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BIOACCUMULATION OF SOME HEAVY METALS IN WATER AND FISH SPECIES (*Oreochromis Niloticus* and *Mugil Cephalus*) FROM SHARKIA AND KAFER EL-SHEIKH FISH FARMS AND THEIR EFFECTS ON SOME BIOCHEMICAL MEASUREMENTS

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Abstract

In the present study, the heavy metals iron, copper, zinc, lead, and cadmium (Fe, Cu, Zn, Pb, and Cd) were determined in water and some tissues of *Oreochromis niloticus* and *Mugil cephalus* in three private fish farms, two at Sahel-El-Heseinia and San El-Hager (Sharkia governorate) and one at Kafer El-Sheikh governorate.

In addition to the study of the effects of bioaccumulation of these metals on some biochemical parameters, the distribution of heavy metals in water samples analyzed were in the order of magnitude as by Fe > Zn > Cu > Cd > Pb. Iron exceed the permissible limits according to WHO (1989). The selected organs, (liver, gills and muscles) accumulated different quantities of the metals. The distribution of that metals in selected organs analyzed were in the order of magnitude as liver > gills > muscles. The lowest concentration levels of cadmium were observed in all tissue of *Oreochromis niloticus* and *Mugil cephalus*, while, lead didn't detect in all fish organs at all farms. No significant difference between bioaccumulation factor (BAF) of all heavy metals in organs of *Oreochromis niloticus* and *Mugil cephalus*. Different levels of heavy metals accumulated by the two fish species but iron & zinc were exceed the maximum permissible limits values of heavy metals for fish tissues as prescribed by some national and international agencies(examples). But WHO (2011) reported that there are no health based guideline-value for both Fe and Zn. Glucose, Total protein, and activities of AST and ALT had significantly different values in the serum between the studied fish.

INTRODUCTION

In recent years, the problems of sewage pollution of inland waters have become a point of local concern (Abdel-Moneim *et al*., 2012). The disposal of the untreated sewage may be harmful concerning its possible hygienic and aesthetical effects and its impact on fauna and flora in the aquatic environment (Bahnasawy *et al.,* 2009). (El-Sheikh *et al*., 2010) mentioned that Egyptian drains receive large quantities of partially treated or untreated domestic and industrial wastewater and other human activities, which in turn ultimately discharge into River Nile, canals, lakes, or seas.

Fish accumulate toxic chemicals such as heavy metals directly from water and diet, and contaminant residues may ultimately reach concentrations hundreds or thousands of times above those measured in the water, sediment and food (Goodwin *et al.,* 2003 & Osman *et al.*, 2007).

Fish is a good bio-indicator because it has a potential to accumulate heavy metals and other organic pollutants (Ahmed and Shubami-Othman, 2010). Besides that, it has high economical value, thus fish are suitable as water quality indicator organism. When Fish is exposed to high concentrations of heavy metals in water it may take up substantial quantities of these metals. Heavy metals can enter from contaminated water and can accumulate into the fish's body by different routes. These metals concentrated at different contents in organs of fish body. Fish accumulates heavy metals in the tissue through absorption and humans can be exposed to heavy metals via food chain. This can cause acute and chronic effects in humans (Dogan and Yilmaz, 2007).

Sultana and Rao (1998) recorded that bioaccumulation of metals reflects the amount of heavy metals ingested by the organism, the way in which the metals are distributed among the different tissues and extent to which the metal is retained in each tissue type.

The study on heavy metal contamination in water and fish is vital to assess the current status of water pollution with heavy metal and threats to human health from heavy metal pollution.

Biochemical characteristics of blood are among the important indices of the status of internal environment of the fish organism (Edsall, 1999).

Biochemical profile in fish has proved to be a sensitive index for evaluation of the fish metabolism under metallic stress. Studies proved that, fish subjected to metals showed reduced levels of proteins, lipids, ALT and AST activities in the fish muscles (Almeida *et al*., 2001). Also, Loskova *et al*., 2002 mentioned that changes in the biochemical blood profile mirror changes in metabolism and biochemical processes of the organism, resulting from the effect of pollutants, and they make it possible to study the mechanisms of the effects of these substances.

The purpose of this study was to measure the current concentrations of the metals (Fe, Cu, Zn, Pb, and Cd) in water and two type of fish (*Oreochromis Niloticus* and *Mugil cephalus*) to determine the edible tissues were within the safe limits for human health or not, in three different fish farms and also their effects on some biochemical measurement.

MATERIALS AND METHODS

Area of study:

The current work was done in three farms. The 1st and 2nd were Sahel El-Husseinya and San El-Hager private fish farms located in Sharkia governorate. Three earthen fish ponds at El- Hussienia farm supplied with waste water driven from Baher El- Baqer drain. San El-Hager farm (Three earthen ponds supplied with mix of water driven from both El-Salam canal (mix of Nile and agric-draining water) and Baher El-Baqer canal. While the 3rd fish farm was a private fish farm located in kafer El-Sheikh governorate supplied with mixed water (industrial, agriculture, waste water) driven from El-Gharbia main drain (ketshner).

Collection and preparing of the samples:

Water and fish samples were collected monthly from the three selected fish farms from August to November, 2014.

Water sampling:

Three replicates surface water samples were collected from each farm using 1L polyethylene sampling bottle during the sampling periods. Bottled samples were taken to laboratory using an ice box at 4^0C. Then, the three replicated samples for each site were composited for all analytical procedures conduct in the laboratory of CLAR after filtration in pre-cleaned one liter bottle acidified by adding 5 ml of concentrated nitric acid and kept for analysis. Each sample of water was put in screw-capped tube till complete dryness. Ten ml of concentrated nitric acid was added to the sample to dryness then diluted to 20 ml with deionized water. The solution was filtrated, transferred to 250 ml volumetric flask, marked and stored refrigerated till analysis for iron, copper, zinc, lead, and cadmium (Fe, Cu, Zn, Pb & Cd).

Fish sampling:

At the same periods and the same sites; three *O. niloticus* and other three *Mugil cephalus* were collected. Blood samples were immediately taken from the caudal vein of the fish using a heparinized syringe and collected into small sterilized plastic tubes. Blood samples were left to coagulate for15-20 min. at room temperature and then centrifuged at 3000 rpm for 10 min. to separate serum which were stored in polyethylene Eppendorf test tubes at -20°C until analysis.

After that, fish samples were carefully dissected to remove the liver, gills and parts of the muscle. Then livers, gills and pieces of edible muscle tissues were oven dried at 85^oC until constant weight (about 18 hours). A 1.0 g dry weight of tissue was ashed in muffle furnace (Thermolyne Corporation, Dubuque, Iowa, USA) for 6 hours. Ash was digested with conc. nitric acids, and diluted with 2N HCL to a constant volume. The samples were filtered into acid-washed volumetric flasks and diluted to 50 mL for elemental analysis. Concentrations of iron, copper, zinc, lead and cadmium were then determined according to APHA (1998) using atomic absorption spectrophotometer (Thermo 6600 Thermo Electron Corporation, Cambridge, UK).

The results of Fe, Cu, Zn, Pb, and Cd were calculated in milligram per kilogram dry weight (mg/kg dry wt).

Biochemical Analyses:

Serum samples were used to estimate the biochemical parameters. The glucose was estimated according to the method of Trinder (1969), while total protein was determined according to young (2001). Aspartate amino transferase (AST) and Alanine amino transferase (ALT) were determined according to the method of Reitman and Frankel (1957).

Statistical analysis:

One way ANOVA and Duncan multiple range test were used to test whether differences among sites and time were significant at $P \leq 0.05$. Correlation coefficients; Pearson correlation (r), between the different parameters were computed. Correlation and all statistical analyses were made using SPSS for windows version 10 (SPSS, Richmond) as described by Dytham (1999).

Bioaccumulation factor (BAF):

The bioaccumulation factor (BAF) is the ratio between the accumulated concentration of a given pollutant in any organ and its dissolved concentration in water according to Authman and Abbas, 2007 using the following equation:

BAF = pollutant concentration in fish tissue (mg/kg)/pollutant (mg/l) in water or sediment.

RESULTS AND DISCUSSION

The results of heavy metals concentration in water samples in the selected farms are given in Table (1).

Iron, Cu and Zinc showed the highest accumulated metals in water of farms II & III compared to farm I due to agricultural drainage waters which containing high concentrations of different pesticides, fertilizers runoff and heavy metals (Authman *et al.*, 2008).

The contamination of water resources and biota by heavy metals of major concern because of their toxicity, persistence and bio accumulative nature (Ikem *et al*., 2003).

Table 1. Mean concentrations (mg/l) of heavy metals in water of different farms from August to November, 2014.

Location Metals	Farm I	Farm II	Farm III
	Sahel El-Husseinya	**San El-Hager**	**Kafer El-Sheikh**
Fe	2.55±0.02[c]	4.04±0.3[b]	7.24±1.15[a]
Cu	0.0٧7±0.0١٧[a]	0.11±0.01[a]	0.13±0.043[a]
Zn	0.48±0.09[b]	1.5±0.33[a]	1.113±0.38[a]
Pb	BDL	BDL	BDL
Cd	0.0039±0.0002[a]	0.0016±0.0٠0٦[b]	0.0044±0.0003[a]

*Means with the same letter within the same row are not significantly different; BDL, below detection limit

It is clear that the distribution of heavy metals in farm III (kafer El-Sheikh) followed by farm II (San El-Hager, Sharkia) had higher contamination when compared to farm I (Sahel El-Husseinya).

According to WHO (1989) and USEPA (1986) standards, the obtained results showed that Fe concentrations were 4.04±0.03 and 7.24±1.15 in water exceed the maximum allowable limits (0.3 m/l) in farm II and III respectively , while Cu, Zn, Cd and Pb not exceed the allowable limits (1, 5, 0.01, 0.05 m/l respectively).

The concentrations of iron in liver, gills and muscles of both *O. niloticus* and *Mugil cephalus* from the selected farms are given in Table (2). The highest value (mg/kg/) of Fe was recorded in liver of *O. niloticus* in farm III (1714 ± 220.2 mg/kg dry wt), while The lowest value recorded in farm I of *O. niloticus (*468.03 ± 44.91 mg/kg) and in *Mugil cephalus the highest value* (854.7±27.18 mg/kg dry wt) were recorded in farm II while the lowest value (486.6.6± 147.6) was recorded in farm I.

In the gills the highest value of Fe was recorded for *O. niloticus* and for *Mugil cephalus* in farm II which were 610.8±99.04 and 485±75.88 mg/kg/wt, respectively. On the other hand, the lowest values were recorded for *O. niloticus* and *Mugil cephalus* (369.4±38.26 & 209.3±20.6 mg/kg dry wt) in farm I, respectively.

Table 2. Mean concentrations of iron (mg/kg) in different organs of *O. niloticus* and *Mugil cephalus* at various sampling sites from August to November, 2014.

Fish species	Organs	Farm I	Farm II	Farm III
		Sahel El-Husseinya	**San El-Hager**	**Kafer El-Sheikh**
Oreochromis niloticus	**Muscle**	1٠5.١±٣0.5٥ ^{b}	404.7±63.5 ^{a}	256±34.37 ^{b}
	BAF	41.21	100.17	35.36
	Gills	369.4±38.26 ^{b}	610.8±99.04 ^{a}	435.6±41.44 ^{b}
	BAF	144.86	150.99	60.17
	Liver	468.03±44.91 ^{b}	675.5±120.7 ^{b}	1714±220.2 ^{a}
	BAF	183.54	167.20	236.74
Mugil cephalus	**Muscle**	132.2±13.83 ^{b}	411.1±47.73 ^{a}	216.9±12.7 ^{b}
	BAF	53.18	101.76	29.96
	Gills	209.3±20.6 ^{b}	485±75.88 ^{a}	390±59.6 ^{ab}
	BAF	82.08	120.05	52.56
	Liver	486.6±147.6 ^{b}	854.7±27.18 ^{a}	807.6±27 ^{a}
	BAF	190.82	211.56	62.55

*Means with the same letter within the same row are not significantly different; BDL, below detection limit BAF means bioaccumulation factor

In the muscle, the highest values of iron were recorded in farm II for both *O. niloticus* and *Mugil cephalus* which was 404.7±63.5 and 411.1±47.73 mg/kg, respectively. In addition, the lowest values were recorded in farm I which were 1٠5.١±٣0.5٥ and 132.2±13.83 mg/kg for *O. niloticus* and *Mugil cephalus,* respectively. On the other hand, the maximum BAF for Fe in liver of *O. niloticus* and *Mugil cephalus* were 236.74 in farm III and 211.56 in farm II, respectively. While the minimum BAF for *O. niloticus* and *Mugil cephalus* were 167.2 and 62.55 in farm II and III, respectively. In gills, the maximum BAF for Fe of *O. niloticus* and *Mugil cephalus* were 150.99 and 120.05, respectively in farm II. The minimum BAF for *O. niloticus* and *Mugil cephalus* were 60.17and 52.56 in farm III. Whilst in muscles, the maximum BAF for Fe of *O. niloticus* and *Mugil cephalus* were 100.17 and 101.76 in farm II. The minimum BAF for *O. niloticus* and *Mugil cephalus* were 35.36 and 29.96 in farm III. The BAF of iron showed no differences between *O. niloticus* and *Mugil cephalus*.

Fe is necessary element in human diet. It plays significant role in metabolic process (Ambedkar and Muniyan, 2012). The observed values of iron levels were exceeded the WHO (1989) and USEPA (1986) recommended standards limits of 30 mg/kg in fish muscle.

The concentrations of copper in liver, gills and muscle of both *O. niloticus* and *Mugil cephalus* from the selected farms are given in Table (3). The concentration were not exceeded the permissible levels according to WHO (1989) and USEPA (1986) standards limits of copper 20 mg/kg in studied fish in all farms, for *O. niloticus* and *mugil cephalus*. Generally the highest values were recorded in liver of *O. niloticus* and *mugil cephalus* followed by gills and the minimum value was recorded in muscles. The highest BAF of Cu was recorded for *O. niloticus* and *mugil cephalus* was 256.92 and 229.09 in fish farms III & II in liver but lowest BAF were recorded in muscles at farm III which was 22.38 &16.77 in *O. niloticus* and *mugil cephalus*. Copper is an essential element that serves as a cofactor in a number of enzymes systems and necessary for the synthesis of hemoglobin (Sivaperumal *et al.*, 2007) but very high intake of Cu can cause adverse health effect problems for most living organism. The highest levels of copper in the different tissues of selected fish species may be due to the presence of domestic waste, Agricultural and industrial waste in the source of water.

Table 3. Mean concentrations of copper (mg/kg) in different organs of *O. niloticus* and *Mugil cephalus* at various sampling sites from August to November, 2014.

Fish species	Organs	Farm I	Farm II	Farm III
		Sahel El-Husseinya	**San El-Hager**	**Kafer El-Sheikh**
Oreochromis niloticus	**Muscle**	3.33±0.36[a]	3.45±0.49[a]	2.91±0.33[a]
	BAF	46.57	31.36	22.38
	Gills	6.07±0.33[a]	5.16±2.77[a]	6.62±0.42[a]
	BAF	86.71	46.91	50.92
	Liver	7.7±1.01[b]	7.09±1.08[b]	33. 4±3.60[a]
	BAF	110	64.45	256.92
Mugil cephalus	**Muscle**	2.61±0.37[a]	2.09±٠.5٤[a]	2.81±0.11[a]
	BAF	37.29	19	16.77
	Gills	6.6±0.68[a]	5.87±0.94[a]	4.74±0.71[a]
	BAF	94.27	53.36	36.46
	Liver	7.99±0.41[b]	25.2±6.79[a]	15.93±2.72[b]
	BAF	114.14	229.09	122.54

*Means with the same letter within the same row are not significantly different; BDL, below detection limit BAF means bioaccumulation factor.

Data in Table (4) show the concentrations of zinc in different organs of *O. niloticus* and *Mugil cephalus* in the selected farms. Zn is the essential mineral for both animals and humans. Zinc was the second abundant metal followed by iron. Like copper zinc is also an essential element in our food. It also showed a protective effect against the Cd and Pb toxicity. Zn is necessary element for embryo development and is important to reproductive organs (Carpene *et al.*, 1994). The highest concentrations of Zn in liver were 573.9±83.3 & 253.77±27.58 mg/kg dry wt of *O. niloticus* and *Mugil cephalus*, respectively recoded in farm II. While, in gills the highest concentration were 209.80±13.49 &134.1±15.39 mg/kg dry wt of *O. niloticus* and *Mugil cephalus* recoded in farm II & farm III, respectively. So we were observed that Zn concentrations recorded in both liver and gills in *O. niloticus* were higher than that were recorded in *Mugil cephalus*. In muscles the highest concentrations of Zn of *O. niloticus* was 199.5±36.11and in *Mugil cephalus* was 90.4±10.17 recorded in farm II. While in muscle, the minimum concentrations of Zn of *O.*

niloticus were 55.1±6.70 and *Mugil cephalus* was 43.53±5.69 recorded in farm I. Also the BAF were recorded of liver, gills and muscle of *O. niloticus* were higher than the BAF were recorded in *Mugil cephalus*. in addition to liver was the highest BAF was recorded followed by gills and the minimum BAF was recorded in both type of fish.

The thresholds of metals in fish can be considered as the concentration level where the metal starts to interfere with the variable physiology of the fish species in such manner that once a particular level of the metal has been sequester in the body, equilibrium is established between the fish burden and the ambience. Also, Olaifa *et al.* (2004) reported that fish species can accumulate heavy metals above the biotic environment to incur bioaccumulation. Species difference in heavy metals bioaccumulation could be linked to difference in feeding habits and behavior of the species (Altindag and Yigit, 2005). According to WHO (1989) and USEPA (1986) all recorded results in the two fish species exceeded the permissible limits for Zn (50 ppm).

Table 4. Mean concentrations of zinc (mg/kg) in different organs of *O. niloticus* and *Mugil cephalus* at various sampling sites from August to November, 2014.

Fish species	Organs	Farm I	Farm II	Farm III
		Sahel El-Husseinya	San El-Hager	Kafer El-Sheikh
Oreochromis niloticus	Muscle	55.1 ± 6.70^{b}	199.5 ± 36.11^{a}	64.24 ± 3.93^{b}
	BAF	114.79	133	57.71
	Gills	137.30 ± 34.06^{ab}	209.80 ± 13.49^{a}	89.99 ± 9.87^{b}
	BAF	286.04	139.87	80.85
	Liver	193.70 ± 9.15^{b}	573.9 ± 83.3^{a}	239.7 ± 20.26^{b}
	BAF	403.54	382.6	215.36
Mugil cephalus	Muscle	43.53 ± 5.69^{b}	90.4 ± 10.17^{a}	72.49 ± 12.3^{b}
	BAF	90.69	55.87	56.15
	Gills	92.64 ± 18.26^{a}	134.1 ± 15.39^{a}	150 ± 31.5^{a}
	BAF	193.40	89.4	135.14
	Liver	103.8 ± 11.37^{b}	253.77 ± 27.58^{a}	212.8 ± 30.8^{a}
	BAF	216.25	169.18	191.7

*Means with the same letter within the same row are not significantly different; BDL, below detection limit BAF means bioaccumulation factor

Data in Table (5) show that Cd concentrations were absent in fish farm I and in other fish farms Cd levels were not exceed the permissible limits according to WHO (1989). Cadmium is highly toxic Non–essential heavy metal and it does not have a role in biological process in living organisms. Thus even in low concentration, cadmium could be harmful to living organisms (Tsui and Wang, 2004).

The highest concentrated values in liver for Cd recorded in farm II for *O. niloticus* and *Mugil cephalus* which were 0.28±0.02 and 0.22±0.08 mg/kg dry wt, respectively. The lowest numbers were 0.14±0.04 and 0.0004±0.00 in farm III for *O. niloticus* and *Mugil cephalus,* respectively.

In gills, the highest value for *O. niloticus* was 0.17±0.04 and 0.17±0.03 for *Mugil cephalus* in farm II. While, the lowest numbers recorded in farm III for *O. niloticus* and *Mugil cephalus* which were 0.04±0.00 and ND, respectively. In muscles, the highest values were recorded in farm II for *O. niloticus* and *Mugil cephalus* which were 0.12±0.006 and 0.057±0.06 mg/kg dry wt, respectively. The lowest concentrations were recorded in farm III (0.032±0.002 and ND) for *O. niloticus* and *Mugil cephalus,* respectively.

The maximum BAF of Cd in liver of *O. niloticus* and *Mugil cephalus* was 175 and 137.5, respectively in farm II; while, the minimum for *O. niloticus* and *Mugil cephalus* were 31.81 and 0.09, respectively at farm III. Also, in gills of *O. niloticus* and *Mugil cephalus,* maximum BAF of Cd were 108.13 and 104.38, respectively in farm II but the lowest was 9.09 in *O. niloticus* and not detected in *Mugil cephalus* in farm III. The maximum BAF of Cd recorded in muscles were 100 and 50 of *O. niloticus* and *Mugil cephalus* in farm II.

The minimum BAF of Cd recorded in muscles of *O. niloticus* and *Mugil cephalus,* was 7.27 and ND, respectively in farm III. From these results, it could be observed the increase of BAF of liver, gills and muscles for *O. niloticus* than that for *Mugil cephalus*. In the literature, heavy metal concentrations in the tissue of freshwater fish vary considerably among different studies (Osman, 2012) possibly due to differences in metal concentrations and chemical characteristics of water from which fish were sampled, ecological needs, metabolism and feeding patterns of fish.

The levels of Cd present in the selected organs of two fish species may be due to industrial and agricultural operations in the investigated areas (Ambedkar and Muniyan, 2011).

Table 5. Mean concentrations of cadmium (mg/kg) in different organs of *O. niloticus* and *Mugil cephalus* at various sampling sites from August to November, 2014.

Fish species	**Organs**	**Farm I**	**Farm II**	**Farm III**
		Sahel El-Husseinya	**San El-Hager**	**Kafer El-Sheikh**
Oreochromis niloticus	**Muscle**	ND	0.12±0.006^{a}	0.032±0.002^{b}
	BAF		75	7.27
	Gills	ND	0.17±0.04^{a}	0.04±0.006^{b}
	BAF		106.25	9.09
	Liver	ND	0.28±0.02^{a}	0.14±0.04^{b}
	BAF		175	31.81
Mugil cephalus	**Muscle**	ND	0.057±0.0٢^{a}	ND^{b}
	BAF		35.63	
	Gills	ND	0.17±0.03^{a}	ND^{b}
	BAF		106.25	
	Liver	ND	0.22±0.08^{a}	0.0004±0.0001^{b}
	BAF		137. 5	0. 1

*Means with the same letter within the same row are not significantly different; BDL, below detection limit BAF means bioaccumulation factor

Lead metal is not detected in all fish samples at all fish farms except in liver for *Mugil cephalus* (0.39±0.02 mg/kg dry wt.) of farm III and this level was not exceeded the WHO (1985) and FEPA (2003) standard limits (2.0 mg/kg).

High accumulations of the Fe, Cu and Zn in water and fish muscles of the selected areas are in agreement with the findings of Shakweer (1998), who concluded that the concentrations of trace metals in various organs of fish reflects the degree of water pollution in the aquatic environments in which such fish are living. Also, Ravera (2001) reported that, if an environment receives metal pollutants, the organisms living in it could take up the pollutants from the water or/and food and concentrate it in their bodies. Fish species living in contaminated waters tend to accumulate heavy metals in their organs and tissues. Various heavy metals are accumulated in fish body in different amount (Jezierska and Witesta, 2001). IN addition, lead and cadmium are taken up passively from the water and deposited in the organisms and hence the organisms contain more quantity of Cd than that in water (Marekert *et al.*, 1997).

Cd and Pb are toxic elements which have no biological functions and show their carcinogenic effect on aquatic biota and humans. Lead toxicity is known to cause musculo-skeletal, renal, ocular, neurological, immunological, reproductive and developmental effects (ATSAR, 1999).

The highest metal values recorded in station II & III may be due to the fact that metal pollutants are discharged into aquatic environment from urban and domestic activities. The relatively higher values obtained from the stations of Sharkia and Kafer El- Sheikh in fish organs may be due to the impact of pollution sources in these areas coming from non treated sewage water and draining water.

The results confirm the differences of heavy metals accumulation in different tissues of fish and the gradual accumulation and increase in the liver, gills and muscles was in the same rang in *O. niloticus* and *Mugil cephalus* at all stations.

Liver of the examined fish contained the highest concentrations of all the detected metals. Also higher metal concentrations in the gills could be due to the element complexion with the mucus that is in gills of *O. niloticus* and *Mugil cephalus*. The highest accumulation levels were in this pattern Fe, Zn, Cu, Cd and Pb. The gills are an important site for the entry of heavy metals that provokes lesions and gills damage (Bols *et al*., 2001).

Muscles appeared to be the least preferred site for the bioaccumulation of metals as the lowest metal concentrations were detected in this tissue. These results were in agreement with many authors who reported that tissue is not an active organ in accumulation of heavy metals (Khail and Faragallah, 2008).

In muscles of *O. niloticus* and *Mugil cephalus*, the highest accumulations levels were as Fe, Zn, Cu, Cd while, Pb was not detected.

Data in Table 6 showed fluctuations in glucose, total protein, AST and ALT in serum of *O. niloticus* and *Mugil cephalus* through the different sites.

Table 6. Changes in serum glucose, Total protein, GOT and GPT of the *O. niloticus* and *Mugil cephalus* at various sampling sites from August to November, 2014.

Fish species	Organs	Farm I	Farm II	Farm III
		Sahel El-Husseinya	San El-Hager	Kafer El-Sheikh
Oreochromis niloticus	Glucose	9.5±1.3[c]	23.07±0.67[b]	59.2±2.05[a]
	T. protein	1.88±0.37[b]	2.40±0.28[b]	3.08±0.23[a]
	AST	55.29±5.55[a]	26.13±5.43[b]	33.29±1.29[b]
	ALT	6.9±2.5[a]	5.07±0.96[a]	4.82±0.97[a]
Mugil cephalus	Glucose	33.68±4.79[b]	72.62±5.16[a]	66.7±2.60[a]
	T. protein	2.08±0.49[a]	2.71±0.38[a]	2.57±0.37[a]
	AST	80.04±9.4[b]	142.2±10.04[a]	80.05±9.50[b]
	ALT	7.84±1.5[b]	20.45±3.5[a]	4.05±2.54[b]

*Means with the same letter within the same row are not significantly different; BDL, below detection limit

These fluctuations may be attributed to the stress effect of pollution in water. Jent *et al*. (1998) mentioned that the higher accumulation of some heavy metals in liver may alter the levels of various biochemical parameters in liver. This may also cause severe liver damage (Ferguson, 1989). The liver, as the major organ of metabolism, comes into close contact with xenobiotic absorbed from the environment and liver lesions are often associated with aquatic pollution.

Glucose is one of the most important sources of energy for the animals and glucose has been studied as an indicator of stress caused by physical factors of particular pollutants (Manush, 2005). In the present study, the increase in serum glucose level of *O. niloticus* and *Mugil cephalus* collected from different fish farms may due to gluconeogenesis to provide energy for the increased species metabolic demands imposed by the poor water quality and heavy metals. Previous investigations proved that, heavy metals such as cadmium modulate the metabolism of carbohydrates, causing hyperglycemia by stimulating the glycogenolysis in some marine and freshwater fish species (Levesque, 2002). Present results confirm this where there are significant positive between glucose levels in blood of *O. niloticus* and *Mugil cephalus* collected from III & II farms and some heavy metals (Fe, Zn, Cu, Cd and Pb) concentrations.

Protein is also one of the important biochemical parameters which have been used to understand the general state of health and biological mechanism of metabolism under pollutant stress (Saravanan, 2011). During stress since gills are the respiratory and osmoregulatory conditions fish need more energy to detoxify the toxicant and to overcome stress. So, due to this, proteins in liver degrade and the serum protein level is altered. Singh and Sharma (1998) reported decline in protein constituent in liver and increase in serum in different fish under stress of pollutants. Results of the present study revealed decreased values of protein, in blood of *O. niloticus* and *Mugil cephalus* collected from different fish farms.

Among the biochemical profiles, monitoring of liver enzymes leakage into the blood has proved to be a very useful tool in liver toxicological studies (Osman *et al*., 2010). The transaminases, AST and ALT are two key enzymes considered as a sensitive measure to evaluate hepato cellular damage and some hepatic diseases (Ibrahim and Mahmoud, 2005). So, in the present study, the increase in AST and ALT transaminases might be attributed to tissue damage, particularly liver (Palanivelu *et al.*, 2005). In different fish species including *O.*

niloticus and *Mugil cephalus* AST and ALT enzymes activity were found to increase in response to heavy metals. Mekkawy (2011). An increase in serum AST and ALT were detected due to high levels of Fe, Zn, Cu and Cd.

In the present study the alterations of these parameters goes in some fish farms which were characteristic in the levels of heavy metals concentrations as a result of pollution stress as previously mentioned by Tayel (2007). Also, level of heavy metals found in the edible tissues (muscles) of the examined fish samples were within the safe limits except iron, zinc, WHO (2011) reported that there are no health based guideline-value for both Fe and Zn. Also, we recommend that must be treatment the water source of darning and domestic water before used in aquaculture.

Table 7. Pearson's correlation coefficient matrix between heavy metals concentration in liver and some biochemical parameters of the *Oreochromis niloticus*.

	Fe	**Cu**	**Zn**	**Pb**	**Cd**	**Glucose**	**protein**	**AST**	**ALT**
Fe	1.00***	0.974**	-0.159	nd	0.136	0.897**	0.753*	-0.367	-0.270
Cu		1.00***	-0.338	nd	-0.026	0.903**	0.683*	-0.215	-0.246
Zn			1.00***	nd	0.788*	-0.139	0.160	-0.742*	-0.106
Pb				nd	nd	nd	Nd	nd	Nd
Cd					1.00***	0.251	0.314	-0.830**	-0.208
Gucose						1.00***	0.716*	-0.426	-0.265
Protein							1.00***	-0.674*	0.175
AST								1.00***	0.169
ALT									1.00***

***** Significant at p<0.05 ****** Significant at p<0.01 ******* Significant at p<0.001 Ns not significant

Correlation between heavy metals in liver and blood parameters in *Oreochromis niloticus* were assessed and presented in Table 7. The correlations between the different metals may result from the similar accumulation behavior of the metals in the fishes and their interactions (Rejomon et al 2010). Noted significant correlations among metals and may reflect a common source of occurrence and indicative of similar biogeochemical pathways for subsequent accumulation in the liver tissue of fish. In the present study, iron is strongly correlated with copper. Zinc, similar to iron showed high correlation with cadmium. No other significant correlation was observed between studied heavy metals. On the other hand, there are strong correlation between iron and copper as well as increased glucose levels. Also, positev and significant correlation between increase total protein and increased iron and copper. No significant between iron, copper, zinc, lead and cadmium and other parameters (AST & ALT).

Table 8. Pearson's correlation coefficient matrix between heavy metals concentration in liver and some biochemical parameters of the *Mugil cephalus***.**

	Fe	**Cu**	**Zn**	**Pb**	**Cd**	**Glucose**	**Protein**	**AST**	**ALT**
Fe	1.00***	0.572	0.581	0.321	0.402	0.844**	0.550	0.474	0.179
Cu		1.00***	0.848**	-0.054	0.316	0.589	0.546	0.503	0.565
Zn			1.00***	0.180	0.425	0.800**	0.377	0.457	0.492
Pb				1.00***	-0.437	0.335	-0.035	-0.474	-0.577
Cd					1.00***	0.569	-0.072	0..823**	0.703*
Glucose						1.00***	0.235	0.541	0.379
Protein							1.00***	0.104	-0.035
AST								1.00***	0.902**
ALT									1.00***

***** Significant at p<0.05 ****** Significant at p<0.01 ******* Significant at p<0.001 Ns not significant

Data in Table 8 showed the correlation between heavy metals concentration in liver and some biochemical parameters of the *Mugil cephalus.* There is highly significant between copper and zinc. In addition, strong correlation between zinc and glucose also between cadmium and AST but significant correlation with ALT and week correlation between liver enzymes metals

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تراكم بعض العناصر الثقيلة في مياه وأسماك المزارع السمكية (البلطي – البوري) بمحافظتي الشرقية وكفر الشيخ وأثرها علي بعض القياسات البيوكيميائية

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الملخـص العربـى

أجريت هذه الدراسة علي مياه وأسماك البلطي النيلي والبوري المجمعة من منطقة صان الحجر وسهل الحسينية بمحافظة الشرقية واحدي المزارع بمحافظة كفر الشيخ ، والتي تعد من أهم مصادر البروتين الحيواني في جمهورية مصر العربية. نظرا لندرة المياة واستخدام مياه الصرف الزراعي في الإستزراع السمكي وكذلك لجوء بعض المزارعين إلي استخدام مياه الصرف الصحي ، الأمر الذي أدى إلى تزايد حالة القلق لدى المستهلك وكذلك حالات النفوق المفاجئة للأسماك في العديد من المزارع السمكية فقد جاءت أهمية هذه الدراسة لبيان حالة المياه والأسماك المستزرعة بهذه المناطق.

تناولت هذه الدراسة تقدير بعض العناصر الثقيلة وهي الحديد ، النحاس ، الزنك ، الرصاص ، الكادميوم في المياه وكذلك كبد وخياشيم وعضلات أسماك البلطي النيلي والبوري المستزرعة في هذه المناطق وإجراء بعض التحاليل البيوكيميائية مثل الجلوكوز و البروتين الكلي وانزيمات GPT & GOT في الدم لبيان احتمال تأثير مثل هذه العناصر على الحالة الصحية للأسماك محل الدراسة.

أوضحت النتائج في عينات المياه والأسماك أن عنصر الحديد كان الأكثر تراكما يليه الزنك ثم النحاس ثم الكادميوم ولم يتم رصد أي تراكمات للرصاص في جميع العينات. وجد أن مستويات عنصري الحديد بالمياه أعلى من الحدود المثلي المسموح بها من قبل منظمة الصحة العالمية ١٩٨٩ أما مستويات النحاس والزنك والكادميوم والرصاص فكانت أقل من الحدود المثلي للمنظمة. أما في الأسماك فقد جاء الكبد في مقدمة الأعضاء الأكثر تراكما للعناصر يليه الخياشيم ثم العضلات وهي الأقل تركيزا للعناصر هذا ولم يكن هناك اختلاف جوهري بين سمكة البلطي والبوري في تراكم تلك العناصر ، فقد تراوحت بين الزيادة والنقصان. وجد أن مستويات الحديد وكذلك الزنك في أنسجة البلطي والبوري كانت أعلي من الحدود المسموح بها طبقا لمنظمة الصحة العالمية ١٩٨٩. كذلك أوضحت الدراسة أن تأثير المتراكم من هذه العناصر في الأسماك أدي إلى حدوث اضطرابات في القياسات البيوكيميائية نظرا لما

تعرض له كبد وخياشيم هذه الاسماك من تلف خصوصا بالمناطق الأكثر تلوثا حيث ارتبطت هذه القياسات بكمية المتراكم من تلكم العناصر في أنسجة هذه الأسماك.

اوضحت الدراسة ان مزرعة محافظة كفر الشيخ وصان الحجر بمحافظة الشرقية هما الأكثر تعرضا للتلوث في حين تعد مزرعة سهل الحسينية بمحافظة الشرقية الأقل عرضة للتلوث الي جانب غياب عنصر الكادميوم فيها غيابا تاما.