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Essential and toxic heavy metals in cereals and agricultural products marketed in Kermanshah, Iran, and human health risk assessment

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ABSTRACT

Levels of some essential and toxic heavy metals such as lead, cadmium, chromium, nickel, zinc and copper in cereals and agricultural products obtained from the markets in Kermanshah city, west Iran, were determined by inductively coupled plasma-optical emission spectrometry (ICP-OES). The average concentrations for lead and cadmium in some cereals were higher than the maximum levels set by the Codex Alimentarius. A potential human health risk assessment was conducted by calculating estimated weekly intake (EWI) of the metals from eating cereals and comparison of these values with provisional tolerable weekly intake (PTWI) values. In combination with recent cereal consumption data, the EWIs of heavy metals were calculated for the Kermanshah population. EWI data for the studied metals through cereal consumption were lower than the PTWI values. Cr, Ni, Zn and Cu levels in all samples analysed were within the ranges reported for similar cereals from various parts of the world.

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Introduction

Essential and toxic heavy metals exist in natural and contaminated environments and cannot be easily detoxified via degradation, resulting in their persistence in the environment. Many of these metals, such as Cd, Pb and Cr, are carcinogens and are involved in several diseases, including Alzheimer's disease, Parkinson's disease, multiple sclerosis, osteoporosis, developmental disorders and failure of several organs (e.g., heart, kidney, lungs, immune system) (Jomova & Valko 2011; Zakir Hossain & Brennan 2011). Some of these metals, such as Ni, Cu and Zn, are essential minerals that are needed for a variety of biomolecules to maintain the normal structure, function and proliferation of cells (Khajeh et al. 2010). In addition, these metals may show potential toxicity in excessive amounts, especially in certain genetic disorders (Zheng et al. 2008).

Rapid and unorganised urban and industrial developments have contributed to the elevated levels of heavy metals in the urban environment of developing countries such as Iran (Maleki & Zarasvand 2008; Naseri et al. 2015), China (Wong et al. 2003), Egypt (Radwan & Salama 2006) and India (Sharma et al. 2008). The growing human population has increased the need for food supply. Rice and cereals are good protein and energy sources. Demand for these products has increased market volumes in the past five decades. Worldwide, people obtain about 27% and 20% of their energy and protein from rice and cereals, respectively (Juliano 1993). It is known that Iranian people consume cereals and rice as a popular and staple food to supply their daily energy (Jahed Khaniki & Zazoli 2005). The Agricultural Research Institute of Iran estimated a weighted average of 40.15 kg (0.110 kg per day) per capita per year consumption of cereals (Naseri et al. 2015). Due to growing demand and lack of sufficient production, a significant proportion of these products are imported from other Asian countries like Pakistan, India, Thailand and China, where soil and groundwater pollution with high level of toxic metals has been reported (Duxbury et al. 2003; Meharg & Rahman 2003; Roychowdhury et al. 2003; Das et al. 2004; Rahman et al. 2013). Increased levels of toxic metals in agricultural soils and their uptake in cereals, rice, vegetables and other food crops have become a serious health issue in these regions (Williams et al. 2006; Islam et al. 2014).

Some studies have indicated excessive amounts of heavy metals in cereals and rice from different countries (Ali & Al-Qahtani 2012; Behbahani et al. 2013; Akinyele & Shokunbi 2015; Cai et al. 2015). These investigations have shown that anthropogenic activities such as

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industrial production, mining and transportation release a high amount of heavy metals into water and soil used in agricultural cultivation. Foods and agricultural products are the principal source of metal exposure for humans. Thus, monitoring of essential and heavy metals, particularly in foods and agricultural products, is very important.

So far there has been very little published information on delineating the contamination scope of bean, grain and cereals in this area, the extent of human exposure and the potential health consequences. In the presented work, the levels of essential and toxic heavy metal concentrations in different types of cereals and agricultural products as consumed in Kermanshah province were determined and an assessment on human health risks due to the consumption of those cereals has been conducted. The results of this study are useful for pollution control and risk management of heavy metals.

Materials and methods

Reagents and solutions

Analytical grade chemicals were employed in the processing of all samples. Nitric acid (HNO_3, 65%, suprapur) and hydrogen peroxide (H_2O_2, 30%) were obtained from Merck (Darmstadt, Germany). The metal standard solutions used for calibration were produced by diluting a stock solution of 1000 mg L^{-1} of the metals as supplied by Sigma Chem. Co. (St. Louis, USA). Ultrapure water, obtained using a Milli-Q system (Millipore, Bedford, MA, USA), was used in all experiments. All plastic and glassware was properly cleaned by soaking them in 2 M nitric acid and rinsed thoroughly with deionised water prior to use. The Certified Reference Materials NCS ZC 73008 Rice, NCS ZC 73009 Wheat (China National Analysis Center for Iron and Steel, Beijing, China) and SRM1568b Rice flour (National Institute of Standards and Technology, Gaithersburg, USA) were used for method validation.

Sample collection

Bean, grain and cereal samples were collected from various brands (imported and domestic cultivated) available in the retail stores of Kermanshah, Iran, in 2014. The samples collected covered both the locally produced and the imported cereals. These samples represent the major brands on the Iranian market. A total of 3–11 different brands were collected within each sample. For each brand, three samples (about 50 g) were randomly selected; 150 packed cereal samples were selected from markets around Kermanshah. Cereal samples were passed through a 50 mesh (<0.30 mm) sieve, sealed in a plastic box and stored at room temperature until analysis.

Sample preparation and analysis

The different cereal samples were cleaned with water and finally washed with distilled water and then dried at 40°C for 100 min. Dried samples were milled to powder with a commercial blender with stainless steel blades (Mixer B-400, BÜCHI, Flawil, Switzerland), homogenised and passed through a 50 mesh (<0.30 mm) sieve. One gram of each homogenised sample was weighed into a porcelain crucible and dry-ashed in a muffle furnace model CWF 1200 (Carbolite Limited, Hope Valley, UK) by stepwise increase in temperature up to 500°C within 3 h and then leaving to ash at this temperature for the next 8 h. The residue was dissolved in a mixture of 8 mL nitric acid (3 M) and hydrogen peroxide (30%) mixture in a 3:1 ratio, filtered into a 25 mL volumetric flask using Whatman filter paper (Whatman Inc., Clifton, NJ, USA), made up to the mark with deionised water and stored under refrigeration at −20°C until analytical determination. Metal analysis was performed simultaneously with an inductively coupled plasma-optical emission spectrometer (ICP-OES), PerkinElmer model 7300 DV (PerkinElmer, Waltham, MA, USA), coupled to a V-groove nebuliser, quartz glass Scott spray chamber and equipped with a charge coupled device (CCD) detector. The experimental conditions are given in Table 1.

Quality assurance

In order to verify that the analytical methodology used was adequate, heavy metals were analysed in three certified reference materials, NCS ZC 73008 Rice, 73,009 Wheat and SRM 1568b Rice flour, under specified conditions. The results indicated good agreement between certified and observed values (Table 2). Relative recoveries observed for the reported certified materials were between 87.5% and 109%. Furthermore, the standard addition technique was performed on the reference material to evaluate the presence of matrix interferences. A comparison of slopes using external standard calibrations versus standard

Table 1. ICP-OES operating conditions.

RF generator power	1.55 kW
Frequency of RF generator	42 MHz
Plasma gas flow rate	16 L min^{-1}
Auxiliary gas flow rate	1.6 L min^{-1}
Viewing height (above coil)	5 mm
Nebuliser pressure	140 kPa
Pump rate	2.5 mL min^{-1}

Table 2. Analytical results of some metals in standard reference materials (µg/g dry weight).

Standard reference material	Metal	Certified	Observed	Recovery (%)
NCS ZC 73008 Rice	Pb	0.08	0.077	96.2
	Cd	0.087	0.082	94.2
NCS ZC 73009 Wheat	Pb	0.065	0.068	104.6
	Cd	0.018	0.019	105.5
SRM1568b Rice flour	Pb	0.008	0.007	87.5
	Cd	0.022	0.024	109

additions in SRM 1568b Rice flour showed good agreement, indicating no matrix effects on the analysis of heavy metals. The limits of detection (LODs) were calculated using the formula $C_L = 3S_b/m$, where S_b is the standard deviation of six replicate blank measurements and m is the slope of the calibration graph. LODs between 0.1 and 0.35 µg kg^{-1} for different metal ions were obtained.

Human health risk assessment

The potential human health risk was assessed by considering the estimated weekly intakes (EWIs) and provisional tolerable weekly intake (PTWI) parameters according to Naseri et al. (2015), who reported a human health risk assessment of some heavy metals in rice available in Shiraz market, Iran. EWI (mg/kg body weight/week) was calculated using the following equation:

$$EWI = C_{\text{cereal}} \times \frac{WC_{\text{cereal}}}{BW}$$

where C_{cereal} = average metal concentration in cereal sample (mg kg^{-1} dry weight), WC_{cereal} = weekly cereal consumption (g $week^{-1}$) per capita for the Iranian population (110 g per capita per day × 7), as described by the Institute of Standards and Industrial Research of Iran (2010), with a BW = average body weight (kg) of the Iranian population of 60 kg. PTWI intake values were taken from Table 4 in Naseri et al. (2015).

Statistical analysis

Descriptive statistics were calculated using Microsoft Office Excel 2010. Univariate and multivariate statistical analyses as one-way ANOVA, inter-metal correlations and principal component analysis (PCA), were performed using SPSS software version 21 (SPSS Inc., Chicago, IL, USA).

Results and discussion

Levels of toxic and essential heavy metals in different cereals

Pb, Cd, Cr, Ni, Zn and Cu (mg kg^{-1} dry weight) concentrations in different types and brands of cereals collected from Kermanshah city are given in the database as mean ± standard deviation (SD) of triplicate analyses.

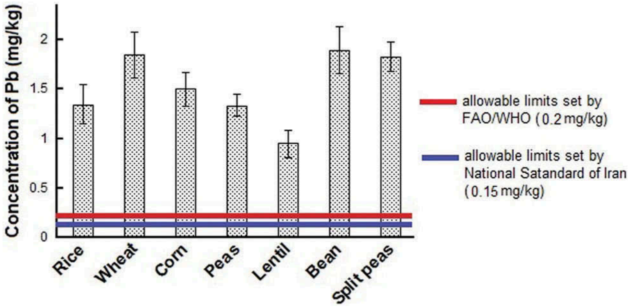

Figure 1. Mean Pb concentrations in cereals marketed in Kermanshah, Iran, compared with limits set by the Codex Alimentarius (1995) and the Institute of Standards and Industrial Research of Iran (2010).

Pb is a very toxic element and chronic exposure, even at low levels, is associated with several health risks (Batista 2012). In this study, Pb levels in different brands of rice samples varied from 0.99 to 2.3 mg kg^{-1}, with a mean concentration of 1.35 mg kg^{-1}. As shown in Figure 1, all mean Pb concentrations were above the limit set by the Codex Alimentarius and the National standards of Iran. However, these results are similar to those obtained by Naseri et al. (2015) and lower than those reported by Mirlohi et al. (2013), Zazoli et al. (2010) and Jahed Khaniki and Zazoli (2005), where Pb levels in wheat, corn, peas, lentil, bean and split peas ranged from 0.54 to 4.89 mg kg^{-1}, 0.70 to 1.95 mg kg^{-1}, 0.90 to 3.23 mg kg^{-1}, 0.74 to 1.36 mg kg^{-1}, 1.26 to 2.96 mg kg^{-1} and 1.45 to 2.44 mg kg^{-1}, respectively. Mean Pb concentrations in cereal samples of different brands of wheat, corn, peas, lentil, bean and split peas were 1.85, 1.50, 1.34, 0.95, 1.90 and 1.83 mg kg^{-1}, respectively (Figure 1). Ekholm et al. (2007) reported Pb in a range of 0.04–0.23 mg kg^{-1} in cereal samples consumed in Finland and Cabrera et al. (2003) reported Pb levels ranging from 0.32 to 0.70 mg kg^{-1} in legumes and from 0.14 to 0.39 mg kg^{-1} in nuts marketed in Spain. Apparently, in these two studies, lower levels were found.

Cd is deemed as an element capable of producing chronic toxicity even when it is present at concentrations of around 1 mg kg^{-1} (Safiur Rahman et al. 2012). The Cd content in different brands of rice samples ranged from 0.041 (Fareidonkenar) to 0.089 mg kg^{-1} (Local Tarom), with an average content of 0.055 mg kg^{-1}. The mean value for cadmium was lower than the limits set by Codex Alimentarius and National standards of Iran, as shown in Figure 2. However, Cd concentrations in Astaneh, Tarom and Local Tarom rice samples were higher than the maximum permitted level in the National standards of

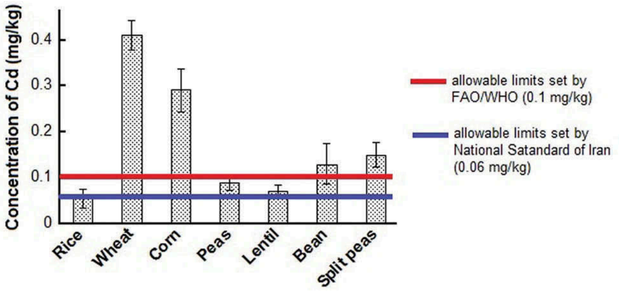

Figure 2. Mean Cd concentrations in cereals marketed in Kermanshah, Iran, compared with limits set by the Codex Alimentarius (1995) and the Institute of Standards and Industrial Research of Iran (2010).

Iran. These results are similar to those obtained by Mirlohi et al. (2013), lower than those reported by Jahed Khaniki and Zazoli (2005) and Naseri et al. (2015) and higher than those reported by Zazoli et al. (2010). The differences could be due to several factors, including sample size, analytical techniques employed, season of the year and agricultural soil. In this study, the levels of Cd in different brands of wheat, corn, peas, lentil, bean and split peas ranged from 0.038 to 1.99 mg kg^{-1}, 0.05 to 1.048 mg kg^{-1}, 0.033 to 0.40 mg kg^{-1}, 0.045 to 0.127 mg kg^{-1}, 0.065 to 0.186 mg kg^{-1} and 0.107–0.205 mg kg^{-1}, respectively. As shown in Figure 2, mean Cd concentrations in different brands of wheat, corn, peas, lentil, bean and split peas were 0.41, 0.29, 0.09, 0.07, 0.13 and 0.15 mg kg^{-1}, respectively. The mean concentrations of Cd in all cereals were higher than the safe limit set by Codex Alimentarius and National standards of Iran, except for peas and lentil. The mean concentrations of Cd in different brands of peas and lentil were lower than the safe limit set by Codex Alimentarius and higher than the safe limit as set by the National standards of Iran. The highest amount of cadmium was found in the Sardari wheat sample (1.99 mg kg^{-1}) and the lowest amount of Cd was found in the Mahidasht peas sample (0.033 mg kg^{-1}). Akinyele and Shokunbi (2015) reported cadmium concentrations in the range of <0.01 mg kg^{-1} in cereals to 0.09 mg kg^{-1} in groundnut. Onianwa et al. (2000) reported cadmium concentrations in the range of 0.06–0.28 mg kg^{-1} in cereals and Ekholm et al. (2007) reported ranges around 0.1 mg kg^{-1} in wheat bran, which are all quite close to those observed in this study.

Cr is an essential mineral for humans and has been related to carbohydrate, lipid and protein metabolism (Berdanier & Zempleni 2009). Cr was detected in all examined cereal samples and its concentration ranged from 0.287 to 1.135 mg kg^{-1}. The minimum and maximum Cr contents were found in Sahneh corn and Azar Shahr split peas, respectively. Mean Cr concentrations in different brands of rice, wheat, corn, peas, lentil, bean and split peas were 0.57, 0.55, 0.74, 0.66, 0.48, 0.65 and 0.75 mg kg^{-1}, respectively. The observed concentrations are similar to previously reported Cr levels in Iranian rice (Mirlohi et al. 2013; Naseri et al. 2015). Akinyele and Shokunbi (2015) found a range of 0.02–0.58 mg kg^{-1} in cereals from Nigeria. In nut and legumes from Spain, Cabrera et al. (2003) reported ranges of 0.25–1.05 mg kg^{-1} and 0.05–0.60 mg kg^{-1}, respectively. This shows that Cr concentrations in the samples analysed are within the range found in the scientific literature.

Ni normally occurs at very low levels in the environment and can cause variety of pulmonary adverse health effects, such as lung inflammation, fibrosis, emphysema and tumours (Forti et al. 2011). Ni was not detected in six samples of rice, one sample of wheat and one sample of corn. The lowest and highest contents of Ni measured were 0.017 mg kg^{-1} (Kozaran peas) and 4.57 mg kg^{-1} (Dineh white bean), respectively. The mean concentrations of Ni in different brands of rice, wheat, corn, peas, lentil, bean and split peas were 0.004, 0.23, 0.10, 0.22, 0.58, 2.50 and 0.10 mg kg^{-1}, respectively. Ni contents of cereals were reported to range from 0.06 to 1.86 mg kg^{-1} by Ekholm et al. (2007). Ni levels in tubers, legumes and cereals have been reported in the range of 0.93–1.79 mg kg^{-1}, 3.47–7.00 mg kg^{-1} and 1.00–1.89 mg kg^{-1}, respectively (Onianwa et al. 2000). Cabrera et al. (2003) also reported Ni levels in legumes and nuts in the range of 0.02–0.35 mg kg^{-1} and 0.10–0.64 mg kg^{-1}, respectively. These data show that our Ni values are within the range reported in other literature references.

Zn and Cu are essential minerals that are needed for a variety of biomolecules to maintain the normal structure, function and proliferation of cells (Khajeh et al. 2010). In addition, these metals may be toxic in excessive amounts, especially in certain genetic disorders (Zheng et al. 2008). Zn concentrations of different types and brands of cereals evaluated in this study ranged from 3.55 to 23.61 mg kg^{-1}. The mean concentrations of Zn in different brands of rice, wheat, corn, peas, lentil, bean and split peas were 7.83, 13.81, 9.20, 10.46, 14.40, 10.99 and 12.45 mg kg^{-1}, respectively. Khajeh et al. (2010) found a range of 8.50–18.40 mg kg^{-1} in cereals from Iran. Zn levels in cereals have been reported in the range of 6.65–46.99 mg kg^{-1} (Akinyele & Shokunbi 2015) and 32.60–70.20 mg kg^{-1} (Cabrera et al. 2003). Onianwa et al. (2001) and Ekholm et al. (2007) also reported Zn levels of 3.54–33.40 mg kg^{-1} and 8.00–89.00 mg kg^{-1} in cereals, respectively. These data suggest that the Zn values obtained in this study are within the range found in literatures. Cu was detected in all examined cereal samples, and its concentration ranged from 0.55 to 6.77 mg kg^{-1}. The minimum and maximum Cu concentrations were found in Sarab Nilofar wheat and Golestan split peas, respectively. The mean concentrations of Cu in different brands of rice, wheat, corn, peas, lentil, bean and split peas were 2.04, 2.13, 1.06, 4.64, 4.23, 4.62 and 5.43 mg kg^{-1}, respectively. The Cu concentrations in cereals were reported in the range of 1.20–3.10 mg kg^{-1} (Khajeh et al. 2010), 1.59–10.56 mg kg^{-1} (Akinyele & Shokunbi 2015), 1.53–3.07 mg kg^{-1} (Onianwa et al. 2001) and 2.00–14.00 mg kg^{-1} (Ekholm et al. 2007). It is clear that on a general note, the Cu values reported in this study are in the range of those reported in literature.

Estimation of the weekly intake of heavy metals from cereals consumption

Exposure of consumers and related health risks are usually expressed as PTWI, a reference value established by Joint FAO/WHO. PTWI for Pb, Cd, Cr, Ni, Zn and Cu is shown in the bottom line of Table 3. Thus, for the estimation of weekly intake of heavy metals through cereal consumption, the mean levels of heavy metals obtained for the different brands of cereal samples analysed in this study were used. Iranian cereal consumption is approximately 110 g per capita per day (Institute of Standards and Industrial Research of Iran 2010). EWI of heavy metals from cereal samples was calculated and is shown in Table 3. As can be observed, bean samples presented higher weekly intake of Pb (24.38 µg kg^{-1} body weight) and Ni (32.08 µg kg^{-1} body weight), and split peas samples presented higher weekly intake of Cr (9.63 µg kg^{-1} body weight) and Cu (69.68 µg kg^{-1} body weight) compared to the other types of cereals analysed. Results showed the highest EWI for Cd and Zn was obtained in wheat samples (5.26 µg kg^{-1} body weight) and lentil samples (184.80 µg kg^{-1} body weight), respectively. The results presented in Table 3 show that the EWI of all heavy metals through cereal consumption is lower than the PTWI.

Table 3. Calculated EWI values for different cereals (µg kg^{-1} body weight).

Cereal type	Pb	Cd	Cr	Ni	Zn	Cu
Rice	17.32	0.70	7.31	0.05	100.48	26.18
Wheat	23.74	5.26	7.06	2.95	177.22	27.33
Corn	19.25	3.72	9.50	1.28	118.06	13.60
Peas	17.20	1.15	8.47	2.82	134.23	59.54
Lentil	12.19	0.90	6.16	7.44	184.80	54.28
Bean	24.38	1.67	8.34	32.08	141.04	59.29
Split peas	23.48	1.93	9.63	1.28	159.77	69.68
PTWI (µg kg^{-1} body weight)	25	7	23.3	35	420	500

Conclusion

This study provides additional data on heavy metal contents of cereals marketed in Kermanshah, Iran, and is useful in risk assessment of consumer exposure to the expected metals. The mean concentrations of Pb and Cd in some cereals were higher than the safe limit set by Codex Alimentarius and National standards of Iran. Furthermore, the EWIs for the studied metals are below the limits set by Codex Alimentarius for metal intake, based on body weight for an average body weight of the Iranian population (60 kg). Thus, the consumption of average amount of these cereals does not pose a health risk to the consumers. The accuracy of the proposed procedure was statistically evaluated by comparing the results obtained for certified materials with their certified values, which resulted to be very good. In Kermanshah, cereals are eaten one or two times a day. Thus, it is necessary that certified agencies monitor and evaluate the heavy metal levels in these products.

Disclosure statement

No potential conflict of interest was reported by the authors.

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