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DETERMINATION OF LEAD AND CADMIUM LEVELS IN SOME OF FRESH WATER FISH MARKETED IN DAMANHOUR CITY

(With 4 Tables)

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تقدير مستويات الرصاص والكادميوم فى بعض أسماك المياه العذبة المسوقة بمدينة دمنهور

إبراهيم على القويعى ، فايزة عبد العزيز التداوى ، هناء رشاد الحوفى

أجريت هذة الدراسة لتقدير تركيزات الرصاص والكادميوم فى ستين عينة من أسماك المياه العذبة (١٥عينة لكل من سمك البلطى والبورى والقرموط والمبروك) المعروضة فى أسواق السمك بمدينة دمنهور باستخدام جهاز المطياف الذرى. سجلت متوسطات تركيزات عنصر الرصاص فى أسماك البلطى والبورى والقرموط والمبروك كالآتى:٠.٣٦ ±٠.٠٩ ، ٠.٧٢ ± ٠.٥٥ ، ١.٣١٥ ±٠.٩٧٥ و ٠.٥٦٨ ± ٠.٠٦٤ جزء من المليون (وزن رطب) بمعدلات ١٣.٣ ٪ فى كل من أسماك البلطى والبورى والقرموط بينما كان معدله فى العينات المفحوصة من سمك المبروك ٣٣.٣ ٪ على التوالى. وكانت نظيراتها لعنصر الكادميوم كالآتى: ٠.٠٠٥±٠.٠٢ ، ٠.٠٠٦±٠.٠١٩ ، ٠.٠١٦ ± ٠.٠٠٣ و ٠.٠١٤± ٠.٠٠٣ جزء من المليون (وزن رطب) بمعدلات ٤٦.٧٪، ٢٠٪، ٦٦.٧٪، ٤٠٪ على التوالي. هذا وقد أوضحت الدراسة أن نسبة العينات المفحوصة والتى لم تتجاوز الحد المسموح به لعنصر الرصاص (٠.١ جزء من المليون) كانت ٨٦.٧٪ تبعا للمواصفة القياسية المصرية لسنة ١٩٩٣ لكل من أسماك البلطى والبورى والقرموط بينما كانت ٦٦.٧٪ لعينات أسماك المبروك. هذا وقد بينت الدراسة أيضا أن جميع العينات المفحوصة لأستبيان تركيزات عنصر الكادميوم لم تتجاوز الحد المسموح به (٠.٠٥ جزء من المليون). وقد تم مناقشة المخاطر الصحية أثر تناول الأغذية التى تحتوى على معدلات عالية من المعادن الثقيلة تتجاوز الحدود المسموح بها محليا ودوليا. وقد أوصت الدراسة باتخاذ التوصيات اللازمة من قبل السلطات الصحية بشأن مستهلكي الأسماك وكذا الإجراءات الحكومية للحفاظ على البيئة.

SUMMARY

Sixty random samples of some fresh water fish species: Tilapia nilotica (Bolti), Mugil cephalus (Bori), Clarias lazera (Karmout) and Cyprinus carpiol (Carp) (15 of each) were collected from fish markets of

Damanhour city for detection of lead and cadmium levels in their muscular tissues using Perkin-Elmer atomic absorption spectrophotometer. The reported results showed that the mean values of lead in the examined samples of the fore-mentioned fish species were 0.360±0.090, 0.720±0.550, 1.315±0.975 and 0.568±0.064 ppm, wet weight with an incidence of 13.3% to each of Bolti, Bori and Karmout, 33.3% for Carp. While that of the cadmium were 0.020±0.005, 0.019±0.006, 0.016±0.003 and 0.014±0.003 ppm wet weight with incidences of 46.7% 20.0%, 66.7% and 40.0%, respectively. The percentages of the examined samples of Bolti, Bori, Karmout and Carp for lead levels which not exceed the permissible limit (0.1 ppm) according to the Egyptian Organization for Standarization and Quality Control "EOSQC" (1993) were 86.7% for each of Bolti, Bori and Karmout, 66.7% for Carp and the values of examined samples for cadmium level not exceed the permissible limit (0.1 ppm) according to "EOSQC" (1993). The puplic health significance and the hazardous toxic effects of these heavy metals were discussed. In addition, recommendations regarding the consumption of fish from the health authorities should be taken for protection of fish consumers. The governmental role is to improve the enforcement of the safety, health and environmental laws.

Key words: *Heavy metals, lead, cadmium, fish, fresh water fish.*

INTRODUCTION

Fish is a valuable source of high quality protein, polyunsaturated fatty acids, minerals and vitamins constituting the major part of the human diet and it is not surprising that numerous studies have been carried out on metals accumulation in different fish species (Gundacker, 2000; Alam *et al.,* 2002). Water pollution is a critical environmental problem facing public health authorities.Industrial wastes, geochemical structure and mining of metals create a potential source of heavy metals pollution in the aquatic environment (Lee and Stuebing, 1990). Metals such as iron, copper, zinc and manganese are essential as they play an important role in biological systems where lead and cadmium are non-essential metals as they are toxic,even in trace amounts.

Heavy metals are recognized as cumulative toxic substances due to slow elimination rates from the body. Most of these pollutants are toxic and cause serious health hazard to human bing depending on their

levels of contamination (Farag *et al.,* 2000). Lead and Cadmium are considered as toxic elements due to their competition with the essential metals for binding sites and also their interference with sulfhydryl groups and structural proteins (Ahmed *et al.,* 1993), and the signs of the toxicity depend on the chemical form of the element, the dosage, the route, the frequency and the duration of administration (Underwood, 1977). Lead is linked to encephalopathy in adults and children. The outcome is frequently fatal in children and those who survive often present irreversible neurological and neuropsychological sequelae (ATSDR, 1999) and also long-term exposure to cadmium leads to several morphological changes in the kidneys (Satarug and Moore 2004). In addition, during woman pregnancy led to reduced birth weight and /or premature birth (Henson and Chedrese, 2004).

Heavy metals have a higher tendency to be incorporated into food chains and become accumulated in tissues and organs of fish and other aquatic organisms in concentrations higher than those in water, and this represents serious health hazards to consumers (Draz *et al.,* 1993).

The present study was conducted to determine the levels of lead and cadmium pollution of commen fresh water fish species marketed in Damanhour city.

MATERIALS and METHODS

A. Collection of samples:

Sixty random fresh-water fish samples of different species (15 each of Tilapia nilotica, Mugil cephalus, Clarias lazera and Cyprinus carpiol) were collected randomly at the consumer level from different fish markets of Damanhour city in El-Boheira governorate during summer of 2010 and each sample was wrapped in a light polyethylene bag placed in ice and taken to the laboratory without delay for analysis of lead (Pb) and cadmium (Cd) levels.

B. Preparation of collected samples:

The collected fish species were washed with tape water several times to remove slime and mud and then with de-ionized water. The samples of each specie taken from the muscles below the dorsal fin and at the middle line were homogenized and kept at fresh polyethylene bags at -20° C for analysis. A measured weight (1gm) was transferred into clean and acid washed scrow-capped digestion tubes. All digestion tubes were identified for examination.

C. Digestion procedure:

Each prepared fish sample was digested according to Tsoumbaris and Papadopoulou (1994). 10ml of mixture of concentratednitric and perchloric acids (1vol.:1vol.) were added to each sample. The tubes were closed and the contents were vigorously shaken and allowed to stand over-night to be cold digested followed by mild increase in temperature till heating to 100° C in a water bath for 3-4 hours to ensure complete digestion of all samples.4-5 drops of hydrogen peroxide 30 % were added to each sample and continue heating process till the brown nitrous gases were expelled and the mixtures become clear. After cooling, each digest was diluted to 25ml with deionized water and filterated through Whatman filter paper No.42. The clear filtrate of each sample was kept in refrigerator to avoid evaporation. All samples and blank solutions were analysed by using Flame Atomic Absorption Spectrophotometry "AAS" (Perkin-Elmer Atomic Absorption Spectrophotometry model 2380, USA) for determination of lead and cadmium concentrations.

Parameters for instrumental instructions of Atomic Absorption Spectrophotometry

Metal	Lamp wave Length (nm)	Slit width (nm)	Lamp Current (ma)	Fuel flow Rate(1/min.)	Burner height (cm)	Detection limit
Lead (Pb)	217	0.7	12	30	8	0.02 ppm
Cadmium (Cd)	228.8	0.7	5	30	8	0.0006 ppm

D.Quantitive determination of heavy metals in the examined samples:
The concentrations of Pb and Cd in the examined samples were calculated according to the following equation: C=R x D/W
Where: C =Concentration of heavy metals (mg/kg) wet weight.
R=Reading of element concentration on digital scale of AAS.
D=Final volume of prepared sample in mls.
W=Weight of the wet sample.

The reading results of the absorbance values of Pb and Cd concentrations were recorded.

RESULTS

Table 1: Lead concentrations (mg/kg,wet weight) in the examined fish samples (n=15 of each).

Fish specie	Examined samples		Minimum	Maximum	Mean±SEM
	NO of +ve	%			
Tilapia nilotica	2	13.3	0.27	0.45	0.36±0.09
Mugil cephalus	2	13.3	0.17	1.27	0.72±0.55
Clarias lazera	2	13.3	0.34	2.29	1.315±0.975
Cyprinus carpiol	5	33.3	0.41	0.79	0.568±0.064

Table 2: Cadmium concentrations (mg/kg,wet weight) in the examined fish samples(n=15 of each)

Fish specie	Examined samples		Minimum	Maximum	Mean±SEM
	NO of +ve	%			
Tilapia nilotica	7	46.7	0.005	0.041	0.020±0.005
Mugil cephalus	3	20.0	0.008	0.029	0.019±0.006
Clarias lazera	10	66.7	0.002	0.034	0.016±0.003
Cyprinus carpiol	6	40.0	0.001	0.026	0.014±0.003

- SEM = standard error of the mean. -Negative samples were under detectable limit of AAS.
- mg/kg = ppm=µg/g. - +ve: samples within detectable limit of AAS.

Table 3: Frequency distribution of lead and cadmium levels in the examined samples according to the permissible limits of *EOSQC (1993)*

Heavy Metal	Fish Samples	Permissible Limit *	More than the Permissible limit		Less than the Permissible limit	
			NO.	%	NO.	%
Lead	Tilapia nilotica	0.1 mg/kg	2	13.3	13	86.7
	Mugil cephalus		2	13.3	13	86.7
	Clarias lazera		2	13.3	13	86.7
	Cyprinus carpiol		5	33.3	10	66.7
Cadmium	Tilapia nilotica	0.1mg/kg	0	0	15	100.0
	Mugil cephalus		0	0	15	100.0
	Clarias lazera		0	0	15	100.0
	Cyprinus carpiol		0	0	15	100.0

*The permissible limit according to Egyptian Organization for Standardization and Quality Control "*EOSQC , 1993*"

Table 4: Estimated daily and weekly intakes of adult person consuming fresh water fishes and the comparison with the recommended National and International permissible tolerable limits in foods.

Metal	Concentration level in examined samples (µg/g)		EWI µg/adult person [a] (EWI µg/kg, b. w.)	Provisional Permissible Tolerable Weekly Intakes (PTWI) (µg/kg ,b.w.)[b]	Permissible Tolerable daily Intakes(PTDI) "µg/day/60kg,b.w." (PTDI,µg/kg , b.w.) [b]	Human Weekly intake of Heavy metals "EOSQC" (1993)
	Examined samples	Mean (µg / g)				
Pb	Bolti	0.360	122.4 (2.04)	25	214.3 (3.57)	50 µg/kg Body weight
	Bori	0.720	244.8 (4.08)			
	Karmout	1.315	447.1 (7.45)			
	Carp	0.568	193.12 (3.22)			
Cd	Bolti	0.020	6.8 (0.113)	7	60 (1.0)	6.7-8.3 µg/kg Body weight
	Bori	0.019	6.46 (0.11)			
	Karmout	0.016	5.44 (0.09)			
	Carp	0.014	4.76 (0.08)			

[a]EWI:estimated weekly intakes from consumption of 12 ounces (approximately 340 grams) of fish every week by adult person of 60 kg body weight according to *US EPA(2004).*

- Values in parenthesis calculated as weekly intake to every kg body weight.
- µg:microgram =0.001 milligram (mg). b.w.:body weight

[b]Joint FAO/WHO Expert Committee on Food Additives(2004) .

DISCUSSION

Several reports describe metals residues in fish from both aquatic and marine species (Yess, 1993; Abou-Arab *et al.,* 1996; Atta *et al.,* 1997) but the concentrations in their tissues vary considerably among different studies possibly due to differences in metal concentrations and chemical characteristics of water from which fish were caught, ecological needs, metabolism and feeding patterns of fish.

Lead (Pb):

Lead causes haematological, gastrointestinal and neurological dysfunctions. Prolonged exposure to Pb may cause also chronic nephropathy, hypertension and reproductive impairment. In addition, Pb inhibits enzymes, alters cellular calcium metabolism and slows nerve conduction (Elinder, 1985). Children under 6 years are especially susceptible to the adverse effects of Pb for several reasons. These include their behavioral characteristics (outdoor activity, less concern for hygienic conditions, hand-to-mouth activities), their higher disposition for lead absorption and the prevalence of nutritional deficiencies (e.g. iron and vitamin D) among children that enhance absorption of Pb from the gastrointestinal tract. As the blood–brain barrier is not yet fully developed in young children, hematological and neurological adverse effects of Pb occur at lower threshold levels than in adults (Air Quality Guidelines for Europe, 2000).

The results obtained in Table 1, showed that the mean values of lead in examined samples of T.nilotica (Bolti), M.cephalus (Bori), Clarias lazera (Karmout) and Cyprinus carpiol (Carp) were 0.360 ± 0.090, 0.720 ± 0.550, 1.315 ± 0.975 and 0.568 ± 0.064 mg/kg,wet weight,with incidence of 13.3% for each of Bolti, Bori and Karmout but was 33.3% for Carp.

These results were agreed with that of El-Atabany (1995) who recorded that the levels of Pb levels in muscles tissue of T.nilotica and M.cephalus were 0.35 ± 0.02 and 0.38 ± 0.01 mg/kg,wet weight. El-Kelish-Hoda (1995) reported that means of Pb levels in muscles tissues of T. nilotica and Carp fish caught freshly from Abbassa fish farm as 0.31±0.04 and 0.33±0.05ug/g,wet weight, also Abd El-Aziz (1996) recorded that T.nilotica in Gharbia governorate contained 0.628ppm Pb. Labib *et al.* (2008) found that the Pb concentrations were ranged from 0.12 to 0.53ppm, wet weight in T.nilotica and C.lazera collected from five districts of Qena governorate, Upper Egypt.

Nearly the same results were registerated by Abd El-Kader *et al.* (1993) where lead concentrations in M.cephalus were ranged from 0.29 to 0.42 ug/g wet weight and attributed these levels to possible pollution through agricultural and industrial re-sources at Assuit governorate.

Higher results were recorded by Abou-Donia (1990) (1.662 ppm) and Abd El-Fatah and Salem (2007) who reported that Pb levels in T.nilotica and M.cephalus collected from Giza markets were 1.015± 0.500 and 1.80 ± 0.61ppm and in samples collected from Cairo markets were 1.10±0.84 and 1.90 ± 1.01ppm, respectively.

The percentages of examined fish samples which was less than the lead permissible limit (0.1 mg/kg) according to Egyptian Organisation for Standarization and Quality Control "EOSQC" (1993) were 86.7% for each of Bolti, Bori and Karmout, and 66.7% for Carp (Table, 3). Since the United States Environmental Protection Agency (EPA, 1999) has never established a reference dose (RfD), a definitive level cannot be asserted for Pb in fish. The advisory limit would likely be 1 ppm or greater. Hodson *et al.* (1984) indicated that the Canadian Pb limit of 10 ppm was discontinued, but that the british limit remains at 2 ppm in fish, fish paste and canned fish (5 ppm in fried and salted fish). Abou-Arab *et al.* (1996) indicated that the FAO limit (1983) was 2.0 ppm.

The calculated weekly intake of lead (Pb) estimated by µg/kg,b.w. for adult person of 60 kg body weight in this study from eating 12 ounces (approximately 340g) of each Bolti, Bori, Karmout and Carp fish according to US EPA (2004) were 2.04, 4.08, 7.45 and 3.22, respectively (Table, 4). These values were lower than the maximum provisional tolerable weekly intake (PTWI) of lead for human stipulated by "E.O.S.Q.C. (1993) which is 50µg /kg body weight. In 1993, the Joint FAO/WHO Expert Committee for Additives and Contaminants (JECFA) reduced the value it had provisionally specified for adults in 1972, for tolerable lead consumption per week (PTWI) from 0.05mg/kg B.W. to 0.025mg (25 µg). The reasons for reduction may be due to, firstly, that research has revealed further harmful potentials in lead and, secondly, that lead contamination has decreased throughout the world.

Cadmium (Cd):

The major route of exposure to Cd for non-smoking general population is via food, the contribution from other pathways to total uptake is small (Goyer, 1991). The International Agency for Research on Cancer (IARC) classifies Cd as Class1 "The agent (mixture) is carcinogenic to humans" (Stanley, 2004). Cadmium, like lead, is a

cumulative poison, i.e. the danger lies primarily in the regular consumption of foodstuffs with low contamination. However, in contrast to lead, the definition of an exact toxicity limit is not possible for cadmium. Although the absorption of Cd is low from gastrointestinal tract, it has a long biological half-life due to the cumulative effect.

The result recorded in Table 2 revealed that the means of cadmium concentrations in examined samples of Bolti, Bori, Karmout and Carp fish were 0.020± 0.005,0.019± 0.006,0.016± 0.003 and 0.014±0.003 mg/kg wet weight, with incidences of 46.7%, 20.0%, 66.7% and 40.0%, respectively.

This result agree with El-Nabawi *et al.* (1987) who reported that the cadmium level in the examined muscles tissues of T.nilotica hunted from Abo-Qir bay,Edku,Maruit lakes was ranged from 0.018 to 0.023 ug/g, El-Atabany (1995) which recorded that the means of Cd levels in muscles samples of T.nilotica and M.cephalus fishered from Al-Manzala lake were 0.09±0.01 and 0.07±0.01mg/kg wet weight, respectively. Nearly similar results of Cd levels in muscles of T.nilotica and Carp caught freshly from Abbassa fish farm were reported by El-kelish-Hoda (1995).

While higher figures were obtained by *Abd El-Fatah and Salem (2007*) who revealed that the mean values of Cd in T.nilotica and Mugil cephalus collected from Giza and Cairo were 0.85± 0.11,1.09 ± 0.416 and 2.0 ± 0.92, 2.51 ± 1.51 ppm, respectively. Also, Labib *et al.* (2008) found that the Cd concentration was ranged from0.32 to 0.42 ppm wet weight of examined samples of T.nilotica and C.lazera.

Table 3 declared that the frequency distribution of cadmium in examined samples of each fish species not exceed the permissible limit (0.1ppm)according to "EOSQC" (1993).The levels of Cd were below the Risk-Based Concentration (RBC)of 1.4 ppm according to United states Environmental Protection Agency (EPA, 1999). However, Gutenmann *et al.* (1988) indicated that a frequently used food safety limit for Cd in food is 2.0 ppm, while by Food and Agriculture Organization (FAO) at (1983) was 0.5 ppm. Moreover, the estimated weekly intakes (µg/kg,b.w.) of Cd from consumption of 340 g of each Bolti, Bori, Karmout and Carp fish were lower than the maximum provisional tolerable weekly intake (PTWI) of Cd stipulated by "E.O.S.Q.C. (1993) and the Joint FAO/WHO Expert Committee for Additives and Contaminants "JECFA" (2004) (Table 4).

Finally,the heavy metals in fresh water fish were attributed to channel water pollution and to the cumulative effects of such metals. It

was very difficult to compare the metals concentrations even within the similar tissues of two different species because of different feeding habits, the difference in the aquatic environments concerning the source and level of water pollution (Papagiannis *et al.,* 2004), growing rates of the species, types of tissues analysed. Using the FAO/WHO (Anonymous, 2003) values of the provisional tolerable weekly intakes of Pb and Cd (25 and 7 ug/g) for a 60 kg adult person were estimated to be 1500 and 420 µg/ person/ week, respectively. The mean concentration of Pb from consumption of Bolti, Bori, Karmout and Carp fish in this study were 0.36, 0.72, 1.315 and 0.568 mg/kg, respectively, and therefore, an adult man of 60 kg body weight could safely consume 4.16, 2.08, 1.14 or 2.64 kg fish meat weekly.

It could be concluded that the incidences and levels of Pb and Cd were almost low and so that the daily and weekly intakes were consequently were not exceeding the national and international permissible limits of human food. Advisory actions should be warned the at-risk populations like children, pregnant and/or lactating women to avoid over consumption of fish. More governmental efforts still be needed and a powerful law incriminate the environmental pollution should be issued.

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