Risk assessment of heavy metals (chromium, nickel, lead, copper, and iron) in fast foods consumed in Isfahan, Iran

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Received: 08/05/2020; Revised: 25/06/2020; Accepted: 07/07/2020; Published: 12/10/2020

Section: This paper was submitted in Food Science and Technology, a section of the J. Bioen Food Sci.

Competing interests: There is not conflict of interest in the research conducted.

Funding: The authors received no specific funding for this work.


Edited by Dr. Victor Hugo Gomes Sales – Federal Institute of Amapá, Brazil.

Review processes: 3032020R02 (Brazil) | 3032020R03 (Brazil)
ABSTRACT - The risk assessment of heavy metals in fast foods have been evaluated in this study. For this purpose, the carcinogenic and the non-carcinogenic hazards of chromium, nickel, lead, copper, and iron concentrations were measured in 40 fast food samples purchased in 2018 from the restaurants in two districts of Isfahan, Iran. Based on the results, only the average concentration of copper was within the permissible limits. For the two groups of children and adults, the highest target hazard quotients for lead were 33.84 and 98.72, respectively. The cumulative hazard index for all the considered metals in both groups was greater than 1. Of all the heavy metals analyzed in the examined food samples, only chromium (in both age groups) and nickel (in children only) showed an incremental lifetime cancer risk. The concentrations of chromium, nickel, lead, and iron in the Mushroom burger, Fried mushrooms, Hamburger with mushroom and cheese, and vegetable pizza were higher than those in other fast foods respectively. Thus, it can be concluded that the mentioned foods are the major sources of heavy metal ingested by humans and their consumption should be closely monitored and managed by the authorities and supervising organizations.


INTRODUCTION

Heavy metals are currently the most common food contaminants identified since they can enter foods through raw materials and/or the utensils used in the production process (Pappalardo, Copat, Ferrito, Grasso & Ferrante, 2017). One of the major problems posed by heavy metals is that they are not usually metabolized in the body. Having entered the body, these elements are not excreted but are instead deposited in adipose tissues, muscles, bones, and joints, resulting in numerous diseases (Wang, Wu, Liu, Liao & Hu, 2017). Some metals, such as lead (Pb), cadmium (Cd), chromium (Cr), and nickel (Ni), are hazardous to human health, as they can result in kidney failure, lung cancer, brain diseases, anemia, skeletal changes, fetal abnormalities, mental retardation, childhood disorders, hearing impairment, immune system dysfunction, abortion, and premature birth. Although metals such as iron (Fe), zinc (Zn) and copper (Cu) are essential nutrients and their deficiencies can cause problems in humans, excessive amounts of these elements could be harmful; therefore, they should be consumed moderately (Pappalardo, Copat, Ferrito, Grasso & Ferrante, 2017; Marín et al., 2018). The assessment of the carcinogenic and non-carcinogenic health hazards of heavy metals, which is a multistage process, was carried out by employing the health risk assessment method of the US Environmental Protection Agency (USEPA). In evaluating the non-carcinogenic effects of heavy metals, a function called target hazard quotient (THQ) was used. THQ is a very useful parameter for assessing the biological hazards of food products contaminated with heavy metals. Also, the incremental lifetime cancer risk (ILCR) parameter is defined as the incremental probability of a person developing any type of cancer over a lifetime exposure to a given daily amount of a carcinogenic metal (Real, Azam & Majed, 2017; Tajdar-Oranj et al., 2018).

Given the environmental and human health concerns arising from heavy metals intake and the essential role of food in the human diet, it is crucial to determine the concentrations of these elements, which are important indicators of the hazards and diseases resulting from the ingestion of these metals, and to set the necessary food quality standards in this respect (Manzoor, Sharma & Wani, 2018).

Fast-food consumption has increased worldwide over the past few decades. Recent studies have shown an increasing trend in fast food consumption by the Iranian population as well as a significant rise in the number of fast-food restaurants in Iran; which ought to be a major health concern for Iranians, especially those living in urban areas. Fast food is cheaper, easier to prepare, and more accessible than fresh food, particularly for less affluent in urban areas. Generally, when food becomes more expensive, people opt for less costly foods, which are often high in caloric content and low in nutrients. The overconsumption of these cheap foods, however, incurs a high cost to society with obesity, which is a risk factor for many non-communicable diseases such as heart diseases, strokes, diabetes, and some cancers (Majabadi et al., 2016). Although extensive
studies have been conducted on seafood (Kawser-Ahmed, Baki, Kundu, Islam, Islam & Hossain, 2016), dairy products (Ismail, Riaz, Akhtar, Amjad, Shahzad & Mujtaba, 2017) and vegetables (Hadayat et al., 2018) with regards to the effects of heavy metals on human health, the present research is the first comprehensive study in Iran that reflects the serious challenges encountered on the issue of food safety. Due to concern of foodstuff safety in specific for the status of food pollutions, estimating the amounts of contaminants entering the body of a living organism, and determining the possible adverse effects of such contaminants on the human body, the objective of this study is to determine the carcinogenic and non-carcinogenic risks of heavy metals (Cr, Ni, Pb, Cu, and Fe) in the fast-food samples obtained from several restaurants located in Isfahan, Iran.

MATERIAL AND METHODS

a) Sampling

A total of 40 fast-food samples were randomly purchased from 19 active restaurants in Isfahan (Iran) in the winter of 2018 and transferred to the laboratory. Wet digestion was used to determine the concentrations of Cr, Ni, Pb, Cu, and Fe metals (AOAC, 1998). The acquired fast food samples were packed in clean polyethylene bags and stored at -20 °C until analysis. In this survey, an attempt was made to select and analyze the kind of fast foods that are favorite to eat for children and adults not only in Iran but also throughout the world (Table 1).

b) Food sample preparation and digestion

Around 5 g of each dried fast food sample was digested using a two-acid mixture (HNO₃:HClO₄ of 1:3) until transparent fumes were obtained. The samples were digested in a microwave digestion system (Aurora Transform 680 model, Canada). Each digested sample was then placed in a standard flask into which Milli-Q water was added to get a total volume of 50 ml. The samples were cooled and filtered using Whatman filter paper 45 and stored in plastic tubes with screw caps. The heavy metal contents of samples were determined using Inductively Coupled Plasma-Optical Emission Spectroscopy (ICP-OES) (Vista-Pro model, made in Germany). Sample blanks (Food samples) were analyzed for Cr, Ni, Cu, Pb, and Fe contents by taking 20 ml of each digestion mixture and performing the same procedure.

c) Risk assessment of heavy metals

Estimated daily intake (EDI) of heavy metals through fast foods

EDI of metals (mg/kg/day), expressed as daily dose over body weight is a parameter used for determining the exposure dose of metals over a specific period. The daily intake of metals depends on the concentration of such metals in a particular food, the daily intake of that food, and consumer body weight, and it is obtained by the following formula (USEPA, 2000):

$$ EDI = \frac{(CF \times IR \times FI \times EF \times ED)}{(BW \times AT)} $$  

In the above equation (1), CF, IR, FI, EF, ED, BW and AT respectively represent the conversion factor (mg/kg), ingestion rate per day (gr/person/day), fraction ingested (presumed as 0.40), exposure frequency (365 days/year), exposure duration (years) (which was considered here as 14 and 70 years for children and adults, respectively), average body weight (24 kg for children and 70 kg for adults), and the average exposure time for non-carcinogens (365 days/year; exposure durations of 14 and 70 years for children and adults, respectively). In this study, the IR values were estimated based on the nutritional labels on food packages (Table 1).
## Table 1. Recommended daily consumption of each examined fast-food in the present study.

<table>
<thead>
<tr>
<th>Code</th>
<th>Food sample</th>
<th>Description</th>
<th>RDC$^1$ (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Fried potatoes</td>
<td>Fried potatoes, salt, black pepper, ketchup, mayonnaise</td>
<td>85</td>
</tr>
<tr>
<td>2</td>
<td>Fried potatoes with mushroom and cheese</td>
<td>Fried potatoes, mushrooms, mozzarella cheese, salt, black pepper</td>
<td>85</td>
</tr>
<tr>
<td>3</td>
<td>Hamburger</td>
<td>One ground beef patty, tomatoes, lettuce, pickles, ketchup, mayonnaise, salt, black pepper</td>
<td>100</td>
</tr>
<tr>
<td>4</td>
<td>Double burger</td>
<td>Two ground beef patties, tomatoes, lettuce, pickles, ketchup, mayonnaise, onion slices, salt, black pepper</td>
<td>100</td>
</tr>
<tr>
<td>5</td>
<td>Cheese burger</td>
<td>One ground beef patty, tomatoes, lettuce, pickles, ketchup, mayonnaise, salt, black pepper, onion slices, cheddar cheese</td>
<td>100</td>
</tr>
<tr>
<td>6</td>
<td>Mushroom burger</td>
<td>Ground beef, mushrooms, tomatoes, lettuce, pickles, ketchup, mayonnaise, salt, black pepper, onion slices, cheddar cheese</td>
<td>100</td>
</tr>
<tr>
<td>7</td>
<td>Hamburger with mushroom and cheese</td>
<td>One ground beef patty, mushrooms, tomatoes, lettuce, pickles, ketchup, mayonnaise, salt, black pepper, onion slices, cheddar cheese</td>
<td>100</td>
</tr>
<tr>
<td>8</td>
<td>Pirashki</td>
<td>Hot dog sausage, lettuce, pickles, tomatoes, process cheese, ketchup, mayonnaise</td>
<td>100</td>
</tr>
<tr>
<td>9</td>
<td>Hot dog</td>
<td>Hot dog sausage, lettuce, pickles, tomatoes, process cheese, ketchup, mayonnaise</td>
<td>100</td>
</tr>
<tr>
<td>10</td>
<td>Special corn</td>
<td>Sweet corn, butter, thyme, lemon juice, mayonnaise, salt, black pepper</td>
<td>100</td>
</tr>
<tr>
<td>11</td>
<td>Corn with mushroom and cheese</td>
<td>Sweet corn, mushrooms, mozzarella cheese, butter, thyme, lemon juice, mayonnaise, salt, black pepper</td>
<td>100</td>
</tr>
<tr>
<td>12</td>
<td>Special snack</td>
<td>Toast bread, soya meat, cheddar cheese, bell peppers, ketchup, mayonnaise</td>
<td>100</td>
</tr>
<tr>
<td>13</td>
<td>Snack with meat and mushroom</td>
<td>Toast bread, soya meat, mushrooms, cheddar cheese, ketchup, mayonnaise</td>
<td>100</td>
</tr>
<tr>
<td>14</td>
<td>Sambusa</td>
<td>Lavash bread, potatoes, onions, ground beef, salt, black pepper</td>
<td>100</td>
</tr>
<tr>
<td>15</td>
<td>Falafel</td>
<td>Falafel, tomatoes, lettuce, pickles, ketchup, mayonnaise, onions, salt, black pepper</td>
<td>100</td>
</tr>
<tr>
<td>16</td>
<td>Special pizza</td>
<td>Beef sausage, mushrooms, onion slices, tomatoes, tomato paste, salt, black pepper</td>
<td>55</td>
</tr>
<tr>
<td>17</td>
<td>Vegetable pizza</td>
<td>Mushrooms, sweet corn, onion slices, tomatoes, carrots, bell peppers, tomato paste, salt, black pepper</td>
<td>100</td>
</tr>
<tr>
<td>18</td>
<td>Fried chicken thigh</td>
<td>Chicken thigh, ketchup, salt, black pepper</td>
<td>100</td>
</tr>
<tr>
<td>19</td>
<td>Fried chicken nuggets</td>
<td>Chicken breast, ketchup, salt, black pepper</td>
<td>100</td>
</tr>
<tr>
<td>20</td>
<td>Fried mushrooms</td>
<td>Mushrooms, ketchup, salt, black pepper</td>
<td>100</td>
</tr>
</tbody>
</table>

$^1$ Recommended daily consumption
Non-carcinogenic risk

THQ is normally used to assess the non-carcinogenic risk and estimate the potential health hazards of heavy metals in fast food. In this work, by using the method proposed by USEPA (Eq. 2), a THQ was obtained for each heavy metal found in the fast-food samples collected from Isfahan restaurants.

\[
\text{THQ} = \frac{\text{EDI}}{\text{RFD}} \quad (2)
\]

In Eq. 2, the oral reference dose (RFD) (mg/kg/day) was 1.5, 0.02, 0.004, 0.04 and 0.7 for Cr, Ni, Pb, Cu, and Fe, respectively. A THQ below 1.0 shows no visible risk; however, consumer health could be endangered if THQ is equal to or greater than 1.0 (USEPA, 2011).

For a mixture of chemical elements in fast food samples, the possible adverse effects on consumer's health are determined by the hazard index (HI), which is obtained by summing the THQ values (USEPA, 2011):

\[
\text{HI} = \sum_{n=1}^{i} \text{THQ}_n \quad (3)
\]

An HI < 1 shows no obvious health risk and an HI > 1 indicates non-cancerous risks to a population exposed to an element.

Incremental lifetime cancer risk

The ILCR, which shows the lifetime risk of cancer due to excessive exposure to Cr, Ni, and Pb, is obtained by multiplying cancer slope factor (CSF), proposed by USEPA, by estimated daily intake:

\[
\text{ILCR} = \text{CSF} \times \text{EDI} \quad (4)
\]

The CSF values (mg/kg/day) for Cr, Ni and Pb are 0.5, 1.7 and 0.0085, respectively. In general, based on the USEPA standards, the carcinogenicity risk of metal is negligible if ILCR values are below \(10^{-6}\) (lifetime cancer risk of 1 in 1,000,000). However, if ILCR values are greater than \(10^{-4}\) (lifetime cancer risk of 1 in 10,000), the carcinogenicity risk will be over the permissible level and thus hazardous to human health. A carcinogenicity risk in the range of \(1\times10^{-6}-1\times10^{-4}\) indicates an acceptable cancer risk under controlled conditions (USEPA, 2006).

d) Statistical analysis

One-way analysis of variance (ANOVA) was used to compare the mean concentrations of heavy metals in the examined fast foods. All the statistical analyses were performed in the SPSS 25.0 software for windows. The significance level was considered as 0.05.

RESULTS

The concentrations of five heavy metals (Cr, Ni, Pb, Cu, and Fe) in the examined fast foods are presented in Table 2 along with the obtained ranges for each metal with method ANOVA. The ranges of heavy metal contents in the examined samples are 0.04–0.93 mg/kg for Cr, 0.05–0.51 mg/kg for Ni, 0.05–0.94 mg/kg for Pb and 0.05–5.11 mg/kg for Cu. Table 2 also shows Fe concentrations of 3.31–51.84 mg/kg in the fast foods obtained from the two sampling sites.

According to the results of this study, the mean concentrations of Cu in all the examined fast foods were significantly lower than the standard levels set by the guidelines of the Food and Agriculture Organization/World Health Organization (FAO/WHO). Only Cheese burger contained a safe level of Cr, and the concentration of this metal in all the other food samples was higher than the permissible level. The concentration of Ni was above the standard FAO/WHO level in one
sample only (Fried mushrooms). The mean concentration of Pb in all the examined fast foods was higher than the safe limit. The highest Pb content was found in “Hamburger with mushroom and cheese” followed by Fried mushrooms, indicating severe Pb contamination in these fast foods. Other than Vegetable pizza and Hamburger, the rest of the fast-food samples had Fe contents below the maximum permissible value set by FAO/WHO.

It should be mentioned that the average concentration of Pb in the examined fast foods was higher than that of other metals.

The EDI of heavy metals in the 20 different fast foods consumed by individuals in the two age groups (children and adults) are listed in Table 3. The EDI levels of Ni, Pb, Cu, and Fe for the two age groups are much lower than the Provisional Tolerable Daily Intake (PTDI) levels presented in Table 3. The toxicity level of heavy metal in the body depends on the daily intake of such metal, which is then compared with PTDI values. PTDI is an estimate of the intake of a given contaminant over a long period, and it is obtained by dividing the assumed maximum national daily intake by the average body weight of a person and expressed as a percentage of the PTDI of the contaminant, which should not exceed the PTDI level determined by FAO/WHO standards (Stoyanova, Sirakov, Velichkova & Staykov, 2015; Saher & Kanwal, 2018).

Table 2. Mean and standard deviation of heavy metals concentrations (mg/kg) in fast-foods collect.

<table>
<thead>
<tr>
<th>Samples</th>
<th>Cr</th>
<th>Ni</th>
<th>Pb</th>
<th>Cu</th>
<th>Fe</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fried potatoe</td>
<td>0.24±0.14</td>
<td>0.10±0.07</td>
<td>0.29±0.34</td>
<td>0.92±1.23</td>
<td>13.61±1.14</td>
</tr>
<tr>
<td>Fried potatoe with mushroom and cheese</td>
<td>0.39±0.36</td>
<td>0.10±0.07</td>
<td>0.29±0.34</td>
<td>2.23±0.72</td>
<td>3.31±1.43</td>
</tr>
<tr>
<td>Hamburger</td>
<td>0.64±0.26</td>
<td>0.21±0.23</td>
<td>0.50±0.63</td>
<td>1.11±0.19</td>
<td>48.10±9.80</td>
</tr>
<tr>
<td>Double burger</td>
<td>0.23±0.05</td>
<td>0.05±0.00</td>
<td>0.51±0.16</td>
<td>2.27±1.18</td>
<td>35.27±4.27</td>
</tr>
<tr>
<td>Cheese burger</td>
<td>0.04±0.00</td>
<td>0.05±0.00</td>
<td>0.05±0.00</td>
<td>0.05±0.00</td>
<td>25.91±0.18</td>
</tr>
<tr>
<td>Mushroom burger</td>
<td>0.93±0.99</td>
<td>0.08±0.04</td>
<td>0.25±0.28</td>
<td>1.43±1.95</td>
<td>28.31±0.39</td>
</tr>
<tr>
<td>Hamburger with mushroom and cheese</td>
<td>0.75±0.48</td>
<td>0.31±0.13</td>
<td>0.94±0.09</td>
<td>1.40±0.53</td>
<td>34.21±4.10</td>
</tr>
<tr>
<td>Pirashki</td>
<td>0.11±0.09</td>
<td>0.21±0.22</td>
<td>0.41±0.51</td>
<td>0.72±0.86</td>
<td>16.08±0.28</td>
</tr>
<tr>
<td>Hot dog</td>
<td>0.51±0.58</td>
<td>0.05±0.00</td>
<td>0.05±0.00</td>
<td>0.31±0.37</td>
<td>13.33±3.78</td>
</tr>
<tr>
<td>Special corn</td>
<td>0.09±0.05</td>
<td>0.18±0.19</td>
<td>0.36±0.43</td>
<td>0.50±0.64</td>
<td>10.54±4.63</td>
</tr>
<tr>
<td>Corn with mushroom and cheese</td>
<td>0.91±0.92</td>
<td>0.15±0.14</td>
<td>0.28±0.32</td>
<td>0.60±0.78</td>
<td>19.03±9.40</td>
</tr>
<tr>
<td>Special snack</td>
<td>0.13±0.00</td>
<td>0.05±0.00</td>
<td>0.05±0.05</td>
<td>0.05±0.00</td>
<td>20.54±0.77</td>
</tr>
<tr>
<td>Snack with meat and mushroom</td>
<td>0.28±0.04</td>
<td>0.05±0.00</td>
<td>0.05±0.05</td>
<td>0.63±0.82</td>
<td>18.67±2.05</td>
</tr>
<tr>
<td>Sambusa</td>
<td>0.24±0.07</td>
<td>0.22±0.24</td>
<td>0.39±0.48</td>
<td>2.28±0.07</td>
<td>24.47±3.18</td>
</tr>
<tr>
<td>Falafel</td>
<td>0.23±0.02</td>
<td>0.20±0.20</td>
<td>0.36±0.43</td>
<td>1.67±0.07</td>
<td>33.87±8.57</td>
</tr>
<tr>
<td>Special pizza</td>
<td>0.48±0.28</td>
<td>0.14±0.12</td>
<td>0.32±0.38</td>
<td>0.94±1.02</td>
<td>22.55±3.97</td>
</tr>
<tr>
<td>Vegetable pizza</td>
<td>0.44±0.41</td>
<td>0.14±0.12</td>
<td>0.12±0.09</td>
<td>0.65±0.18</td>
<td>51.84±4.66</td>
</tr>
<tr>
<td>Fried chicken thigh</td>
<td>0.08±0.04</td>
<td>0.16±0.03</td>
<td>0.08±0.04</td>
<td>0.89±0.46</td>
<td>16.95±4.47</td>
</tr>
<tr>
<td>Fried chicken nugget</td>
<td>0.07±0.03</td>
<td>0.18±0.01</td>
<td>0.08±0.04</td>
<td>0.96±0.31</td>
<td>14.54±2.65</td>
</tr>
<tr>
<td>Fried mushroom</td>
<td>0.47±0.03</td>
<td>0.51±0.01</td>
<td>0.58±0.75</td>
<td>5.11±0.74</td>
<td>27.93±10.89</td>
</tr>
<tr>
<td>P value</td>
<td>0.001</td>
<td>0.01</td>
<td>0.01</td>
<td>0.002</td>
<td>0.03</td>
</tr>
</tbody>
</table>

Regulatory limits of metals in foods

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>0.050</td>
<td>0.025</td>
<td>10</td>
<td>48</td>
</tr>
</tbody>
</table>

(FAO/WHO, 2010)
Table 3. Estimated daily intake of heavy metals (mg/kg/day) for adults (70 kg) and children (24 kg) from fast-foods consumption in Isfahan urban.

<table>
<thead>
<tr>
<th>Samples</th>
<th>Chromium (Cr)</th>
<th>Nickel (Ni)</th>
<th>Lead (Pb)</th>
<th>Copper (Cu)</th>
<th>Iron (Fe)</th>
<th>Chromium (Cr)</th>
<th>Nickel (Ni)</th>
<th>Lead (Pb)</th>
<th>Copper (Cu)</th>
<th>Iron (Fe)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fried potatoe</td>
<td>0.02</td>
<td>0.01</td>
<td>0.03</td>
<td>0.11</td>
<td>1.66</td>
<td>0.08</td>
<td>0.03</td>
<td>0.10</td>
<td>0.32</td>
<td>4.85</td>
</tr>
<tr>
<td>Fried potatoe with mushroom and cheese</td>
<td>0.04</td>
<td>0.01</td>
<td>0.03</td>
<td>0.27</td>
<td>0.40</td>
<td>0.13</td>
<td>0.03</td>
<td>0.10</td>
<td>0.79</td>
<td>1.18</td>
</tr>
<tr>
<td>Hamburger</td>
<td>0.09</td>
<td>0.03</td>
<td>0.07</td>
<td>0.15</td>
<td>6.78</td>
<td>0.26</td>
<td>0.08</td>
<td>0.21</td>
<td>0.46</td>
<td>19.78</td>
</tr>
<tr>
<td>Double burger</td>
<td>0.03</td>
<td>0.007</td>
<td>0.07</td>
<td>0.32</td>
<td>5</td>
<td>0.09</td>
<td>0.02</td>
<td>0.21</td>
<td>0.95</td>
<td>14.81</td>
</tr>
<tr>
<td>Cheese burger</td>
<td>0.01</td>
<td>0.007</td>
<td>0.007</td>
<td>0.007</td>
<td>3.73</td>
<td>0.21</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td>10.88</td>
</tr>
<tr>
<td>Mushroom burger</td>
<td>0.13</td>
<td>0.01</td>
<td>0.03</td>
<td>0.21</td>
<td>4</td>
<td>0.39</td>
<td>0.03</td>
<td>0.10</td>
<td>0.60</td>
<td>11.89</td>
</tr>
<tr>
<td>Hamburger with mushroom and cheese</td>
<td>0.10</td>
<td>0.04</td>
<td>0.13</td>
<td>0.20</td>
<td>4.92</td>
<td>0.31</td>
<td>0.13</td>
<td>0.39</td>
<td>0.58</td>
<td>14.37</td>
</tr>
<tr>
<td>Pirashki</td>
<td>0.01</td>
<td>0.03</td>
<td>0.05</td>
<td>0.10</td>
<td>2.31</td>
<td>0.04</td>
<td>0.08</td>
<td>0.17</td>
<td>0.30</td>
<td>6.75</td>
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<td>0.007</td>
<td>0.007</td>
<td>0.04</td>
<td>1.91</td>
<td>0.21</td>
<td>0.02</td>
<td>0.02</td>
<td>0.13</td>
<td>5.59</td>
</tr>
<tr>
<td>Special corn</td>
<td>0.01</td>
<td>0.02</td>
<td>0.05</td>
<td>0.07</td>
<td>1.51</td>
<td>0.03</td>
<td>0.07</td>
<td>0.15</td>
<td>0.21</td>
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<tr>
<td>Corn with mushroom and cheese</td>
<td>0.13</td>
<td>0.02</td>
<td>0.04</td>
<td>0.08</td>
<td>2.74</td>
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<td>0.06</td>
<td>0.11</td>
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</tr>
<tr>
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<td>0.007</td>
<td>0.007</td>
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<td>0.007</td>
<td>0.09</td>
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<td>0.01</td>
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<td>2.44</td>
<td>0.03</td>
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<td>0.03</td>
<td>0.37</td>
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<td>Fried chicken nugget</td>
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<td>0.01</td>
<td>0.13</td>
<td>2</td>
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<td>Fried mushroom</td>
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<td>0.007</td>
<td>0.08</td>
<td>0.73</td>
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<td>0.21</td>
<td>0.24</td>
<td>2.14</td>
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</table>

(PTDI - mg/kg/day): Cr: 0.1, Ni: 5, Pb: 3.57, Cu: 3, Fe: 48 (FAO/WHO, 2016)

For adults, the Cr levels were higher than PTDI values in just 2 samples: Mushroom burger and “Corn with mushroom and cheese”. However, the Cr levels for children were higher than PTDI limits in “Fried potatoes with mushroom and cheese”, Hamburger, Cheese burger, Mushroom burger, “Hamburger with mushroom and cheese”, Hot dog, “Corn with mushroom and cheese”, “Snack with meat and mushroom”, Vegetable pizza, and Fried mushrooms.

The THQ values for heavy metals are separately presented in Figures 1 and 2 for the two different age groups.

According to these two figures, the highest THQ values for adults (33.84) and children (98.72), associated with toxic Pb levels, are obtained by consuming 100 g of “Hamburger with mushroom and cheese” per day.

Regarding the other examined fast foods, the THQ ranges for adults are 1.8–33.84 for Pb, 0.006–0.08 for Cr, 0.36–3.6 for Ni, 0.18–18.4 for Cu and 0.53–10.93 for Fe; and for children, they are 5.25–98.72 for Pb, 0.01–0.26 for Cr, 0.02–10.5 for Ni, 0.52–53.66 for Cu, and 1.68–31.11 for Fe. Accordingly, the THQ values of heavy metals in the fast foods consumed in Isfahan are in the following order: Cr < Ni < Cu < Fe < Pb.
Madani et al. (2020)
Risk assessment of heavy metals (chromium, nickel, lead, copper, and iron) in fast foods consumed in Isfahan, Iran

Figure 1. Target Hazard Quotient for adults of heavy metals due to the consumption of fast-foods in Isfahan urban.

Figure 2. Target Hazard Quotient for children of heavy metals due to the consumption of fast-foods in Isfahan urban.
It should be noted that except for Cr, for which the THQ value in all the fast food samples is less than 1.0 for both age groups, the THQ values of other metals in most samples are higher than 1.0 for both adults and children.

Figure 3 illustrates the HI values for the total intake of fast foods consumed by adults and children. As is observed, the HI values of all fast food samples for the two age groups (children and adults) are higher than 1.0. This indicates that fast-food consumers are at risk of developing non-cancerous diseases in the long run.

Figure 3. Hazard Index for adults and children of heavy metals due to the consumption of fast-foods in Isfahan urban.

The ILCR values of heavy metals in the fast foods consumed in Isfahan are presented in Table 4 for children and adults separately. The results indicate that the levels of Cr (in both age groups) and Ni (in children only) is higher than the permissible limits. On the other hand, the Pb contents of food samples are at critical levels ($1 \times 10^{-4})$ except in 7 samples (Cheese burger, Hot dog, Special snack, “Snack with meat and mushroom”, Vegetable pizza, Fried chicken thighs, and Fried chicken nuggets), in which they are within the permissible limit ($1 \times 10^{-5}$), and except in “Hamburger with mushroom and cheese”, in which the Pb level is above the permissible limit in adults. These risk values indicate that long-term consumption of fast foods would lead to cancer in adults and children, but the carcinogenicity risks of Cr, Ni, and Pb are greater for children.
DISCUSSION

Concentrations of heavy metals

Cr

Overall, the amount of Cr was significantly higher in a Mushroom burger than in the other food products examined in the present study. It should be noted that the amount of Cr was within the permissible limits in 5% of the samples. These findings are fully consistent with the results of a study by Islam & Desk (2018) conducted in Bangladesh on commonly consumed foods (meat, eggs, fish, milk, vegetables, and fruits). In this survey, Cr concentrations in all the food groups (except chili) were higher than the maximum allowable concentration of Cr in foods. A study by Sultana, Rana, Yamazaki, Aono & Yoshida (2017) on vegetables and fruits in Bangladesh reported Cr concentrations exceeding the permissible limit. Also, Nuapia, Chimuka & Cukrowska (2018) showed that the mean concentrations of Cr in cabbage and bean samples are higher than the allowed limits.

As shown in Table 1, the two main ingredients of the Mushroom burger are meat and mushrooms, which can increase the levels of toxic metals in this sandwich.

The accumulation of toxic metals in mushrooms is mainly influenced by the ecosystem, type of soil, the acidic substances used in the cultivation of mushrooms, and mushroom species; so the increase of heavy metals concentrations in mushrooms is in many ways different from that in other plants. According to different studies, mushrooms have a texture that can readily absorb heavy metals, especially Cr. Therefore, the high levels of Cr found in mushrooms can be due to the high amount of Cr in the materials used for their cultivation (Sithole, Mugivshisa, Amoo & Olowoyo, 2017).

Recent studies have shown that food processing is one of the causes of food contamination with heavy metals. Meat processing, packaging, and unhealthy sales methods have been reported as causes of heavy metal contamination in red meat. On the other hand, feeding livestock and poultry with forage and feed contaminated with heavy metals can be regarded as another source of heavy metal contamination (Yakup, Sabow, Saleh & Mohammed, 2018).
According to Table 2, the permissible level of Ni in foods has been specified as 0.5 mg/kg. The results of this study show that 95% of the examined food samples contained Ni at levels below the maximum limit recommended by FAO/WHO; except Fried mushrooms, whose Ni content was above 0.5 mg/kg.

Ni is toxic at high concentrations and can cause gastrointestinal bleeding, hemolysis, acute renal failure, chest pain, encephalopathy, sperm reduction, pulmonary fibrosis, and lung cancer (Charles, Ogbolosingha & Afia, 2018). Currently, the consumption of edible mushrooms is on the rise. In developed countries, mushrooms have long been used in various foods because of their high protein levels and the rare minerals they contain. Heavy metals concentrations are significantly higher in mushrooms than in other vegetables and fruits; this shows that mushrooms have a very effective mechanism for the easy intake of heavy metals. Thus, the high levels of toxic elements found in mushrooms may outweigh their nutritional benefits, to the extent that eating even a single mushroom may endanger human health (Türkmen & Budur, 2018).

Stoyanova, Sirakov, Velichkova & Staykov (2015) reported a higher concentration of Ni in different species of marine fish than the permissible level set by food safety standards. A study by Kawser-Ahmed, Baki, Kundu, Islam, Islam & Hossain (2016) examined the hazards of heavy metals in Bangladesh river fish and showed that the average concentration of Ni in these fish is higher than the maximum permissible level recommended by FAO/WHO. Furthermore, the findings of Latif et al. (2018) are consistent with the results of the present study concerning the mean concentration of Ni in vegetables.

Pb

Given the significant difference between the mean concentrations of Pb found in the examined fast foods in Isfahan and the permissible limit of lead, these foods were contaminated with Pb.

Based on the results, 100% of the tested fast foods had Pb levels above the limit set by FAO/WHO. The highest concentration of Pb was observed in “Hamburger with mushroom and cheese”, followed by Fried mushrooms, Double burger, and Hamburger.

Nowadays, consumers show a lot of interest in meat products, which can be contaminated with metals at the manufacturing and processing stages. With technological advances and the easier preparation of these foods in the shortest possible time and with the more favorable taste and the cheaper price of such products in comparison to meat, they have become increasingly popular among consumers. However, failure to tackle health issues such as polluted drinking water and feed used for livestock could potentially contaminate these meat products with environmental contaminants such as heavy metals (Abedi, Zabihzadeh, Eskandari & Ferdowsi, 2018).

Pb, a known toxic element, could enter the body through air, water, and food and cause great damage to the nervous system, kidneys, liver, bone marrow, and blood. Other negative effects of increased Pb levels in the body include hemoglobin biosynthesis and anemia, hypertension, kidney damage, abortion, and premature infants, nervous system disorder, brain damage, male infertility, and decreased learning ability. This toxic metal cannot be eliminated by washing or cooking (Seyfferth, McClatchy & Paukett, 2016).

Analyzing the concentration of Pb in some meat products, Bamuwamye, Ogwok & Tumuhairwe (2015) reported that Pb levels in 3 samples (grilled pork, beef, and chicken) are higher than the permissible limit. Kawser-Ahmed, Baki, Kundu, Islam, Islam & Hossain (2016) studied the heavy metal contents of 5 fish species in Bangladesh and showed that the average concentration of Pb in these food sources is higher than that specified in the Food Standards Agency (FSA) guidelines; which is consistent with the findings of the present study. Sobhanardakani (2018) demonstrated that Pb levels are not within the acceptable range in 80% of processed meats. Besides, Marín et al. (2018) reported a range of 0.0192–0.0958 mg/kg for the mean concentration of Pb in foods. However, the Pb levels found in the present research are higher than the standard level. As shown in Table 2, the examined meat products had the highest amount of lead among the fast-food samples. In addition, the concentration of Pb in meat products was above the level recommended by FAO/WHO. So, the food consumers in Iran, and fact in the
whole country, are recommended to cut down on the consumption of red meat to reduce their lead intake.

**Cu**

According to the FAO/WHO standards, the permissible limit for Cu in fast foods is 10 mg/kg. All the fast-food products examined in this study had Cu levels lower than this value. Rajeshkumar & Li (2018) reported Cu concentrations of 0.21–1.45 mg/kg in fish, which is lower than the values found in the present research. Latif et al. (2018) found Cu concentration of 65 mg/kg in vegetables, which is much higher than the values obtained in the present study. In their study of metals contained in poultry meat, Bayomi, Darwish, Elshahat & Hafez (2018) concluded that the average concentration of Cu in poultry is within the permissible limit proposed by FAO/WHO.

**Fe**

Based on the results of this study, 10% of the examined fast foods produced in Isfahan contained Fe levels higher than the limit suggested by FAO/WHO. The highest concentration of Fe came from Vegetable pizza, followed by Hamburger.

Fe plays a major role in several functions and processes within the body, including the formation of blood cells; however, in high doses, it becomes a toxic element. Two types of Fe generally exist in food sources: heme and non-heme. Heme Fe, found only in meat, fish, and poultry, is absorbed better than non-heme Fe found in vegetables, enriched foods, supplements, and acidic foods. Moreover, vitamin C and other acids naturally found in fruits, juices, and some vegetables help increase the absorption of Fe. Fe at high concentrations could lead to liver cancer, cardiac arrhythmias, and diabetes (Alturiqi & Albedair, 2012; Sharma, Katnoria & Nagpal, 2016).

Therefore, considering the higher concentration of Fe in Vegetable pizza compared to the other fast food products tested, the consumption of such foods can increase the level of Fe in the body.

Vegetables are essential components of the human diet, and if they are contaminated with heavy metals, their consumption could pose a risk to human health. The accumulation of heavy metals in vegetables is affected by many factors including the concentration of heavy metals in soil, the composition of phosphate, organic and nitrogen fertilizers added to the soil, and the application of some pesticides and herbicides. Thus, to prevent the contamination of agricultural products with heavy metals, it is necessary to measure the concentrations of nutrients in the soil regularly to avoid excessive application of fertilizers that generally contain heavy metals as well (Sharma, Katnoria & Nagpal, 2016; Sultana, Rana, Yamazaki, Aono & Yoshida, 2017).

In a study on fish, meat and meat products, Alturiqi & Albedair (2012) showed that the concentration of Fe exceeds the acceptable level suggested by FAO/WHO and the European Commission (EC); this, by the way, is in line with the findings of the present study. By analyzing the concentrations of some heavy metals in grilled meats in Awka, Nigeria, Bamuwamye, Ogwok & Tumuhairwe (2015) obtained average Fe levels of 5.58–24.55 mg/kg; which is lower than the Fe concentration found in the present study. A study by Sultana, Rana, Yamazaki, Aono & Yoshida (2017) on vegetables reported an average Fe level of 23,474.75 mg/kg. The highest amount of Fe found in our samples was much lower than that reported by Sultana, Rana, Yamazaki, Aono & Yoshida (2017).

**EDI of heavy metals in fast foods for adults and children**

The EDI of Ni, Pb, Cu, and Fe in the fast foods examined in our study were lower than the maximum permissible limits set by the joint committee of FAO/WHO. Also, according to the results of this study, Cr concentrations for adults and children were smaller than the levels suggested by PTDI in 90% and 50% of the tested fast foods, respectively. Moreover, for both age groups, the concentrations of Cr in Mushroom burger and “Corn with mushroom and cheese” were above 0.1; which suggests that these food products could pose a serious health hazard in terms of Cr.
contamination.

In a similar survey, Stoyanova, Sirakov, Velichkova & Staykov (2015) found much lower EDI of Zn, Cd, Pb, and Ni from fish than the PTDI values. In a study by Mohamed, Haris & Brima (2017) on four staple foods (rice, wheat, meat, and chicken) in Saudi Arabia, the EDI of Pb from all tested samples were found to be within the allowed range. A study by Ismail, Riaz, Akhtar, Amjad, Shahzad & Mujtaba (2017) titled ‘Intake of Heavy Metals through Milk and Toxicity Assessment’ showed that only the EDI of Pb and Cd are lower than the PTDI values. Darwish, Atia, Khedr & Eldin (2018) reported lower EDI of Pb and Cd from meat than the PTDI levels. Also, a study by Pappalardo, Copat, Ferrito, Grasso & Ferrante (2017) on the heavy metal content of canned tuna found lower EDI of Cd, mercury (Hg) and Pb than the proposed PTDI values.

Non-carcinogenic health hazards of heavy metals in fast foods for adults and children

Based on the results illustrated in Figures 1 and 2, Pb poses the highest hazard to the health of fast food consumers in Isfahan.

It should be noted that the THQ value obtained for each element in the fast foods consumed in Isfahan was the same for adults and children, with Pb and Cr having the highest and lowest THQs, respectively. The main reason for the low THQ of Cr in both age groups could be its relatively high RfD value (Tajdar-Oranj et al., 2018).

Among the fast foods tested, “Hamburger with mushroom and cheese” posed the highest risk for non-cancerous diseases such as cerebrovascular, dermatologic and respiratory illnesses, diabetes, and hypertension.

According to the results of previous studies, in addition to meat and mushroom, heavy metals have also been found in the cheese used in some fast foods. Metals can find their way into cheese at various stages of production. Livestock feed contaminated with heavy metals, contaminated water used by livestock and contaminated containers, and processing equipment in dairy farms could be the potential sources of heavy metals in the milk and cheese produced by dairy companies. Moreover, due to the affinity of heavy metals with caseins and fats, clotting increases the amount of these metals in cheese relative to other dairy products (Christophoridis, Kosma, Evgenakis & Bourliva, 2019).

Since the highest THQ has been reported for “Hamburger with mushroom and cheese” followed by Fried mushrooms, the consumption of these fast foods is not recommended more than once a week. Results of studies also plainly show that exposure to Pb can be minimized if fast food consumption is restricted to once a week.

The HI values of all the considered metals in the examined fast foods were found to be much higher than 1.0 for both children and adults; implying that both age groups are at risk of non-cancerous diseases. Yet, the values of this parameter were considerably higher for Fried mushrooms and “Hamburger with mushroom and cheese”; which is indicative of the adverse effects of these foods, especially on children.

Bamuwamye, Ogwok & Tumuhairwe (2015) reported the maximum HI value for roast beef, with a higher value for children than adults. These findings are consistent with the findings of the present study. An investigation by Kawser-Ahmed, Baki, Kundu, Islam, Islam & Hossain (2016) on the non-carcinogenic hazards of heavy metals in fish in Bangladesh indicated that the HI values associated with fish consumption are above 1.0. Pappalardo, Copat, Ferrito, Grasso & Ferrante (2017) found a THQ of higher than 1.0 for Hg in canned tuna. Sultana, Rana, Yamazaki, Aono & Yoshida (2017) reported the highest THQ level for metals in fruits and vegetables; which in the present study is higher for children than adults. Evaluating the health risks of noodle samples collected from Iranian supermarkets, Tajdar-oranj et al. (2018) revealed that both adults and children are at risk of non-carcinogenic diseases through exposure to aluminum (Al). Also, Igbiri, Udowelle, Ekhator, Asomugha, Igweze & Orisakwe (2018) reported that the THQ and HI values of Pb, Ni, and Cu in edible mushroom samples exceed the permissible ranges.
ILCR for adults and children through the consumption of fast foods

Based on the results of ILCR obtained in this study, the intake Cr followed by Ni and Pb from fast foods is associated with increased cancer risk in adults and children. In all the samples studied, Cr level was higher than the permissible limit for both age groups.

For adults, Ni concentrations were above the permissible limit in 75% of the tested foods and highly hazardous (1x10^-4) in 25% of the food samples; but for children, Ni levels were above highly hazardous (1x10^-4) in all the examined samples.

Similarly, for adults, Pb concentrations were above the permissible level in 5% of the examined fast food samples, highly hazardous (1x10^-4) in 60% of the samples, and average (1x10^-5) in 35% of the tested samples; however for children, Pb contents were above the permissible limit in 40% of the examined food samples and highly hazardous (1x10^-4) in 60% of the samples. Thus, it can be concluded that frequent exposure to low doses of Cr and Ni can lead to many types of cancers in the long run.

Cr can enter the body through eating or breathing, with the former being more important. Cr (III) is less absorbed in the stomach than Cr (IV). Trivalent Cr is an essential element in diet and it is naturally found in many vegetables, fruits, meats, and grains; so it is beneficial for health if consumed this way. However, a high intake of hexavalent Cr can cause gastrointestinal bleeding, hemolysis, acute renal failure, sperm reduction, pulmonary fibrosis, and lung cancer (Charles, Ogbolosingha & Afia, 2018). Based on this evaluation, the type of Cr found in the examined fast food samples is Cr (IV).

A study by Kawser-Ahmed, Baki, Kundu, Islam, Islam & Hossain (2016) revealed that high Ni accumulation in all types of fish could lead to cancer. Sultana, Rana, Yamazaki, Aono & Yoshida (2017) found that in all the samples examined, the highest carcinogenicity risk comes from the accumulation of arsenic (As) and Cd in fruits and vegetables. Charles, Ogbolosingha & Afia (2018) indicated that the cancer risk of Cr, Ni, and Pb was higher than 1x10^-4; which is in line with the findings of this study. Moreover, Igbiri, Udowelle, Ekhator, Asomugha, Igweze & Orisakw (2018) revealed that Ni concentration in edible mushrooms posed the highest cancer risk.

CONCLUSION

This study has found the average concentrations of heavy metals in the fast foods generally consumed in the city of Isfahan (Iran) to be in the following order: Pb > Cr > Fe > Ni > Cu. The results show that despite the high levels of Pb in some fast food samples, these foods have a lower EDI than the PTDI level suggested by FAO/WHO. Therefore, considering the potential health risks associated with the consumption of fast foods contaminated with heavy metals, proper measures should be taken to avoid long-term exposure to such products. In this research, only the THQ for Cr was lower than 1.0 in all the examined samples, and the highest THQ value belonged to Pb. On the other hand, HI was greater than 1.0 for all the metals in the considered fast foods. The highest THQ and HI values for metals in food samples were obtained for children. Also, Cr showed the highest ILCR for children. The highest concentrations of metals were found in the Mushroom burger, Fried mushrooms, “Hamburger with mushroom and cheese” and vegetable pizza. Thus, it is recommended that environmental studies be carried out on soil and agricultural products regularly and that the concentrations of metals, and especially heavy metals, in soil and crops be annually measured to control their contamination and to reduce the levels of heavy metals in vegetables such as mushrooms. There is also a need for careful monitoring of the amounts of fertilizers and pesticides used in farms. Given the increasing consumption of cultivated mushrooms in Iran, more attention should be paid to the conditions and environment of mushroom farms. Since heavy metals enter the meat and dairy products (frequently consumed as burgers and cheese) through the environment, air, water, and contaminated livestock feed, it is recommended that these contaminants be periodically and regularly monitored. Although the presence of a toxic element in food does not mean that the food must be banned, proper planning for the consumption of foods that may contain heavy metals, especially mushrooms, meat and dairy products, can prevent or reduce the harm inflicted by these elements in the body.
Decloration of Conflicting Interests

The authors have declared that there is no conflict of interest.

REFERENCES


