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Concentration of some heavy metals in rice types available in Shiraz market and human health risk assessment

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ABSTRACT

This investigation was conducted to survey the levels of some heavy metals such as cadmium, lead, chromium, nickel and cobalt in domestic cultivated and imported rice sold on the Shiraz – Iran markets. The potential human health risk assessment was conducted by considering estimated weekly intake (EWI) of toxic metals from eating rice and compared calculated values with provisional tolerable weekly intake (PTWI). The mean values for lead and cadmium in domestic cultivated and imported rice were considerably higher than allowable limits set by FAO/WHO. In combination of recent rice consumption data, the estimated weekly intakes of toxic element were calculated for Iranian population. EWI for cadmium, nickel, chromium through imported and domestic cultivated rice consumption was lower than the PTWI. The EWI for lead were considerably higher than other measured toxic metals. The highest mean level of EWI for lead was observed in some imported rice samples (25.76 μg/kg body weight).

1. Introduction

Food contamination by heavy metals has become a serious problem in recent years because of their potential accumulation in biosystems through contaminated water and soil sources (Lokeshwari & Chandrappa, 2006).

Living organisms require varying amounts of essential heavy metals. Iron, cobalt, copper, manganese, molybdenum, and zinc are required for biochemical processes. Excessive levels can be harmful to human health. Other heavy metals such as mercury, cadmium, chromium, and lead are toxic metals and their accumulation over the time in the living organisms can cause serious effects (Onsanit, Ke, Wang, Wang, & Wang, 2010).

Like many organisms, heavy metals cannot be detoxified by the humans' body mechanism (Castro-González & Méndez-Armenta, 2008). Instead, they tend to accumulate in different tissues such as liver, muscles and bone and threaten the health of humans. Therefore, the heavy metals are among the most of the pollutants which have received attention in many countries and considered the most dangerous category of pollutants in the nutritional compounds.

The growing human population has increased the need for food supply. Rice is good protein and energy source. Demand for this product has increased in market in past five decades. Worldwide, people obtain about 27% and 20% of their energy and protein from rice, respectively (Juliano, 1993).

It is known that Iranian people consume rice as a popular and staple food to supply their daily energy (Jahed Khaniki & Zazoli, 2005). The Rice Research Institute of Iran estimated a weighted average of 40.15 kg (0.110 kg per day) per capita per year consumption of rice. Due to growing demand and lack of sufficient production, a significant proportion of this product is imported from the other countries.

Some studies have indicated excessive amount of heavy metals, especially for cadmium, chromium, and lead in rice grains from different countries (Batista, 2012; Jung, Yun, Lee, & Lee, 2005; Zhang, Watanabe, Shimbo, Higashikawa, & Ikeda, 1998). These investigations have shown that anthropogenic activities such as industrial production, mining and transportation release a high amount of heavy metals to the water and soil sources used in the rice cultivation. Rice cropped even from non-polluted areas may be contaminated because of fertilizers, pesticides and sewage that are used in farm, containing cadmium, lead and other heavy metals (Jahed Khaniki & Zazoli, 2005; Nriagu & Pacyna, 1988).

Iran is dependent on importing of rice. As a result of imposed economic sanctions, inexpensive rice is imported from India, Pakistan and Thailand. However the contents of heavy metals in imported rice types have not been reported by healthcare organizations. In some investigations controversial reports have been published regarding heavy metal contamination of imported rice

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(Cheraghi, Mosaed, Lorestani, & Yousefi, 2012; Malakootian, Yaghmaeian, Meserghani, Mahvi, & Danesh pajouh, 2011; Mirlohi, Morekian, Azadbakht, & Maracy, 2013).

The objective of this study was to investigate and monitor amounts of cadmium, lead, chromium, nickel and cobalt in different types of rice (domestic cultivated and imported) available in Shiraz – Iran Market. Furthermore the levels of heavy metals in the selected rice samples were compared to the allowable limits set by FAO/WHO and the national standards of Iran. Based on the obtained data, estimated weekly intake (EWI) of toxic metals were calculated and compared with provisional tolerable weekly intake (PTWI).

2. Materials and methods

2.1. Sampling

Rice samples were collected from various brands (imported and domestic cultivated) available in retail stores of Shiraz, Iran in 2013. The samples collected covered both the locally produced and the imported rice. These samples represent the major brands on the Iran market. A total of 3–7 different brands were collected within each major group. 210 packed rice samples (weighing from 15 to 20 kg) were selected from markets around Shiraz. For each brand, five samples (for about 250 g) were randomly selected. Rice samples were passed through a 50 mesh (<0.30 mm) sieve, sealed in a plastic box and stored at room temperature until analysis.

The details of the samples are shown in Table 1. The area of production was often only vaguely described, e.g. just stated as being of India, Thailand, and Pakistan.

2.2. Samples preparation and analysis

In this investigation atomic absorption spectrophotometry after dry ashing was performed based on AOAC official method 999.11 (Jorhem, 2000). Rice samples were dried at 105 °C for 48 h. Rice samples were ashed at 450 °C under gradual increase (⩽50 °C/h). 6 M HCl (1 + 1) is added, and the solution was evaporated to dryness. The residue is dissolved in 0.1 M HNO_3 (Jorhem, 2000). The digested solution was analysed for Pb, Cd, Cr, Ni and Co, contents by flame and graphic furnace atomic absorption spectrometer (Shimadzu AA-680, Kyoto, Japan; Shimadzu MUV-IA, Kyoto, Japan). All of the samples were digested as triplicate. Each sample was analysed three times. Concentrations of trace metal in samples were expressed in terms of mg kg^{-1} on a dry weight basis (Jorhem, 2000).

2.3. Quality control procedures

All Chemicals, reagents and standards for heavy metals with maximum purity were obtained from Merck, Germany. High purity de-ionised water used in sample and solution preparation was obtained from Agricultural Biotechnology Research Center, Shiraz University. A clean laboratory and laminar-flow hood capable of producing class II, was used for preparing solutions. Plastic bottles and vessels were cleaned by soaking in 8% (v/v) HNO_3 for 48 h, rinsed three times with de-ionised water and dried in a class II laminar flow hood before use. All operations were performed in, Central Laboratory of Natural Resource and Environment Department of Shiraz University.

A rigorous quality control program was implemented, which included reagent blanks, replicate samples and a certified international reference material of SRM1568b (rice flour, National Institute of Standards and Technology, Gaithersburg, USA). The accuracy of the laboratory result was within the 87.5% confidence intervals of the stated reference values (Table 2).

The mean blank value was deducted from the readings before the result was calculated. The batch blank was used to decide if the results of the batch were acceptable or not, i.e. if the blank was unacceptably high the batch was re-analysed (EN 13804).

2.4. Human exposure assessment

The potential human health risk assessment was conducted by considering the following parameters according to Onsanit et al. (2010). The estimated weekly intakes (EWI), provisional tolerance weekly intake (PTWI) was jointly established by FAO/WHO (2004). The EWI (mg/kg body weight/week) was calculated using the following equation: EWI = C *rice* × [wc *rice*/body weight].

Where C *rice* = average trace metal concentrations in rice (mg kg^{-1} dry weight), wc *rice* = weekly rice consumption (g $week^{-1}$) *per capita* for the Iranian population (110 g per capita per day × 7) as described by Institute of Standards and Industrial Research of Iran (2010), and body weight = average body weight (kg) of the Iranian population (60 kg). The values are expressed in mg $week^{-1}$ $Person^{-1}$.

2.5. Statistical analysis

SPSS, version 21 (Stanford, California, USA), was used for the statistical analysis. Data from the different chemical measurements were subjected to one-way analysis of variance. Statistical significant difference was set at 5%. The comparison of the means was performed using Duncan method. One Sample *t*-test applied to compare the mean values of each element with the available standard ($p < 0.05$).

3. Results and discussion

The levels of heavy metals (Cd, Pb, Cr, Ni and Co) for domestic cultivated rice (3 brands) and imported rice (7 brands) have been represented in Table 3.

Table 1
Types of rice found on Shiraz – Iran market and included in this survey.

Name	Grain	Domestic cultivated	Imported	Imported from
Kamfirooz	Short grain (*japonica*)	***		
Tarom	Short grain (*japonica*)	***		
Kohmare	Short grain (*japonica*)	***		
Padideh	Long grain (*Indica*)		***	India
Abdalsalam	Long grain (*Indica*)		***	India
Abdolsaeid	Long grain (*Indica*)		***	India
Maryam	Long grain (*Indica*)		***	India
Tajmahal	Long grain (*Indica*)		***	India
Khatereh	Long grain (*Indica*)		***	India
Thailand	Long grain (*Indica*)		***	Thailand

Table 2
Chemical results of some elements in SRM1568b (rice flours, NIST, USA) (μg/g dry weight).

Elements	SRM1568b (rice flours, NIST, USA)		
	Certified (μg/g)	Observed (μg/g)	(Observed/certified)·100 (%)
Pb	0.008	0.007	87.5
Cd	0.022	0.020	90
Cr	–	–	–
Co	0.0177	0.019	107
Ni	–	–	–

Each element was analysed three times and means data reported in observed column.

3.1. Toxic elements in domestic cultivated rice samples

Several studies have shown that exposure to heavy metals is associated with serious damage to human health such as anaemia, hypertension and the serious effect in kidneys, lungs, bones (Jahed Khaniki & Zazoli, 2005). Some studies are identifying rice as a potential source of heavy metals exposure in some regions (Batista, 2012; Lin, Wong, & Li, 2004; Shimbo et al., 2001; Zhang et al., 1998).

Cadmium levels in domestic cultivated rice samples varied from 0.27 to 0.47 (mg kg^{-1}). Kamfirooz rice presented the highest mean levels (0.47 mg kg^{-1}) against 0.27 mg kg^{-1} in Kohmare rice. The mean values for cadmium were considerably higher than the allowable limits set by FAO/WHO and National standards of Iran as shown in Fig. 1.

Jahed Khaniki and Zazoli (2005) found an average of 40.8 mg kg^{-1} in rice samples collected in north of Iran. Zazoli, MOhseniBandpei, Ebrahimi, and Izanloo (2010) reported cadmium was not detectable in rice samples collected in other areas of Iran (Babol). Unlike the results of this investigation, Mirlohi et al. (2013) had reported the concentrations of cadmium in rice samples collected from Yazd province was below the permissible limit.

Lead is a very toxic element and the chronic exposure, even at low levels, is associated to several health risks effects (Batista, 2012). In this study, the levels of lead in domestic cultivated rice samples varied from 0.71 to 1.28 mg kg^{-1}. As shown in Table 3, there is no significant difference between lead contents of different domestic cultivated rice samples. These values are much lower than the values observed for rice samples cultivated in the other areas of Iran, reported by Mirlohi et al. (2013), Zazoli et al. (2010), Jahed Khaniki and Zazoli (2005). However as shown in Fig. 2, the average concentrations of lead in samples were above the safe limit set by FAO/WHO and National standards of Iran.

In this investigation concentration of the other heavy metals such as chromium, cobalt, and nickel was evaluated. Nutritional requirements or recommended dietary allowances for these elements have not been established. The SCF stated clearly that the data were not sufficiently conclusive to justify setting any recommended intakes (SCF, 1993). However, commission regulation of European Union (2006) stated the intake of nickel and chromium from the average diet is estimated to be about 75 and 150 μg/day (about 1.25 and 2.5 μg/kg body weight/day) respectively.

Chromium levels in the domestic cultivated rice samples varied from 0.33 to 0.44 mg kg^{-1}. Despite the non statistical significance (Table 3), Kamfirooz rice presented the highest mean levels followed by Tarom and Kohmare samples. The mean value for all samples was above the European Union stated as the average diet. The values found in the present study are quite close to those observed for rice samples in the other study conducted by Sinha et al. (2006).

Nickel is essential for the catalytic activity of some plant and bacterial enzymes. However, biochemical functions of nickel have not been demonstrated in humans and mammals. Nickel levels in domestic rice samples analysed in the present study varied from 0.72 to 0.79 mg kg^{-1}. There is no significant difference between Ni contents of different domestic rice samples. The values found in the present study are much higher than the EU average diet.

Cobalt levels in domestic rice samples analysed in the present study varied from 0.13 to 0.41 mg kg^{-1}. There was no significant difference between the Tarom and Kamfirooz rice samples analysed whilst Kohmare rice presented the lowest mean levels (0.13 mg kg^{-1}).

3.2. Estimation of the weekly intake of toxic elements from domestic rice consumption

Before importing rice from the other countries, domestic cultivated rice was the predominant type of grain consumed in Iran. Thus, for the estimation of weekly intake of toxic elements through rice consumption, we have used the mean levels of the toxic elements obtained for the domestic rice samples analysed in this study. Iranian rice consumption is approximately 110 g per capita per day (Institute of Standards & Industrial Research of Iran, 2010). Estimated weekly intake of toxic elements from domestic rice samples was calculated and shown in Table 4. As can be observed, Kamfirooz rice samples presented higher weekly intake of Cd (5.3 μg/kg body weight), Cr (5.7 μg/kg body weight), Ni (10.2 μg/kg body weight) and Co (1.6 μg/kg body weight) compared to the other types of rice analysed. Results showed the highest estimated weekly intake of lead obtained in Kohmare sample (16.49 μg/kg body weight).

The Codex committee on food additives and contaminants of the joint FAO/WHO food standards program had proposed draft levels for typical daily exposure and theoretical tolerable weekly intake (PTWI) for some of heavy metals. Provisional tolerable weekly intake for the Cd, Pb, Cr and Ni is shown in bottom of Table 4. The result presented in Table 4 shows the estimated

Table 3
Heavy metal contents in domestics and imported rice types (μg/g dry weight).

Rice type	Name	Cd	Pb	Cr	Ni	Co
Domestic	Tarom	0.34 ± 0.01^{B}	0.95 ± 0.21	0.39 ± 0.03	0.78 ± 0.12	0.34 ± 0.05^{A}
Domestic	Kohmare	0.27 ± 0.05^{C}	1.28 ± 0.30	0.33 ± 0.03	0.72 ± 0.04	0.13 ± 0.04^{B}
Domestic	Kamfirooz	0.41 ± 0.02^{A}	0.71 ± 0.15	0.44 ± 0.03	0.79 ± 0.12	0.41 ± 0.05^{A}
Imported	Maryam	0.34 ± 0.02^{B}	1.61 ± 0.37^{A}	0.39 ± 0.02B^{C}	0.87 ± 0.08^{A}	0.11 ± 0.05^{C}
Imported	Abdossaeid	0.29 ± 0.01^{B}	2.00 ± 0.15^{A}	0.36 ± 0.16^{C}	0.90 ± 0.02^{A}	0.31 ± 0.01^{AB}
Imported	Tajmahal	0.36 ± 0.07^{B}	1.62 ± 0.18^{A}	0.38 ± 0.05^{C}	0.89 ± 0.03^{A}	0.17 ± 0.05^{BC}
Imported	Khatereh	0.43 ± 0.03^{A}	0.96 ± 0.21^{B}	0.46 ± 0.06^{ABC}	0.81 ± 0.03^{A}	0.40 ± 0.10^{A}
Imported	Padideh	0.44 ± 0.02^{A}	0.92 ± 0.22^{B}	0.53 ± 0.01^{AB}	0.79 ± 0.06^{A}	0.44 ± 0.06^{A}
Imported	Thailand	0.44 ± 0.03^{A}	0.76 ± 0.19^{B}	0.47 ± 0.07^{ABC}	0.67 ± 0.03^{B}	0.33 ± 0.06^{AB}
Imported	Abdossalam	0.48 ± 0.03^{A}	0.88 ± 0.45^{B}	0.55 ± 0.05^{A}	0.65 ± 0.07^{B}	0.29 ± 0.11^{AB}

Note: results are means ± standard deviation. Means within the same column that have no common letters are significantly different ($p < 0.05$).

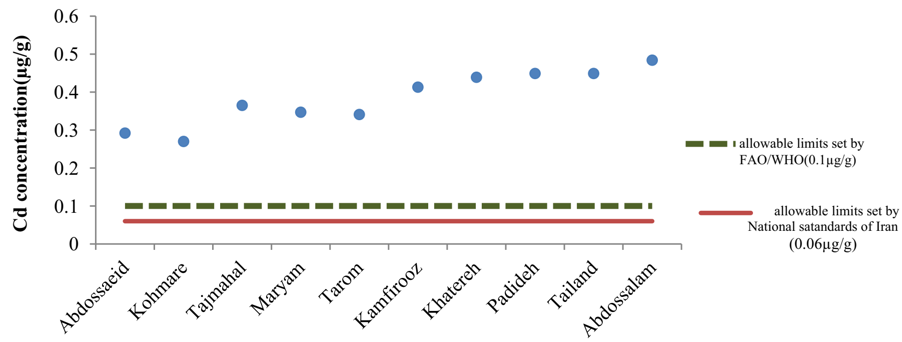

Fig. 1. Cd levels in comparison with allowable limits set by FAO/WHO (2004) and Institute of Standards and Industrial Research of Iran (2010).

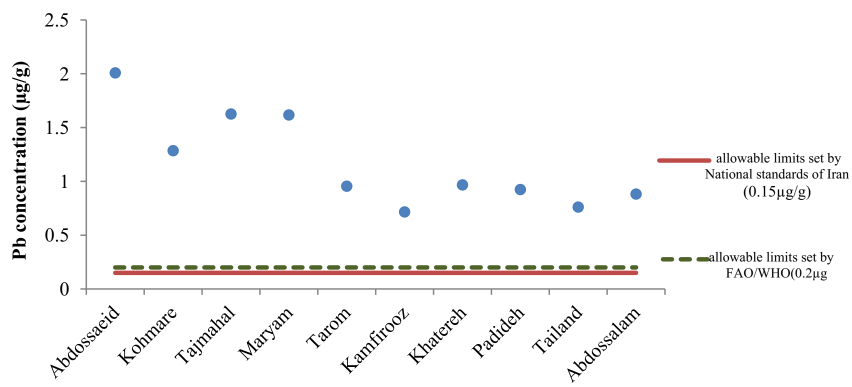

Fig. 2. Pb levels in comparison with allowable limits set by FAO/WHO (2004) and Institute of Standards and Industrial Research of Iran (2010).

Table 4
Calculated EWI values for domestics and imported rice types (µg/kg body weight).

Rice type	Name	Cd	Pb	Cr	Ni	Co
Domestic	Tarom	4.376	12.256	5.095	10.038	4.402
Domestic	Kohmare	3.465	16.491	4.325	9.323	1.668
Domestic	Kamfirooz	5.300	9.189	5.762	10.252	5.300
Imported	Maryam	4.453	20.751	5.069	11.248	1.489
Imported	Abdossaeid	3.747	25.769	4.658	11.624	3.978
Imported	Tajmahal	4.684	20.867	4.928	11.501	2.220
Imported	Khatereh	5.634	12.410	5.955	10.471	5.133
Imported	Padideh	5.762	11.858	6.891	10.247	5.685
Imported	Thailand	4.453	20.751	5.069	11.248	1.489
Imported	Abdossalam	3.747	25.769	4.658	11.624	3.978
PTWI* (µg/kg body weight)		7	25	23.3	35	N*

Note: EWI: estimated weekly intake for toxic elements, based on WHO guidelines.
* There is no PTWI set for cobalt but is used an MTDI (Maximum Tolerable Daily Intake) of 100 µg/kg body weight (=700 µg/kg body weight in a week) for it.

weekly intake of all heavy metals through domestic rice consumption is lower than the provisional tolerable weekly intake.

3.3. Toxic elements in imported rice samples

The composition of heavy metals in imported rice samples is shown in Table 3. The contents of investigated elements in samples were found to be in the range of 0.29–0.48 mg kg^{-1} for cadmium, 0.76–2.00 mg kg^{-1} for lead, 0.36–0.55 mg kg^{-1} for chromium, 0.65–0.90 mg kg^{-1} for nickel and 0.11–0.44 mg kg^{-1} for cobalt. According to these data, lead had the highest concentration, followed by nickel, chromium, cadmium and cobalt, respectively.

Fig. 1 shows, the mean cadmium concentration of imported samples was much higher than the current allowable limits set by FAO/WHO (2004) and Institute of Standards and Industrial Research of Iran (2010). The highest cadmium concentration was observed in Khatereh, Padideh, Thailand and Abdolssalam rice (0.43, 0.44, 0.44 and 0.48 mg kg^{-1}, respectively). Lin et al. (2004) found an average of 0.02 mg kg^{-1} Cd in rice samples collected in Taiwan. In previous survey on imported rice, a mean of 0.041 mg kg^{-1} was reported by Mirlohi et al. (2013).

The variation in Cd levels, possibly indicating influence of other parameters like *e.g.* variation in Cd concentrations in soil, fertilizer, water supply or differences in Cd uptake between genotypes (Jahed Khaniki & Zazoli, 2005; Jorhem et al., 2008).

Lead as a toxicologically relevant element has been brought into the environment by man in extreme amounts, despite its low geochemical mobility and has been distributed worldwide (FAO/WHO, 2009). Plant food may be contaminated with lead through its uptake from ambient air, water and soil (Jahed Khaniki & Zazoli, 2005). Lead levels in imported rice samples varied from 0.76 to 2.00 mg kg^{-1}. Maryam, Tajmahal and Abdoosaeid rice types presented the highest mean levels against the other rice brands. The mean values for lead in all imported samples were considerably above the allowable limits (0.2 µg/g and 0.15 µg/g), as shown in Fig. 2. Similar finding was reported by Mirlohi et al. (2013). In the study of Mirlohi et al. the average concentration of lead in Indian rice samples was higher than the permission levels (254.55 ± 77.2 µg/kg).

As previously mentioned, nutritional requirements or recommended dietary allowances for chromium, cobalt, and nickel have not been established. Nickel is known to be responsible for cancer (oral and in-testinal), depression, heart attacks, kidney dysfunction, low blood pressure, malaise, muscle tremors and paralysis, nausea, skin problems and vomiting (Lokeshappa, Shivpuri, Tripathi, & Dikshit, 2012). Nickel was the second most abundant element which was found in imported rice samples. The nickel level was observed to be highest in Abdoosaeid, Tajmahal, Maryam, Khatereh, and Padideh. These samples had higher concentrations than Thailand and Abdossalam. According European Union (2006); the intake of nickel from imported rice was much higher than the average diet (75 μg/day).

Similar to the present results, high concentration of nickel in different types of rice consumed in Nigeria was reported previously. Umar, Ugonor, and Kolawole (2013) found an average of 3.3 mg kg^{-1} in wild rice samples collected in Kaduna State and Emumejaye (2014) reported nickel was in range of 0.05–2.37 mg/kg in rice samples Consumed in Delta State (Nigeria).

The mean chromium content of imported rice samples is shown in Table 3. The mean chromium level in Maryam, Abdoosaeid and Tajmahal samples had no significant difference ($\geqslant$40 mg kg^{-1}). The highest concentration of Cr observed in Abdossalam rice (55 mg kg^{-1}). The observed concentrations are similar to previously reported chromium levels in Spain rice. Schuhmacher, Domingo, Llobet, and Corbella (1994) found a range of 0.31–0.65 mg kg^{-1} in rice from southern Catalonia, Spain. Singh, Sharma, Agrawal, and Marshall (2010) reported a range of 0.27–7.29 mg kg^{-1} in rice from the wastewater irrigated site of a dry tropical area of India. Some studies have indicated excessive amount of heavy metals including Zn, Cu, Pb, Mn, Ni, Cr, Cd, in crops and vegetables is associated with sewage and industrial wastewater used for irrigation in agriculture activities (Gupta, Khan, & Santra, 2008; Sharma, Agrawal, & Marshall, 2007; Sinha et al., 2006). In some brands of rice consumed in Delta state, Emumejaye (2014) reported a mean level of 0.5 mg kg^{-1}. Commission regulation of European Union (2006) reported the intake of chromium from the average diet is estimated to be about 150 μg/day (2.5 μg kg^{-1} body weight day^{-1}). In comparison to this estimation, the mean value for all imported rice was above the EU stated as the average diet.

The cobalt levels in imported rice sold on the Shiraz market ranged between 0.11 and 0.044 mg kg^{-1}, average 0.30 mg kg^{-1}, with no significant difference between Abdossalam, Tajmahal, Abdossaeid, Khatereh, Padideh and Thailand (Table 3). The lowest level of cobalt was observed in Maryam rice brand.

3.4. Estimation of the weekly intake of toxic elements from imported rice consumption

The calculated EWI of cadmium, lead, chromium, nickel and cobalt were based on the average consumption of rice (110 g day^{-1}) by Iranian people (according to the Institute of Standards and Industrial Research of Iran) is shown Table 4.

Estimated weekly intakes of cadmium in imported rice samples varied from 3.74 to 5.76 (μg/kg body weight). Padideh rice presented the highest mean levels (5.76 μg/kg body weight) against 3.74 (μg/kg body weight) in Abdossalam and Abdossaeid rice.

The EWI for lead was considerably above the other measured toxic metal (11.85–25.76 μg/kg body weight). The highest mean levels of EWI for lead was observed in the Abdossalam and Abdossaeid rice samples (25.76 μg/kg body weight).

EWI of chromium, nickel and cobalt in the imported rice samples were found to be in the range of 4.65–6.89 (μg/kg body - weight) for Cr, 10.24–11.62 (μg/kg body weight) for Ni and 1.48–5.68 (μg/kg body weight) for Co.

Provisional tolerable weekly intake for the investigated metals is shown in Table 4. According to the results shown in Table 4, the estimated weekly intake of cadmium, nickel, chromium through imported rice consumption was lower than the provisional tolerable weekly intake. Comparing the results in Table 4 about lead content of imported rice, it appears that the obtained values of some samples (Abdossalam and Abdossaeid) were higher than the provisional tolerable weekly intake.

4. Conclusion

As results of this investigation, the mean values for lead and cadmium in domestic and imported rice samples were considerably higher than the allowable limits set by FAO/WHO. The estimated weekly intake for lead in some brands of imported rice was considerably above than provisional tolerable weekly intake. The event was not observed in the domestic rice. According to the results of this study approximately 65.9% of the daily intake of cadmium comes from rice. Health risk will increase with consumption of other contaminated food that was not evaluated in this study. So, measurement of heavy metals in the complete diet over a specified period is recommended. According to the reports of the Institute of Standards and Industrial Research of Iran, rice is a dominating staple food in the diet of Iranian population. In the Shiraz, rice is the main food eaten one or two times a day. Thus, it is necessary that certified agencies monitor and evaluate the heavy metal levels in domestic and imported rice regularly.

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