Health risk assessment of potentially toxic elements in common cultivated rice (Oryza sativa) emphasis on environmental pollution

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ABSTRACT
Heavy metal accumulation and contamination is a global serious dilemma due to toxicity, abiotic characteristics, abundant resources, and cumulative behavior of heavy metals. This evaluation aimed at measuring and evaluating seven heavy metals (Ni, Cu, As, Zn, Pb, Cr, and Cd) content in rice (Oryza sativa) and associated health risks was carried out for consumers in Khuzestan Province, Iran. We choose 20 stations in 5 strategic regions of rice cultivation. The average carcinogenic risk of arsenic by rice consumption in Khuzestan Province was estimated to be 3 adults and 4 children per 10,000 populations. The average concentrations of heavy metals in rice and rice field soils were as follows: In rice: Ni (0.38 ± 0.25), Cu (5.29 ± 3.04), As (0.087 ± 0.066), Zn (17.89 ± 3.35), Pb (0.17 ± 0.08), Cr (0.69 ± 0.25) and Cd (0.058 ± 0.027) mg kg⁻¹. In soils: Ni (49.32 ± 16.71), Cu (36.69 ± 19.74), As (5.02 ± 3.42), Zn (71.20 ± 12.72), Pb (10.99 ± 4.299), Cr (81.23 ± 22.31), and Cd (0.18 ± 0.072) mg kg⁻¹, respectively. Nickel showed the highest standard deviation of the SPI with an average of 2.38 ± 0.81. The average CPI for rice was in the following descending order: Cd (0.53 ± 0.31) > As (0.36 ± 0.068) > Cd (0.69 ± 0.25) > Pb (0.84 ± 0.41) > Cr (0.69 ± 0.25) > As (0.65 ± 0.52) > Cu (0.95 ± 0.45). Cadmium and nickel respectively showed the highest and lowest BAF with an average of 0.302 and 0.0067. According to the US Environmental Protection Agency (USEPA) classification, the overall HI for adults in four regions was at Level 3 (medium) and for children in two regions at Level 4 (high). The corresponding values for cadmium were 5 adults and 8 children per 100,000 populations. This means that rice consumption in Khuzestan Province could cause cancer during the lifetime of rice consumers.

1. Introduction
One of the most consumed grains across diverse agricultural crops, in the world is Rice (Oryza sativa) (Malakootian et al. 2011, Shokrzadeh and Rokni 2013, Djahed et al. 2018). Rice is the essential diet of more than a moiety of humanity in Asian countries, African and South & North American countries (Mehria 2013, Al-Saleh and Abduljabbar 2017, Sharma et al. 2018). As well as, rice is one of the staple ingredients of the repast among the various foodstuffs in Iran (Naseri et al. 2015).

Cardinal trajectory of heavy metals intake by mankind has occurred the dietary intake via infected cuisines (Miri et al. 2017, Chen et al. 2018). HMs are one of the dangerous pollutants of the environment that they have prolix biological half-lives, are non-biodegradable, and someone’s are toxicant even at tinge concentrations (Sanchooli Moghaddam et al. 2016, Ghasan et al. 2017, Ghasemi et al. 2017). HM toxicity can be decrease or damaged central and intellectual nervous function. Continuity long-term exposure with some HMs (or their compounds) may ensue cancer (Salakinkop and Hunshal 2014, Miri et al. 2018a).

Pollution caused by the accumulation of toxic metals is one of the first ventures to soil, air and water (Gholizadeh et al. 2019). Given the central role of soil and water in the plants’ growth and crop production, plants and their products are a sect of the pollution in their own context (Arunakumara et al. 2013). Soil may be contaminated due to the accumulation of heavy metals (HMs) through human activities (anthropogenic) such as mining, farming, fertilizers, irrigation with wastewater, sewage sludge on agricultural lands, construction, livestock manure, use of pesticides and industrial processes (Khan et al. 2015, Miri et al. 2016, Al-Saleh and Abduljabbar 2017, Momtazan et al. 2019, Idani et al. 2020). As HMs are non-bio-degradable, they stay for a long time in environmental ecosystems. They enter rice paddies with chemical fertilizers and...
irrigation water and thus comer the human nutritional cycle. Among the essential agricultural products, rice is a special one because of the high uptake and accumulation rate of HMs (Chaney et al. 2004). The entrance of these metals into the food chain and their critical concentrations may afford metabolic, neurological disease and physiological contrary effects on living organisms. several investigations administrated in Iran have revealed that rice contamination is provided by some HMs (Naseri et al. 2015). 95% of total rice in the world is produced in Asia also Iran is appropriated an area of 630 hectares to cultivate this crop that is achieved annually about 2.3 million tons from these lands, Khuzestan province shares an average of 300,350 thousand tons, which is about 13.63% of the total rice production of Iran (Yearbook 2017).

The high potential of Khuzestan Province for production of diverse, strategic and industrial crops on the one hand and the effect of industrial activities such as refineries, petrochemical complexes, steel industries, sugarcane factories, and oil and gas companies in this province; improper use of fertilizers and pesticides and incorrect irrigation practices on the other hand (Rastmanesh et al. 2016). Significantly increase the importance of this research work. Accordingly, this study aims at determining contamination levels of nickel, arsenic, chromium, zinc, copper, lead, and cadmium in Khuzestan Province’s rice and rice field soils to assess the health risk of rice consumption for children and adults. The second objective is to determine the bioaccumulation factor (BAF) of the mentioned HMs in rice grains.

2. Materials and methods

2.1. Study areas

Khuzestan Province is one of the 31 provinces in Iran, is located in the southwest of Iran, with an area of 63238 km². It borders Iraq on the west and the Persian Gulf on the south (Khaefi et al. 2016, 2017, Goudarzi et al. 2018, 2019, Dastoorpoor et al. 2019, Idani et al. 2020).

According to the 2016 report of the Iranian Statistics Center, this province has 24 counties, 64 cities, and 132 villages (Yearbook 2017). Khuzestan Province had a population of 4711000 in 2016.

The Karun River, as the most important river in Iran, passes through large areas of Khuzestan Province and is closely associated with a group of essential industries including metallurgical, petrochemical, and petroleum industries and with a large part of urban and agricultural wastewater. Therefore, the presence of any metallic elements, especially heavy metals, in the wastewater entering this river can be considered a potential risk and a potentially contaminating factor (Diagomanolin et al. 2004).

The present study is a study estimating the health risk assessment of potentially toxic elements in common cultivated rice (Oryza sativa) during the 2018–2019 in Khuzestan Province. In the present study, we choose 20 stations of paddy fields in five strategic regions of rice cultivation counties in Khuzestan Province. Bavi, region 1, Shadegan, region 2, Baghmalek, region 3, Behbahan, region 4, and Dezful, region 5 (respectively in the center, southwest, east, southeast, and north of the province) were selected to estimate the risk of rice consumption. The selected counties are marked in Figure 1. These counties were in different geographical directions to represent the province better. Four paddy fields in each county were selected for sampling.

2.2. Sampling

After selecting the fields in each county, soil samples were taken from the paddy fields at a depth of 0–30 cm (the root zone) by a steel shovel in harvest time. Four rice and four soil samples were taken from each field. Sampling method were randomly and considering three replications, a total of 160 soil samples and the same number of rice samples were taken from the 20 sampling stations. The samples were put in polyethylene bags and transferred to the laboratory.

2.3. Sample preparations and chemical analysis

At the first Rice, samples were leached with deionized water three times to dispose of impurities and dust and withered for 72 h in an oven. Then they were ground with blender and passed through a 1 mm mesh sifter afore they were digested. Thence, 0.5 gr of each powdered sample was digested by HNO₃ (4 ml) (≥69% w/w) and 1 ml of H₂O₂ (≥30% v/v) in a microwave oven (Milestone, FKV, MLS 1200, Italy).

Afterward, the subsequent heating plan was applied with the microwave oven: 250 W for 1 min, 0 W for 2 min, 250 W for 5 min, 400 W for 5 min, and 600 W for 5 min. The confected sample was subtilized with 25 ml Milli-Q water; after that, for analysis, we used mass spectrometry with inductively coupled plasma (Agilent 7500cx). To appraise the precision of the analytical manners, convenient, adduced recourse materials (NIST-SRM-1568b) was done analyzed under an identical situation as the samples were. The collected soil samples were dried in air and winnow
through a 2-mm rustproof steel mesh to dispose of plant roots and rocks. Following digestion of soil samples with hydrochloric acid (HCl) and nitric acid (HNO₃) in a ratio of 3:1 (HNO₃: HCl), the total concentrations of Pb, Cr, Cu, Ni, Cd, Zn, and As were defined using inductively coupled plasma (ICP) optical emission spectroscopy (ICP-OES). Dehumidified samples were ground in a rustproof steel grinder (<0.25 mm), and the total intents of the above-mentioned heavy metals were determined using ICP-OES. For certifying the measurement precision and accuracy in the entire analytical procedure, samples were analyzed in triplicate. Synchronously, blank reagent determinations were used to rectify the appliance readings.

After evaluating 20 samples using standard calibration solutions, the ICP-MS was tuned and calibrated (Djahed et al. 2018). All statistical analyses were accomplished by Microsoft Excel 16 and IBM SPSS Statistics 22.

2.4. Data analysis

The measurement or estimation of uncertainty toxicants parameters and lack of precise knowledge can be can be by risk assessment (Qu et al. 2016, Djahed et al. 2018). In this regard, there are two approaches to exert a health risk assessment model – deterministic and stochastic approaches. Prepares information on the uncertainty in the model outputs and describes uncertainty in the model inputs are the most specifications the stochastic approach (Benke and Hamilton 2008, Djahed et al. 2018). Monte Carlo Simulation (MCS) is the most widely used for the stochastic approach (Kentel and Aral 2004, Farzadkia et al. 2015). Considerable improvements in the scientific rigor of risk assessment, analyze and control the uncertainty existing in the input parameters are guaranteed by employing MCS (Qu et al. 2016, Djahed et al. 2018). First, the desired input parameters and MCS were simulated after iterations the exposure and risk models.

2.5. Pollution assessment

The pollution threshold for each HM in the soils was defined using the formula for the Single Pollution Index (SPI) obtained from Equation (1):

\[ \text{SPI} = \frac{\text{Toxicant concentration in soil}}{\text{Threshold concentration}} \]

Figure 1. Location of the sampling area, Khuzestan province, Iran.
Here, $C_i$ is the concentration of the HM $i$, and $B_i$ is the pollution limen of the HMi (geochemical background: (Weissmannová and Pavlovský 2017)).

Since no comprehensive standard of soil quality has been developed in Iran, Class 2 Environmental Quality Standard for Soils in China was used in this section (Hu et al. 2017). Table 2 exhibitions the assortments of the single pollution index (SPI).

The pollution levels of the HMs in rice were appraised exerting the crop pollution index (CPI).

The CPI for each heavy metal was determined by dividing its concentrations in the rice samples by its standard CPI value (Equation (2)).

$$\text{CPI} = \frac{C_i}{S_i}$$  \hspace{1cm} (2)

where $C_i$ represents the evaluated concentration of the heavy metal $i$ in rice and $S_i$ the standard value suggested by FAO/WHO and the National Chinese Standard (Hu et al. 2017). They were used because of the determination of more HMs compared to JECFA.

The bioaccumulation factor (BAF) of each plant was used to estimate the conduction of HMs from soil to herbage. This factor can be obtained from Equation (3):

$$\text{BAF} = \frac{C_r}{C_s}$$  \hspace{1cm} (3)

Here, $C_r$ and $C_s$ are the concentration of each HM in rice and the corresponding soil sample, respectively (Zhou et al. 2016).

### 2.6. Health risk assessment

The health risk is assessed using quantitative and qualitative procedures. In this research, the qualitative risk appraisal procedure was used hazard quotient (HQ) index for non-carcinogenic effects (Equation (5)) (Islam et al. 2016, Djahed et al. 2018, Hussaina and Abdullaha 2018).

$$\text{HQ} = \frac{\text{EDI}}{RfD}$$  \hspace{1cm} (5)

where HQ (dimensionless) indicated the hazard quotient index; $RfD$ (mg kg$^{-1}$day$^{-1}$) is the reference dose of the heavy metal (for As, Cd, Cr, Cu, Pb, Ni and Zn 0.0003, 0.0005, 0.003, 0.04, 0.0035, 0.02 and 0.3 (mg kg$^{-1}$day$^{-1}$), respectively) & EDI (mg kg$^{-1}$day$^{-1}$) is the estimated daily intake; and. Moreover, for the risk assessment of a syntax of chemicals, the individual HQs are added together to form the hazard index (HI). If the worthiness of HQ and, or HI oversteps 1, there may be potential non-carcinogenic effects on health, whereas HQ and, or HI $< 1$ means inhabitants are not maybe to experience any health risks as an issue of disposal to HMs (Djahed et al. 2018, Hussaina and Abdullaha 2018, USEPA 2018). The hazard index (HI) was calculated using Equation (6):

$$\text{Hazard Index} = \sum_{n=1}^{N} \frac{\text{EDI}_n}{RfD_n}$$  \hspace{1cm} (6)

The carcinogenic risk was accounted using Equation (7) (Islam et al. 2016, Hussaina and Abdullaha 2018).

$$\text{Cancer Risk} = \text{EDI} \times SF$$  \hspace{1cm} (7)

In Equation (7), EDI (mg kg$^{-1}$day$^{-1}$) considers the estimated daily intake of the carcinogenic HM, and the SF (mg kg day$^{-1}$) related to its slope factor. The estimated value for the carcinogenic risk (CR) is the probability that if an individual will develop any type of cancer from lifetime exposure to carcinogenic
3. Results and discussion

The proceedings in various industries can enhance the condensations of HMs in soils. These contaminants can accumulate in crops and import the food chain (Huang et al. 2013, Satpathy et al. 2014). We can presume rice one of the principal sources that comfort the entrance of HMs into human corporality. In this investigation, concentrations of seven HMs (Cd, Zn, As, Cr, Pb, Cu, and Ni) were measured in rice and rice field soils in various regions of Khuzestan Province, Iran.

### 3.1. Descriptive statistics of HMs in soil and rice

#### 3.1.1. Pb concentrations

Lead is the production of anthropogenic activities such as gasoline production, and influences humans in different ways. It affects the focal and lateral nervous systems, causes kidney disorders and hepatic system, delays mental and physical development in children, inhibits synthesis of hemoglobin, and results in vascular and cardiac injury (Huang et al. 2013, Rahman et al. 2014).

In the present research, the mean measured Pb concentration in rice at all stations was 0.17 ± 0.08 mg kg⁻¹ with the minimum condensation of 0.02 mg kg⁻¹ recorded in region 5 (Dezful) and the maximum condensation of 0.33 mg kg⁻¹ in region 2 (Shadegan) (Table 1). Statistical analysis using Tukey’s test revealed significant differences between regions 1, 2, and 3 with regions 3 and 4 in terms of lead concentration in rice samples \((p<.01)\). The consequences of this analysis are presented in Figure 2.

Similar studies in Iran reported an average lead concentration of 0.64 ± 0.3 mg kg⁻¹ in rice grown in

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**Table 1. Summary statistics of HM concentrations in soil and rice (mg kg⁻¹) (N = 60).**

| Content    | Cr (mg kg⁻¹) | Pb (mg kg⁻¹) | Cd (mg kg⁻¹) | As (mg kg⁻¹) | Cu (mg kg⁻¹) | Zn (mg kg⁻¹) | Ni (mg kg⁻¹) |
|------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|
| Mean (Rice)| 0.69         | 0.17         | 0.058        | 0.087        | 5.29         | 17.89        | 0.38         |
| Std.       | 0.25         | 0.08         | 0.027        | 0.066        | 3.04         | 3.35         | 0.25         |
| Median     | 0.7          | 0.19         | 0.06         | 0.06         | 4.1          | 16.95        | 0.23         |
| CV (%)     | 0.36         | 0.49         | 0.48         | 0.75         | 0.57         | 0.18         | 0.68         |
| Min        | 0.2          | 0.02         | <LOD         | <LOD         | 1.8          | 12.9         | 0.11         |
| Max        | 1.3          | 0.33         | 0.13         | 0.21         | 12.6         | 24.6         | 0.9          |

**Food Safety Standard**

- Pb: 0.06 mg kg⁻¹
- Cr, Cd, As, Cu, Zn, Ni: 10 mg kg⁻¹

**ISRI**

- Pb: 0.06 mg kg⁻¹
- Cr, Cd, As, Cu, Zn, Ni: 10 mg kg⁻¹

**Background value**

- Pb: 0.05 mg kg⁻¹
- Cr, Cd, As, Cu, Zn, Ni: 5 mg kg⁻¹

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**Table 2. Descriptive statistics and classification of the SPIs in the soil-pollution grade (N = 60).**

| Item               | Cr (%) | Pb (%) | Cd (%) | As (%) | Cu (%) | Zn (%) | Ni (%) |
|--------------------|--------|--------|--------|--------|--------|--------|--------|
| Mean               | 1.44   | 0.30   | 1.11   | 0.87   | 1.58   | 0.82   | 2.38   |
| Std.               | 0.403  | 0.119  | 0.458  | 0.602  | 0.901  | 0.149  | 0.810  |
| Median             | 1.41   | 0.310  | 1.05   | 0.62   | 1.22   | 0.76   | 2.36   |
| CV (%)             | 27.93  | 39.39  | 40.98  | 69.01  | 56.72  | 18.17  | 34.01  |
| Min                | 0.70   | 0.15   | 0.39   | 0.24   | 0.70   | 0.63   | 1.28   |
| Max                | 61.95  | 0.25   | 1.94   | 2.28   | 3.71   | 1.13   | 3.79   |
| Pollution degree   | Cr (%) | Pb (%) | Cd (%) | As (%) | Cu (%) | Zn (%) | Ni (%) |
| Safety (≤1.0)      | 13.3   | 100    | 38.3   | 58.3   | 36.7   | 78.4   | 0      |
| Slight pollution (1.0 < SPI ≤ 2.0) | 76.7  | 0      | 61.7   | 31.7   | 38.3   | 21.6   | 41.6   |
| Mild pollution (2.0 < SPI ≤ 3.0)  | 10.0   | 0      | 0      | 10     | 14.6   | 0      | 33.2   |
| Moderate pollution (3.0 < SPI ≤ 5.0) | 0     | 0      | 0      | 10     | 14.6   | 0      | 25.2   |
| Severe pollution (SPI > 5.0)         | 0     | 0      | 0      | 0      | 0      | 0      | 0      |
Kashan (Rabbani et al. 2015) and 0.075 ± 0.07 mg kg\(^{-1}\) in rice produced in Lorestan Province (Jafari et al. 2018).

Various studies have demonstrated that lead concentrations in crops that cultivated in uncontaminated soils scarcely invaded 1.0 mg kg\(^{-1}\) (Kwon et al. 2017, Jafari et al. 2018). Similar research was reported that the average condensation of Pb in rice cultivated in the north of Iran was 11.5 mg kg\(^{-1}\) (Zazouli et al. 2010), which was significantly higher than the mean of concentration recorded in this research. Moreover, the average lead contents in rice grown on wetland rice fields were 17.13 ± 0.4 mg kg\(^{-1}\) in Ropar (India) (Sharma et al. 2018).

The mean lead concentration in the soils of the studied regions in the present research was 10.99 ± 4.299 (with a minimum of 5.6 and a maximum of 19 mg kg\(^{-1}\)) (Table 1).

### 3.1.2. Cd concentrations

Cd is one of the mobile and most noxious elements among the toxic HMs. It has introduced as a carcino- gen by the International Agency for Research on Cancer (IARC) (Kwon et al. 2017). Besides, this HM is a
significant invoice in bone lesions, pulmonary insufficiency, hypertension, renal disturbances, and cancer (Huang et al. 2013, Zazouli et al. 2010).

The average measured Cd concentration in the studied regions was $0.058 \pm 0.027 \text{mg kg}^{-1}$ with a maximum of $0.13 \text{mg kg}^{-1}$ in region 2 and a minimum lower than the limit of detection (LOD) in region 5 (Table 1).

Figure 2 compares various Cd concentrations in rice produced in the five studied regions. Regions 1 and 2 were not significantly different in terms of Cd concentration in the rice grains, but there were significant differences between the other three regions in this respect ($p<.01$).

In a similar investigation, Jafari et al. compared rice produced in seven Iranian provinces during 2000–2007. The maximum and minimum Cd concentrations in rice of $0.64 \pm 0.05 \text{mg kg}^{-1}$ and $0.013 \pm 0.0007 \text{mg kg}^{-1}$ were respectively observed in Kashan and Kermanshah Province (Jafari et al. 2018).

In an investigation, Zazouli et al. reported that Cd was not detected in the cultivated rice of paddy lands in the north of Iran (Zazouli et al. 2010).

In Eastern China, a mean of Cd content was reported $0.57 \pm 0.44 \text{mg kg}^{-1}$ by (Chen et al. 2018). Moreover, in a research conducted in Sweden, the mean Cd content in rice was $0.024 \text{mg kg}^{-1}$, which was identical to that acquired in the present study approximately (Jorhem et al. 2008).

In this study, the average Cd concentration in the soils of Khuzestan Province was $0.18 \pm 0.072 \text{mg kg}^{-1}$ with a minimum of $0.05 \text{mg kg}^{-1}$ in region 5 and a maximum of $0.32 \text{mg kg}^{-1}$ in region 2 (Table 1). Phosphate fertilizers were the primary origin of Cd in the soils of rice fields. Many studies have shown increased concentrations of Cd in field soils caused by overuse of Cd-containing phosphate fertilizers (Cai et al. 2012).

### 3.1.3. As concentrations

All types of grains accumulate arsenic. Due to the domination of anaerobic conditions in the soil of paddy fields, rice accumulates arsenic more efficiently (Huang et al. 2013, Kwon et al. 2017). Further, investigations have exhibited that condensation of rice circa 10 times more arsenic than other crops (Islam et al. 2016). The utilization of agrochemicals such as hormones, fungicides, insecticides, and fertilizers in paddy fields is the primary source of this HM and can increase its concentration in soils (Kongsri et al. 2016). Chronic arsenic poisoning may have severe effects on health including kidney, bladder, lung, prostate and skin cancers, hyperkeratosis, melanosis, peripheral vascular disease, restrictive lung disease, ischemic heart disease, and hypertension (Islam et al. 2016, Kongsri et al. 2016, Rebelo and Caldas 2016).

In the present investigation, the average arsenic concentrations measured in the five regions in Khuzestan Province was $0.087 \pm 0.066 \text{mg kg}^{-1}$ with a minimum less than LOD in regions 4 and 5 and a maximum of $0.21 \text{mg kg}^{-1}$ in region 3 (Table 1). Statistical analyses revealed significant differences between the concentrations of this HM in the various stations (Figure 2).

Adibi et al. conducted a study in Kermanshah Province in Iran and reported that the average content of this HM in rice was $0.046 \pm 0.002 \text{mg kg}^{-1}$, which was close to those measured in this study (Jafari et al. 2018). Djahed et al. carried out a survey in Iranshahr and reported an average concentration of $0.369 \pm 0.94 \text{mg kg}^{-1}$ for arsenic in rice grown in this region (Djahed et al. 2018). This value is very different from those obtained in other parts of Iran (including the present study, probably because of the large quantity of rice smuggled from abroad into that region.

In a study conducted on rice sold in Spanish and Portuguese markets, it reported that the mean content of As in white rice samples was $0.17 \text{mg kg}^{-1}$ (Pinto et al. 2016). In similar study, the average concentration of arsenic was $0.098 \text{mg kg}^{-1}$ in various brands of rice samples in Turkey (Gunduz and Akman 2013). In another research on rice cultivated in China, it found an average concentration of $0.05 \text{mg kg}^{-1}$ for this heavy metal (Fang et al. 2014). Concentrations of arsenic reported in other studies are almost close to those found in this study. The average concentration of this HM in soils of Khuzestan Province’s paddy fields was $5.02 \pm 3.42 \text{mg kg}^{-1}$ with a maximum of $14.1 \text{mg kg}^{-1}$ in region 2, and a minimum of $1.3 \text{mg kg}^{-1}$ in region 5 (Table 1).

### 3.1.4. Cu concentrations

Exposure to Copper may afford renal inconvenience, irritation of the eyes, mouth, nose, stomachaches, dizziness, insomnia and drastic gastrointestinal effects (Rahman et al. 2014, Ullah et al. 2017).

In the present research, the average Cu condensation in rice produced in Khuzestan Province was $5.29 \pm 3.04 \text{mg kg}^{-1}$ with a maximum of $12.6 \text{mg kg}^{-1}$ in region 2, and a minimum of $1.8 \text{mg kg}^{-1}$ in region 4.

Statistical analyses indicated no significant differences in Cu concentrations between regions 3, 4 and 5, but these regions individually differed significantly...
from regions 1 and 2 in terms of Cu concentration (Figure 2).

Similar research conducted in Iran reported the following Cu concentrations in rice: $1.1 \pm 0.03 \text{mg kg}^{-1}$ in Tehran Province (Kashian and Fathivand 2015) and $22.8 \pm 0.03 \text{mg kg}^{-1}$ in Lorestan Province (Jafari et al. 2018).

The mean content of Cu in rice consumed in Iranshahr was $3.095 \pm 0.439 \text{mg kg}^{-1}$ (Djahed et al. 2018).

In a study conducted on the East Coast of India, was reported Cu concentrations were in the confinement of $0.1–0.23 \text{mg kg}^{-1}$ (Satpathy et al. 2014). An analogous study, the mean content of Copper in rice grown close to an industrial area in China, was $2.64 \text{mg kg}^{-1}$ (Cao et al. 2010).

The Cu concentrations measured in this study were higher than those that were reported in the studies as mentioned earlier. However, on average, Cu concentrations did not exceed the maximum permissible level (Table 1).

The average Cu concentration in the soils of Khuzestan Province measure in this study was $36.69 \pm 19.74 \text{mg kg}^{-1}$ with a maximum of $86.2 \text{mg kg}^{-1}$ in region 2 and a minimum of $13.6 \text{mg kg}^{-1}$ in region 4 (Table 1). The presence of Cu in soils mainly reflects industrial activities or direct use of pesticides in crops (mainly insecticides and fungicides) (Roca-Perez et al. 2010).

3.1.5. Ni concentrations
Nickel is an indispensable element in some animal species, and researchers have recommended it may be the principal for human nutrition. The USEPA has assorted nickel carbonyl as a Group B 2, nickel sub sulfide, and nickel refinery dust as Group A, human carcinogens, a probable human carcinogen (Islam et al. 2016, USEPA 2018).

Condensation of Ni in rice and rice field soils were assessed in this research. The average Ni concentration in rice produced in Khuzestan Province was $0.38 \pm 0.25 \text{mg kg}^{-1}$ with a maximum of $0.9 \text{mg kg}^{-1}$ in region 2 and a minimum of $0.11 \text{mg kg}^{-1}$ in region 5. Statistical analyses indicated that the five regions were significantly distinct in terms of Ni concentration except for region 3 ($p<.01$).

The average Ni concentration in a similar study conducted in Shiraz was $0.76 \pm 0.101 \text{mg kg}^{-1}$ (Malakootian et al. 2011). In China, Hu et al. reported that the average Ni concentration in crops such as rice and other cereals was about $0.36 \pm 0.49 \text{mg kg}^{-1}$ (Hu et al. 2017). Ni contents measured in this investigation were close to those consequences in the mentioned investigations. The average nickel concentration in rice field soils in this study was $49.32 \pm 16.71 \text{mg kg}^{-1}$ with a maximum of $80.3 \text{mg kg}^{-1}$ in region 2 and a minimum of $25.6 \text{mg kg}^{-1}$ in region 5.

The nickel in agricultural soils probably has anthropogenic origins such as atmospheric precipitation caused by exhaust emissions and industrial activities that have been reported as the primary source of soil contamination with nickel in many documented studies (Ağca and Özdel 2014). Besides, the fact that nickel accompanies copper in agricultural soils is often due to the use of urban wastewater for irrigating agricultural fields (Zhao et al. 2014).

3.1.6. Cr concentrations
Chromium is much used in abundant industrial procedures and hence is a contaminant of a multitude of environmental systems (Cohen et al. 1993). Chromium compounds are applied in chrome plating, industrial welding, pigments and dyes, wood preservation, and leather tanning. However, exposure to chromium through Cr (VI)-containing environmental compounds is realized to occasion multiorgan toxicity such as asthma and allergy, renal damage, cancer of the respiratory tract in humans (Goyer and Clarkson 1996, Joint FAO/WHO Expert Committee on Food Additives and World Health Organization).

The average Cr concentration in rice measured in this study was $0.69 \pm 0.25 \text{mg kg}^{-1}$ with a maximum of $1.2 \text{mg kg}^{-1}$ in region 2 and a minimum of $0.2 \text{mg kg}^{-1}$ in region 5.

Based on the statistical analyses, all the regions, except for regions 3 and 4, were significantly different in terms of Cr concentrations in rice. According to the study by Jafari et al. conducted on rice in Iran, the average Cr concentration in rice was $0.24 \pm 0.06 \text{mg kg}^{-1}$ in Mazandaran Province and $0.36 \pm 0.03 \text{mg kg}^{-1}$ in Fars Province (Jafari et al. 2018).

In a study accomplished in China, Cr concentration in rice was $0.74 \pm 0.44 \text{mg kg}^{-1}$ (Hu et al. 2017). Cr concentrations in the present study were somewhat higher than those in the mentioned studies, but they were lower than the maximum permissible level in most cases.

The average Cr concentration in the soils at all the stations was $81.23 \pm 22.31 \text{mg kg}^{-1}$ with a maximum of $123.1 \text{mg kg}^{-1}$ in region 2 and a minimum of $39.4 \text{mg kg}^{-1}$ in region 5 (Table 1).

3.1.7. Zn concentrations
Zinc is a necessary element for plant nutrition. In totality, concentrations of Zn in crops are in the
confinement of 1.2–73 mg kg\(^{-1}\), lowest fruits and grains, and the highest in leafy plants.

The average Zn concentration in rice produced in Khuzestan Province was 17.89 ± 3.35 mg kg\(^{-1}\) with a maximum of 24.60 mg kg\(^{-1}\) in region 2 and a minimum of 12.9 mg kg\(^{-1}\) in region 3. Based on the statistical analyses and Tukey’s multiple comparison test, there were no significant discrepancies between regions 1, 3, and 5 in terms of Zn concentrations, but regions 2 and 4 and the combined regions 1, 3, and 5 differed significantly in terms of Zn concentrations (Figure 2). According to Jafari \textit{et al.}, Zn concentrations in rice in Tehran and Lorestan Provinces in Iran were 20.7 ± 0.31 and 28.6 ± 12.39 mg kg\(^{-1}\), respectively (Jafari \textit{et al.} 2018). Hu \textit{et al.} represented in a similar study that the average Zn concentration in agricultural products in China’s Yangtze Delta was 14.22 ± 10.93 mg kg\(^{-1}\) (Hu \textit{et al.} 2017).

Several types of research represented contain Zn in rice grains produced in different countries. In universal, Zn concentration in rice grains sampled from different conventional markets was approximately 20 mg kg\(^{-1}\) (Antoine \textit{et al.} 2012). Besides, Roychowdhury \textit{et al.} and Zhao \textit{et al.} also issued Zn concentrations in cultivated rice in uncontaminated lands were about 20.7 and 12.7 mg kg\(^{-1}\), respectively (Roychowdhury \textit{et al.} 2003, Zhao \textit{et al.} 2009). Although, lofty levels of Zn were issued in cultivated rice near mining sites with a mean of 43.2 mg kg\(^{-1}\) (Kwon \textit{et al.} 2017). Zn concentrations in rice field soils in this investigation were almost analogous to those found in the mentioned studies. The average Zn concentration in the agricultural soils of the five study regions was 71.20 ± 12.72 mg kg\(^{-1}\) with a maximum of 98 mg kg\(^{-1}\) in region 2 and a minimum of 54.6 mg kg\(^{-1}\) in region 5 (Table 1).

### 3.2. Assessment of HMs in soil and rice samples

#### 3.2.1. SPI

Pursuant to the soil quality standards of China (CEPA 1995, Hu \textit{et al.} 2017), Class II can be applied as the limen value of health protection for humans. Table 2 presents the SPI values for the different HMs in descending order. The average SPI values were in the following descending order: Ni (2.38 ± 0.81) > Cu (1.58 ± 0.9) > Cr (1.44 ± 0.40) > Cd (1.11 ± 0.458) > As (0.87 ± 0.602) > Zn (0.82 ± 0.149) > Pb (0.30 ± 0.119).

Soil contamination in the study regions was assessed by classifying the contamination into four levels. Table 2 ranks the SPI degrees of soil pollution. As shown, 100% (Pb), 78.4% (Zn), 58.3% (As), 38.3% (Cd), 36.7% (Cu) and 13.3% (Cr) contents in the soil samples were at the safety level. In addition, 76.7% (Cr), 61.7% (Cd), 41.6 (Ni), 38.3% (Cu), 31.7% (As) and 21.6% (Zn) contents in the soil samples were at the slight pollution level. Moreover, 33.2% (Ni), 14.6% (Cu), 10% (As and Cr) concentrations in the soil samples at the mild pollution level; and 25.2% (Ni) and 10.4% (Cu) contents in the soil samples were at the moderate pollution level. The contents of Ni in the study regions were substantially high, especially in the southern regions of Khuzestan Province, where soil salinity and EC levels are higher, because of the uncontrolled use of fertilizers by farmers. In other cases, levels of contamination with HMs were at the safety or slight pollution levels.

#### 3.2.2. CPI

CPI values for the rice samples were calculated using Equation (2), and Table 3 lists inferential statistics, the number of contaminated samples, and corresponding percentages for each HM.

The average CPI value in descending order was Cd (0.94 ± 0.45) > Ni (0.92 ± 0.64) > Pb (0.84 ± 0.41) > Cr (0.69 ± 0.25) > As (0.65 ± 0.52) > Cu (0.53 ± 0.31) > Zn (0.36 ± 0.068). This is different from the order for soil SPI values. This indicates differences in the accumulation capacities of the crop for the different HMs, and many other factors, including pH, organic matter, and soil phosphorus and oxygen contents, which can turn influence plant capacity for absorbing HMs (Hu \textit{et al.} 2017).

Table 3 demonstrates that the contents of Cd, Ni, Pb, As, Ni, Cr, and Cu in descending order of 28.3, 20, 16.7, 15, 11.6, and 8.3% in the rice samples exceeded the maximum permitted levels only the Zn concentrations did not exceed the maximum permissible level in any of the rice samples.

| Item | Cr | Pb | Cd | As | Cu | Zn | Ni |
|------|----|----|----|----|----|----|----|
| Mean | 0.69 | 0.84 | 0.94 | 0.65 | 0.53 | 0.36 | 0.92 |
| Std  | 0.25 | 0.41 | 0.45 | 0.52 | 0.31 | 0.068 | 0.64 |
| Median | 0.65 | 0.87 | 1.011 | 0.39 | 0.38 | 0.34 | 0.55 |
| CV (%) | 35.88 | 49.24 | 48.41 | 80.77 | 58.22 | 18.99 | 70.04 |
| Min  | 0.23 | 0.13 | 0.25 | 0.079 | 0.20 | 0.26 | 0.29 |
| Max  | 1.22 | 1.56 | 1.89 | 1.48 | 1.22 | 0.48 | 2.08 |

| Number of polluted samples | 7 | 10 | 17 | 9 | 5 | 0 | 12 |
| Percent of polluted samples (%) | 11.6 | 16.7 | 28.3 | 15 | 8.3 | 0 | 20 |
3.2.3. Translocation from soil to rice
The BAF values and inferential statistics of the samples calculated by Equation (3). The average values of the BAF in a descending order were Cd (0.302 ± 0.045) > Zn (0.25 ± 0.0089) > Cu (0.118 ± 0.32) > Pb (0.015 ± 0.0048) and As (0.015 ± 0.0064) > Cr (0.0085 ± 0.0018) > Ni (0.0067 ± 0.003). This indicates that Cd is easily absorbed by the plants, and its deficient concentration in the soils is of great importance. Besides, competition between the HMs in the soil influenced their levels of absorption by the plants. This is consistent with the intuitions of other similar researches (Hu et al. 2017).

3.3. Health risk assessment of HMs
3.3.1. Estimation of EDI
Estimated daily intake of the HMs through rice consumption at each station was determined using Equation (4) to calculate the hazard quotients (HQs) of exposure to these HMs at the 20 stations in the five study regions. Diagrams in Figure 3 presents the average EDI amounts for adults and children.

The World Health Organization (WHO) has determined daily intake levels of these HMs for people weighing 60 kg as Cd: 0.06; As: 0.26; Cu: 30; Zn: 45; Ni: 0.2; Cr: 0.2; Pb: 0.214 (mg kg⁻¹) (Kwon et al. 2017,

Figure 3. Comparing values of EDI for adults (column graphs) and children (line graphs), furthermore WHO suggested amounts of PTDI for adults (mg kg⁻¹ day⁻¹).
The WHO then introduced these suggested PTDI values for the studied HMs: Cr: 0.0033; Pb: 0.0035; Cd: 0.001; As: 0.002; Cu: 0.5; Zn: 0.75; Ni: 0.005 (mg kg\(^{-1}\) day\(^{-1}\)). As shown in Figure 3, at all stations, the values of EDI for adults were less than those suggested by the FAO/WHO. The results of this study are consistent with those of other studies carried out on rice produced in Iran (Kwon et al. 2017, Jafari et al. 2018, Eskola et al. 2020). However, estimates of health risks of these HMs must consider their long term consumption because of long-term health problems caused by these heavy metals. In 2015 Rezaei et al in Birjand, Iran studied the variation and probabilistic risk assessment of exposure to HMs (Fallahzadeh et al. 2018). Based on result the Hazard Index (HI) values for the children and teens groups were 1.02 and 2.02, respectively, which was more than 1 (Fallahzadeh et al. 2018).

### 3.3.2. Non-carcinogenic risk

Equation (5) was used for a one year to compute the non-carcinogenic risk of the studied HMs. The estimated HQ values for adults and children living in the five studied regions are separately listed in Table 4.

The HQ values for all the HMs were less than 1 except for arsenic in the three stations in Shadegan and Baghmalek, which exceeded 1 for adults and 2 for children.

The origin of pollution in Baghmalek is probably the use of water wells for irrigating rice fields. Previous studies in this region showed that the water in these wells contained large amounts of As and that the concentration of this HM in irrigation water of some rice fields influenced the quality of the produced rice (Kolahkaj and Batalebloie 2007, Bortey-Sam et al. 2015, Kolahkaj et al. 2017).

Since the rice fields in Shadegan receive water downstream of the Karun River and large volumes of urban and industrial wastewater enter upstream of the river, the irrigation water in this region is severely polluted (Diagomanolin et al. 2004). Therefore, the value of HQ for As exceeded 1 for adults and 2 for children. In addition to As, the HQ of Cr was also higher than unity both for children and for adults. The non-carcinogenic risk of chromium for the three age groups in the study area is as children > teens > adults. In other similar studies also, the age group of children was introduced as a high-risk population in terms of health (Wu et al. 2011, Fallahzadeh et al. 2018).

Non-carcinogenic risk of the HMs for children increases much more because of their higher intake of the HMs compared to their body weights (Bortey-Sam et al. 2015). In Bavi, the HQ of Cr for adults was less than 1 (at level 2), but it was more significant than unity for children and reached level 3 chronic risk.

These results show that As and Cr accounted for the most significant part of the potential non-carcinogenic risk for the children and the adults in the studied regions.

By Equation (6), HI values were calculated for each region by considering the sum of HQ amounts. The HI values were at the medium level for the adults at all the stations in Bavi, Shadegan, Baghmalek, and Behbahan.

However, HI values were at a high level (Level 4) in all stations of Shadegan (region 2) and one station in Baghmalek (region 3). The radar charts in Figure 4 shows the effect of each HM on the HI values for the adults and children in the perusal regions.

### 3.3.3. Carcinogenic risk

According to the USEPA classification, As is in group A (carcinogens), and the IARC has introduced Cd as a carcinogenic compound. The concentrations of these two HMs in the various regions were calculated using Equation (7). Table 4 presents the results separately for children and adults.

According to the result of on Table 4, the average carcinogenic risk (CR) of As for children and adults is respectively 4.59E-04 and 3.07E-04 put this HM in the group with moderate carcinogenic risk (Table 4).

The average carcinogenic risk of Cd for adults and children is respectively 5.43E-05 and 8.04E-05, put this HM in the group with low carcinogenic risk (Table 4). Therefore, one can conclude that the consumption of rice produced in Khuzestan province can increase the risk of cancer in the long-term. These consequences are consistent with those found by Djahed et al. in Iranshahr and with those of similar studies concerning the higher carcinogenic risk for children compared to adults (Bortey-Sam et al. 2015, Djahed et al. 2018). In 2018, Miri et al. studied potential Probabilistic risk assessment of exposure to fluoride in most consumed brands of tea. They demonstrated that there were significant risks of exposure to fluoride in most of studied brands of tea for children (Miri et al. 2018b).

In this study, the risk of consuming rice with high values of HQ and CR in children was significantly higher than in adults indicating the greater sensitivity of children to contaminated rice compared to adults. This finding was consistent with those of similar studies (Bortey-Sam et al. 2015, Chen et al. 2020).
4. Conclusions

Whereas, rice is one of the main foods in Khuzestan Province, consumption of rice contaminated with HMs may cause hygienic and health hazards for the people in this region. Therefore, a priority should be given to the continuous monitoring of HM concentrations in rice. According to the results, severe pollution was not found in any of the soils in the study regions. However, due to bio-accumulation of these metals in plants and, of course, because of their accumulation in the human body, even low and average levels of soil contamination can cause plant contamination and endanger the health of rice consumers.

In areas with high concentrations of HMs in the soils and with high enrichment coefficients of HMs, the potential harm to human health can be reduced by controlling contamination sources, adjusting and modifying agronomic practices, modifying planting patterns, making proper use of agricultural fertilizers and, or even through changing land-use patterns. According to USEPA estimates, the estimated average carcinogenic risk of As via consumption of rice produced in Khuzestan Province was 3 adults and 4 children per 100,000 people. The corresponding value for cadmium was 5 adults and 8 children per 100,000 people. This means that the consumption of rice in Khuzestan Province may cause cancer during the lifetime of rice consumers. Therefore, one can conclude that the use of rice in this province can increase the risk of cancer.

Soil properties strongly influence the dynamics of heavy metals in soil and their uptake by plants, and play a significant role in the bioavailability of these metals. Future studies should place more emphasis on soil properties. Besides, for a detailed analysis of the health risks of edible products, it is necessary to prepare questionnaires to obtain the per capita consumption of these products more reliably.

Table 4. Non-carcinogenic risk (HQ and HI) of HMs and carcinogenic risk (CR) of As and Cd in cultivated rice of Khuzestan province, Iran (Adult & children).

| Sample site/Region | Item | Cr | Pb | Cd | As | Cu | Zn | Ni | HI | Cancer risk (As) | Cancer risk (Cd) |
|-------------------|------|----|----|----|----|----|----|----|----|----------------|-----------------|
| Total Ave. (Adult)| Mean | 1.0486 | 0.1316 | 0.5826 | 0.6448 | 0.6359 | 0.2026 | 0.1146 | 3.3611 | 0.00029 | 0.00111 |
| Region 1 Std. (Bavi) | Mean | 0.7083 | 0.0889 | 0.3937 | 0.4305 | 0.4296 | 0.1347 | 0.077 | 2.2637 | 0.000194 | 7.48E-05 |
| Min | 0.1573 | 0.0249 | 0.0320 | 0.0734 | 0.0408 | 0.0114 | 0.0116 | 0.0745 | 3.31-05 | 6.10E-06 |
| Max | 0.9087 | 0.1112 | 0.4402 | 0.5277 | 0.4875 | 0.1444 | 0.0991 | 2.3289 | 0.000238 | 0.000084 |
| Region 2 Std. (Shadeghan) | Mean | 0.8655 | 0.1941 | 0.4487 | 1.2847 | 0.6395 | 0.1869 | 0.0913 | 3.7019 | 0.000578 | 8.51E-05 |
| Min | 0.14618 | 0.0273 | 0.0972 | 0.2663 | 0.1111 | 0.0158 | 0.0096 | 0.3443 | 0.000128 | 1.83-05 |
| Max | 0.6070 | 0.1693 | 0.3725 | 0.8611 | 0.4937 | 0.1652 | 0.0859 | 3.3999 | 0.000398 | 0.000207 |
| Region 3 Std. (Baghmalek) | Mean | 0.5235 | 0.1560 | 0.1778 | 1.2500 | 0.1666 | 0.1086 | 0.0169 | 1.7826 | 0.000238 | 0.000099 |
| Min | 0.0995 | 0.0416 | 0.0218 | 0.4869 | 0.0435 | 0.0129 | 0.0031 | 0.4662 | 0.000218 | 4.13E-06 |
| Max | 0.6378 | 0.1983 | 0.2032 | 1.555 | 0.2395 | 0.1841 | 0.0287 | 2.8024 | 0.00007 | 0.000038 |
| Region 4 Std. (Behbahan) | Mean | 0.5475 | 0.1354 | 0.2988 | 0.3125 | 0.1956 | 0.1717 | 0.0278 | 1.6907 | 0.000140 | 5.67E-05 |
| Min | 0.0529 | 0.0120 | 0.0415 | 0.0513 | 0.0526 | 0.0126 | 0.000111 | 0.00181 | 2.32E-05 | 7.86E-05 |
| Max | 0.6039 | 0.1524 | 0.3366 | 0.3888 | 0.2395 | 0.1841 | 0.0237 | 2.8024 | 0.00007 | 0.000038 |
| Region 5 Std. (Dezfoul) | Mean | 0.3252 | 0.0169 | 0.1100 | 0.1284 | 0.1671 | 0.1277 | 0.0161 | 0.9135 | 0.000175 | 0.000604 |
| Min | 0.1096 | 0.0205 | 0.0266 | 0.0347 | 0.0265 | 0.0096 | 0.0011 | 0.9098 | 1.56E-05 | 5.06E-05 |
| Max | 0.1975 | 0.0193 | 0.0762 | 0.0833 | 0.1375 | 0.1163 | 0.0148 | 0.8117 | 0.000037 | 0.000014 |
| Total Ave. (children) | Mean | 0.4515 | 0.0628 | 0.1337 | 0.1666 | 0.1895 | 0.1386 | 0.0173 | 1.0086 | 0.000078 | 0.000225 |
| Region 1 Std. (Bavi) | Mean | 0.7083 | 0.0889 | 0.3937 | 0.4305 | 0.4296 | 0.1347 | 0.077 | 2.2637 | 0.000194 | 7.48E-05 |
| Min | 0.2877 | 0.0286 | 0.1128 | 0.1253 | 0.2035 | 0.1750 | 0.0219 | 1.2030 | 0.000056 | 0.000021 |
| Max | 0.1597 | 0.0303 | 0.0394 | 0.0522 | 0.0391 | 0.0148 | 0.0012 | 0.0489 | 0.00009 | 0.000034 |
| Region 2 Std. (Shadeghan) | Mean | 0.1037 | 0.227 | 0.5757 | 1.4722 | 0.7604 | 0.2022 | 0.1585 | 3.9734 | 0.000663 | 0.000109 |
| Min | 0.08681 ± 0.13 | 0.1816 ± 0.043 | 0.4231 ± 0.042 | 1.2026 ± 0.023 | 0.4988 ± 0.09 | 0.2243 ± 0.018 | 0.0691 ± 0.012 | 3.276 ± 0.14 | 0.000459 | 8.04E-05 |
Also, significant policies and measures should be taken to control and reduce the concentration of heavy metals pollutants in the foodstuffs of this region. Educating people, improving the quality of cultivation methods and controlling pesticides emissions are the most significant policies about reduce effects risk HMs.

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