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## Trace elements in animal-based food from Shanghai markets and associated human daily intake and uptake estimation considering bioaccessibility



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### ABSTRACT

The concentrations of four human essential trace elements [iron (Fe), manganese (Mn), zinc (Zn), chromium (Cr)] and non-essential elements [cadmium (Cd), lead (Pb), arsenic (As), and mercury (Hg)] in eighteen animal-based foods including meat, fish, and shellfish collected from markets in Shanghai, China, were analyzed, and the associated human daily intake and uptake considering bioaccessibility were estimated. The mean concentration ranges for eight trace elements measured in the foods were 3.98–131  $\mu\text{g g}^{-1}$  for Fe, 0.437–18.5  $\mu\text{g g}^{-1}$  for Mn, 5.47–53.8  $\mu\text{g g}^{-1}$  for Zn, none detected–0.101  $\mu\text{g g}^{-1}$  for Cr,  $2.88 \times 10^{-4}$ – $2.48 \times 10^{-2}$   $\mu\text{g g}^{-1}$  for Cd,  $1.18 \times 10^{-3}$ – $0.747 \mu\text{g g}^{-1}$  for Pb, none detected– $0.498 \mu\text{g g}^{-1}$  for As, and  $8.98 \times 10^{-4}$ – $6.52 \times 10^{-2}$   $\mu\text{g g}^{-1}$  for Hg. The highest mean concentrations of four human essential elements were all found in shellfish. For all the trace elements, the observed mean concentrations are mostly in agreement with the reported values around the world. The total daily intake of trace elements via ingestion of animal-based food via an average Shanghai resident was estimated as  $7371 \mu\text{g d}^{-1}$  for the human essential elements and  $13.0 \mu\text{g d}^{-1}$  for the human non-essential elements, but the uptake decreased to  $4826 \mu\text{g d}^{-1}$  and  $6.90 \mu\text{g d}^{-1}$ , respectively, after trace element bioaccessibility was considered. Livestock and fish for human essential and non-essential elements, respectively, were the main contributor, no matter whether the bioaccessibility was considered or not. Risk estimations showed that the intake and uptake of a signal trace element for an average Shanghai resident via ingestion animal-based foods from Shanghai markets do not exceed the recommended dietary allowance values; consequently, a health risk situation is not indicated.

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### 1. Introduction

Animal-based food like meat and fish are important for human because they provide protein, minerals, and essential polyunsaturated fatty acids (Guérin et al., 2011). World consumption of meat and fish has increased due to their health benefits. Despite their recognized benefits, meat and fish may represent a risk for human health since they can accumulate organic pollutants (Wang et al., 2012) and heavy metals (Medeiros et al., 2012). Contamination with heavy metals of animal-based food is a serious threat to human health because of their toxicity, persistence, bioaccumulation, and biomagnification (Kannan et al., 2007; Chary et al., 2008).

Food safety is growing public concern due to the potential accumulation of heavy metals.

Metals, such as iron (Fe), manganese (Mn), zinc (Zn), and chromium (Cr) play important role in biological systems. A deficiency of one or more of them can lead to various pathologic conditions such as malnutrition (Amiard et al., 2008). Therefore they are classed as human essential trace elements (HEEs). However, excessive intake of HEEs can cause adverse effect on human health (Bragigand et al., 2004). Mercury (Hg), cadmium (Cd), lead (Pb), and arsenic (As) are toxic, even in trace amounts. They are classified as human non-essential trace elements (HNEs) (Kannan et al., 2006). These toxic elements have been widely detected in different environmental matrices (Li et al., 2012; Gentès et al., 2013). The toxic elements in the environment may be ingested by animals and stored in their bodies, eventually resulting in the contamination of animal-based food. It has been reported that humans are mainly exposed to heavy metals through ingestion of

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food, especially animal-based food. For example, Guérin et al. (2011) reported that fish can contribute significantly to dietary human exposure to trace elements in France. The animal-based foods like fish have been employed as bioindicators of environment contamination of inorganic compounds (Guérin et al., 2011; Medeiros et al., 2012). Therefore, the measurement of trace elements in animal-based food is very important to evaluate the possible exposure to human by ingestion of contaminated food because of their adverse effects on human health.

In recent years, much attention has been focused on the human health risk assessment of heavy metals in food (Chary et al., 2008; Du et al., 2012). However, these assessments are mostly based on the total amount of heavy metals. In fact, in the human gastrointestinal tract, ingested contaminants in food are not necessarily absorbed. Those bound to the food matrices will be excreted via feces. The total amount of an ingested contaminant in the diet does not always reflect the amount of that is available to consumers (Amiard et al., 2008). To improve human exposure assessment, some researchers have used contaminant bioaccessibility, which is obtained via *in vitro* methods simulating the human gastrointestinal digestion process, as the intestinal absorption rate to evaluate the human daily intake of contaminants (Amiard et al., 2008; Ramos et al., 2012). Nevertheless, the scientific basis for adequate use of “bioaccessibility” in the assessment of human risks is weak. There is therefore a pressing need to evaluate the human daily intake considering bioaccessibility of contaminants to improve health risk assessment.

Shanghai, one of the biggest commercial and industrial cities, is located in eastern China. There are more than 23 million residents. In our previous studies, the levels and daily intake and uptake of organic pollutants in food collected from Shanghai markets were reported (Yu et al., 2012a, b). At the same time, several studies have been widely conducted to investigate the spatial distribution of metal levels in sediment, soil, and plants in the region of Shanghai (Cao et al., 2008; Li et al., 2012; Ji et al., 2012). Only few studies have ever taken aquatic organisms like fish and crabs in rivers of Shanghai area (Du et al., 2012; Zhao et al., 2012). So far, the data of trace elements in animal-based foods like livestock and poultry from Shanghai markets have not been reported. Therefore, the present study aimed: (1) to determine the concentrations of trace elements in various animal-based foods collected from markets in seven urban and three suburban districts in Shanghai; (2) to evaluate the human daily intake and uptake of trace elements, and the associated health risk via food consumption for an average Shanghai resident considering bioaccessibility of trace elements.

## 2. Materials and methods

### 2.1. Sampling and sample preparation

A total of 175 animal-based food samples, which were the same as those used in our previous studies (Yu et al., 2012a), were collected from markets in seven urban and three suburban districts in Shanghai from September 2008 to June 2009. The food items included two types of livestock meat: pork and beef; two types of poultry meat: chicken and duck; four types of seawater fish including silver pomfret (*Trachinotus blochii*), reeves shad (*Tenualosa reevesii*), smallhead hairtail (*Lepturacanthus savala*) and large yellow croaker (*Pseudosciaena crocea*), seven types of freshwater fish including grass carp (*Ctenopharyngodon idellus*), Wuchang fish (*Megalobrama amblycephala*), crucian carp (*Carassius auratus*), bighead carp (*Aristichthys nobilis*), northern snakehead (*Channa argus*), mandarin fish (*Siniperca chuatsi*) and largemouth bass (*Micropterus salmoides*), and three types of shellfish including clam (*Macra chinensis*), snail (*Bellamya*) and Pacific white shrimp (*Penaeus vannamei*).

A wet digestion method with some modifications was used (Türkmen and Ciminli, 2007) to measure the concentrations of Cd, Pb, Fe, Mn, and Zn. Briefly, for each analysis, 2 g of powdered dry sample was added to a 100 mL Erlenmeyer flask, 30 mL of concentrated HNO<sub>3</sub> was added slowly, and the sample was soaked

overnight. Thereafter, 4 mL of 30 percent H<sub>2</sub>O<sub>2</sub> was added to the flask and the sample was soaked again for 4–6 h. The flask containing the sample solution then was heated at 250 °C to evaporate the solution slowly to near dryness. Next, 10 mL of concentrated HNO<sub>3</sub> was added to the residue and the solution was evaporated slowly to near dryness again. The digestion process using HNO<sub>3</sub> was repeated until all organic materials in the sample was completely digested. After cooling down, the digested residue was transferred to a 25 mL volumetric flask containing deionized water filtered through a 0.45 μm nitrocellulose membrane filter. The resulting sample solution was stored at 4 °C until analysis.

For the measurement of Hg, As, and Cr concentrations, the samples of a given type of animal-based foods were pooled, and the same amount of sample was used for each animal-based food. A microwave digestion method was used (Cui et al., 2011). Briefly, for each sample, 0.2 g dry sample was digested using a microwave system with 4 mL of 65 percent HNO<sub>3</sub> and 3 mL of 30 percent H<sub>2</sub>O<sub>2</sub> in a Teflon-lined vessel under controlled pressure. The sample was heated to 70 °C and held for 5 min, to 100 °C and held for 5 min, and to 120 °C and held for 5 min. All the digested samples were cooled down to room temperature. The samples then were filtered through 0.45 μm nitrocellulose membrane filters and diluted to 25 mL in volumetric flasks with deionized water. The sample solutions were stored at 4 °C until analysis.

### 2.2. Instrumentation analysis

Quantifications of Fe, Mn, and Zn concentrations were performed using an inductively coupled plasma-atomic emission spectrometry (ICP-AES) (LEEMAN Prodigy, USA). The concentrations of Pb, Cd, and Cr were determined using Graphite furnace atomic absorption spectrometry (GFAAS) (ZEEEnit600/650, Germany). The Hg and As concentrations were measured using atomic fluorescence spectrometry (AFS) (AFS-9130, USA).

### 2.3. Quality assurance and quality control

To monitor any interference during the sample treatment, a procedural blank was run for each batch of experiment (twelve samples). The values obtained from the blanks were subtracted from the sample values. To examine the reproducibility of the result, generally, a selection of samples were re-analyzed after every two batches of samples. The mean relative standard deviations of replicate samples were generally less than twenty percent. The accuracy of the method was tested using trace element-spiked samples in our previous study (Hao et al., 2013), in which the mean recovery rates of the spiked trace elements was (105.7 ± 8.1) percent with ranging from 92.6 percent to 121.3 percent. The method detection limits (MDLs) for the trace elements were as follows: Hg 0.000005 μg mL<sup>-1</sup>, As 0.00005 μg mL<sup>-1</sup>, Cd 0.000015 μg mL<sup>-1</sup>, Pb 0.0009 μg mL<sup>-1</sup>, Cr 0.0001 μg mL<sup>-1</sup>, Fe 0.08 μg mL<sup>-1</sup>, Mn 0.016 μg mL<sup>-1</sup>, and Zn 0.02 μg mL<sup>-1</sup>.

To prevent contamination of the samples, all laboratory-ware were soaked in five to ten percent HNO<sub>3</sub> solution for at least 48 h and rinsed five times with water and then five times with deionized water prior to use.

### 2.4. Calculation

The daily intake (DI, μg d<sup>-1</sup>) and uptake (DU, μg d<sup>-1</sup>) of trace elements for an average Shanghai resident via food consumption was calculated according to the following equations:

$$DI = C \times m \quad \text{and} \quad DU = C \times m \times AR$$

where C (μg g<sup>-1</sup>) is the concentration of an individual trace element, m (g d<sup>-1</sup>) is the food consumption rate for an average resident, AR (percent) is the intestinal absorption rate of the trace element, and it is estimated using bioaccessibility measured via *in vitro* test mimicking human gastrointestinal tract.

In the present study, the consumption rates were set at 57.8, 25.2, 40.8, and 14.5 g d<sup>-1</sup> for livestock, poultry, fish, and shellfish for an average Shanghai resident, respectively (Yu et al., 2011). The bioaccessibility values of the trace elements are summarized in Table 1. The bioaccessibility of the eight trace elements in animal-based foods varied in a large range of 20–97 percent.

## 3. Results and discussion

### 3.1. The concentrations of trace elements

Concentrations of the trace elements are expressed in μg g<sup>-1</sup> ww (wet weight) and are given in Table 2 in the format of means, standard deviations and ranges. The composition profiles of the trace elements measured in the eighteen types of foods were illustrated in Fig. 1.

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