

Original Article**Determination of Iron and Chromium Levels in Canned Fish Produced in Factories of Khuzestan Province, Southwest of Iran**

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ABSTRACT

Background: The heavy metal pollutions were accumulated in aquatic animals such as fish. Whereas consumption of canned fish is increased in many countries, contaminated fish meat would make a hazard to food security and public health. In this study, the levels of iron and chromium were measured in canned fish products in Khuzestan, Iran, in 2015.

Methods: Forty-six of canned fish composite samples were analyzed for levels of iron and chromium after dry digestion and then determined by atomic absorption spectrometry.

Results: The mean concentrations of A and B canned brands for iron were 4.6504348 and 0.1908696 and for chromium were 1.36030435 and 0.67629565, respectively. There were significant differences in the iron and chromium levels between two brands of canned fishes ($P < 0.05$). Varieties of canned fishes were within FAO/WHO, U.S. FDA, U.S. EPA and U.K for iron and chromium.

Conclusion: According to US EPA health criteria for carcinogens, there was no health risk to chromium in canned fish.

Keywords: Atomic Absorption Spectrometry, Chromium, Canned Fish, Heavy Metals, Iran, Iron.

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INTRODUCTION

Fish and other aquatic foods are widely consumed in many parts of the world due to their high protein content, minerals, lip soluble vitamins, and omega fatty acids are known to support good health [1]. Among aquatic foods, fish are currently consumed, therefore, they are a connecting link for the transfer of toxic heavy metals in human beings [2, 3]. Fish are ideal indicators of heavy metals contaminations in aquatic systems [4], because they are constantly exposed to alloys in polluted waters [1, 5] and occupied different trophic levels [4] also may accumulate heavy metals, and represent one of the major sources of heavy metals for human [6, 7].

“The pollution of the aquatic environment has become a worldwide problem during recent years” [2]. The most important forms of pollutions are heavy metals because of their ability to bioaccumulation in marine organisms, their toxic potential and ability to transfer to human through the food chain [8]. Heavy metals are important from the viewpoint of their essential and toxicity, thus

they can be assorted as potentially toxic, probably essential and essential [1]. Foodstuffs are most important way to entrance heavy metals to the human body. Canned fish contaminated with heavy metal can arise from water pollutions and commercial processing like canning [1].

Iron is an essential metal because it has an important role in function and structure of hemoglobin, myoglobin, cytochromes and iron-containing enzymes. Iron deficiency is a widely spread around the world eventuated anemia, mental retardation, weakness of immune system. Iron overload has been attached with multiple pathological manifestations, including liver and heart disease, cancer, neurodegenerative disorders, diabetes, hormonal abnormalities and immune system abnormalities [9].

Chromium is in two major valence state including hexavalent chromium (VI) and trivalent chromium (III). Hexavalent chromium is widely known to cause carcinogenicity, genotoxicity, allergic dermatitis etc. [10]. Unlike hexavalent chromium, trivalent chromium is an essential metal

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for insulin proper function [11]. Levels of heavy metals in fish and canned fish have been widely reported in the literature [1, 6, 12, 13]. Several agencies and organizations such as the Joint FAO/WHO Expert Committee recommended permissible tolerable weekly intakes (PTWIs) as a guideline for food additives, certain contaminants and dietary exposure of trace elements in sea foods [14]. Since, canned fish is consumed in many parts of the world like USA, Portugal, Turkey, and Iran [12], therefore, it is important to monitor the levels of heavy metals in canned fish to ensure that they are safe to consumers.

This study purposed to assess the levels of iron and chromium in canned fish samples produced and marketed in Ahvaz City Khuzestan, Iran by Graphite Furnace Atomic Absorption spectrometry (GF-AAS) for Chromium and Flame Atomic Absorption Spectrometry (FAAS) for iron. Results of the present study were compared with international standards.

MATERIALS AND METHODS

Sampling

Overall, 138 canned fish samples were collected from two brands (69 samples for each brand were gathered: ("A Co" and "B Co"). The canned fish were produced in 2014 and their expiration date was two years after the production. Three samples were obtained during each company visit and mixed together to form a composite and homogenized sample. All experiments were performed in Jan and Feb, 2015.

Reagents

All glassware was soaked in 10% nitric acid for 24 h and then was washed with double deionized water. All dilutions were produced by double deionized water with resistivity 18.0 MΩ cm (ELGA LabWater, Wycombe Bucks, UK). High purity HNO₃ was used for diluted the solutions and digestion the samples (Merck, Germany). The

element standard solutions were prepared from stock solutions of iron and chromium (1000 mg/l in 0.1M HNO₃) from Merck. Solutions of 2g/L Mg (NO₃)₂ and 3g/L Pd(NO₃)₂ (Merck, Germany) in 15% (v/v) HNO₃, were used to prepared chemical modifier.

Instruments

A Varian Spectra AA-240FS atomic absorption spectrometer (Varian Australia, Pty Ltd, Mulgrave, Victoria) with deuterium background corrector and equipped with auto sampler was used for metal quantifications. Determination of chromium was carried out by GF-AAS, using argon as inert gas. All measurements were performed using pyrolytic graphite-coated tubes and furnace programs (Table 1). Iron determination was done in an air-acetylene flame. According to recommend by the manufacturer, the operating parameters were set for iron determination by FAAS as follows: lamp current; 5 mA, fuel; acetylene, support, air, and wavelength; 248.3 nm. Instrument parameters for chromium analysis by GF-AAS were adjusted as follows: lamp current, 7 mA; spectral bandwidth, 0.2R nm; wavelength, 357.9 nm and maximum Absorbance, 2. Furnace opera

Sample Preparation

Amount 5 gr of composite samples were placed in a porcelain crucible, and then were burned by the heater. After that samples were transferred to the electrical furnace. The furnace temperature was raised to 420°C until a white or gray ash residue obtained. The gray residues were dissolved in 2 ml of nitric acid and then heated until the acid is evaporated. The sample transferred to furnace again to obtain the white ash. The white ash was dissolved in diluted nitric acid and the volume was made up to 25 ml, and 1 ml was removed for subsequent analysis of chromium by GF-AAS. The iron measurements were performed with the remaining volume of sample solutions by FAAS.

Table 1. Furnace operating conditions for chromium.

| Step no. | Temp.(°c) | Time (sec) | Gas flow (l/min) | Gas type | Read command |
|----------|-----------|------------|------------------|----------|--------------|
| 1 | 85 | 5 | 3.0 | Normal | No |
| 2 | 95 | 40 | 3.0 | Normal | No |
| 3 | 120 | 10 | 3.0 | Normal | No |
| 4 | 1000 | 5 | 3.0 | Normal | No |
| 5 | 1000 | 1 | 3.0 | Normal | No |
| 6 | 1000 | 2 | 0 | Normal | No |
| 7 | 2600 | 1.2 | 0 | Normal | Yes |
| 8 | 2600 | 2 | 0 | Normal | Yes |
| 9 | 2600 | 2 | 3.0 | Normal | No |

Statistical Analysis

Data were shown as mean±standard error of the mean (S.E.M). The program of SPSS 20.0(Chicago, IL, USA) version was used for statistical calculations. Mann-Whitney Test and Tukey's post-hoc test were used for comparing. Probability <0.05 was considered to be statistically significant.

RESULTS

Based on ICH guideline the limit of quantification (LOQ) and the limit of detection

(LOD) of the test methods were determined. Measured LOD and LOQ of chromium in the sample solutions were 0.9 and 3.0 ng/g and they were 0.3 and 1 µg/g for iron, respectively.

Measuring of all the metal concentrations was performed on a wet weight basis. The results of heavy metals measurements in different brands of canned fish are shown in Table 2. The Figures 1 and 2 show the comparative levels of Fe and Cr in the various varieties of tuna fish type.

Table 2. The results of chromium and iron in two brands of canned fish samples (ng/g).

| Matrix | Element | N (Composite) | Mean | Std. Deviation | Minimum | Maximum | 95% Confidence Interval for Mean | | SEM |
|--------|----------|------------------|---------|-------------------|---------|---------|-------------------------------------|----------------|--------|
| | | | | | | | Lower Bound | Upper Bound | |
| A | Iron | 23 | 4.6504 | 20.7317 | 0.1200 | 99.7500 | -4.3146 | 13.6155 | 4.3228 |
| | Chromium | 23 | 1.3603 | 2.7966 | 0.0000 | 13.4037 | 0.1509 | 2.5696 | 0.5831 |
| B | Iron | 23 | 0.19086 | 0.1532 | 0.0900 | 0.8500 | 0.12458 | 0.2571 | 0.0319 |
| | Chromium | 23 | 0.6762 | 2.2347 | 0.0285 | 10.7600 | -0.2901 | 1.6426 | 0.4659 |

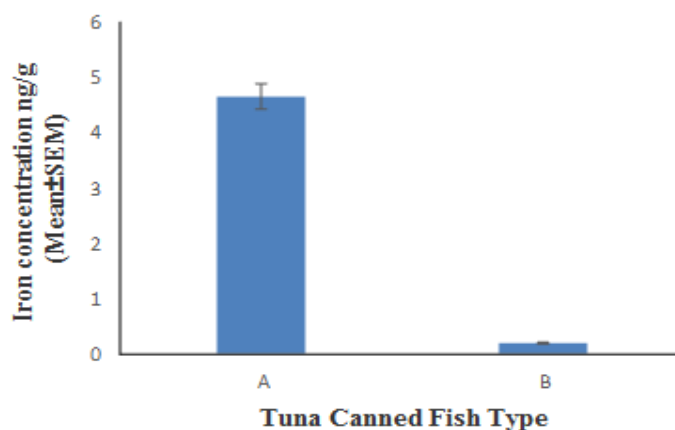


Figure 1. Comparative levels of iron in two canned fish brands, A and B.

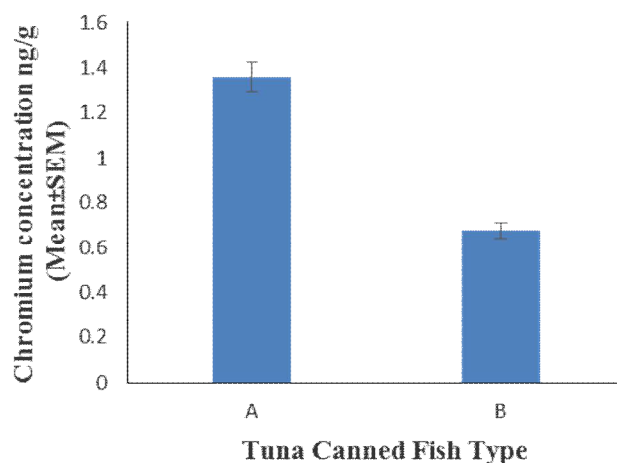


Figure 2. Comparative levels of chromium in two canned fish brands, A and B.

DISCUSSION

There are many factors affected on the heavy metals level in the fish among, the concentrations of pollutions in the water and period exposure of fish to this polluted water, water chemistry, any pollution due to handling and processing of fish, fish species feeding customs, fish gender, and weight. The pH of the canned product, the quality of the lacquer coatings of canned products, oxygen concentration in the headspace, quality of coating and place of storage place may control the metal levels in canned fish for those metals found in the canning material [8, 12].

Chromium

One of the essential micronutrients in human diet is Cr (III) that has a role in carbohydrates, lipids and proteins metabolism through the insulin function, however, Cr (VI) has potential to cause cancer [8, 12, 15]. The range of Cr levels in fish have been suggested to be 0.06-0.84 $\mu\text{g/g}$ [16], 0.10-1.60 $\mu\text{g/g}$ [17], 0.310-0.73 $\mu\text{g/g}$ [18], 0.2-1.87 $\mu\text{g/g}$ [19], 0.04-1.75 $\mu\text{g/g}$ [20] for muscles of fish from the Black Sea coasts, the Turkish seas, the western coast of the United Arab Emirates, Indian fish markets and the Marmara, Aegean and Mediterranean Sea, respectively.

There were significant differences in the Cr concentration across different brands of canned fish. According to the Institute of Medicine, there is no upper tolerable intake level for chromium but permissible tolerable intakes (PTI)/day (mg /day/60 kg body weight) of this element is 20 and 30 mg/day, respectively [21]. The Cr concentrations for the different brands of canned fish were generally within the PTI recommended by FAO/WHO, U.S. EPA reference dose (RfDs) and U.S. FDA reference daily intake (RDI)[3, 22]. Moreover, chromium concentration of canned fish was analyzed in this study is within US EPA health criteria for carcinogens (8 mg/kg) [23].

Iron

Iron is an essential metal for the body and deficiency of iron put out anemia. Whereas mammals can not to excrete excess iron, overload of iron can produce slow and progressive failure of various organs [8]. The range of iron levels of muscles of fish between 59.6-73.4 $\mu\text{g/g}$ [24], 68.6-163 $\mu\text{g/g}$ [25], 30-160 $\mu\text{g/g}$ [26] for muscles of fish from the Mediterranean Sea, the Black and Aegean Seas and the Black Sea coasts, respectively. Besides, the range of iron levels in canned sardines

marketed in USA and Brasilia have been reported to be 4.83-29.2 $\mu\text{g/g}$ [23] and 21.0-88.8 $\mu\text{g/g}$ [27], respectively. Iron PTWI value is 5600 mg kg⁻¹ body weight per week according to the Joint Expert Committee on Food Additives of the FAO and WHO [14]. The iron concentrations for the different brands of canned fish were generally within the PTWI recommended by FAO/WHO,U.S. EPA reference dose (RfDs) and U.K guidance (17mg/day) [22].

CONCLUSION

Existing of significant differences in the metal concentrations between two brands of canned fish was pronounced. They were safe according to FAO/WHO, U.S. FDA, U.S. EPA and U.K for iron and chromium. In addition, there was no health risk chromium concentration in canned fish analyzed based on US EPA health criteria for carcinogens. Consequently, consumption of the studied heavy metals has not serious health risk in the two brands canned fish. In order to help safeguard the health of humans, more research of seafood quality is required in many countries. The results of this study will help at ensuring the safety of the foodstuff and minimizing human exposure.

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