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Toxicological assessment of inorganic arsenic and zinc content in button mushrooms

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ABSTRACT

Nowadays, discharge of toxic heavy metals into the environment is an increasing global concern. In light of this and that the per capita consumption of mushrooms has increased in Iran, the current study was carried out to investigate the levels and health-risk assessment of elements arsenic (As) and zinc (Zn) through consumption of button mushrooms marketed in Kermanshah city in 2016. In this descriptive study, a total of 30 samples from 10 brands of edible mushrooms were collected from the market basket of Kermanshah city. After preparation and processing of the samples in the laboratory, the contents of the studied elements were determined using ICP-OES. Also, all statistical analyses were performed using the SPSS version 19 statistical package. Based on the results, the contents (mg/kg) of arsenic and zinc with an average of 65.23±13.57 and with an average of 66.23±2.80 respectively were higher than the maximum permissible limit (MPL). Also, the results showed that all the computed values of health risk index (HRI) of both elements in adults, but only Zn in children, were within safe limits (HRI<1). On the other hand, the HRI values of children with an average of 1.47 were more than the safe limit and led to a potential health risk under the current mushrooms consumption per capita for this age group. Therefore, serious attention to the discharge of chemicals into the environment and monitoring of the residue levels of pollutants, especially toxic heavy metals and pesticides in the foodstuffs, is recommended.

Keywords: mushroom; toxicological assessment; arsenic; zinc; health risk

Introduction

Since mushrooms are able to biodegrade the substrate and therefore use agricultural production wastes, they play an important role in the ecosystem. Fruiting bodies of mushrooms are valued for their texture, flavor, and nutritional properties. Also, uncontaminated mushrooms have been known as therapeutic foods, useful in preventing diseases including hypercholesterolemia, hypertension, and cancer.[1-3] Therefore, for these functional characteristics of mushroom, especially their ability to accumulate toxic heavy metals as common pollutants present in the environment, a survey of their chemical composition is important.[3,4]

Nowadays, to provide an adequate supply of food for the world's growing population and also in the control of pests and insect-borne diseases in crops, the use of chemical fertilizers, heavy metal-based pesticides, and organic fertilizers as an agricultural inputs is essential.[5] Therefore, discharge of chemicals, particularly toxic heavy metals, into the environment in this way has caused an increasing global concern. Toxic heavy metals pollution is a major problem concerning the quality of the environment. Different metalloids or heavy metals such as As, Cd, Hg, Pb, and Ni are toxic, but, due to many elements such as Cr, Cu, Fe, Mn, and Zn known as enzyme activators, these groups of elements are vital for metabolic processes in low amounts. Nevertheless, these elements become toxic as the level increases.[4,6]

Weathering of sedimentary and igneous rock is the major source of arsenic as a naturally-

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occurring element. However, the smelting of non-ferrous metals, mining activities, production of energy from fossil fuels, and also applications of agricultural inputs including arsenical pesticides or herbicides, are the anthropogenic origin of this element.[7-9] Black foot disease, cancers of the kidney, liver, lung, bladder, prostate, colon, and skin are the most important diseases related to arsenic.[10,11] However, zinc is known as an essential functional and structural element in several cellular processes, but an excessive intake of this element can be critical in its effects on growth, and plays a main role in cancer while also harming some physiological activities.[12,13]

A review of the related literature has revealed the potential of mushrooms to accumulate pollutant compounds present in the environment at trace contents, particularly heavy metals.[4,14-16] In this regard, the results of other studies have showed that the fruiting bodies of mushrooms have the ability to accumulate heavy metals.[1-4,6,17-24]

The aim of risk assessment as part of risk analysis is a study of adverse health effects associated with exposure to a chemical. Therefore, identification and collection of information about the chemicals health hazards, exposure of humans to the hazardous substances, and the relationships between exposure, dose, and the serious impacts of chemicals on human health are the requisites of risk assessment.[25] Mushrooms form an important part of human diet in many countries in the world,[4] and their consumption is growing rapidly in Iran with 880 g consumption per capita. Therefore, the aim of the current research is to investigate As and Zn concentrations of the fruiting bodies of button mushrooms collected in Kermanshah city, in order to provide information on the possible health risks associated with the consumption of mushrooms. This was done through evaluation of the daily intake of metal (DIM), the health risk index (HRI), and total HRI (THRI) of Ar and Zn, respectively.

Materials and Methods

Sample collection

In this study, 30 samples of 10 different brands of mushrooms (*Agaricus bisporus*) were purchased from different markets in Kermanshah city in 2016 and were used for analysis of levels of As and Zn.

Chemical analyses

After washing the mushroom samples with distilled water, 1 g of the cap of each sample was placed in a porcelain crucible and ashed in an oven at 420–450 ºC for 24 h. Then, ashed material was dissolved in 2 mL concentrated nitric acid (ultrapure grade, Sigma-Aldrich), evaporated to dryness, heated again for 3 h to 450 ºC, dissolved in 3 mL mixture of concentrated nitric and sulfuric acid (2:1 v/v), and diluted with double distilled water (ddH_2O) up to 25 mL. Also, a blank digest was carried out in the same way. Finally, for the As and Zn analyses with three replications, a Varian710-ES inductively coupled plasma-optical emission spectrometry was used in this study.[26]

Statistical analysis

The obtained results were statistically analyzed in the current study as follows:
The normality of the distribution of data was analyzed using the Shapiro-Wilk test. Then the variance homogeneity was analyzed using an analysis of variance parametric test with a DMS post hoc and Duncan multiple range test. Also, the mean contents of elements were compared with the international standard using a one-sample test. The statistical calculations were done using statistical package SPSS version 20.0 (SPSS Inc., Chicago, IL, USA).

Potential health risk assessment

In this study, the risks to human health posed by chronic exposure to heavy metals were assessed. The average daily intake of metal was evaluated for the study of potential health risk assessment, in accordance with Eq. 1:[27]

$$DIM=\frac{Cmetal \times Cfactor \times Dfood\ intake}{Baverage\ weight} \quad (1)$$

where C_{metal}, C_{factor}, and $D_{food\ intake}$ represent the heavy metal concentrations in analyzed foodstuffs (mg/kg), conversion factor (0.085) and daily intake of mushrooms (2.40×10^{-3} kg per person per day), respectively.[28,29] Also, $B_{average\ weight}$ indicates average body weight equaling 70.0 kg and 15.0 kg for adults and children, respectively.[30,31]

The health risk index for button mushrooms' consumers was evaluated using Eq. 2:[27]

$$HRI=\frac{DIM}{RfD} \quad (2)$$

In this equation, DIM indicates daily intake of metal (mg) and RfD refers to reference dose of metal (mg/kg/day). The oral reference doses were 0.0003 for As, and 0.30 for Zn. Also, an HRI<1 means the mushroom consumption is assumed to be safe.[32-34]

Total HRI (THRI) of elements for the mushrooms was evaluated in accordance with Eq. 3:[27]

$$THRI= HRI\ (toxicant\ 1)+ HRI\ (toxicant\ 3) + \cdots + HRI\ (toxicant\ n) \quad (3)$$

Results and Discussion

Nowadays, the use of metal-containing pesticides, micronutrient and organic fertilizers, including arsenic-based pesticides, zinc sulfate and compost for the cultivation of mushrooms, can lead to the accumulation of trace elements and toxic heavy metals in their fruiting body. Based on the results, among the studied mushroom samples, arsenic was detected in amounts ranging from 40.0 mg/kg to 85.0 mg/kg, and zinc ranged from 61.0 mg/kg to 72.0 mg/kg (Table 1).

Table 1. Residual levels of examined elements in edible mushroom sample (mg/kg, dry weight)

Sample	Metal Concentration	
	As	Zn
1	74.33 ± 5.13^{d}	66.0 ± 4.36^{a}
2	63.33 ± 8.02^{bc}	67.33 ± 3.06^{a}
3	57.67 ± 5.86^{b}	66.33 ± 3.06^{a}
4	44.0 ± 5.29^{a}	64.33 ± 0.58^{a}
5	42.0 ± 2.64^{a}	68.0 ± 2.65^{a}
6	69.33 ± 3.79^{cd}	68.0 ± 5.29^{a}
7	77.33 ± 7.51^{d}	64.33 ± 0.58^{a}
8	77.67 ± 5.86^{d}	66.33 ± 1.53^{a}
9	75.0 ± 4.36^{d}	66.33 ± 0.58^{a}
10	71.67 ± 1.15^{cd}	65.33 ± 3.79^{a}
Min.	42.0	64.33
Max.	77.67	68.0
Mean	65.23	66.23
S.D.	13.57	2.80

Comparing the element contents in mushroom samples with the MPL (mg/kg) (0.10 for As and 50.0 for Zn) established by FAO/WHO[35,36] showed that the mean contents of both elements observed in all samples were higher than the permissible limits. Duncan's test results (different roman letters are presented in each column of Table 1) indicate statistical significant differences (*P*<0.05) in the contents of As and Zn between some brands of mushrooms. Based on the results, no statistically significant differences were observed in the Zn concentration between all brands (samples) of mushrooms.

In addition, all the calculated HRI values of arsenic and zinc in adults, and Zn in children were within the safe limits (HRI<1) in the current study (Table 2). Furthermore, the THRI values in edible mushrooms, which varied from 0.39 to 0.83 for adults and from 1.81 to 3.85 for children, were also within the safe limit (THRI<1) only for adults. Therefore, it can be concluded that, through only consuming button mushrooms under the mentioned consumption rate, children might have potential significant health risk.

Table 2. Daily intakes of metals (DIM, mg) for ndividual heavy metal

	Adults		Children	
Brand	As	Zn	As	Zn
1	0.0002	0.0002	0.001	0.0009
2	0.0002	0.0002	0.0009	0.0009
3	0.0002	0.0002	0.0008	0.0009
4	0.0001	0.0002	0.0006	0.0009
5	0.0001	0.0002	0.0006	0.0009
6	0.0002	0.0002	0.0009	0.0009
7	0.0002	0.0002	0.001	0.0009
8	0.0002	0.0002	0.001	0.0009
9	0.0002	0.0002	0.001	0.0009
10	0.0002	0.0002	0.001	0.0009

Table 3. Health risk index (HRI) for individual heavy metals caused by the button mushroom

	Adults		Children	
Brand	As	Zn	As	Zn
1	0.722	0.0006	3.37	0.003
2	0.615	0.0006	2.87	0.003
3	0.560	0.0006	2.61	0.003
4	0.427	0.0006	1.99	0.003
5	0.408	0.0007	1.90	0.003
6	0.673	0.0007	3.14	0.003
7	0.751	0.0006	3.51	0.003
8	0.754	0.0006	3.52	0.003
9	0.728	0.0006	3.40	0.003
10	0.696	0.0006	3.25	0.003

Since inorganic arsenic is acutely toxic, intake of large quantities of inorganic arsenic leads to some adverse health effects, including gastrointestinal symptoms, severe disturbances of the central nervous systems and cardiovascular disease, and eventually death.[26,37] In the present study, the mean contents of As in mushroom samples with 65.23 ± 13.57 mg/kg were much higher than the MPL. In this regard, Zhang et al. reported that the mean levels of As in caps of *Laccaria* mushrooms consumed in Yunnan, China were 87.80 ± 69.50 mg/kg.[38] Also, the results of other studies showed that the As contents (mg/kg) in caps of mushrooms consumed in Ljubljana, Slovenia, were in the range of 41–182,[39] in caps of mushrooms consumed in Domzale, Slovenia, they were in the range of 109–200,[40] and in caps of mushrooms consumed in Slovenia they were in the range of 0.80–129.[41] Chen et al. also reported the content of As in the wild edible mushrooms from China being in the range of 0.44–1.48 mg/kg.[26]

Zn has an important role in biological systems and plays an essential role in the structure of RNA, DNA, and protein synthesis, and also is necessary for the functioning of various enzymes that are known as essential nutrients. The major symptoms of deficiency of this element are slow maturation and delayed growth.[24] Based on the results of this study; the mean levels of Zn in caps of mushroom samples with 66.23 ± 2.80 mg/kg were generally lower than MPL. The mean contents of Zn (mg/kg) in the literature have been reported as 98.50 ± 34.75 in wild edible mushrooms in Soguksu National Park, Turkey,[3] 38.83 ± 2.15 in *Pleurotus ostreatus*, 102.15 ± 33.28 in *Lycoperdon perlatum,* and 42.14 ± 2.45 in *Fistulina hepatica* from industrial areas of Romania,[4] 4.35 ± 3.0 in wild mushrooms consumed in Zimbabwe,[42] 64.62 ± 25.37 in wild edible mushroom samples consumed in Canakkale Province, Turkey,[24] and 43.84 ± 8.70 in wild edible mushrooms from Jordan.[43] Zinc content of mushroom samples in other study ranged from 30 mg/kg to 150 mg/kg.[15]

As shown in Table 2, the HRI values of arsenic and zinc for adults and the HRI values of Zn for children were less than 1, while the HRI value of As for children was higher than the safe limits. Here the average HRI value in edible mushroom samples was 0.316 for adults and 1.47 for children. Therefore, it can be admitted that children in the target population might have potential significant non-carcinogenic risks through only consuming analyzed button mushrooms from Kermanshah city. In this regard, Nharingo et al. reported that the consumption by children of 300 g of fresh *Amanita loosii* and *Cantharellus floridulus* per day less than 16 kg harvested from Rail Block forest in Zimbabwe would cause health problems.[42] Also, Zhang et al. after analyzing the As content in caps of *Laccaria* mushrooms consumed in China, reported that due to As levels being high in the fruit bodies of the studied samples, consumption of these foods may pose risks to human health.[38]

Conclusions

Based on the results, the average HRI values of analyzed mushrooms for children of 1.47 were higher than the safe limits (HRI<1), and lead to potential health risks for children through consumption of mushrooms under the current consumption rate. Therefore, serious attention to the discharge of chemicals, especially toxic heavy metals and pesticides, to the environment and their content or residue monitoring in the foodstuffs, is recommended.

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