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Different analytical methods for estimating the heavy metals in fish products available in the Iraqi local markets by atomic absorption technique

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ABSTRACT

The present study aims to estimate heavy metals (Cadmium, Lead, Copper, Zinc and Iron) by different analytical methods in local and imported fish products such as the following samples: (White tuna meat small pieces) Thailand produce, (Sardines with vegetable oil pepper, slices without heads) produced by Morocco, (Fish fillet without bones) Vietnam production (live Carp fish from ponds and live Carp fish from the Tigris River) Iraq production, and (Tuna cut small) Turkey production, The concentrations of above-mentioned elements in the target samples were determined by the flame atomic absorption device . Concentrations of heavy metals were estimated at (ppm) as follows : Cadmium values (0.48 - 2.05 ppm) , While Lead values (0.66- 2.90 ppm), The values of Zinc (2.99 - 19.14 ppm) , Copper values (0.15 - 2.66 ppm) , The values of Iron (15.47-3.28 ppm), The results showed that the best method of extraction was the method of digestion which showed better results than the other two methods (ultrasound method and Ashing method). The results showed that the levels of Cadmium and Copper in some products are higher than the limits allowed by the World Health Organization (WHO) and World Food Organization (FAO) while the values of Lead, Iron and Zinc were within the limits allowed .

Introduction

Environmental studies have received the attention of researchers and specialists in the field of environmental pollution. There have been many studies dealing with environmental issues and problems, especially such the components of the environment (soil, water, air) contaminated with natural, chemical and biological contaminants, leading to a defect in the components of the environment , the human is the main factor in the imbalance in the components and elements of the environment in terms of industrial activity, population expansion and the expansion of cities that have a devastating impact on the environment, On this basis, environmental pollution is defined as the change in the installation of one of the main elements in the environment as a result of unacceptable behavior by man, causing environmental pollution to achieve his personal interests[1] .

The exacerbation of the problem of food contamination, global concern and the pressures

exerted by environmental protection organizations on industries have necessitated the contribution of industry to find solutions to these problems modern industrial projects have begun to reduce pollution of food , products foods packaged in metal containers are susceptible to chemical contamination for several reasons, including directly through food plants contaminated with heavy metals such as Lead, Cadmium, Chromium, Zinc and Copper and other elements through their volatility, movement and physical transition across the food chain in organic chemical formulations or no organic free soil layers, water , plant and the air thus leads to health problems in the future case of increased consumption of these contaminated food plants fresh or canned by the community [2] .

Heavy elements can be defined as those that increase their density about 5 g /cm³ more than five times the water density associated with pollution and toxicity [3] .

Fish contain essential Amino acids, Fatty acids, Protein, Carbohydrates, Vitamins and heavy elements are important for the human body fish and aquatic food are link to transport heavy metals to humans [4]. According to World Food Organization (FAO), fish are formed about 16% from the global human intake of animal protein [5].

Since fish is an essential component of the human diet, it is not surprising that there are aspects of quality safety that are of particular importance over the past several decades. The concentration of heavy metals in fish has been widely studied in different places around the world because fish is an important source of heavy metals for humans [6].

The main interest was in commercial edible species where it is important to study the contents of heavy elements in fish analysis whether it is acceptable for human consumption or may be higher than the recommended legal limits [7].

Many studies have shown that the liver is the most abundant tissue in the accumulation of heavy

elements from any other tissue because the liver removes toxins and deposits them before they are distributed in the body [8].

Canli and Atli found (0.37 - 0.79) ppm cadmium, (2.98 - 6.12) ppm lead, (2.19 - 4.41) ppm copper, (16.48 - 37.39) ppm zinc and (19.60-78.40) ppm iron in six species of fish collected from the Northeast Mediterranean [9]. Voegborlo et al. found (0.18) ppm cadmium and (0.218) ppm lead in Tuna [10]. Salman et al. found (1.17, 27.96, 79.15, 100.58) ppm for cadmium, copper, zinc and Iron respectively in canned Tuna [11]. Geogieva et al. found (0.63, <1, 0.21, 8.25) ppm for cadmium, lead, copper, and zinc respectively in the Scardinius Erythrophthalmus L. [12]. While Jaccob found cadmium (0.15 - 0.21) ppm, copper (1.87 - 2.72) ppm, iron (1.89 - 3.05) ppm and zinc (23.9 - 36.1) ppm in fish products [13].

Materials and methods

1- Materials Chemical using

Table (1): Track All chemicals used in the work

Subject Name	Chemical formula	purity and concentration	Company
Nitric acid	HNO ₃	69%	Fluka
Sulfuric acid	H ₂ SO ₄	95-97%	BDH
Pyrochloric acid	HClO ₄	60 %	BDH
Hydrochloric acid	HCl	37%	Fluka
Water free of ions	DI-H ₂ O		Chemistry Lab
Standard solution for heavy elements	Cd ⁺² Fe ⁺² Cu ⁺² Pb ⁺² Zn ⁺²	1000 ppm	Merck

2. Collection of samples

The samples mentioned in table (2) collected randomly from the local markets. All sample data were recorded from the date of production,

completion, brand, type of packaging and country of production. If no extraction is possible, and to avoid some changes caused by storage, samples are kept in the refrigerator and at low temperature.

Table (2): shows the sample information used in the study

Sample	Product name	marking	Country of Origin	Packing type	Production Date	Expiry date
A	White TunaMeat	Al-Taghziah	Thailand	metal can	12\2018	12\2019
B	Sardine	GEAMIC	Morocco	metal can	19\12\2019	19\12\2020
C	Slices fish filet	Al-Fakher	Vietnam	Pocket nylon	27\8\ 2018	25\5 \ 2019
D	Fish carp basin	local	Iraq	Fresh	Fresh	Fresh
E	Fish of the Tigris River	local	Iraq	Fresh	Fresh	Fresh
F	Tuna fish	SuperFresh	Turkey	metal can	29\5\2018	28\5\2020

3. Methods of work

Heavy elements were estimated in the target sample by using of atomic absorption device measured in the Department of Chemical Engineering at the University of Tikrit after preparing the samples follow ways:

A - Method of Digestion [14] :

Take 10 grams of the sample after mixed in a round flask. Then we add 20 mL of acid mixture (HNO₃:H₂SO₄) ratio (1:1), and 20 mL of DI-H₂O to the mixture in a round flask. Then heat (reflux) using the heat source for one hour under a temperature of 80 °C until the solids completely dissolve and become pure solution, filtering the solution to get rid of the solid minutes hanging in the solution then

transfer the solution to a volume flask of 100 ml and complete the size to the mark by DI-H₂O, samples are ready for measurement by the atomic absorption device.

B - Method of Ashing [15] :

Take 10 grams of the sample after mixed in a ceramic jar. Then we dry them in the drying oven for one hour under a temperature of (105 °C) until the sample dried completely. The sample will be transferred to the burn furnace. Then the temperature gradually increases to (550 °C) about (2.5 - 3) hours until being ash. Then we transfer the ash to a conical flask, adding to the ash 10 ml of acidic mix (HNO₃:HClO₄) ratio (1:1), 10 ml from DI-H₂O, until completely dissolve the ash, filtering the solution then transfer

the solution to a volume flask of 100 ml and complete the size to the mark by DI-H₂O. The samples are ready for measurement by the atomic absorption device.

C - Method of Ultra- Sonic [16]

Take 10 grams of the sample after mixed in a volumetric flask. Then add 10 ml from HNO₃ and 20 mL (DI-H₂O). Then put in ultrasonic device with a temperature of (80°C) for one hour until the solution becomes clear. Filtering the solution, then transfer the solution to a volume flask of 100 ml and complete

the size to the mark by DI-H₂O. The sample is ready for measurement by the atomic absorption device.

4- Analysis of samples and mechanical conditions in atomic absorption spectrometry

Atomic absorption technology was used to estimate heavy metals (Cu, Fe, Pb, Zn, Cd) in the liquid phase. Calibration curves were used to determine concentrations of heavy metals. The operating conditions of the atomic absorption device are shown in table(3)

Table (3): shows the automatic conditions that must be met to measure the heavy elements by atomic absorption device

Measurement requirements	Heavy elements studied				
	Cd	Pb	Fe	Cu	Zn
Wave length (nm)	228	283	248	324	213
High burner (mm)	8.0	8.0	8.0	8.0	8.0
Intensity of lamp current (mA)	4.0	5.0	6.0	4.0	5.0
Type the lamp	HCL	HCL	HCL	HCL	HCL
The amount of fuel (NL /h)	50.0	50.0	50.0	50.0	50.0
Flame Type	Air-acetylene	Air-acetylene	Air-acetylene	Air-acetylene	Air-acetylene

**HCL=Hollow Cathode lamp

5- Calibration curves:

From the standard solution and using the dilution law, the number of standard solutions for the studied heavy elements was prepared the calibration curves for the heavy elements as shows in figures (1-5)

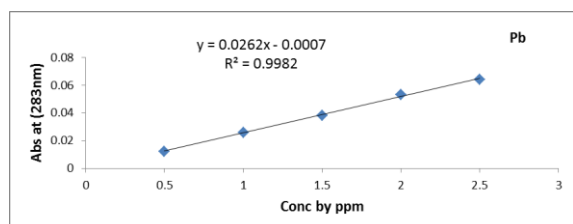


Figure (1): the calibration curve of the Lead

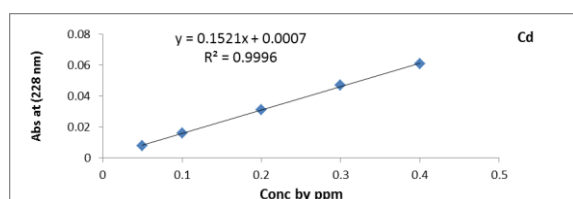


Figure (2): The calibration curve of the Cadmium

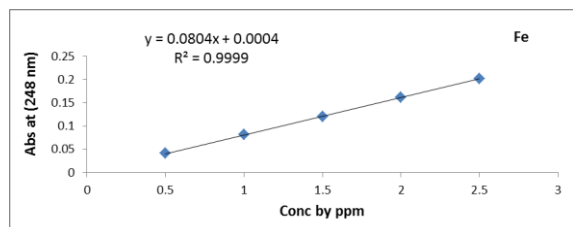


Figure (3): The calibration curve of the Iron

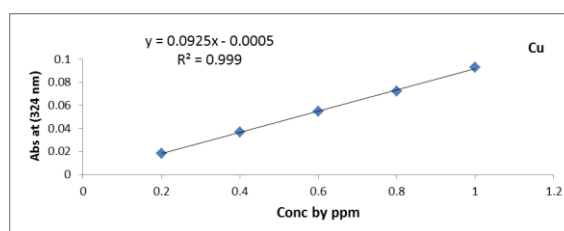


Figure (4): The calibration curve of the Copper

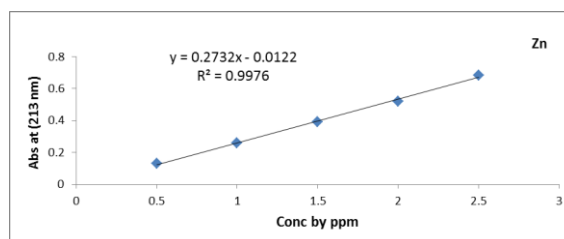


Figure (5): The calibration curve of the Zinc

6. Statistical analysis

Results were statistically analyzed using ANOVA (F-Test). Computed averages were compared for transactions using (Dunkin) Multipliers and the probability level ($P \leq 0.05$) according to the Minitab-Ver-17 statistical program.

7- Results and discussion

Table (4) shows the results of the concentration of the heavy metals obtained. The table presents the results of each of the target sample in the study and the results of each technique used in the methods of work

Table (4): The measurement results by(ppm)

Product Code	Extraction method	Heavy elements studied				
		Zn	Cu	Fe	Pb	Cd
A	Ultra-Sonic	5.80 ± 1.59 a	0.65 ± 0.23 a	4.86 ± 0.68 c	ND	0.83 ± 0.04 a
	digestion	5.34 ± 0.71 a	0.33 ± 0.08 a	4.26 ± 0.27 c	ND	1.94±0.04a
	Ashing	4.19 ± 0.44 a	0.44 ± 0.10 a	5.10±0.34 c	ND	0.64±0.04a
B	Ultra-Sonic	9.62 ± 1.20 a	1.48 ± 0.53 a	8.59±0.34 c	ND	0.83±0.08 b
	digestion	11.40 ± 1.58 a	1.07 ± 0.41 a	9.31±0.17 c	ND	2.05±0.02a
	Ashing	19.14 ± 2.50 a	1.38 ± 0.42 a	15.21±0.86 c	ND	1.01±0.06a
C	Ultra-Sonic	3.09 ± 1.51 a	1.83 ± 0.01 a	10.63±0.23 c	2.90±0.34 b	0.48±0.00 b
	digestion	11.60 ± 0.58 a	2.66 ± 0.10 a	15.47±0.23 c	0.96±0.27 b	0.96±0.00b
	Ashing	2.99 ± 0.78 a	1.33 ± 0.10 a	3.28±0.37 c	1.93±0.14b	0.48±0.00b
D	Ultra-Sonic	5.83 ± 0.33 a	0.31 ± 0.12 a	9.83±0.26 c	1.93±0.12b	0.97±0.04a
	digestion	7.54 ± 0.51 a	0.21 ± 0.14 a	12.47±0.31 c	2.40±0.53b	0.68±0.02a
	Ashing	9.12 ± 2.50 a	0.15 ± 0.15 a	10.67±0.17 c	0.66±0.34 b	0.50±0.00b
E	Ultra-Sonic	4.83±0.32 a	0.29±0.42 a	9.64±0.23 c	1.23±0.86 b	0.67±0.06a
	digestion	7.05±1.55 a	0.19±0.11ba	11.47±0.53c	2.10±0.12b	0.58±0.04a
	Ashing	8.99 ± 0.45 a	0.09 ± 0.12 a	9.57 ± 0.12 c	0.56±0.53b	0.48±0.42ab
F	Ultra-Sonic	5.83 ± 0.49 a	0.75 ± 0.42 a	4.96 ± 0.14 c	1.93 ± 0.42 b	0.87±0.14a
	digestion	5.36 ± 1.61 a	0.34 ± 0.14 a	4.46±0.68 c	1.34±0.26 b	1.91±0.53a
	Ashing	4.21± 0.32 a	0.46 ± 0.53 a	5.32± 0.34 c	0.88 ± 0.37 b	0.66 ± 0.12 a
Average mean		7.32	0.77	8.61	1.56	0.91
Pooled St Dev		4.023	0.745	3.939	0.981	0.424
Limited allowed ⁽¹⁷⁾		40	3	30	6	2
P-Value		0.550	0.850	0.770	0.506	0.023
F-Value		0.62	0.16	0.27	0.71	4.89

ND = Under Detection Limit

(a b c) similar letters in one column mean no significant differences while different letters mean significant differences

1- Cadmium

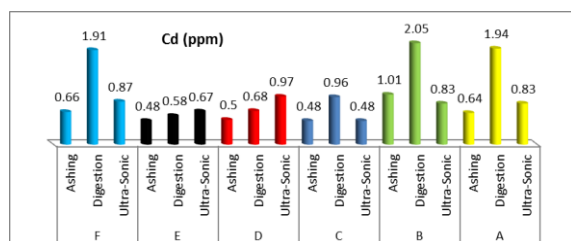


Figure (6): The concentration of Cadmium in the samples

Figure (6) shows the concentration of cadmium in the target samples. The above Figure shows that the cadmium concentration in the (B) sample was the highest concentration (2.05 ppm) according to the

digestion technique, which is much higher than the previous studies and slightly higher than the Iraqi standards. The concentration of cadmium was (0.48 - 0.97) ppm by ultra-sonic technique, while (0.48-1.01ppm) in ashing technique, these results agreed with the results of [9] and [11]. Then follow the sample (B) Samples (A) and (F) with the highest concentrations (1.94ppm) and (1.91ppm), respectively according to the technique of digestion, which is slightly higher than what researchers found [9] and [11] while the concentration in other techniques (0.87-0.66 ppm) for the sample (F) and (0.83-0.64 ppm) for sample (A) using techniques Ultra-Sonic and Ashing respectively, while the Cadmium concentrations in the other samples within the Iraqi standards and compatible with the findings of [10], [12] and [13].

2- Lead

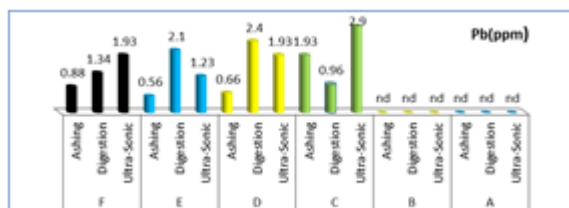


Figure (7): The concentration of Lead in the samples

The above Figure illustrates the absence of sample (A) and (B) of Lead, whose concentration is below the detection limit (nd) of the device, which is (< 0.01 ppm). This finding is consistent with that found [12]. The highest concentration of lead was found in the form of (C) which was (2.9 ppm) in digestion technique and this agrees with the finding of [9] while disagree with [10]. This difference could be attributed to the type of packaging or extraction technique used for preparation. The typical concentration was found in sample (C), (D) and (E) which was (2.4 ppm) and (2.1 ppm) respectively. According to digestion technique the sample (F) was little different with other samples with the highest concentration of lead through ultra-sonic reached (1.39 ppm) and this was within the levels allowed according to Iraqi standards. The ashing technique has relatively little concentration due to high temperature to about 550°C resulting in loss of heavy elements.

3 – Iron

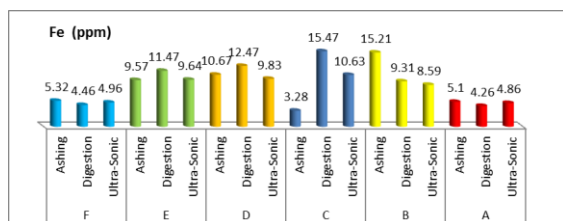


Figure (8): The concentration of Iron in the samples

The above Figure shows that the highest concentration of Iron was found in sample (C) which was (15.47 ppm) in the digestion technique. The concentration of Iron in the samples (D) and (E) was (12.47 ppm) and (11.47 ppm) respectively in the technique of digestion and this was much lower than what was found [11] and higher than what was found [13]. The highest concentration in ashing techniques was in the sample (B) (15.21 ppm) which is much lower than what was found [11] and higher than found [13]. The lowest concentration of Iron in sample (A) was (4.26 ppm) which was agreed with [13]. However, there is no difference between the concentration of Iron in other samples and in any of the techniques in which the concentration of iron in the target samples was within the Iraqi standards and within the limits allowed by the World Health Organization (WHO) and the Food and Agriculture Organization (FAO).

4- Copper

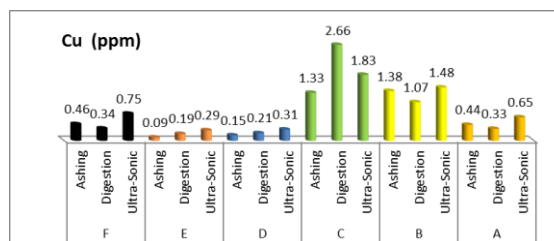


Figure (9): The concentration of Copper in the samples

The above Figure shows that the highest concentration of copper was found in sample (C) which was (2.66 ppm) in the digestion technique. The highest concentration through the techniques of ashing and ultra-sonic where it reached (1.33 ppm) and (1.83 ppm) respectively in sample (C). These results were agreed with these which are founded [9] and [13], and much lower than of [11]. These results are higher than results [12]. The result samples (A), (D), (E), and (F) were agreed with their founded [12]. Which reached the concentration of copper in them (0.09-0.75 ppm) in all the above three separation techniques, we found that the concentration of copper in all the varieties was within the levels allowed by the World Health Organization (WHO), the Food and Agriculture Organization (FAO) and the Iraqi Standards.

5. Zinc

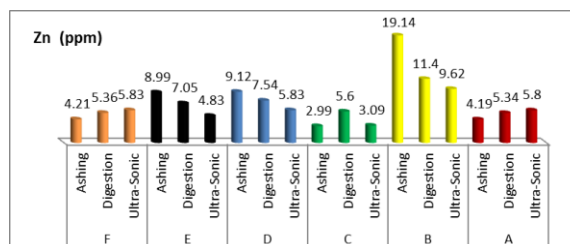


Figure (10): The concentrations of Zinc in the samples

The results shows the highest concentration of Zinc was found in sample (B) was (19.14 ppm) by using ashing technique these results are in agreed with many studies like [9], and are not agreement with other studies like [11,12] and [13]. The concentrations of Zinc in samples (A), (C), and (F) were somewhat similar to all the above results. The results for Zinc were within the limits allowed by the World Health Organization (WHO), the World Food Organization (FAO) and Iraqi standards.

Conclusion

- The results showed that the samples with metallic casing (A, B and F) had the highest contamination of cadmium, followed by samples with nylon bags (F) while local samples (D and E) were less polluted.
- As of Lead the highest ratio in sample (C) and local samples (D and E) was the lowest ratio while the two samples (A and B) were lead free
- For Iron, samples with metallic casing are the highest proportion of the rest of the samples, followed by samples of nylon bags, while local samples have the lowest ratio

- Copper is also no different from the previous results, with metal-coated samples having the highest proportion of copper while the local ratio is lower
- For Zinc, metal cans were the highest proportion followed by samples with nylon bags, while local was the lowest proportion

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- Most of the results were slightly above the permissible limits, especially those imported and canned. This shows that the elements may move from the metal cover to samples or contamination during the process of package. Therefore, we must depend on local foods because they are more safe than canned .

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طرق تحليلية مختلفة في تقدير العناصر الثقيلة في منتجات الاسماك المتوفرة في الاسواق المحلية العراقية بواسطة تقنية الامتصاص الذري

حمد فرحان موسى الجبوري ، سرحان علي سلمان العزاوي

قسم الكيمياء ، كلية العلوم ، جامعة تكريت ، تكريت ، العراق

الملخص

تهدف هذه الدراسة لتقدير العناصر الثقيلة (الكاديوم، الرصاص، النحاس، الخارصين والحديد) بطرق تحليلية مختلفة في منتجات الاسماك المحلية والمستوردة الحية والمعلبة والتي سحبت بصورة عشوائية من الاسواق المحلية ومن هذه العينات هي: (لحم تونا أبيض قطع صغيرة) إنتاج تايلند، (سردين بالزيت النباتي المفلفل شرائح بدون رؤوس) إنتاج المغرب، (شرائح سمك فيليه بدون عظام) إنتاج فيتنام، (سمك كارب حي من الاحواض وسمك كارب حي من نهر دجلة) إنتاج العراق، (سمك تونا قطع صغيرة) إنتاج تركيا، حددت تراكيز العناصر المذكورة اعلاه في هذه العينات المستهدفة بواسطة جهاز الامتصاص الذري اللهب، فكانت تراكيز العناصر الثقيلة مقدره بـ (ppm) على النحو التالي: حيث تراوحت قيم الكاديوم (0.48 - 2.05) بينما كانت قيم الرصاص (0.66 - 2.90) وقيم الخارصين (2.99 - 19.14) وقيم النحاس (0.15 - 2.66) وقيم الحديد (3.28 - 15.47). من خلال النتائج أن افضل طريقة للاستخلاص هي طريقة الهضم التي اظهرت نتائج افضل من الطريقتين الاخرى (طريقة الموجات فوق الصوتية وطريقة الترميد)، اظهرت النتائج أن مستويات الكاديوم والنحاس في بعض المنتجات اعلى من الحدود المسموح بها بحسب منظمة الصحة العالمية (WHO) ومنظمة الأغذية والزراعة العالمية (FAO) بينما كانت قيم الرصاص والحديد والخارصين ضمن الحدود المسموح بها. أن وجود هذه المعلبات والمنتجات الملوثة بالعناصر الثقيلة في الاسواق العراقية يعني غياب الرقابة الصحية والبيئية على ما ينتج وما يستورد مما يهدد السلامة والامن الغذائي الوطني بشكل مباشر .