The Determination of Some Heavy Metals in Different Selected Diets

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Abstract: The present study aims to determine the concentrations of the selected heavy metals in different food categories. Fifty-seven food samples were collected and digested using a mixture of analytical grade of strong mineral acids. The digested solution analyzed by ICP-AES and AAS techniques to determine quantitatively the concentration of lead, Arsenic, cadmium, copper and nickel in the investigated samples. The levels of the interested metals were compared with the maximum acceptable levels of heavy metals contents in food items according to the Codex alimentarius and the European Commission regulations in order to evaluate the potential risks and contamination. The study inducted that most of heavy metals in food samples were within acceptable levels except some rice samples had more arsenic concentration than the acceptable levels

Keywords: Heavy Metals, Food, AAS, ICP-AES, Codex.

INTRODUCTION

The awareness about the safety of food is increasing in several parts of the world. Many chemical compounds, such as acrylamide, pesticides, nitrosamines, and heavy metals, are considered as toxic contaminants when they occur at certain levels in the food (1). Many studies have shown that heavy metals have toxic effects even at very low concentrations. Heavy metals are natural components of the Earth's crust and cannot be degraded nor destroyed (2). They enter the human body through food, water and air. The consequence of heavy metal pollution can be hazardous to man through his food. Therefore, it is important to monitor heavy metals in aquatic environments such as water, sediment and biota (3-8).

Some elements including arsenic, cadmium and chromium are carcinogenic. Others, such as lead and mercury have a health implication and been associated with developmental abnormalities including autism in children when their levels in food proceed the acceptable (9, 10). The problem of heavy metals in food is rather complicated since their levels in food depend on several factors ranging from environmental conditions to the methods of production and processing. Plant might absorb heavy metals from soil, water or air.

Food might be easily contaminated during processing. High metals' concentration like lead (Pb), cadmium (Cd), copper (Cu) and nickel (Ni) in food have been correlated with the metal pollution in soil, air and water. Heavy metals are dangerous in their form of captions and highly toxic when bonded to their short chains of carbon atoms. Therefore, controlling the heavy metal concentration in food like cereals, fish and vegetables should be made to ensure their safety (11,12).

Heavy metals are ubiquitous; therefore, they tend to bio-accumulate and, hence, cause an increase in their concentration in a biological system. The interested heavy metals include lead, Arsenic, cadmium, copper and nickel. The levels of the interested metals in the selected food will be interpreted to determine the toxicity of such heavy metals according to the international regulations and recommendations like Codex alimentarius commission (13, 14) and the European Commission regulations (15, 16).

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Many instrumental analytical methods may be employed to measure the concentration level of heavy metals in various samples. The most predominant techniques are atomic absorption spectrometry (AAS), atomic emission/fluorescence spectrometry (AES/AFS), inductively coupled plasma mass spectrometry (ICP-MS), inductively coupled plasma optical emission spectrometry (ICP-OES), X-ray fluorescence (XRF) and anodic striping voltammetry (AVS) (17-23). Different digestion methods of food samples were investigated before to determine the best method for the recovery of the heavy metals (24-27).

The present study aims at determining the amount of the selected heavy metals in different food categories. These food samples include cereals, breads, rice, canned tuna, soft drinks and processed meat. The amount of heavy metals in the selected diets will be identified by two methods and compared with the maximum levels of the interested metals according to the codex and European Commission standards. This investigation compares the result of the induced coupled plasma-atomic emission spectroscopy to the atomic absorption spectroscopy of heavy metals analysis in different types of food.

MATERIAL AND METHODS

Material Used

Nitric acid (69% w/w) and sulphuric acid (98% w/w) for trace analysis were purchased from VWR (USA). The 1000 ppm stock standard solutions of Ni, Cu, As, Pb and Cd were purchased from Merck (Germany). The working standard solutions were prepared from stock solution by dilution using deionized water. All the used reagents were of analytical grade. Various food samples were bought from local markets in Amman city.

Instrumental Devices

Atomic Absorption Spectrophotometer WFX-210 (Rayleigh, China) with oxygen air-acetylene flame analysis technique was used to analyse the digested solutions. The wavelength range is between 190 and 900 nm. Flame temperature is continuously adjustable between 2300-2950 °C, which makes it possible to choose the best atomization temperature for different elements. Integrated flame/graphite furnace atomization system, changeable with flame emission burner, was involved. Hollow cathode lamps were used as sources of radiation and the background correction was provided by a deuterium lamp.

The inductively coupled plasma- atomic emission spectrometer (ICP-AES) is used for the analysis of the digested solutions. The Shimadzu ICPE-9820 multi-type ICP atomic emission spectrometer and UAG-1 ultrasonic nebulizer were used. The ICP-AES introduces a sample solution by spraying them into plasma which is generated by an induction coil supplied with a high-frequency current. The measurement conditions are indicated in Table 1.

Table 1: The analytical conditions of ICP-AES technique.

Instrument	ICPE-9820
Radio frequency power	1.2 kW
Plasma gas Flowrate	10 L/min
Auxiliary gas Flow rate	0.6 L/min
Carrier gas Flowrate	0.7 L/min
Sample introduction	Nebulizer 10
Misting chamber	Cyclone chamber
Plasma torch	Mini Torch
Observation	Axial (AX) / Radial (RD)

Investigated Diet Samples

Many categories of food samples were investigated in this study. Table 2 lists the cereal and bread samples. The different investigated rice samples are listed in Table 3. Table 4 shows the different canned tuna samples. The investigated processed meat samples are listed in Table 5. Table 6 lists the different investigated soft drink samples.

Sample Digestion of Grains, Breads and Rice

The samples were ground in the laboratory by using an electrical grinder. The digestion process was performed by using analytical grades of nitric acid (69% w/w) to recover the metal contents from the samples. Four grams of grained samples were mixed with 20 ml of nitric acids on a hot plate at 50 °C under the fume hood for three hours (28). All the obtained solutions from the digested samples were filtered by vacuum filtration and analyzed by the induced coupled plasma-atomic emission spectroscopy (ICP-AES) and atomic absorption spectroscopy (AAS).

Sample Digestion of Processed Meat and Canned Tuna

These samples were ground in the laboratory by using the electrical grinder. The digestion process was performed by using analytical grades of nitric acid and sulphuric acid in the ratio 3:1 to recover the metal contents from the samples.

Four grams of grained samples were mixed with 20 ml of a mixture of nitric acids and sulphuric acid on a hot plate at 50 ^{O}C under the fume hood for 3 hours. All the obtained solutions from the digested samples were filtered by vacuum filtration and analysed by the ICP-AES and AAS.

Sample Digestion of Soft Drink Samples

Prior to analysis, the samples were digested according to the method of Wallace (29). 10 ml of 69% concentrated nitric acid were added to 25 ml of the sample, and the mixture was evaporated on a hot plate in the fume hood until the brown fumes disappears leaving white fumes.

50 ml of distilled water was then added, and the solution was concentrated by evaporation on a hot plate to 25 ml. Subsequently, additional 25 ml of distilled water were added to make up the volume to 50 ml. All the obtained solutions from the digested samples were filtered by the vacuum filtration and analysed by the ICP-AES and AAS.

Determination of the Interested Metals in AAS Technique

The filtered solutions obtained from the digestion process were analyzed by flame atomic absorption spectroscopy (FAAS) or graphite furnace atomic absorption spectroscopy (GFAAS) -only in As analysis- and by ICP-AES. Table 7 indicates the analysis conditions of the interested metals by the ICP-AES and the AAS techniques.

Table 2: The cereal (C) and bread investigated samples

Sample ID	Sample description
C1	Wheat flour
C2	Ground rice
C3	Coarse Semolina
C4	fine semolina
C5	Soft Farik
C6	Groats
C7	Oats
C8	Chickpeas
C9	Bean
C10	Barley
C11	Biscuits
C12	Ground corn
C13	Brown lentil
C14	Vicia
C15	Bread (automatic oven)
C16	Lupine
C17	Whole grain Wheat
C18	Lima bean
C19	Bread (Taboon)
C20	Bread from whole grain wheat

Table 3: The investigated rice (R) samples

Sample ID	Sample description
R21	American rice (long)
R22	American rice (short)
R23	White basmati rice
R24	Bangladesh rice
R25	Indian rice
R26	puffed Bangladesh rice
R27	Chinese rice
R28	Thailand rice
R29	Iraqi rice
R30	Red Seri lank rice
R31	American rice
R32	Samba Seri lank rice

Table 4: The different investigated canned tuna (CT) samples

Sample ID	Sample brand
CT33	Sun bell
CT34	Postman
CT35	Maxim's
CT36	Durra
CT37	Goody
CT38	Carrefour
CT39	Alalali
CT40	Sun shine
CT41	Sunny sea

Table 5: The investigated processed meat (PM) samples

Sample ID	Sample description
PM42	Guinea fowl meat
PM43	Chicken meat
PM44	National Chicken meat
PM44	Roost beef meat
PM45	Exeter corned beef
PM46	Beef luncheon meat
PM47	Beef luncheon meat
PM48	Hot dog sausages
PM49	Hot dog sausages

Table 6: The different soft drink (SD) samples

Sample ID	Sample description
SD50	Shani drink
SD51	Mirinda drink
SD52	Vimto drink
SD53	Parley drink (Barbican)
SD54	Pepsi cola
SD55	Sprite light
SD56	Energy drink (Pison)
SD57	Coca cola Light

Table 7: The analysis conditions of ICP-AES and AAS and techniques

The metal	current of hollow cathode lamp (mA)	Wavelength (nm)	
		AAS	ICP-AES
As	9.0	193.7	193.7
Cd	8.0	228.8	214.43
Cu	9.0	324.7	224.7
Ni	7.0	232	231.6
Pb	9.0	283.3	220.35

RESULTS AND DISCUSSION

The Metal Contents of Investigated Samples by the ICP-AES and AAS

To analyse samples by the ICP-AES and the AAS techniques, the calibration curve of each element must be determined first with good linear regression. Figures 1-5 show the obtained calibration curves by AAS measurements of the interested metals. These elements include lead Pb, nickel Ni, cadmium Cd, arsenic As and copper Cu, respectively.
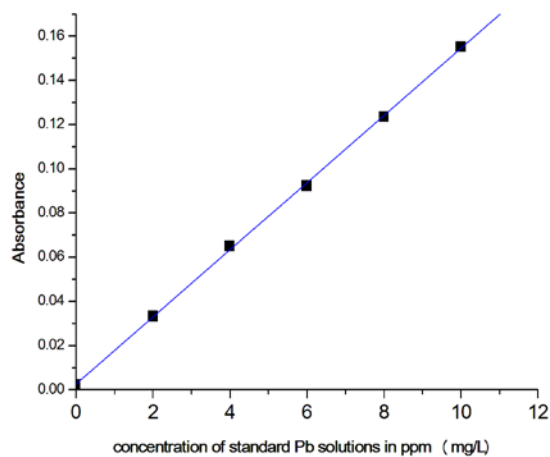
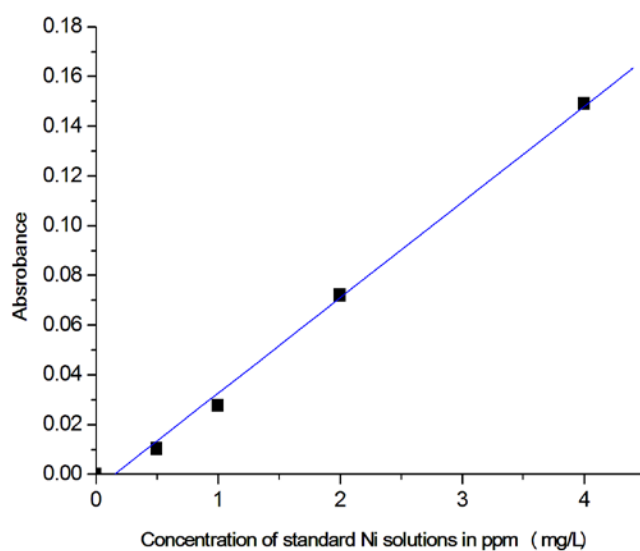Figure 1: The calibration curve of standard Pb solutions by the AAS technique

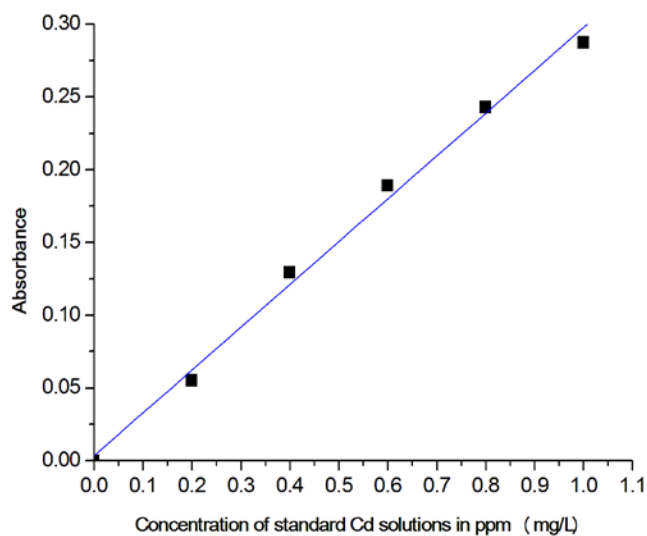Figure 2: The calibration curve of standard Ni solutions by the AAS technique

Figure 3: The calibration curve of standard Cd solutions by the AAS technique
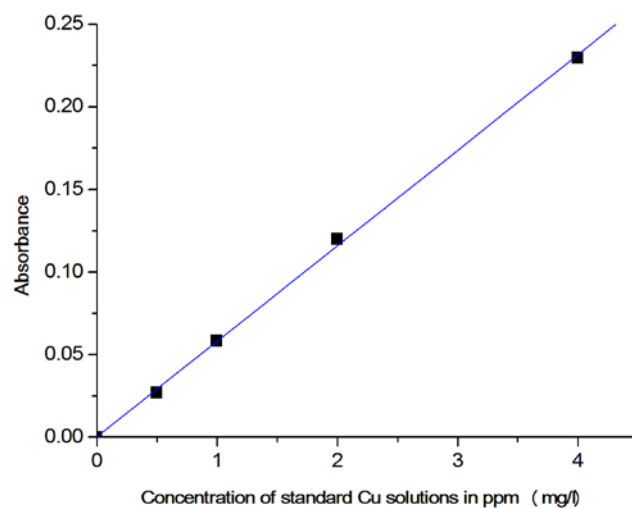
Figure 4: The calibration curve of standard Cu solutions by the AAS technique
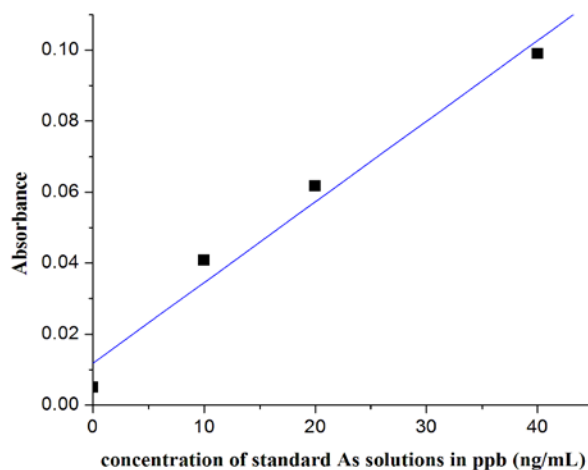

Figure 5: The calibration curve of standard As solutions by the AAS technique

The working calibration curves of each element of interest have been obtained also by using ICP-AES method. Figures 6-10 shows the calibration of Pb, Ni, Cd, Cu and As, respectively.
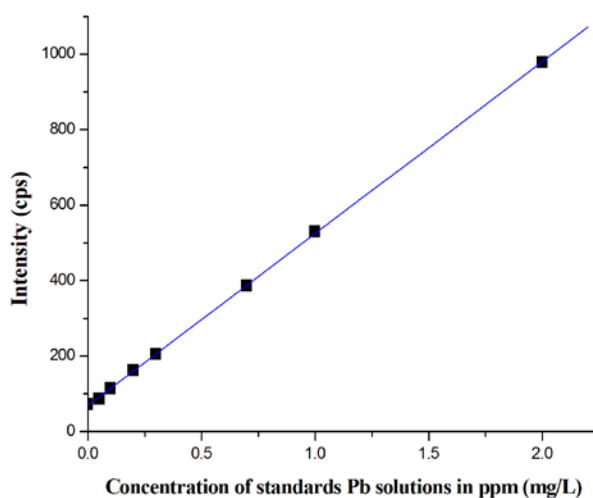

Figure 6: The calibration curve of standard Pb solutions by the ICP-AES technique
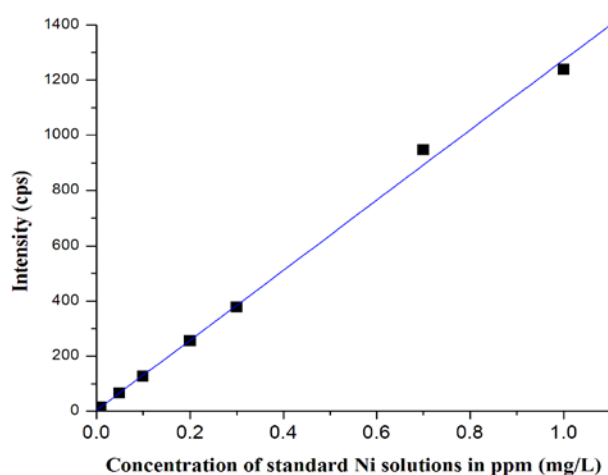
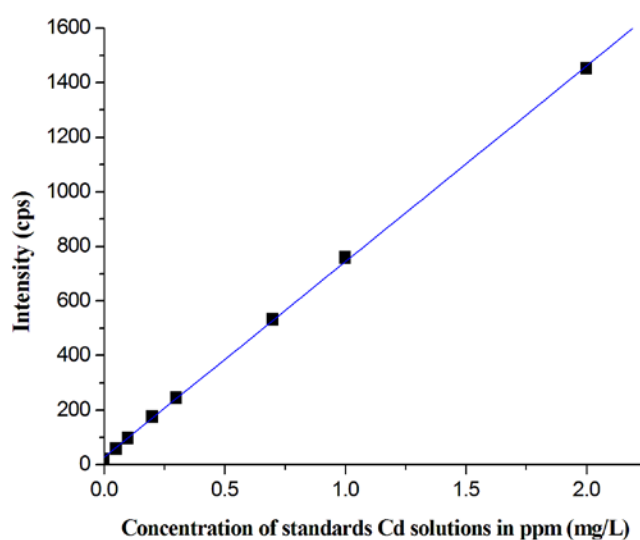Figure 7: The calibration curve of standard Ni solutions by ICP-AES technique

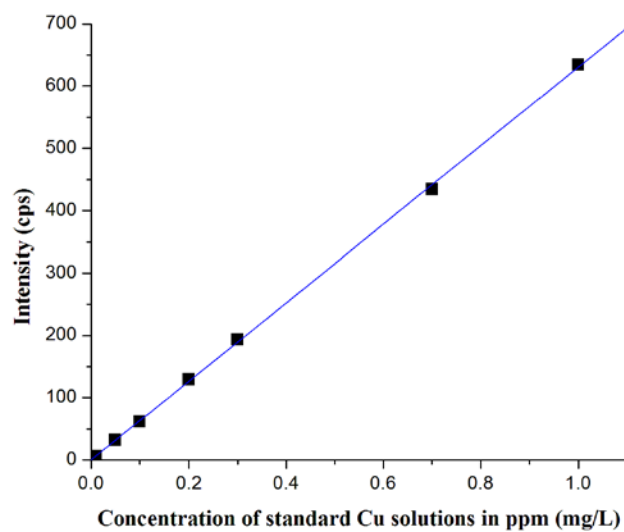Figure 8: The calibration curve of standard Cd solutions by the ICP-AES technique

Figure 9: The calibration curve of standard Cu solutions by the ICP-AES technique
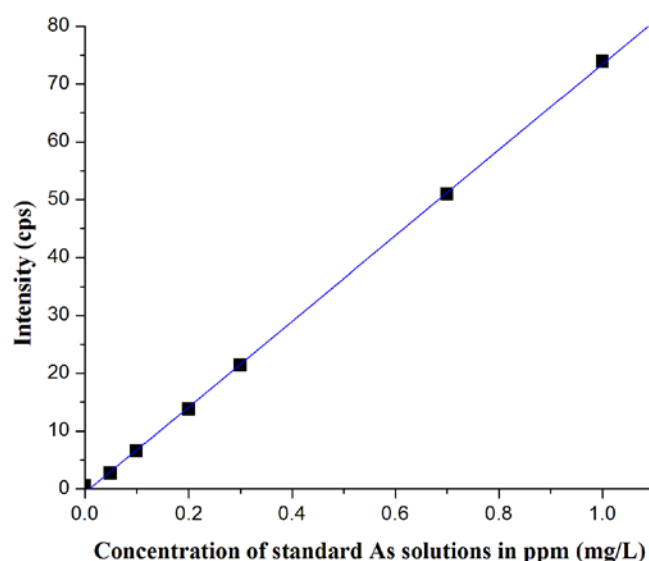
Figure 10: The calibration curve of standard As solutions by the ICP-AES technique

The correlation coefficient values that listed in Table 8 show the degree of linear association between the concentration of standards and absorption of the radiation. It is clear from the correlation coefficient values that the linearity of the obtained calibration curves from ICP-AES was better than that curves resulted from the AAS technique.

Table 8: The correlation coefficients of the obtained calibration curves from the AAS and ICP-AES measurements

The calibration curve	Correlation coefficient (r^2)	
	AAS	**ICP-AES**
Pb standards	0.9770	0.9999
Ni standards	0.9993	0.9995
Cd standards	0.9972	0.9999
Cu standards	0.9995	0.9995
As standards	0.9756	0.9996

The sensitivity of the analytical method can be described by values of Limit of Detection (LoD) and Limit of Quantification (LoQ). The LoD is the lowest concentration of analyte that can be detected and reliably distinguished from zero but not necessarily quantified.

The LoQ can be defined as the lowest concentration of analyte that can be determined quantitatively with an acceptable level of precision. To determine the minimal concentrations that the instrument can detect and measure quantitatively, the LoD and the LoQ of each calibration curve were calculated. The LoD values were calculated as 3.3 times of standards deviation (SD) of the blank solution, and the LoQ values were calculated as 10 times of SD value of the blank solution. The values of LoD and LoQ of each interested metal for both techniques (ICP-AES and AAS) are listed in Table 9. The LoD and LoQ values were lower in ICP-AES than in AAS. The ICP-AES technique provides higher detection sensitivity compared to AAS technique (30).

Table 9: The calculated values of LoD and LoQ of the AAS and ICP-AES measurements

The metal	**LoD** (ppm)		**LoQ** (ppm)	
	AAS	**ICP-AES**	**AAS**	**ICP-AES**
Pb	0.18	0.02	0.6	0.1
Ni	0.06	0.009	0.2	0.05
Cd	0.06	0.02	0.2	0.1
Cu	0.06	0.06	0.2	0.1
As	0.04	0.01	0.1	0.1

The contents of Pb, Cu and Ni in the investigated cereal and bread samples are listed in the Table 10. The results from both techniques (ICP-AES and AAS) were in a good consensus.

The concentration of Cd and As in the investigated samples were not detected in all cereal and bread investigated samples. The arsenic contents were determined in the rice samples only. The Arsenic contents in the investigated rice samples are indicated in the Table 11. The contents of Pb, Cu, Cd and Ni in the investigated canned tuna are listed in the Table 12. Table 13 lists the concentrations of metals of interest in the selected processed meat samples. The Ni and As contents are not detected in all processed meat samples. Table 14 lists the concentrations of interested metal in the soft drink samples.

Table 10: The metal contents in the cereal and bread investigated samples

Sample ID	Cu conc.*		Pb conc.		Ni conc.	
	Mean ± SD		Mean ± SD		Mean ± SD	
	ICP-AES	AAS	ICP-AES	AAS	ICP-AES	AAS
C1	4.4± 0.06	5± 0.1	0.08 ±0.03	n.d	n.d	n.d
C2	1.9	1.5	0.09	n.d	0.34± 0.05	0.5± 0.1
C3	1.4	1.5	0.08	n.d	0.05	n.d
C4	1.8	1.5	0.05	n.d	n.d	n.d
C5	3.3	3.1	0.09	n.d	0.09	n.d
C6	2.6	3	0.07	n.d	0.17	0.3
C7	2.6	3	0.15	0.2	0.26	0.3
C8	5.2	5	0.13	0.2	2.22	2.5
C9	25	20	0.9	1.0	1.38	1.5
C10	3.3	3	0.09	n.d	0.50	0.5
C11	1.4	1.5	0.09	n.d	0.01	n.d
C12	1.3	1.5	0.07	n.d	0.16	0.2
C13	5.0	6	0.1	n.d	0.53	0.6
C14	4.1	5	0.1	n.d	0.42	0.5
C15	1.4	1.0	0.09	n.d	n.d	n.d
C16	5.1	5	0.11	n.d	1.46	1.5
C17	5.4	5	0.09	n.d	0.21	0.2
C18	4.4	5	0.55	0.7	0.38	0.4
C19	1.4	1.5	0.09	n.d	0.12	0.1
C20	2.0	1.8	0.05	n.d	0.17	0.2

*all concentration units present in the table are in mg/L (ppm)
 n.d: not detected, SD: standard deviation

Table 11: The Arsenic contents in the investigated rice samples

Sample ID	As conc. (mg/L)	
	Mean ± SD%	
	ICP-AES	GF-AAS
R21	0.20 ± 0.03	0.24 ± 0.05
R22	0.06 ± 0.01	0.09 ± 0.03
R23	0.03 ± 0.01	0.04 ± 0.02
R24	0.04 ± 0.01	0.06 ± 0.02
R25	0.05 ± 0.01	0.06 ± 0.02
R26	0.06 ± 0.01	0.08 ± 0.03
R27	0.73 ± 0.04	0.81 ± 0.06
R28	0.21 ± 0.03	0.28 ± 0.04
R29	0.52 ± 0.03	0.65 ± 0.06
R30	0.40 ± 0.03	0.43 ± 0.04
R31	0.32 ± 0.02	0.42 ± 0.04
R32	1.16 ± 0.05	1.50 ± 0.08

Table 12: The metal contents in the investigated canned tuna samples

Sample ID	Cu conc.*		Cd conc.		Pb conc.		Ni conc.	
	ICP-AES	AAS	ICP-AES	AAS	ICP-AES	AAS	ICP-AES	AAS
CT33	0.37± 0.05	0.4± 0.08	0.18± 0.05	0.2 ± 0.1	0.52 ± 0.05	0.7 ± 0.08	0.2 ± 0.04	0.3± 0.08
CT34	0.14	0.3	0.03	n.d	0.05	n.d	0.6	0.3
CT35	0.24	0.4	0.12	0.1	0.31	0.2	0.6	0.5
CT36	0.14	0.5	0.06	n.d	0.09	n.d	0.7	0.5
CT37	0.17	0.4	0.05	n.d	0.09	n.d	0.1	0.4
CT38	0.21	0.5	0.10	0.1	0.12	n.d	0.1	0.4
CT39	0.20	0.4	0.11	0.1	0.11	n.d	0.1	0.5
CT40	0.09	0.4	0.09	0.1	0.11	n.d	n.d	0.5
CT41	0.13	0.4	0.09	0.1	0.25	0.3	0.1	0.5

*all concentration units present in the table are in mg/L (ppm), n.d: not detected

Table 13: The metal contents in the investigated processed meat samples

Sample ID	Cu conc.*		Pb conc.		Cd conc.	
	ICP-AES	AAS	ICP-AES	AAS	ICP-AES	AAS
PM42	0.37± 0.06	0.6± 0.1	0.52± 0.05	0.7± 0.08	n.d	n.d
PM43	0.23	0.5	0.05	n.d	0.18± 0.04	0.1± 0.06
PM44	0.31	0.4	0.31	0.5	0.03	n.d
PM45	0.25	0.5	0.09	n.d	0.12	0.1
PM46	0.27	0.5	0.09	n.d	0.05	n.d
PM47	0.29	0.5	0.13	0.3	0.05	n.d
PM48	0.15	0.4	0.12	n.d	0.10	0.1
PM49	0.23	0.5	0.12	n.d	0.09	n.d
PM50	0.18	0.4	0.21	n.d	0.09	n.d

*all concentration units present in the table are in mg/L (ppm), n.d: not detected

Table 14: The metal contents in the investigated soft drink samples

Sample ID	Cu conc.*		Pb conc.		Ni conc.		Cd conc.	
	ICP-AES	AAS	ICP-AES	AAS	ICP-AES	AAS	ICP-AES	AAS
SD51	0.06 ± 0.02	0.4 ± 0.1	0.57± 0.06	0.30± 0.1	n.d	0.36± 0.1	0.01± 0.008	0.04± 0.02
SD52	0.05	0.3	0.12	0.28	n.d	0.16	0.10	0.03
SD53	0.06	0.3	0.37	0.71	0.4± 0.05	0.53	0.01	0.03
SD54	0.06	0.4	0.08	0.50	0.02	0.70	0.05	0.08
SD55	0.06	0.3	0.10	0.37	0.05	0.48	0.01	0.03
SD56	0.02	0.1	0.39	0.43	0.03	0.44	0.02	0.05
SD57	0.06	0.1	0.08	0.09	0.02	0.35	0.01	0.03

*all concentration units present in the table are in mg/L (ppm), n.d: not detected

The maximum levels for heavy metals contents in foodstuffs according to the codex commodity and EU commission standards are shown in Table 15. When comparing the metals contents in the selected food samples with the maximum levels of the interested metals according to the codex and EU commission standards (Table 15), some samples have more levels than the allowed. For example, Samba Seri lank rice had six times arsenic concentration than the accepted concentration (0.2 ppm for arsenic).

Table 15: The maximum levels for As, Pb, Cd and Sn contents in foodstuffs according to the codex commodity and EU commission standards

Metal	Maximum level (mg/kg)	
	Codex commodity	EU commission
As	0.2 (Rice)	-
Pb	0.2 (cereal grains) 0.3 (fish) 0.5 (Luncheon meat) 0.1 (canned juice)	0.2 (cereals and legumes) 0.3 (fish) 0.1 (Meat) 0.05 (juice)
Cd	0.1 (Cereal grains) 0.4 (rice) 0.2 (wheat)	0.1 (Cereals) 0.1 (tuna) 0.05 (meat) 0.2 (wheat and rice)
Sn	250 (Canned foods) 150 (Canned beverages) 50 (Luncheon meat)	200 (Canned foods) 10 (Canned beverages) 50 (Luncheon meat)

CONCLUSION

The ICP-AES and AAS techniques were used to identify the contents of Pb, Ni, Cd, Cu and As in seventy-five diet samples. The working calibration from the ICP-AES and AAS measurements shows a very good linear relationship with low values of LoD and LoQ. The two analysis methods gave relatively close results with a preference to the ICP-AES technique. Some Seri lank and Chinese rice samples had more Arsenic concentration than that allowed according to the international requirements. Therefore, strict inspection should be carried out on these rice brined.

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