root and rice grain samples were oven-dried (at 85 °C), and 0.5 g of the dried sample was placed in a digestion tube and 10 ml of the HNO_3:$HClO_4$ acid mixture (3:1) was added. The acid digestion was continued at 250 °C for 30 min [20]. The total heavy metal concentrations of the digested soil and plant samples were estimated using ICP-OES (Thermo Scientific ICAP7400 DUO).

Data analysis

Statistical analyses were performed using the MINITAB statistical software package (MINITAB 17.1.0 version). The two-way ANOVA was followed to test the significant difference ($p < 0.05$) among the means. Then, the Tukey's honestly significant difference test was used to conduct the pairwise comparisons for the significant

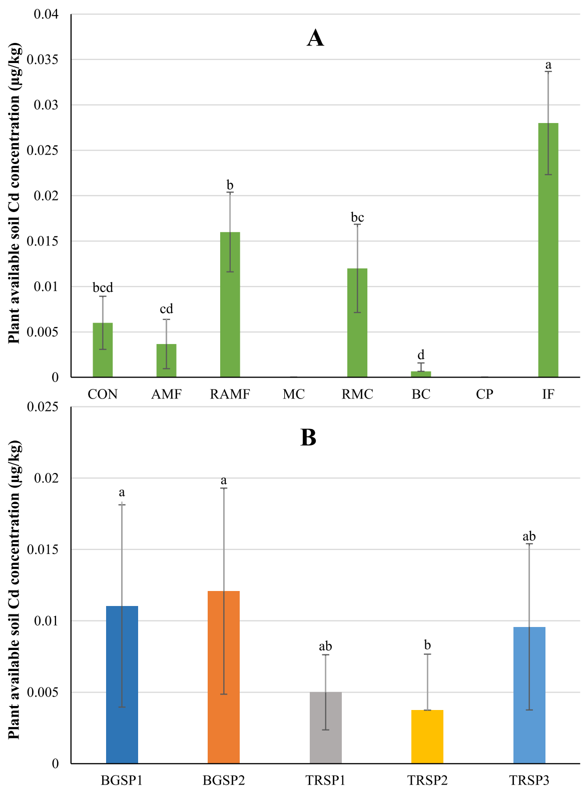

Fig. 1 (A) Plant available soil Cd (µg/kg) concentration with the amended biofertilizers, natural amendments, and chemical fertilizer applications. (B) Plant available soil Cd (µg/kg) concentration with the growth of different rice varieties. Error bars indicate 95% confidence intervals. Means that do not share a same letter are significantly different at $p = 0.05$. CON, no biofertilizer or amendment addition; AMF, addition of AMF; RAMF, rock phosphate + AMF addition; MC, addition of mixed microbial culture; RMC, rock phosphate + mixed microbial culture; BC, Addition of biochar; CP, Addition of compost; IF, addition of recommended dose of chemical fertilizer.

cases. The significance level (α) 0.05 was used for all the statistical tests.

RESULTS AND DISCUSSION

Heavy metals in soil

Plant available cadmium

The results indicated that soil Cd levels were significantly different ($p < 0.05$) in variety (V) and the treatments (T) (Fig. 1A and Fig. 1B). However, V × T interaction was not statistically significant for soil Cd ($p > 0.05$). The highest plant available soil Cd was observed in BGSP2 rice variety (0.012 ± 0.001 µg/kg). Elevated concentrations of soil available Cd were found in chemical fertilizer amended (IF) plots, despite the rice variety (0.028 ± 0.001 µg/kg). However, the maximum concentration of soil Cd (0.028 ± 0.001 µg/kg) was far lower than the maximum standard level of Cd by the Codex Alimentarius Commission, 200 µg/kg. The phosphate fertilizers particularly addition of triple superphosphates could be the cause of elevated plant available soil Cd, as 2.3 to 46 mg/kg of Cd was found in triple superphosphates in Sri Lanka [21].

The available soil Cd concentrations in compost (CP) and mixed microbial culture amended (MC) treatments were found to be lower than the ICP-OES detection limit for Cd (< 0.01 µg/kg). Compost facilitates Cd immobilization by adhering to its humic substances and carboxyl, carbonyl, and phenolic organic functional groups [21]. Compost could affect the physicochemical properties of soil and, hence, enhance the adsorption of heavy metals by soils indirectly [18]. However, some researchers believe that long-term application of compost may promote the mobilization of heavy metals and mineralization of humic substances leading to the release Cd to soil and water [22]. Soil Cd concentration varied with the amended rock phosphate, particularly in RAMF (0.016 ± 0.002 µg/kg) and RMC (0.012 ± 0.001 µg/kg) treatments of the present study. The results also revealed that in RAMF and RMC treatments, the same rock phosphate has added very low amount of Cd to the soil.

Plant available lead

Plant available soil Pb concentration varied significantly ($p < 0.05$) with rice varieties, amendments, and interactions (Fig. 2A). Treatment TRSP2 × IF (0.046 ± 0.005 µg/kg) showed the highest soil Pb concentration (Fig. 2A). Compost has different affinities to different heavy metals, and it was reported as of Pb > Cu > Cd > Zn [23]. Results of the present study also indicated that compost and biochar effectively mitigated the negative effects of Pb in agricultural soils.

Plant available arsenic

The plant available soil As concentration varied significantly among rice varieties, amendments, and variety × amendment interaction. It was further revealed that soil with amended chemical fertilizer (IF) showed the highest soil available As (Fig. 2B). Considering the V × T interaction, higher levels of soil available As concentrations were recorded in TRSP3 × IF (0.66 ± 0.11 µg/kg) (Fig. 2B). However, the highest value was lower than the maximum threshold value of 200 µg/kg As in soil, determined by the Codex Alimentarius Commission. Plant available soil As concentrations were low in the present study ranging from 0.66 ± 0.11 µg/kg to the ICP-OES undetectable levels (Fig. 2B). However, Takahashi et al [24] reported that in flooded rice fields, soil organic matter mobilize As as arsenide and subsequently uptake by plants. However, biochar has been reported to adsorb heavy metals, including Pb, As, and Cd. Furthermore, Mo-

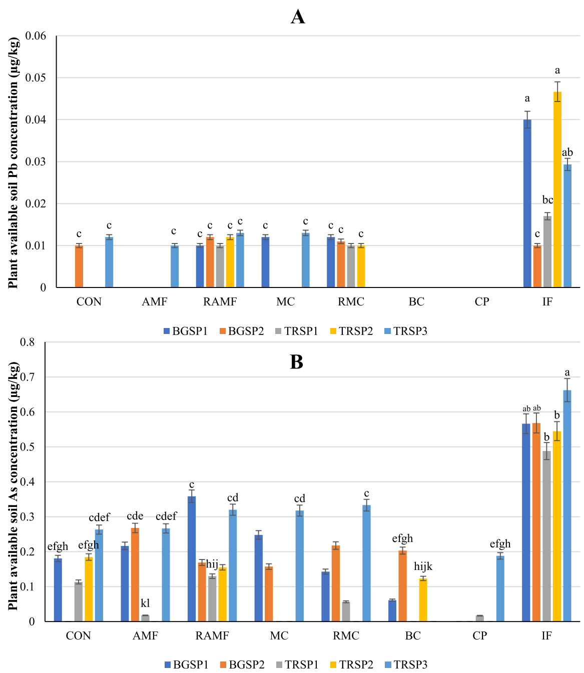

Fig. 2 (A) Plant available soil Pb (µg/kg) and (B) plant available soil As (µg/kg) concentrations with the amended biofertilizers, natural amendments, and chemical fertilizer applications and growth of the different rice varieties. Other details are as described in Fig. 1.

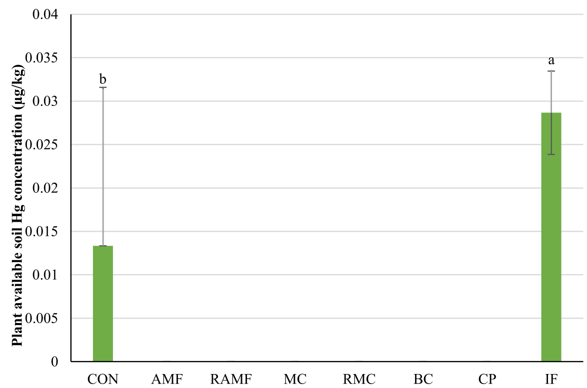

Fig. 3 Plant available soil Hg (µg/kg) concentration with the amended biofertilizers, natural amendments, and chemical fertilizer applications. Other details are as described in Fig. 1.

han et al [9] reported that oak bark biochar removed approximately 70% of the As in solution. Metal adsorption ability of biochar mainly occurs by ion exchange mechanisms [25]. Chandrajith et al [21] have reported the mean As concentrations in some paddy soils of wet zone (0.9 mg/kg) and dry zone (0.7 mg/kg) in Sri Lanka, even higher than the maximum limit. However, the mean As concentrations from this study were far lower than the As concentrations recorded by Chandrajith et al [21].

Plant available mercury

Statistical analysis revealed that there was no significant difference ($p > 0.05$) of soil available Hg among the rice varieties and the V × T interactions (Fig. 3). It was evident that plant available Hg (II) concentrations showed a significant difference ($p < 0.05$) among the control and the amended treatments. However, Hg concentrations estimated in the present study ranged from 0.029 ± 0.001 µg/kg (IF) to the ICP-OES undetectable levels.

Heavy metals in rice roots

Cadmium in rice roots

Mean root Cd concentration in rice roots significantly varied ($p < 0.05$) among rice varieties, treatments, and interactions. Among interactions, the highest root Cd levels were observed in all rice varieties with IF compared with the other treatments and the control (Fig. 4A). The highest mean of total root Cd was observed in the interaction TRSP2 × IF (3.59 ± 0.01 µg/kg). In rice roots of AMF, RAMF, RMC, CP amended, and CON subplots, Cd levels were below the detection limit of ICP-OES (Fig. 4A).

However, it has been reported that AMF often permits in higher concentrations of heavy metals in roots but lower concentrations in shoots by Wu et al [26]. In the AMF colonized roots, heavy metals mainly accumulated in intraradical fungal structures rather than in root cells. In the contrary, AMF have been shown in some studies to reduce heavy metal absorption by roots.

Lead in rice roots

Statistical analysis revealed that Pb concentration in roots varied significantly ($p < 0.05$) among rice varieties, treatments, and interactions. The highest mean root Pb concentration was observed in TRSP3 × IF (5.93 ± 0.05 µg/kg) (Fig. 4B). It was also observed that in roots of TRSP3 variety, Pb accumulation was higher than the other varieties except TRSP2 with IF (Fig. 4B). The root Pb concentrations of the rice varieties also depended on the plant available soil Pb (Fig. 4A and Fig. 4B).

Arsenic in rice roots

Statistical analysis revealed that root As concentration varied significantly ($p < 0.05$) among rice varieties, treatments, and V × T interactions. The highest root As concentration was observed in TRSP3 × IF (0.38 ± 0.05 µg/kg) (Fig. 4C). Furthermore, the roots of improved rice varieties, BGSP1 and BGSP2, in CON and IF contained approximately similar amounts of As (Fig. 4C). Arsenic concentration of roots of all rice varieties in CP amended subplots were below the detection limit of ICP-OES.

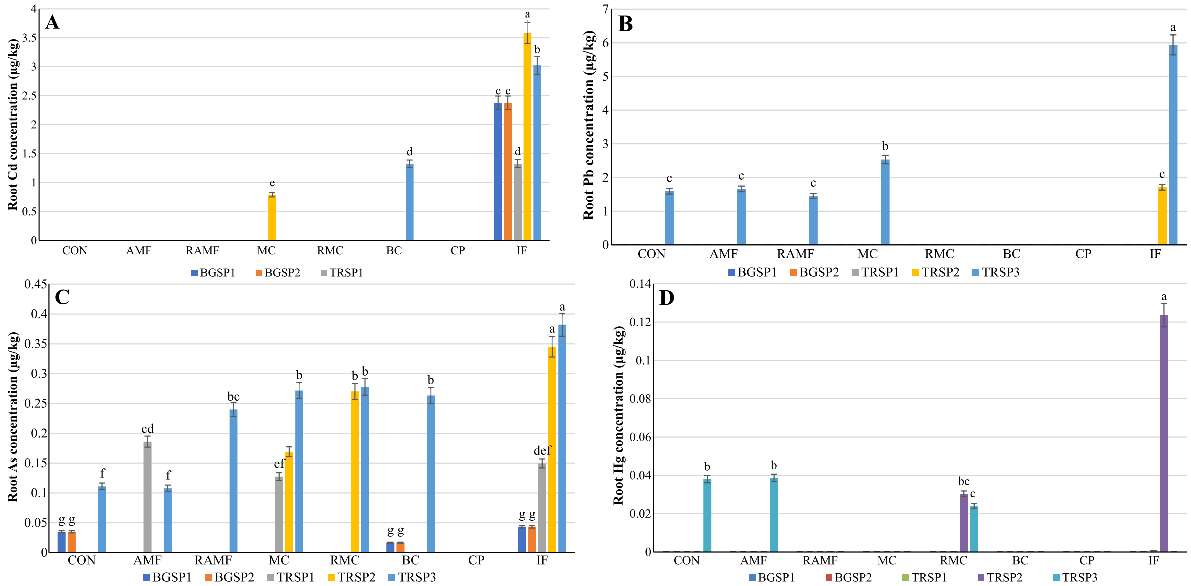

Fig. 4 (A) Mean root Cd (μg/kg), (B) mean root Pb (μg/kg), (C) mean root As (μg/kg), and (D) mean root Hg (μg/kg) concentrations with the amended biofertilizers, natural amendments, and chemical fertilizer applications and growth of the different rice varieties. Other details are as described in Fig. 1.

Mercury in rice roots

Concentration of Hg in roots varied significantly ($p < 0.05$) among rice varieties and V × T interactions, but not among treatments. The highest Hg concentration was observed in TRSP2 × IF (0.12 ± 0.02 μg/kg) (Fig. 4D). Furthermore, Hg in rice roots of RAMF, MC, BC, and CP amended subplots were below the detection limit of ICP-OES.

Heavy metals in rice grains

Cadmium in rice grains

It was revealed that Cd concentration of the rice grains in all rice varieties and experimental conditions of the present study was below the detection limit of the ICP-OES. Furthermore, it was clearly indicated that Cd concentration in rice grains was poorly correlated with soil extractable cadmium. According to Grant et al [27], the application of P fertilizers containing 20 to 50 mg/kg of Cd led to significant increases in the Cd concentration of the soil, which may lead to higher crop Cd accumulation. Previous studies also revealed that the rice grains Cd levels depend on the cultivars [28]. According to Payus and Talip [29], rice grains were concentrated with the highest cadmium levels as compared to other part of the rice plant such as the root, stem and shoot. However, in accordance with the present study, Liu et al [28], has reported that the average Cd accumulation in rice roots were much higher than rice grains.

Codex committee of the Food and Agriculture Organization of the United Nations (FAO) published that the maximum permissible level of Cd in polished rice grain is 0.2 mg/kg [30]. Ji et al [31], reported that the cadmium concentration in brown rice ranged from 0.03 mg/kg to 0.96 mg/kg with an average as 0.15 ± 0.17 mg/kg. Previous studies in Sri Lanka reported that Cd concentration of rice grains was greater than the safe level of 0.2 mg/kg [30]. Yuan et al [32] reported that organic matter applied soil effectively immobilized dissolved soil cadmium in flooded conditions in rice production.

The rice plants of all the experimental subplots were subjected to the submerged conditions throughout the growth. This may have been the reason for not having detectable levels of Cd in rice grains in the present study. The phenomenon was confirmed by the findings of Sriprachote et al [33] that the increased Cd content of harvested grains were observed with the decreased submergence of rice plants in surface water during the growing season. Cadmium ions can be transformed to an insoluble cadmium sulfide, which is hardly absorbed by rice plants.

Lead in rice grains

Lead concentration in rice grains in the present study varied with different rice varieties subjected to the treatments. Statistical analysis revealed that Pb concentration in rice grains varied significantly ($p < 0.05$) among rice varieties, treatments, and the interactions. However, Pb content of rice grains was high in IF amended plots (Fig. 4A). The highest grain Pb was observed in TRSP3 × IF (13.0 ± 1.16 μg/kg), followed by BGSP2 × IF, TRSP2 × IF, BGSP1 × IF, and TRSP1 × IF

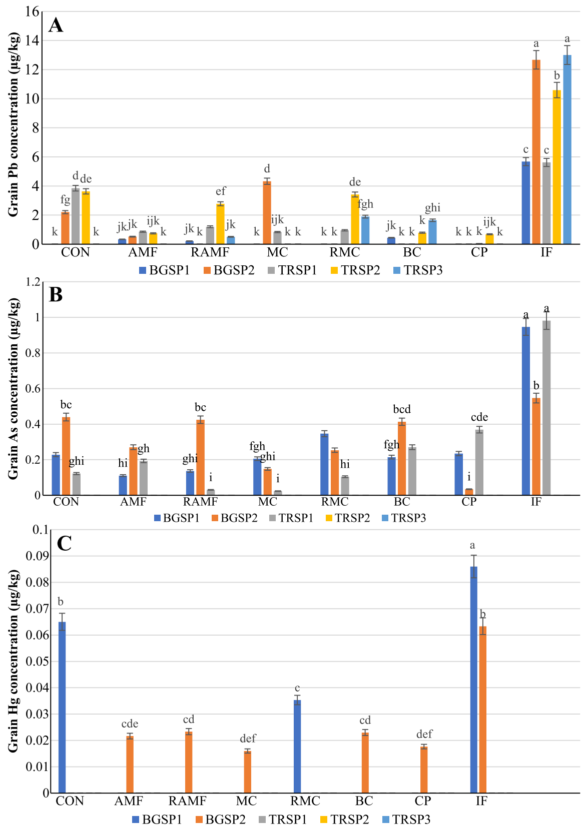

Fig. 5 (A) Mean grain Pb (μg/kg), (B) mean grain As (μg/kg), and (C) mean grain Hg (μg/kg) concentrations with the amended biofertilizers, natural amendments, and chemical fertilizer applications and growth of the different rice varieties. Other details are as described in Fig. 1.

(Fig. 5A). Furthermore, the grains of BGSP2, TRSP2, and TRSP1 rice varieties grown in CON plots also contained Pb in the concentrations of 2.21 ± 0.003 μg/kg, 3.63 ± 0.29 μg/kg, and 3.85 ± 0.03 μg/kg, respectively (Fig. 5A). It was also observed the BGSP1 and BGSP2 rice varieties grown in CP subplots possessed very minute amounts of Pb in their grains ranging from 0.68 μg/kg to undetected levels in the present study. Lead can strongly bind with organic and colloidal materials, and it is believed that only trace amounts of the lead in soil are soluble and thereby available for plant uptake in CP amended plots. It was recorded that the affinity of humus for heavy metals followed the order of Pb > Cu > Cd > Zn [34]. The common safety threshold of Pb is also set at 200 μg/kg for rice grain [30]. Although the total grain Pb concentration in the present study was always below the safety threshold limit, the organic amendments might potentially help further reduce the Pb content [35].

Arsenic in rice grains

Mean grain As concentration varied significantly ($p < 0.05$) among rice varieties, treatments and V × T interactions in the present study (Fig. 5B). Among the interactions, the highest As concentration was observed in TRSP1 × IF (0.98 ± 0.001 μg/kg) followed by BGSP1 × IF, BGSP2 × IF, BGSP2 × CON, BGSP2 × RAMF, BGSP2 × BC, and TRSP1 × CP (0.37 ± 0.05 μg/kg) (Fig. 5B). Grain As concentration of TRSP3 variety was below the detection limit of ICP-OES under the experimental conditions of the present study.

Rice can be considered as one of the main sources of inorganic As in which humans are consumed [36]. However, none of the rice grain samples in the present study showed total As levels exceeded the threshold of 200 μg/kg recommended for polished rice by the Codex Committee on Food Additives and Contaminants [30]. The mean As level was previously reported as 43 μg/kg in rice grain of improved variety from Sri Lanka [37].

Mercury in rice grains

The total Hg levels of the rice grains in the present study (Fig. 5C) was far less than the grain Hg levels recorded in China, ranging from below the detection level to 70 μg/kg [38]. Mercury in rice is also present as methyl mercury (MeHg) which is more toxic than inorganic forms [Hg(II)]. However, only total Hg was measured in the present study, and the exact percentage of MeHg in rice was not estimated, as it can be varied from 3.5% to 40% [39]. The Hg maximum limit (20 μg/kg) fixed for cereals by Chinese legislation is far more than the Hg concentration of the rice grain in the present study. Furthermore, US Environmental Protection Agency (EPA) has set a reference dose for methyl mercury of 0.1 μg/kg body weight per day, and the World Health Organization (WHO) has set the dose at 1.6 μg/kg body weight per week [33].

CONCLUSION

Organic soil management is an effective soil remediation technique for rice soils to neutralize the toxic heavy metals (e.g. Pb, Hg, Cd) and metalloids (As) that could lead to a harvest with minimum heavy metal contaminations. Natural soil amendments such as compost, biochar and biofertilizers, which reduce the heavy metal concentrations of soil and rice grains, could be recommended for soil application instead of chemical fertilizers for both traditional and improved rice varieties. Furthermore, addition of rock phosphate also makes a difference in heavy metal availability in the soil and the rice grains compared with the chemical fertilizers.

Acknowledgements: This research was funded by Rajarata University of Sri Lanka (RJT/RP&HDC/2013/App/R/02), Mihintale, Sri Lanka; Yunnan Fundamental Research Projects (grant no. 202001AU070114), CAS President's International Fellowship Initiative (PIFI) young staff under the grant no. 2020FYC0002 and the National Natural Science Foundation of China (NSFC Grant 41807524 and 31851110759).

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