

SHORT REPORT

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Presence of arsenic in Sri Lankan rice

Channa Jayasumana^{1*}, Priyani Paranagama², Saranga Fonseka², Mala Amarasinghe², Sarath Gunatilake³ and Sisira Siribaddana⁴

Abstract

Background: Arsenic and heavy metals are implicated in causation of CKDu among farmers in dry zone of Sri Lanka. Rice has been identified as a major source of arsenic in research carried out in other countries. We analyzed 120 samples of new improved varieties (NIVs) and 50 samples of traditional varieties (TV) of rice for total arsenic content.

Findings: Rice cultivated in Sri Lanka is contaminated with arsenic. Agrochemical dependent NIVs contain considerable amount (20.6 -540.4 µg/Kg) of arsenic. There is no difference between the arsenic content in NIV rice samples from areas where there is high or low prevalence of CKDu. TVs that are cultivated without using agrochemicals contain significantly less arsenic (11.6 - 64.2 µg/Kg). However, it is evident that the TVs also contain toxic metals if they are grown with fertilizers and pesticides.

Conclusion: A high proportion of arsenic in rice exists in the inorganic form. Sri Lanka is a nation with high per capita consumption of rice. Codex Alimentarius recommends the maximum allowable limit for inorganic arsenic in rice as 200 µg/kg. Assuming that 70% of the total arsenic content exists in the inorganic form, this corresponds to a level of about 286 µg/kg of total arsenic. As such, 11.6% of the samples of NIVs exceeded this maximum recommended level in polished rice. Inorganic arsenic is a non-threshold carcinogen. Research should be focused on developing rice varieties that do not retain arsenic within the rice grain.

Keywords: Arsenic; Rice; Chronic kidney disease; Sri Lanka; Agrochemicals

Findings

Arsenic (As) has been a known poison for thousands of years and is classified as a class one non-threshold human carcinogen (European Food Safety Authority 2009). Arsenic was not identified as an environmental pollutant in Sri Lanka until recently. Arsenic was identified as a possible etiological factor for the newly emerging epidemic of Chronic Kidney Disease of unknown origin (CKDu), a tubulo-interstitial nephritis among paddy farmers in dry zone of Sri Lanka (Jayasumana et al. 2013; 2014). Analytical studies have shown that a significant amount of arsenic in biological samples (urine, hair and nails) of these CKDu patients (Jayasumana et al. 2013; Jayatilake et al. 2013). Hence, we decided to analyze rice grown in Sri Lanka for arsenic. Analysis was carried out at the department of chemistry, University of Kelaniya and again at the Institute for Integrated Research in Materials, Environments and

Society (IIRMES) lab, California State University, Long Beach (CSULB), USA.

A total of 170 rice samples were collected from seven different locations to polythene zip bags (Figure 1). There were reported cases of CKDu in and around the sample collection centers from Padaviya, Sripura and Mahawilachchiya but not in at other locations (Kurunegala, Mihinthale, Moneragala and Gampaha). Hundred and twenty of these samples were Newly Improved Varieties (NIV) of rice and were collected directly from paddy farmers. Other 50 samples consisted of 5 different types of traditional varieties (TV) of rice, all cultivated at Kurunegala and Sripura without using agrochemicals but with plant extracts for pest control and with the use of natural fertilizers. Six samples of TVs were purchased from a supermarket in Colombo. They were marketed by a private company and produced, using agrochemicals. Samples were ground using an agate mortar passed through 0.3 mm sieve before analysis. Atomic absorption spectrometry (AAS) with hydride generation system (GBC 3000) and background corrector (GBC 932 plus GBC

* Correspondence: jayasumanalk@yahoo.com

¹Faculty of Medicine & Allied Sciences, Rajarata University of Sri Lanka, Saliyapura 50008, Sri Lanka

Full list of author information is available at the end of the article

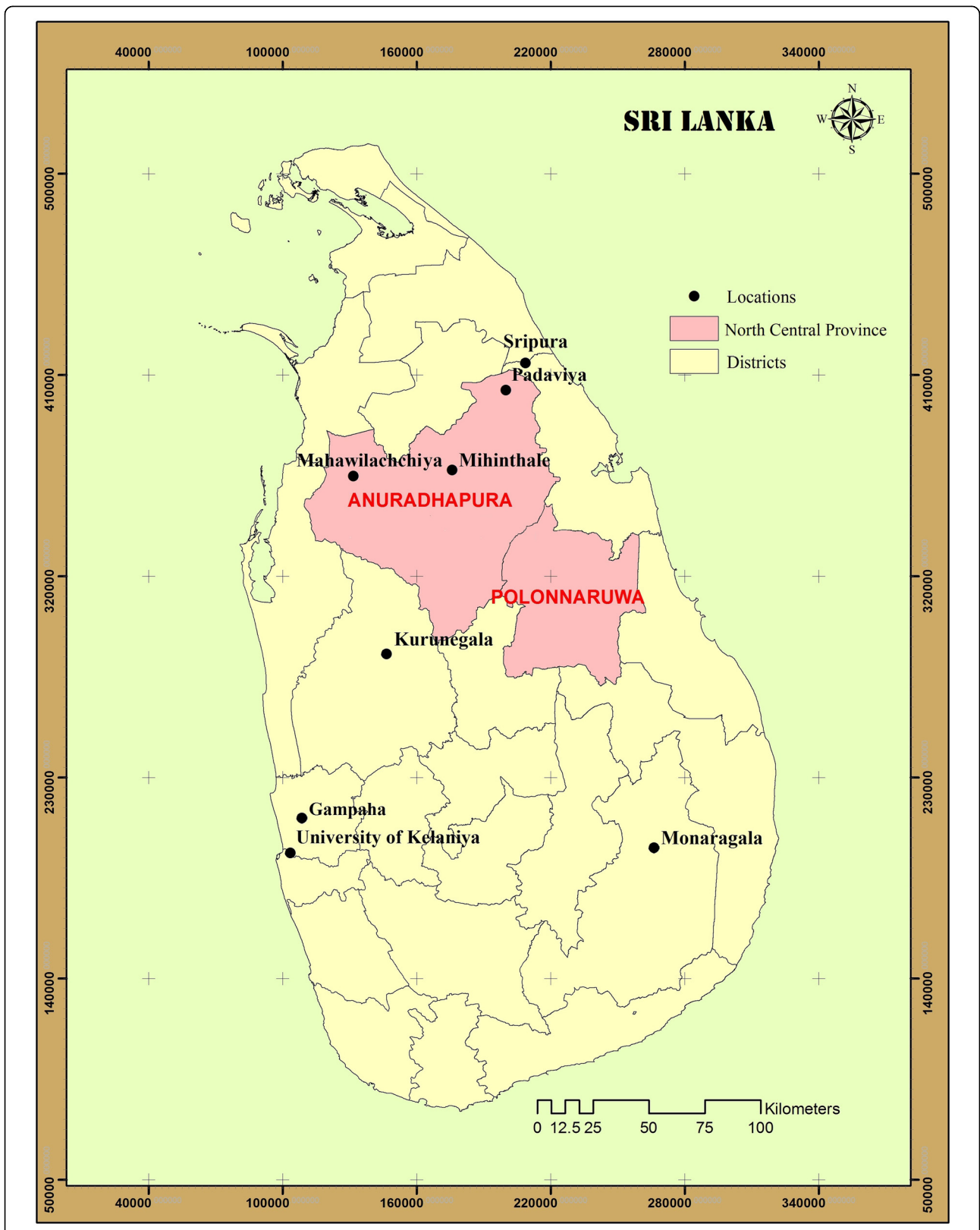
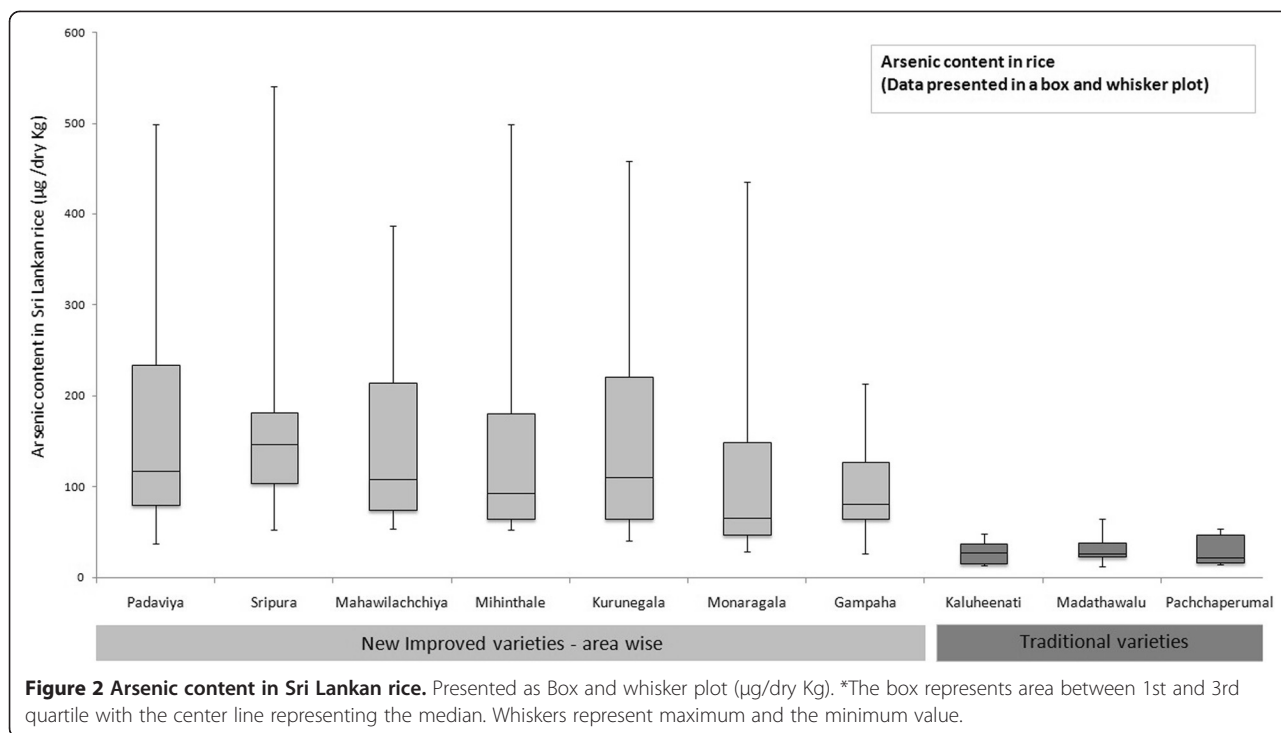


Figure 1 Sample collection sites in a district map of Sri Lanka.



scientific equipment's, VIC, Australia) was used in University of Kelaniya to detect arsenic. Method detection limit (MDL) was 10 µg/Kg. Inductively Coupled Plasma Mass Spectrometer (ICP-MS; HP 4500, Agilent Technologies, Palo Alto, CA) equipped with a quadrupole analyzer and octopole collision/reaction cell was used to detect arsenic and other trace metals at IIRMES lab, CSULB (EPA method 6020 m). Mercury was detected by cold vapor atomic fluorescence spectroscopy using EPA method 245.7m. MDL was 1 µg/g for aluminum and iron, 0.01 µg/g for mercury and 0.025 µg/g for all other trace metals. Samples tested at IIRMES laboratory are not split samples of those tested in Sri Lanka. Data analysis was done using Microsoft excel 2007.

In 1998, rice has been identified as potentially important source of arsenic to humans for the first time (Yost et al.

1998) and is the largest dietary source of inorganic arsenic (Tsuji et al. 2007). Arsenic accumulation in rice is a newly recognized disaster for South-East Asia (Meharg 2004). Rice is particularly susceptible to arsenic accumulation compared to other cereals because it is grown anaerobically in paddy fields that are flooded (Meharg and Zhao 2012). Inorganic arsenic in the soil is inter-converted between the arsenite [As III] and arsenate [As V]. Arsenite predominates in anaerobic environments in contrast to arsenate, which is seen predominantly under aerobic conditions. However, organic monomethylarsonic acid (MMA) and dimethylarsinic acid (DMA) is also present in rice (Meharg and Hartley-Whitaker 2002). Around 50% of total arsenic in rice is inorganic and the remainder is DMA (Heikens 2006). In South Asian countries the percentage of toxic inorganic arsenic is relatively higher (Rahman and Hasegawa 2011).

Table 1 Arsenic content in Sri Lankan rice (µg/dry Kg)

| | New improved varieties - area wise | | | | | | | Traditional varieties | | |
|--------------------------------|------------------------------------|---------|------------------|------------|------------|------------|---------|-----------------------|--------------|-----------------|
| | Padaviya | Sripura | Maha Wilachchiya | Mihinthale | Kurunegala | Monaragala | Gampaha | Kalu Heenati | Mada Thawalu | Pachcha Perumal |
| | N = 20 | N = 17 | N = 25 | N = 17 | N = 19 | N = 11 | N = 11 | N = 10 | N = 10 | N = 10 |
| Minimum | 37.2 | 52.4 | 54.2 | 52.4 | 40.2 | 28.8 | 26.6 | 12.8 | 11.6 | 14.4 |
| Maximum | 498.6 | 540.4 | 387.6 | 498.4 | 458.4 | 435.2 | 212.8 | 48.6 | 64.2 | 54.2 |
| Mean | 162.8 | 186.1 | 144.3 | 141.2 | 152.6 | 129.3 | 96.2 | 28.1 | 30.5 | 28.5 |
| Median | 116.8 | 146.2 | 108.4 | 92.5 | 110.6 | 65.4 | 80.6 | 27.6 | 26.7 | 22.4 |
| Daily As intake from rice (µg) | 51.7 | 59.1 | 45.6 | 44.9 | 48.5 | 41.1 | 30.6 | 8.9 | 9.7 | 9.1 |

Table 2 Trace metal values of traditional varieties of rice-ICP-MS analysis ($\mu\text{g}/\text{dry g}$) please note that the units are different from Table 1

| | Al ¹ | Sb | As ¹ | Ba | Be | Cd ¹ | Cr ² | Co | Cu ² | Fe ² | Pb ¹ | Mn ³ | Hg ¹ | Mo ² | Ni ³ | Se ² | Ag | Sr | Tl | Sn ¹ | Ti | V ² | Zn ² |
|-------------------------|-----------------|-----|-----------------|-----|----|-----------------|-----------------|-----|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----|-----|----|-----------------|-----|----------------|-----------------|
| Kalu Heenati (n = 6) | 4.3 | 0.1 | ND | 0.7 | ND | ND | 0.7 | 0.3 | 2.3 | 7.7 | ND | 9.3 | ND | 0.7 | 1.3 | 0.2 | 0.2 | 0.3 | ND | 0.1 | 0.5 | 1.0 | 11.6 |
| Kuruluthuda (n = 4) | 1.5 | 0.1 | ND | 0.6 | ND | ND | 0.7 | 0.3 | 1.8 | 3.1 | ND | 5.2 | ND | 0.4 | 0.6 | 0.2 | ND | 0.2 | ND | 0.1 | 0.3 | 0.9 | 7.15 |
| Madathawalu (n = 2) | 3.3 | 0.1 | ND | 0.6 | ND | ND | 0.7 | 0.3 | 2.4 | 4.8 | ND | 8.0 | ND | 0.4 | 1.4 | 0.2 | ND | 0.2 | ND | 0.1 | 0.3 | 0.9 | 10.9 |
| Kahawanu (n = 2) | ND | 0.1 | ND | 0.3 | ND | ND | 0.7 | 0.3 | 2.2 | 2.1 | ND | 4.0 | ND | 0.6 | 0.8 | 0.2 | ND | 0.1 | ND | 0.1 | 0.2 | 1.0 | 6.45 |
| Kalu Heenati* (n = 2) | 2.0 | 0.1 | 0.1 | 1.0 | ND | ND | 0.8 | 0.3 | 2.0 | 12.0 | ND | 16.9 | ND | 0.6 | 2.5 | 0.2 | ND | 0.3 | ND | 0.1 | 0.6 | 1.2 | 17.0 |
| Madathawalu* (n = 2) | 2.4 | 0.1 | 0.1 | 3.4 | ND | ND | 0.6 | 0.3 | 1.8 | 7.8 | ND | 17.0 | ND | 0.7 | 1.4 | 0.3 | ND | 0.4 | ND | 0.1 | 0.4 | 1.0 | 17.3 |
| Pachchaperumal* (n = 2) | 7.5 | 0.1 | 0.1 | 1.2 | ND | ND | 0.7 | 0.4 | 2.3 | 7.4 | ND | 11.1 | ND | 0.4 | 9.5 | 0.3 | ND | 0.8 | ND | 0.1 | 0.5 | 1.0 | 12.9 |

¹Toxic elements ²Essential elements and trace elements (in overdose some may be toxic) ³Probably essential elements. (http://apps.who.int/iris/bitstream/10665/37931/1/9241561734_eng.pdf).

*non-organically cultivated.

Rice is the staple food of majority of Sri Lankans and is grown widely in the dry zone. In 2010 annual per capita rice consumption in Sri Lanka was 116 Kg (<http://www.agridept.gov.lk/images/stories/site/PDF/Publication/English/BOOK/proposedplan.pdf>). In the 1960s the International Rice Research Institute based in Philippines, introduced the new improved varieties (NIV). Development of Bg34-8 at Bathalagoda- a major rice research institute in Sri Lanka with a yield potential of 7 t/ha became very popular, replacing the widely grown traditional variety Pachchaperumal. Within 60 years, NIVs have replaced TVs making them almost extinct. Nevertheless, research has shown that TVs are richer in protein, iron, antioxidants, anti-amylase, anti-glycation and glycation reversing activities in comparison to the NIVs (Gunaratne et al. 2013; Premakumara et al. 2013).

We were unable to perform arsenic speciation due to non-availability of facilities and the lack of funds. However, arsenic speciation is important as it varies with type of rice (Booth 2008). According to the Codex Alimentarius, the maximum allowable limit for inorganic arsenic in rice is 200 $\mu\text{g}/\text{kg}$ (<http://www.fao.org/news/story/en/item/238802/icode/>). Assuming that 70% of the total arsenic content exists in the inorganic form, this corresponds to a level of about 286 $\mu\text{g}/\text{kg}$ of total arsenic. As such, 11.6% of the samples of NIVs exceeded this maximum recommended level in polished rice. Person (60 kg of body weight) eating rice obtained from Sripura area consumes approximately 1 $\mu\text{g}/\text{kg}/\text{day}$ of arsenic. Assuming 70% of this is inorganic arsenic the daily exposure would approximate to 0.7 $\mu\text{g}/\text{kg}$. Although this value exceeds the United States environmental protection agency reference (0.3 $\mu\text{g}/\text{kg}/\text{day}$) (http://www.epa.gov/teach/chem_summ/Arsenic_summary.pdf) it is below the value recommended by joint FAO/WHO expert committee on food additives (2-7 $\mu\text{g}/\text{kg}/\text{day}$) for the avoidance of potential cancers of skin, lung and bladder (<http://www.who.int/ipcs/features/arsenic.pdf>). Highest amount of arsenic (540.4 $\mu\text{g}/\text{kg}$) was reported in a sample obtained at Sripura, a farming

colony where the CKDu epidemic originated in the 1990s. (Table 1 and Figure 2).

Our measurements reflected the total arsenic content (both inorganic and organic) and the tested NIVs were polished rice but TV were rice with bran. The Codex Alimentarius standard is established for polished rice and not for brown rice. Rice bran was found to contain approximately seven to nine time higher concentrations of total and inorganic arsenic respectively than those found in the corresponding polished rice (Narukawa et al. 2014; Ruangwises et al. 2012). Our results are similar to those of Chandrajith et al. (2011) who reported an arsenic content of (90–260 $\mu\text{g}/\text{kg}$) in an analysis of 10 samples of rice done by ICP-MS in two other endemic areas for CKDu. However, an earlier analysis done by Instrumental Neutron Activation followed by high-resolution γ -ray spectrometry which revealed an arsenic content of 34–92 $\mu\text{g}/\text{kg}$ in rice (Jayasekera and Freitas 2005). There is no difference between the arsenic content in NIVs from the dry zone and wet zone (Gampaha). Compared to NIVs, arsenic content in TVs cultivated without agrochemicals is low, when tested with AAS, (Table 1). These levels are lower than the MDL when tested with ICP-MS method (Table 2). MDL for arsenic in ICP-MS was 25 $\mu\text{g}/\text{kg}$. However, the TVs cultivated using agrochemicals contains 100 $\mu\text{g}/\text{kg}$ of arsenic (Table 2). Furthermore, a higher amount of Fe, Mn, Ni and Zn were also detected in non-organically cultivated TVs than the organically cultivated TVs. Out of the TVs, Madathawalu showed the highest amount of Ba, Mn and Zn while Kalu Heenati contained the highest amount of iron. None of the tested TVs contained Be, Cd, Pb, Hg and Tl.

These results imply that the arsenic in rice grown in Sri Lanka, originates most probably from the agrochemicals. We have already identified phosphate fertilizers as a main source of arsenic in Sri Lanka (Fernando et al. 2012). Arsenic accumulates in soil following repeated long-term application of contaminated fertilizers. Arsenic in TVs of rice produced without agrochemicals may originate from

already contaminated soil. Farmers started the cultivation of TVs again without agrochemicals only two seasons prior to our study. Arsenic content in Sri Lankan rice is not as high as in West Bengal, Bangladesh and other East Asian countries (Zhu et al. 2008; Rahman and Hasegawa 2011). However, there is no CKDu similar to Sri Lanka in these countries. Therefore, arsenic in rice cannot be linked directly to the CKDu although the additive effects of arsenic in combination with other heavy metals cannot be ruled out. However, given the high per capita consumption, the adverse effects due to arsenic in rice cannot be overlooked, as it is a non-threshold carcinogen and is linked to many non-communicable diseases (Tchounwou et al. 2003; Kapaj et al. 2006). Jasmine rice from Thailand and Basmati rice (both TVs) from South Asia contain the least amount of arsenic (Potera 2007). The arsenic in the rhizosphere is absorbed through ubiquitous aquaglycoproteins and phosphate channels in the rice roots (Mukhopadhyay et al. 2014). Activity of these proteins and channels and many other factors determines whether rice plant is hyper-tolerant or a hyper-accumulator and whether the rice grain contains excessive amounts of arsenic or not (Srivastava et al. 2012).

Water management and rhizosphere manipulation can alter the arsenic concentration in the rice (Meharg and Zhao 2012). Arsenic content in the rice also can be reduced by rinse washing and cooking it in large amount of water (Raab et al. 2009).

Abbreviations

CKDu: Chronic kidney disease of unknown origin; NIV: New improved varieties; TV: Traditional varieties; MDL: Method detection limit; MMA: Monomethylarsonic acid; DMA: Dimethylarsinic acid.

Competing interests

The authors declare that they have no competing interests.

Author's contributions

CJ, PP, MA and SF designed and performed the experiment; CJ, SS and SG analyzed the data; CJ and SS wrote the first draft of the manuscript. All authors read and approved the final manuscript.

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Author details

¹Faculty of Medicine & Allied Sciences, Rajarata University of Sri Lanka, Saliyapura 50008, Sri Lanka. ²Faculty of Science, University of Kelaniya, Colombo 11600, Sri Lanka. ³Department of Health Science, California State University Long Beach, Long Beach, CA 90840, USA. ⁴Faculty of Medicine & Allied Sciences, Rajarata University of Sri Lanka, Saliyapura 50008, Sri Lanka.

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