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Full Length Research Paper

Evaluation of some heavy metals in *Pangasius hypothalmus* and *Tilapia nilotica* and the role of acetic acid in lowering their levels

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Concentrations of cadmium, mercury, manganese, phosphorus, lead and zinc in samples of Basa fish (*Pangasius hypothalmus*) and some fresh water fish (*Tilapia nilotica*, named Bolti and Karmout) collected from Kafer-El-Zayat were determined. Karmout fish contained the highest level of metals where phosphorus level was 3.45 µg/g, lead (1.51 µg/g), zinc (1.03 µg/g), cadmium (0.17 µg/g), manganese (0.08 µg/g) and mercury (0.007 µg/g). In Bolti fish, phosphorus level was 4.03 µg/g, lead (0.83 µg/g), zinc (0.62 µg/g), cadmium (0.12 µg/g), manganese (0.08 µg/g) and mercury (0.004 µg/g). In Basa fish, phosphorus level was 4.06 µg/g, lead (0.79 µg/g), zinc (0.62 µg/g), cadmium (0.12 µg/g), manganese (0.07 µg/g) and mercury (0.004 µg/g). Cadmium and lead levels in all examined fish were higher than permissible safety level of human use (0.1 ppm). Pre-treatment of fish muscles by acetic acid (5%) for a period of 15 min resulted in marked reduction of metals in these fish. When the Basa fish was immersed in 5% acetic acid, the cadmium level in the flesh was reduced from 0.12 to 0.07 µg/g (41.6%) and lead level was reduced from 0.83 to 0.41 µg/g (51.9%). In Bolti fish, pre-treatment of the fleshy parts by acetic acid reduced cadmium level from 0.12 to 0.07 µg/g (58.3%) and lead was reduced from 0.83 to 0.02 µg/g (24%). The same result was observed in Karmout fish, where pre-treatment with acetic acid lowered the cadmium level from 0.17 to 0.05 µg/g (29.4%) and lead was lowered from 1.51 to 0.82 µg/g (54.3%).

Key words: Basa fish, *Tilapia nilotica*, Karmout, acetic acid, heavy metals.

INTRODUCTION

Fish had long been regarded as a desirable and nutritional source of high quality protein and generous supply of minerals and vitamins. During the last few decades, great attention has been paid to the possible dangers of heavy metal poisoning in human due to the consumption of contaminated fish.

Industrial and agricultural discharges such as coal and oil combustion, phosphate fertilizers, plastics and pesticides are considered the major sources of heavy metal pollutants of water. Fish absorbed heavy metals from water through the gills, skin and digestive tract. The heavy metals of the most wide spread concern to human health are lead, copper, mercury and cadmium (Kris-Etherton et al., 2003; Chen et al., 2007; Din et al., 2008; Bhouri et al., 2010). Heavy metals are recognized as toxic substances due to their low rate of elimination from the consumer body; either man or animals (WHO, 1992; Harris, 2007; Wafaa et al., 2003).

Mriagu (1988) suggested that, over one billion human beings are currently exposed to elevated concentrations of toxic metals and metalloids in the environment and several million people may be suffering from subclinical metal poisoning. Heavy metals in freshwater is a matter of concern because of their toxic potential ability to be accumulated in food chain. The adverse toxic effects caused by cadmium and lead (Wafaa et al., 2003), mercury, and zinc (Abd El-Nasser et al., 1996) are widely

Abbreviations: **PFA,** Berfluoroalkoxy; **PTFE,** polytetrafluoroethylene; **ICP-OES,** inductively coupled plasma-optical emission spectroscopy; **QC,** quality control; **MDL,** method detection limit.

Table 1. Results for reference material of International Atomic Energy Agency (IAEA - 407).

Element	Reference material	
	Certified value	Observed value
Zn (mg kg^{-1})	67.1 ± 3.8	66.9 ± 5.5
Cu (mg kg^{-1})	3.3 ± 0.4	3.2 ± 0.7
Fe(mg kg^{-1})	146 ± 14	151 ± 6.5
Mn (mg kg^{-1})	3.5 ± 0.3	3.7 ± 0.7
Mg (g kg^{-1})	2.7 ± 0.1	2.6 ± 0.9
Na(g kg^{-1})	13.1 ± 0.6	12.7 ± 0.8
K(g kg^{-1})	13.1 ± 1.2	11.9 ± 1.0
Ca(g kg^{-1})	27.0 ± 1.8	28.3 ± 1.5

recognized for their detrimental effects on human health. Metals also, may act as allergens, mutagens or carcinogens (Goyer, 1992).

Many investigators (Mahdy et al., 1993; Jehan et al., 1999; Zienab, 2001; Wafaa, 2003; Turkmen et al., 2008; Vinodehini and Narayanan, 2008) have studied the presence of heavy metals in surface waters and in fish. Some authors investigated the removal of heavy metals from water (Gado and Midany, 2003). Up till now, methods of removal of heavy metals and other pollutants from fish tissue have not been well investigated.

The aim of this study is to measure some heavy metals (cadmium, mercury, manganese, phosphorus, lead, and zinc) in Basa fish (*Pangasius hypothalmus*) and some fresh water fish (*Tilapia nilotica*, named Bolti and Karmout). Also, the beneficial effect of acetic acid in lowering the levels of these heavy metals in edible fish tissue was studied.

MATERIALS AND METHODS

Sampling

Basa fish were collected from different sites in the markets with different exporters from Vitenam in the form of frozen Basa filet 20% glazing. Fresh water fish (Bolti and Karmout) were collected from Kafer-El-Zayat from February to April, 2009. The total weights of the samples after collection were measured to the nearest gram (g) before dissection. The mean weight of Karmout fish was 800 ± 50.6 g (that is, average age of 9 to 10 months) while the mean weight of Bolti fish was 350 ± 20.4 g (that is, average age of 4 to 5 months). The average weight of Basa filet was 175 g. Approximately, 100 mg sample of muscle and 500 mg of fat from each fish were dissected, washed with distilled water, weighed, packed in polyethylene bags and stored at -18°C for chemical analysis.

Chemical analysis

Digestion procedure of fish muscle and fat

The muscle and fat samples analyzed in this experiment were digested as follows: The sample was taken in a 6 ml perfluoroalkoxy (PFA) tube, which was cleaned by boiling in a diluted (2-fold) aqua regia for 8 h, the polytetrafluoroethylene (PTFE) ball was put as the cover cap on the top of the tube. After adding 1 ml of concentrated HNO_3 to the muscle sample and HNO_3 (H_2O_2) to the fat sample, they were heated on an aluminum heating-block at 190°C for 2 h. After removing the PTFE ball, the sample was further heated almost to dryness at 200°C. Finally, the residue was dissolved with 5 ml of 0.1 M HNO_3, which contained the internal standard elements (Ge, Rh and Re; 10 ng ml^{-1} each). The goal of every digestion process was therefore the complete solution of the analytes and the complete decomposition of the sold (matrix) while avoiding loss or contamination of the analyte; thus the obtained samples solutions were subjected to the inductively coupled plasma-optical emission spectroscopy (ICP-OES) measurements as the final analysis solution. Heavy metals were assayed using the induced ICP-OES and the results were given as µg/g dry wt.

Data obtained from the experiments were analyzed and the results were expressed as mean ± S.D. The results were evaluated using Student's t-test. Values of $P < 0.001$ were considered statistically significant.

Quality control to check accuracy

Quality control (QC) is an important step to measure the presence of trace elements for reference materials of International Atomic Energy Agency (IAEA – 407 and A-13), as shown in Tables 1 and 2. The QC sample was run at a frequency of once in every 10 samples; to check for the calibration accuracy and the instrument drift. The results within 10% of the known QC values were deemed acceptable. Blank samples were run in duplicate in each analysis batch in a randomized order and were used to calculate the method detection limit (MDL).

RESULTS AND DISCUSSION

Monitoring of some heavy metals in edible parts of Basa fish and some fresh water fish (*Tilapia nilotica* named Bolti and Karmout) was carried out. The flesh is one of the ultimate parts for heavy metals accumulation. The highest concentration of cadmium, mercury, manganese, lead and zinc were found in Karmout fish.

Concentrations of lead and cadmium in Basa fish and *T. nilotica* named: Bolti and Karmout were higher than the permissible level recommended by EQSQC (1993). Bolti fish contained the highest level of phosphorus and least

Table 2. Results for reference serum sample of International Atomic Energy Agency (IAEA) (A-13).

Element	Measured value (µg/L)[a] ± SD	Certified value (µg/L)
Li	5.91 ± 0.12	6.10
B	61.23 ± 1.48	63.00
Na	315.00 ± 5.87	308.00
Mg	20.02 ± 0.22	20.00
Al	19.92 ± 0.35	19.00
K	165.77 ± 2.61	167.00
Ca	95.11 ± 0.81	94.00
Cr	0.26 ± 0.005	0.26
Mn	3.73 ± 0.07	3.60
Fe	1.28 ± 0.04	1.30
Co	0.08 ± 0.0.0008	0.08
Cu	366.24 ± 7.01	370.00
Zn	1058.51 ± 18.03	1070
Se	25.45 ± 0.41	26.00
Br	2013 ± 31	2040
Ru	64.18 ± 1.11	66
Cd	0.074 ± 0.0008	0.074
Sn	1.21 ± 0.01	1.20
I	47.56 ± 0.53	47.00
Cs	0.36 ± 0.007	0.36
Hg	0.16 ± 0.002	0.16

Table 3. Heavy metals analysis in flesh of different fish (µg/g dry weight) (treated with HCl only).

Heavy metal	Basa		Bolti		Karmout		Permissible level*
	Mean	SD	Mean	SD	Mean	SD	
Cd	0.12	0.011	0.12	0.005	0.17	0.02	0.1
Hg	0.004	0.00	0.004	0.00	0.007	0.002	0.1
Mn	0.07	0.005	0.08	0.01	0.08	0.009	NAD
Ph	4.06	0.05	4.03	0.03	3.45	0.26	NAD
Pb	0.79	0.05	0.83	0.04	1.51	0.128	0.1
Zn	0.62	0.015	0.62	0.01	1.03	0.05	50

The values were statistically significant at $p < 0.001$; NAD, No available data; *Egyptian Organization for Standardization and Quality Control (EOSQC, 1993); SD, standard deviation.

level of zinc (Table 3). There was no significant change between heavy metals in the flesh or fat for each type of fish (Tables 3 and 4).

There was a marked reduction of heavy metals concentration after the addition of acetic acid to the flesh of Basa, Bolti and Karmout fish as shown in Tables 5, 6 and 7. When Basa fish was immersed in 5% acetic acid for 15 min, the cadmium level was reduced from 0.12 µg/g in the flesh to 0.07 µg/g in the homogenate (41.6%) and the lead level was reduced from 0.83 to 0.41 µg/g (51.9%). In the Bolti fish, pre-treatment of the fleshy parts by acetic acid reduced cadmium level from 0.12 to 0.07 µg/g (58.3%) and the lead level was reduced from 0.83 to 0.02 µg/g (24%). The same finding was observed in Karmout fish, where pre-treatment with acetic acid lowered cadmium level from 0.17 to 0.05 µg/g (29.4%) and lead level was lowered from 1.51 to 0.82 µg/g (54.3%). A considerable amount of heavy metals was found in the liquid of these fishes.

The natural aquatic system has been extensively contaminated with heavy metals released from domestic, industrial, agricultural and other man-made activities (Conacher et al., 1993). Among animal species, fish are the inhabitants that cannot escape from the detrimental effects of these pollutants (Clarkson, 1998; Dickman and Leung, 1998; Olaifa et al., 2004).

Table 4. Heavy metals analysis in fat of different fish (µg/g dry weight) (treated with HCl only).

Heavy metal	Basa		Bolti		Karmout		Permissible level*
	Mean	SD	Mean	SD	Mean	SD	
Cd	0.078	0.019	0.136	0.01	0.17	0.04	0.1
Hg	0.002	0.0004	0.004	0.0006	0.007	0.002	0.1
Mn	0.78	0.017	0.08	0.01	0.82	.008	NAD
Ph	2.36	0.16	4.11	0.02	3.33	0.55	NAD
Pb	0.53	0.087	0.81	0.02	1.49	0.43	0.1
Zn	0.86	0.07	0.61	0.02	1.00	0.09	50

*Egyptian Organization for Standardization and Quality Control (EOSQC, 1993); NAD, No available data; SD, standard deviation.

Table 5. Heavy metals in flesh of Basa (not pre-treated with acetic acid) and Basa homogenate and its liquid (pre-treated with acetic acid).

Heavy metal	Basa (not pre-treated) (µg/g dry weight)		Basa (pre-treated with acetic acid)				
			Homogenate (µg/g dry weight)			Liquid (µg/sample)	
	Mean	SD	Mean	Percent	SD	Mean	SD
Cd	0.12	0.011	0.05	(41.6)	0.03	0.07	0.04
Hg	0.004	0.01	0.002	(50)	0.001	0.002	0.001
Mn	0.07	0.005	0.05	(71.4)	0.004	0.02	0.02
Ph	4.06	0.05	2.32	(57.1)	0.013	1.62	0.34
Pb	0.79	0.05	0.41	(51.9)	0.03	0.37	0.22
Zn	0.62	0.015	0.28	(45.1)	0.016	0.34	0.12

SD, Standard deviation.

Table 6. Heavy metals in flesh of Bolti fish (not pre-treated with acetic acid) and Bolti homogenate and its liquid (Pre-treated with acetic acid).

Heavy metal	Bolti (not pre-treated) (µg/g dry weight)		Bolti (pre-treated with acetic acid)				
			Homogenate (µg/g dry weight)			Liquid (µg/sample)	
	Mean	SD	Mean	Percent	SD	Mean	SD
Cd	0.12	0.005	0.07	(58.3)	0.004	0.05	0.003
Hg	0.004	0.001	0.001	(25)	0.001	0.003	0.001
Mn	0.08	0.01	0.02	(25)	0.01	0.06	0.01
Ph	4.03	0.03	2.11	(52.3)	0.02	1.9	0.11
Pb	0.83	0.04	0.2	(24)	0.03	0.63	0.02
Zn	0.62	0.01	0.41	(66)	0.02	0.21	0.03

SD, Standard deviation.

The principle behind this study was the analysis of different types of fish to their metal contents in order to monitor metal excess in their tissues. Two species of fresh water fish (*T. nilotica,* named Bolti, and Karmout) caught from River Nile at Kafer El-Zayat, were used to detect the presence of heavy metals and its accumulation in their edible tissues. Also, the samples from Basa fish were taken. Bolti and Karmout were chosen as they are used extensively by millions of people in Egypt. Basa fish was analyzed because of many suspicions about their level of pollution being imported from Vietnam. Basa, Bolti and Karmout fish were chosen from the average weights to be sure of suitable age and period of exposure to pollutants and to be among edible size.

The mean values of cadmium level in the fleshy parts of examined fish recorded in Table 3 were 0.12, 0.12, and 0.17 µg/g for Basa, Bolti and Karmout, respectively. These values were over the permissible limits (0.1 ppm)

Table 7. Heavy metals in flesh of Karmout fish (not pre-treated with acetic acid) and Karmout homogenate and its liquid (pre-treated with acetic acid).

Heavy metal	Karmout (not pre-treated) (µg/g dry weight)		Karmout (pre-treated with acetic acid)				
			Homogenate (µg/g dry weight)			Liquid (µg/sample)	
	Mean	SD	Mean	Percent	SD	Mean	SD
Cd	0.17	0.02	0.05	(29.4)	0.01	0.12	0.01
Hg	0.007	0.002	0.003	(42.8)	0.001	0.004	0.005
Mn	0.08	0.003	0.02	(25)	0.001	0.06	0.001
Ph	3.45	0.026	1.1	(31.9)	0.15	2.28	0.155
Pb	1.51	0.12	0.82	(54.3)	0.03	0.69	0.02
Zn	1.03	0.05	0.52	(50.4)	0.04	0.51	0.01

recommended by the Egyptian Organization for Standardization and Quality Control (EOSQC, 1993). Cadmium level in Bolti fish was equal to that reported by Khallaf et al. (1994) in the study, where 0.12 µg/g was observed in the same area and fish. Similar results were reported by Seddek et al. (1996) in River Nile fish and Celik and Oehlenschlager (2007), where the cadmium levels varied from 0.1 to 0.8 ppm. Lower results were reported by Hamza et al. (1996) and Shokrzadeh et al. (2004); the recorded cadmium levels varied from 0.01 to 0.09 ppm. On the other hand, Tariq et al. (1994) and Juma et al. (2002) reported higher levels of cadmium which varied from 2.0 to 15.8 ppm. Also, the cadmium level in Karmout was higher than that reported by Khallaf et al. (1994) at the same location.

High incidence of cadmium in Bolti and Karmout fish caught from River Nile at Kafer El-Zayat may be due to the contamination of the fish from industrial waste of factories in the town. Mercury levels in all examined fishes were 0.004, 0.004 and 0.007 µg/g in Basa, Bolti and Karmout flesh respectively; 0.002, 0.004 and 0.007 µg/g in Basa, Bolti and Karmout fat respectively. Fortunately, these values were below permissible level (50 ppm) approved by EOSQC (1993). This may be related to the absence of mercurial pollutant at high risk. No information about the maximum permissible levels of manganese or phosphorus in the fish tissue of Egyptian standards was available EOSQC (1993).

The results of lead level obtained in this study were 0.79, 0.83 and 1.51 µg/g in Basa, Bolti and Karmout flesh, respectively and 0.53, 0.81 and 1.49 µg/g in Basa, Bolti and Karmout fat, respectively. These values are in harmony to those reported by Seddek et al. (1996) and Marouf and Dawoad (2006). The recorded levels ranged from 0.42 to 0.74 ppm. However, lower results were reported by Suppin et al. (2005). The registered levels varied from 0.04 to 0.076 ppm. On the other hand, higher results were recorded by Adekunle et al. (2004) and Saleh (2004). It was clear that the values obtained were over the permissible limits (0.1 ppm) recommended by the Egyptian Organization for Standardization and Quality Control (EOSQC, 1993).

Unfortunately, lead level in Bolti and Karmout exceeded that reported by Khallaf et al. (1994) at the same area and types of fish (0.05, 0.25 ppm respectively) which indicated more pollution of the aquatic field and more industrial pollution since 1994 to 2009. This could be related to more pollution after an excess of 15 years. Higher level in Karmout may be due to affinity of Karmout for retaining metals in tissue being older in age, with more chance for heavy metal bioaccumulation.

Concerning zinc, the data were 0.62, 0.62 and 1.03 µg/g in Basa, Bolti and Karmout flesh respectively and 0.86, 0.61 and 1.0 µg/g in Basa, Bolti and Karmout fat respectively. These values were lower than the results obtained by Jaffar et al. (1988) in the edible part of fresh water fish which ranged from 1.87 to 50.6 µg/g. According to Celik and Oehlenschlager (2004), zinc level was 8.6 µg/g. Celik and Oehlenschlager (2005) reported that zinc level was 9.73 µg/g. Mean levels of zinc in all examined fish muscles were lower than the permissible limit (50 ppm) recommended by EOSQC (1993).

Interestingly, there was a marked reduction of heavy metal levels in fleshy parts of the fish pre-treated with 5% acetic acid (similar to house-hold concentration). When Basa fish was immersed in 5% acetic acid (15 min), the cadmium level was reduced from 0.12 µg/g in flesh to 0.07 µg/g in homogenate (41.6%) and the lead level was reduced from 0.83 to 0.41 µg/g (51.9%). In Bolti fish, pre-treatment of fleshy parts by acetic acid reduced the cadmium level from 0.12 to 0.07 µg/g (58.3%) and the lead level was reduced from 0.83 to 0.02 µg/g (24%). The same finding was observed in Karmout fish, where pre-treatment with acetic acid lowered the cadmium level from 0.17 to 0.05 µg/g (29.4%) and the lead level was lowered from 1.51 to 0.82 µg/g (54.3%).

Reduction of heavy metals in fish pre-treated with acetic acid could be due to formation of insoluble acetate salts of these metals. Fish liquid contained a considerable amount of metals, leaked with liquid by filtration, and thus should be discarded to minimize metal exposure.

Gado and Midany (2003) reported a marked reduction of heavy metals in water of fish ponds after the addition of slaked lime and attributed this beneficial effect to the action of calcium carbonate that forms insoluble complexes with heavy metals (Dodge and Theis, 1979). In comparison with our trial, Satoh (1999) provided a method of removing heavy metals from a heavy metal containing substance; the method comprises of the following steps: 1) Addition of water and acid to a heavy-metal-containing substance to form a liquid having a pH equal to or lower than 4, and stirring the liquid; 2) extraction of heavy metals substance in the liquid, as heavy metal ions; 3) addition of alkali to the liquid to increase the pH of the liquid to 10 or more; and precipitating the heavy metal ions out of the liquid as a carbonate by bringing bubbles of carbonic acid gas into contact with the liquid having a pH equal to or higher than 10 (www.freepatentsonline.com/5958248.htm).

The method adopted seemed to be much easier and applicable. It needed only good immersion of fish in a diluted solution of acetic acid (5%) for 15 min; then getting rid of the liquid part (containing most of the metals) and repeated washing of the fish with tap water.

Conclusions

The result indicated that the heavy metal contamination definitely affected the aquatic life of different fish. Hence, a scientific method of detoxification is essential to improve the health of these fish in any stressed environ-mental conditions. Addition of acetic acid to fish tissue helped to reduce level of heavy metals. Further research studies on methods of removal of heavy metals from water and fish should be conducted.

REFERENCES

Abd El-Nasser M, Shaaban AA, Seham M, Aly, Manal M, Sayed (1996). Levels of some heavy metals in fish caught from river Nile at Assiut governorate, Egypt Assiut Vet. Med. J., (34): 68 -83.

Adekunle IM, Akinyemi MF (2004): Lead levels of certain consumer products in Nigeria. Smoked fish from Abeokuta. Food Chem. Toxicol., 42(9): 1463 -1468.

Bhouri AM, I Bouhlel, L Chouba, M Hammami, M El Cafsi, A Chaouch (2010): Total lipid content, fatty acid and mineral compositions of muscles and liver in wild and farmed sea bass (*Dicentrarchus labrax*), Afr. J. Food Sci., 4(8): 522 - 530,

Celik U, Oehlenschlager J (2004). Determination of zinc and copper in fish samples collected from North East Atlantic, Turkey. Food Chem., 87(3): 343-347.

Celik U, Oehlenschlager J (2005). Zinc and copper content in marine fish samples collected from eastern Mediterranean sea Turkey. European Food Research and Technology,220(1): 37-41.

Celik U, Oehlenschlager J (2007). High contents of Cd, Pb, Zn and Cu in popular fishery products sold in Turkish supermarkets. Food Control,18(3): 258-261.

Chen YQ, Edwards JJ, Kridel SJ, Thornburg T, Berquin IM (2007). Dietary fat gene interactions in cancer. Cancer. Metastasis. Rev. 26: 535-551

Clarkson TW (1998). Human toxicology of mercury. J. Trace. Elem. Exp. Med., 11 (2-3): 303-317.

Conacher HB, Page BD, Rayan JJ (1993). Industrial chemical contamination of food(Review).Food Addit Contam.,10(1): 129-143

Dickman MD, Leung KM (1998): Mercury and organochlorine exposure from fish consumption in Hong Kong. Chemosphere, 37(5): 991-1015.

Din JN, Harding SA, Valerio CJ, Sarma J, Lyall K, Riermersma RA, Newby DE, Flapan AD (2008). Dietary intervention with oil rich fish reduces platelet monocyte aggregation in man. Artherosclerosis, 197: 290-296

Dodge EE, Theis TL (1979): Effects of chemical speciation on the uptake of copper by *Chironomus tetans*. Environ. Sci. Technol., 13: 1287-1288.

Egyptian Organization for Standardization and Quality Control (EOSQC) (1993). Maximum level for heavy metal contamination in food E.S. No. 2360.

Gado MS, Midany SA (2003):Studies on some heavy metals pollutants in cultured *Oreochromis niloticus* fish at Kafer El-Sheikh Governorate. Kafer El-Sheikh Vet. Med. J., 1 (2) 83-95.

Goyer RA (1992): Toxic effects of metals. In: Amdur, M.O., Doull, J. and Klassen, C.D. (Eds) Casarett and Doull, s Toxicology, The basic science of poisons. Pergamon press, pp. 623-680.

Hamza CA, Romeo M, El-Aded A (1996). Heavy metals in different fishes from the Middle Eastern Coast of Tunisia. Bull. Environ. Contam. Toxicol., 56: 766-773.

Harris WS (2007). Omega-3 fatty acids and cardiovascular disease: A case for omega-3 index as a new risk factor. Pharmacol. Res., 55: 217-223

Jaffar M, Ashraf M, Rosool A (1988). Heavy metal contents in some selected local fresh water fish and relevant waters. Pak. J. Sci. Ind. Res., 31(3):189-193.

Jehan R, Daoud A, M Amin, Menha M, Abd El- Khalek (1999). Residual analysis of some heavy metals in water and *Oreochromis niloticus* fish from polluted areas. Vet. Med. J. Giza. 47 (3): 351-365.

Juma H, Battah A, Salim M, Tiwari P (2002). Arsenic and cadmium levels in important fresh and frozen fish in Jordan. Bull. Environ. Contam. Toxicol., 68(1): 132-137.

Khallaf MF, Neverty FG, Tonkhy TR (1994). Heavy metal concentration in fish and water of the River Nile and fish farms In: National conference on the River Nile, Assiut University, Egypt.

Kris-Etherton P, Harris WS, Appel LJ (2003). Fish consumption, fish oil, omega-3 fatty acids, and cardiovascular disease. Arterioscler. Thromb. Vasc. Biol., 23: 20-31

Mahdy AA, Tork IY, Aref MA (1993). Heavy metal pollution in drinking water and methods of removals. Alex. J. Vet. Sci.9 (1): 65-69.

Marouf HA, Dawoud AS (2006). Evaluation of heavy metals content in freshwater cray fish in Damietta. J. Egypt. Vet. Med. Assoc., 66, (3): 217-225.

Mriagu JO (1988). A silent epidemic of environmental metal poisoning? Environ. Pollut., 50: 161-189.

Olaifa FG, Olaifa AK, Onwude TE (2004). Lethal and sublethal effects of copper to the African Cat fish (*Clarias gariepnus*). Afr. J. Biomed. Res., 7: 65-70.

Saleh EA (2004). Monitoring of some heavy metals residues in some fish. Issn 110-2047, Alex. J. Vet. Sci., 21(1).

Seddek ASH, Salem DA, El-Sawi NM, Zaky ZM (1996). Cadmium, lead, nickel, copper, manganese and fluorine levels in River Nile fish. Assiut Vet. Med. J., 34: 95.

Shokrzadeh M, Ebadi AG, Heidari R, Zaree S (2004). Measurement of lead, cadmium and chromium in five species of most consumed fish in caspian sea, Iran. Int. J. Biol. Biotechnol., 1(4): 673-675.

Suppin D, Zahlbruckner R, Krapfenbauer CC, Hassan HC, Smulders FGM (2005). Mercury, lead and cadmium content of fresh and canned fish collected from Austrian. Emahrung, 29(11): 456-460

Tariq J, Ashraf M, Jaffar M (1994). Assessment of pollution of rivers Jehlum and sutlej pakistan through trace metals in fish sediment and water. Toxicol. Environ. Chem., 43: 169.

Turkmen M, Turkmen A,Tepe Y (2008). Metal contamination in five fish species from black, Marmara, Aegean and Mediterranean seas, Turkey. J. Chil. Chem. Soc., 53 (1): 1-8

Vinodehini N (2008). Bioaccumlation of heavy metals in organs of fresh water fish *Cyprinus carpio* (common carp). Int. J. Environ. Sci. Tech., 5(2): 179-182.

Wafaa E, Saleh G, Dief M (2003). Some studies on heavy metal

pollution in water and fishes. Egypt. J. Basic Appl. Physiol., 2 (2):185-200.
World Health Organization (1992): Environmental health criteria. No.134. Cadmium. WHO. Geneva.
www.freepatentsonline.com/5958248.htm.
Zienab I, Soliman M, F Galab (2001): A study on lead pollution in water and tilapia fish in Manzala Lake. J. Egypt Vet. Med. Assoc., 61(6C): 71-76.