



Impact of Some Heavy Metals on Muscles, Hematological and Biochemical Parameters of the Nile Tilapia in Ismailia Canal, Egypt

* Mohamed Y. M. Aly^{1*}, Dalia M. El-Gaar², Ghada S. Abdelaziz³

^{1,2}Pollution Laboratory, ³Chemistry Laboratory, Freshwater and Lakes Division, National Institute of Oceanography and Fisheries, Cairo, Egypt

*Corresponding Author: redwanmuhammad151@gmail.com

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ABSTRACT

Ismailia Canal is one of the most important irrigation and drinking water resources. It is one of the most important branches of the Nile River in Egypt. It is the main source of drinking and irrigation water for many cities. In a four- month study period (April- July 2022), *O. niloticus* fish samples were collected every two weeks from 4 sites. Abu-Z'abaal, Belbies, Abbassa region and El-Tal Al-Kapeir City were selected to study. The samples were chemically analyzed for the detection of heavy metals (Fe, Zn, Cu, Cd and Pb). El-Tal Al-Kapeir City site was more polluted than the other sites. The average concentration of Fe in the water of Abbassa region exceeded the permissible limit, while Pb surpassed the permissible range in all regions for the water samples prescribed by EOS (1993) and ESC (1983). The concentration of heavy metals evaluated in the muscles of the fish under study was generally lower than the levels issued by WHO (1999), EOS (1993). The mean values of RBC, Hb, Hct and MCV in the blood of the Nile tilapia decreased from Abu Z'abaal City to El-Tal Al-Kapeir city. On the contrary, MCH and MCHC increased from Abu Z'abaal City to El-Tal Al-Kapeir City. The mean concentrations of all the biochemistry parameters increased in the blood serum of fish collected from El-Tal Al-Kapeir City compared to those sampled from Abu Z'abaal City.

INTRODUCTION

Ismailia Canal is one of the most affected ecosystems by pollution resulting from human activity. It extends eastward for a distance of approximately 125km from the Nile River in the Al-Mazalat region north of Cairo to Ismailia City on the Suez Canal (Geriesh *et al.*, 2008). It was divided into two branches: one in the north to supply the city of Port Said and the second in the south to supply the city of Suez. The width of the Canal is about 30-70 m. The average depth of the water is 2.8 meters. The average velocity of water is 0.28m/ s (Ibrahim *et al.*, 2009). In the final development phase, the canal discharge reached a value of 433.56 m³/s (Mohamed, 2008). The upstream portion of Ismailia Canal (from Cairo to Abu-Z'abaal, western side) includes the largest industrial zones in the region. Ismailia Canal acts as an effluent stream along these sites. Chemical leakages and fallen dusts from these industries pollute the canal water (Geriesh *et al.*, 2008). Previous studies reported that

Ismailia Canal is polluted with heavy metals, organic contaminants, agricultural chemicals and indicator bacteria of sewage pollution (**Ralph & Ramadan, 2008**). Heavy metals are natural trace components of the aquatic environment, but their levels have been increased due to industrial waste, geochemical structure, agricultural and mining activities (**Singh *et al.*, 2007**). All these sources of pollution affect the physicochemical properties of water, sediments and biological components, thus the quality and quantity of fish stocks (**Al-Rawi, 2005**). Heavy metals enter the environment through various natural methods and human activities and can accumulate in fish and other organisms causing a serious and widespread environmental problem due to their toxicity. The untreated industrial and sewage wastes arising from industries and metropolitan activities polluting River Nile (**Kalay & Canli, 2000**).

Heavy metals are dealt with a special care and great importance due to their highly toxic effect on fish in addition to fish tendency to accumulate high amounts of heavy metals in their tissues (**Marcovecchio, 2004**). Fish absorbed heavy metals from water through the gills, skin and digestive tract. The heavy metals of the widest spreading concern to human health are lead, mercury and cadmium (**Chen *et al.*, 2007; Din *et al.*, 2008**). Heavy metals resist chemical and biological transformation and accumulate in the tissues of liver, kidney and nerve causing toxicity. Uptake of heavy metals through food chain in aquatic organisms may cause various pathological disorders including hypertension, sporadic fever, renal damage or cramps in human (**Gabriel *et al.*, 2006**). Hematological and biochemical variables have become promising biomarkers in measuring the effects of aquatic pollution in fish since blood parameters respond to low doses of pollutants. Blood parameters are considered as good physiological biomarkers of the whole body, and therefore, they are important in diagnosing the structural and functional status of fish exposed to environmental pollutants (**Seriani *et al.*, 2011**).

Hematological and biochemical variables are nonspecific in fish response to environmental stressors. However, they may provide important information in studies evaluating the impact by influencing the general physiology and health status of fish (**Mekkawy *et al.*, 2011**). Several researchers have investigated hematologic changes of pollutants in fish (**Sayed *et al.*, 2011; Heier *et al.*, 2013**). The use of blood biomarkers in fisheries research is rapidly increasing as they are very important in environmental monitoring for toxicological research and in predicting health conditions of fish (**Bitten-Court *et al.*, 2003**). Eventually, for these reasons, heavy metals load in fish has become an important worldwide concern, not only because of the threat posed on fish but also the health risks associated with fish consumption (**Begum *et al.*, 2013**).

Consequently, this study aimed to determine the amount of heavy metals in the water and muscles of fish (*Oreochromis niloticus*) and assess their effect on hematological and biochemical parameters at four stations of the Ismailia Canal, starting from Abu Zaabal area to Al-Tal Al-Kapir along this canal.

MATERIALS AND METHODS

1. Study area

Water and fish samples were collected every two weeks during the trial period from April to July during the year 2022. Four stations (1-4) were chosen as sampling sites along the area from Abu Z'abaa City to the city of El-Tal Al-Kabir, which has the largest amount of pollution sources. With the help of the fishermen, samples were gathered from the areas under study, where sources of pollution were monitored in each region that was described as follows:-

Site 1 (Abu Z'abaa City): Source of pollution: Fertilizer companies; industrial waste (Abu Z'abaa Fertilizer Company), where some untreated factory waste is discharged directly to the Ismailia Canal. It lies between Latitude $30^{\circ} 16^{\prime} 28^{\prime\prime}$ and Longitude $31^{\circ} 22^{\prime} 44^{\prime\prime}$

Site 2 (Belbies city): Source of pollution: sewage and industrial wastes from the city of the 10th of Ramadan, lying between Latitude $30^{\circ} 24^{\prime} 57^{\prime\prime}$ and Longitude $31^{\circ} 34^{\prime} 33^{\prime\prime}$.

Site 3 (Abbassa area): The source of pollution is the fish farm waste and agricultural wastewater. These wastes are discharged directly to the Ismailia Canal. The area is located between Latitude $30^{\circ} 32^{\prime} 04^{\prime\prime}$ and Longitude $31^{\circ} 42^{\prime} 35^{\prime\prime}$.

Site 4 (El-Tal Al-Kapeir City): The source of pollution is the agricultural drainage waste, which is discharged directly to the Ismailia Canal from the various agricultural drains located along the Ismailia Canal in the city. It is located between Latitude $30^{\circ} 32^{\prime} 47^{\prime\prime}$ and Longitude $31^{\circ} 52^{\prime} 09^{\prime\prime}$.

The details of surface water and fish sampling from the study sites are presented in Table (1) and Fig. (1).

Table 1. Source of pollution, latitude and longitude of surface water sampling locations along Ismailia Canal

Station	Source of pollution	Latitude	Longitude
Abu Z'abaa city	Industrial waste fertilizer companies (Abu Z'abaa fertilizer Company)	$30^{\circ} 16^{\prime} 28^{\prime\prime}$	$31^{\circ} 22^{\prime} 44^{\prime\prime}$
Belbies city	sewage and industrial waste	$30^{\circ} 24^{\prime} 57^{\prime\prime}$	$31^{\circ} 34^{\prime} 33^{\prime\prime}$
Abbassa region	Agricultural and fish farm waste	$30^{\circ} 32^{\prime} 04^{\prime\prime}$	$31^{\circ} 42^{\prime} 35^{\prime\prime}$
El-Tal Al-Kapeir city	Agricultural drainage waste and industrial waste	$30^{\circ} 32^{\prime} 47^{\prime\prime}$	$31^{\circ} 52^{\prime} 09^{\prime\prime}$

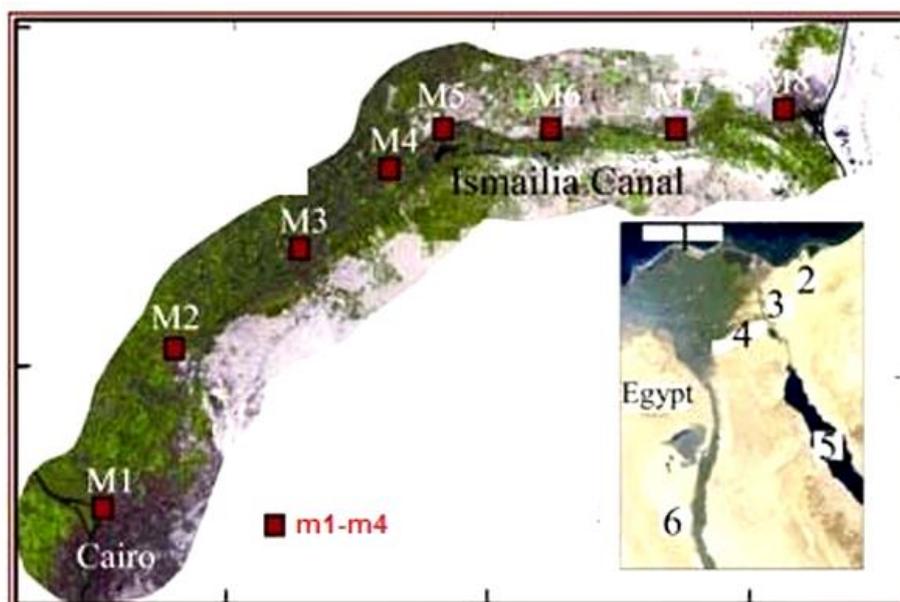


Fig. 1. The water and fish sampling locations of Ismailia Canal

2. Collected data from samples

Samples were collected for each site to determine different data in terms of its sources of pollution. Fish samples were collected; blood samples were taken from them at the time of their acquisition, and the necessary steps were taken to preserve them until analysis, as well as water samples using the methods used to preserve them until analysis.

3. Water samples

Surface water samples were taken from the four stations under study by a PVC vertical water sampling device at a depth of half a meter from the water surface. Samples of 1L were placed in polyethylene bottles and transported to the laboratory for heavy metal analysis. All water samples were collected from each site in three samples each time during the experiment.

4. Clinical examination of fish samples

The collected fish were clinically examined for any external abnormalities in the body. The existence of pathological lesions in the muscles of fish; fish individuals were alive to take blood samples, and the average lengths and weights at the time of taking blood sample are all presented in Table (2).

Table 2. Average lengths and weights of *O. niloticus* collected from the four sites along the Ismailia Canal during the experimental period

Site	Number	Variable	Minimum values	Maximum values	Mean
Abu Z'abaa City	40	Weight (g)	85.7	195.4	140.5
		Length (cm)	18.3	24.7	21.5
Belbies City	40	Weight (g)	75.9	181.20	128.5
		Length (cm)	16.8	23.41	20.1
Abbassa region	40	Weight (g)	92.6	152.90	122.7
		Length (cm)	18.20	20.7	19.4
El-Tal Al-Kapeir City	40	Weight (g)	76.31	166.3	121.3
		Length (cm)	15.6	21.2	18.4

5. Fish samples collection

About 160 live fish (*Oreochromis niloticus*) samples were collected biweekly during the experimental period from April to July 2022, using networks of selected sites (40 samples from each site). Total fish weights ranged from 15.6- 24.7cm for TL and 75.9– 195.4g in weight). For water samples, they were kept in a one-liter polyethylene bottle in an ice box and analyzed in the laboratory.

6. Blood samples

Two blood samples were collected. First sample was freshly collected in glass tubes containing anticoagulant (Na-EDTA) for hematological analysis. Hemoglobin concentration (Hb), hematocrit value (Hct) and red blood cells (RBCs) count were determined using automated hematological analyzer, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC), calculated according to **Dacie and Lewis (2001)**.

The second blood sample was collected and left to coagulate for 15 – 20min prior to centrifugation for 20min at 3000rpm to obtain the serum for biochemical analysis (**Osman et al., 2018**). Colorimetric determinations of the selected biochemical parameters were performed using spectrophotometer (Jasco-V530). The absorbency of the detected sample was examined at an appropriate wavelength ranging from 340 to 546nm according to the parameter tested. Alanine aminotransferase (ALT, U/I), aspartate aminotransferase (AST, U/I), cholesterol (mg/dl), glucose (mg/dl), creatinine (mg/dl), Urea (mg/dl) and uric acid were determined using commercial diagnostic kits (bioMérieux, France), following the manufacturer's instruction.

7. Statistical analysis

Descriptive statistical analysis (Mean \pm SD) was performed using a computer program of SPSS (version17.0 for windows).

RESULTS

1. Heavy metals concentrations in water and fish muscles

The present study assessed the concentration of five heavy metals (Fe, Zn, Cu, Cd and Pb) in water collected from the four sites along the area were chosen from Abu Z'abaa to El-Tal Kabeir (Table 3). Iron (Fe) values fluctuated within range (211.7 to 471.41 µg/l). The highest value was recorded at Abbassa (471.41 µg/l). Zinc (Zn) concentrations fluctuated between 51.86 and 139.46 µg/l, where the highest value was recorded at Al-Tall Al-Kabeir. While, the lowest value was recorded in Abu Z'abaa. For copper (Cu), the highest value was in that of the Al-Tall Al-Kabeir (38.8 µg/l) and the lowest was found in Abu Z'abaa (29.5 µg/l). These results showed a slight increase in copper in the Belbies, compared to the other places. Cadmium (Cd) values fluctuated in a narrow range (2.9 to 4.4 µg/l). The lowest value was recorded in Abu Z'abaa (2.9 µg/l) and the highest was found in Al-Tall Al-Kabeir (4.4 µg/l). Lead (Pb) values fluctuated within range (15.9 to 22.5 µg/l). The highest value was recorded at Abbassa (22.5 µg/l) and the lowest was found in Abu Z'abaa (15.9 µg/l).

The results of heavy metals concentrations in the muscle of the fish (*O. niloticus*,) are presented in Table (3). Compared to the other metals, Iron (Fe) showed higher values at all sites, ranging from 31.4 to 71.6 µg/kg. Its highest value was in Abbassa region, and its lowest value was recorded in Abu Z'abaa. Zinc (Zn) concentrations fluctuated between 13.3 and 28.2 µg/kg, where the maximum values were recorded at Al-Tall Al-Kabeir, and its lowest value was recorded in Abbassa. Copper (Cu) showed simple variation ranging from 2.83 to 5.17 µg/kg. The highest concentrations were recorded in Abbassa and the lowest in Abu Z'abaa. Cadmium (Cd) showed slight differences between the four sites, ranging between 1.4 and 1.9 µg/kg. The lowest concentration was recorded in Abu Z'abaa and the highest concentration in Al-Tal Al-Kabeir. In general, cadmium concentrations fluctuated between sites but still showed a slight increase from Abu Z'abaa towards Al-Tal Al-Kabeir. Lead (Pb) concentrations showed obvious difference between the four sites. The highest value was recorded in Abbassa (7.8 µg/kg), and the lowest value was recorded in Abu Z'abaa (4.7 µg/kg).

2. Maximum permissible concentrations of heavy metals in water and fish muscles

The concentrations of heavy metals in water and fish are presented in Table (4). The average concentration of Fe in Abbassa surpassed the permissible limit but Pb exceeded the permissible limit in all regions prescribed by **EOS (1993)**, with a note according to Egyptian chemical standards (**ECS, 1994**). The maximum permissible limits of heavy metal in water are 5mg for Zn, 1mg/l for Cu, 0.01mg/l for Cd, 0.05mg/l for Pb and 1mg/l for Fe (**ECS, 1994**). In the current study, levels of five heavy metals from four sites were within the permissible limits. The measured heavy metal concentrations in fish muscles were generally lower than levels reported in **EOS (1993)** and **WHO (1999)**.

3. The proportions of heavy metals in water and fish muscles

Sequence of heavy metal concentrations from four sites were ordered as follows: Fe>Zn>Cu> Pb>Cd in water at all stations, while it was sequentially ordered to be Fe >

Zn>Pb> Cu> Cd in muscles at Abu Z'abaa and Belbies. Whereas, it was Fe> Zn> Pb> Cd> Cu in Abbassa region and El-Tal Al-Kapeir City (Table 5).

4. Concentration of heavy metals according to their percentages at sites

The sequence of concentrations of heavy metals in water according to their presence in the sites was as follows: iron and lead: Abbassa >El-Tal Al-Kapeir > Belbies > Abu Z'abaa; copper and cadmium: El-Tal Al-Kapeir > Abbassa > Belbies > Abu Z'abaa. While, zinc: El-Tal Al-Kapeir > Belbies > Abbassa > Abu Z'abaa.

While, in the muscles of the fish, the sequence was as follows: iron and lead: Abbassa >El-Tal Al-Kapeir > Belbies > Abu Z'abaa; zinc: El-Tal Al-Kapeir> Abu Z'abaa > Belbies > Abbassa; copper: El-Tal Al-Kapeir > Abbassa > Belbies > Abu Z'abaa and cadmium: El-Tal Al-Kapeir > Belbies > Abbassa > Abu Z'abaa (Tables, 6 & 7).

Table 3. Heavy metal concentration (Mean \pm SD) in water ($\mu\text{g/l}$) and fish muscle ($\mu\text{g/kg}$ dry wt.) samples from the studied locations

Metal	Abu Z'abaa		Belbies		Abbassa		El-Tal Al-Kapeir	
	water	muscles	water	muscles	water	muscles	water	muscles
Iron (Fe)	211.7 \pm 19.51	31.44 \pm 7.12	256.4 \pm 23.4	35.37 \pm 7.31	471.41 \pm 43.74	71.69 \pm 15.3	291.12 \pm 28.4	41.6 \pm 9.25
Zinc (Zn)	51.86 \pm 3.53	27.3 \pm 2.32	118.56 \pm 5.53	14.3 \pm 3.11	87.3 \pm 3.54	13.3 \pm 2.49	139.46 \pm 6.67	28.2 \pm 4.77
Copper (Cu)	29.5 \pm 2.4	2.83 \pm 0.4	31.31 \pm 3.22	3.65 \pm 0.6	33.6 \pm 4.62	4.37 \pm 0.72	38.8 \pm 3.1	5.17 \pm 1.59
Cadmium (Cd)	2.9 \pm 0.33	1.47 \pm 0.35	3.1 \pm 1.17	1.8 \pm 1.8	3.3 \pm 0.26	1.68 \pm 0.44	4.4 \pm 2.34	1.94 \pm 2.2
Lead (Pb)	15.9 \pm 1.22	4.7 \pm 1.05	18.9 \pm 1.14	5.76 \pm 1.15	22.5 \pm 2.58	7.8 \pm 2.95	21.2 \pm 1.19	6.4 \pm 1.31

Table 4. Maximum permissible limit (MPL) of heavy metals in water ($\mu\text{g/l}$) and fish (mg/kg wet wt.) according to international standards

Source	Fe	Zn	Cu	Pb	Cd
ECS (1994) water	1000	5000	1000	500	100
EOS (1993) water	300	5000	1000	10	10
WHO(1999) fish	43.0	60.0	3.0	0.214	0.1
EOS (1993) fish	30	40	20	2.0	0.5

*Permissible limits according to guidelines of EOS (1993) and ESC (1994) in water; WHO (1999) and EOS (1993) in fish.

Table 5. Sequence of heavy metal concentrations in water samples and *O.niloticus* muscles from four sites in the Ismailia Canal

Site	Water	Fish muscles
Abu Z'abaa City	Fe > Zn > Cu > Pb > Cd	Fe > Zn > Pb > Cu > Cd
Belbies City	Fe > Zn > Cu > Pb > Cd	Fe > Zn > Pb > Cu > Cd
Abbassa region	Fe > Zn > Cu > Pb > Cd	Fe > Zn > Pb > Cu > Cd
El-Tal Al-Kapeir City	Fe > Zn > Cu > Pb > Cd	Fe > Zn > Pb > Cu > Cd

Table 6. Sequence of heavy metals in water relative to sites

Heavy metal	Site
Iron (Fe)	Abbassa >El-Tal Al-Kapeir > Belbies > Abu Z'abaa
Zinc (Zn)	El-Tal Al-Kapeir > Belbies > Abbassa > Abu Z'abaa
Copper (Cu)	El-Tal Al-Kapeir > Abbassa > Belbies > Abu Z'abaa
Cadmium (Cd)	El-Tal Al-Kapeir > Abbassa > Belbies > Abu Z'abaa
Lead (Pb)	Abbassa >El-Tal Al-Kapeir > Belbies > Abu Z'abaa

Table 7. Sequence of heavy metals in Fish muscles relative to sites

Heavy metal	Site
Iron (Fe)	Abbassa >El-Tal Al-Kapeir > Belbies > Abu Z'abaa
Zinc (Zn)	El-Tal Al-Kapeir > Abu Z'abaa > Belbies > Abbassa
Copper (Cu)	El-Tal Al-Kapeir > Abbassa > Belbies > Abu Z'abaa
Cadmium (Cd)	El-Tal Al-Kapeir > Belbies > Abbassa > Abu Z'abaa
Lead (Pb)	Abbassa >El-Tal Al-Kapeir > Belbies > Abu Z'abaa

4. Hematological and biochemical variables

The value of hematology (RBC, HG, HT, MCV, MCH, MCHC and WBC) and biochemistry (AST, ALT, glucose, cholesterol, creatinine, urea and uric acid) are presented in Tables (8, 9) as mean values \pm standard deviation. The mean hematological values obtained for the four sectors are presented in Table (8). The statistical analysis of these variables exhibited significant differences among the selected sites. The mean values of RBCs, Hb, Hct, and MCV in the blood of the Nile tilapia decreased from Abu Z'abaa City to El-Tal Al-Kapeir City. In contrast, MCH and MCHC in the blood of fish increased from Abu Z'abaa City to El-Tal Al-Kapeir City. The mean concentrations of cholesterol, alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), glucose, uric acid, creatinine and urea in the blood serum of the Nile tilapia are shown in Table (9). Differences were observed in the mean values of all the detected biochemical variables among the selected sites. The mean concentrations of all the biochemical parameters increased in the blood serum of fish collected from El-Tal Al-Kapeir City, compared to those sampled from Abu Z'abaa City (Table 9).

Table 8. Hematological concentrations (mean \pm SD) in the Nile tilapia blood samples collected from different sites of Ismailia Canal, Egypt

Parameter	Abu Z'abaa	Belbies	Abbassa	El-Tal Al-Kapeir
WBC ($10^3/\mu\text{l}$)	48.9 \pm 5.9	46.3 \pm 8.4	38.1 \pm 5.3	34.4 \pm 6.1
Hct (%)	38.4 \pm 6.3	35.9 \pm 7.6	21.6 \pm 4.1	19.5 \pm 4.8
Hb (g/dl)	12.4 \pm 3.8	11.5 \pm 3.1	11.3 \pm 2.9	7.6 \pm 1.3
RBC ($10^6/\mu\text{l}$)	1.9 \pm 0.76	2.2 \pm 1.16	1.8 \pm 1.19	1.3 \pm 0.16
MCV (fl)	144.4 \pm 25.9	131.8 \pm 22.1	125.1 \pm 14.2	118.1 \pm 14.7
MCH (Pg)	41.6 \pm 5.11	42.12 \pm 4.4	51.82 \pm 5.66	57.3 \pm 5.15
MCHC (g/dl)	26.52 \pm 4.34	28.51 \pm 4.21	37.43 \pm 5.85	36.04 \pm 5.31

Evaluation of the hemogram involved the determination of the white blood cell count (WBC), hematocrit (Hct), hemoglobin concentration (Hb), red blood cell count (RBC), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC).

Table 9. Biochemical parameters concentrations (mean \pm SD) in the Nile tilapia blood serum collected from different sites of Ismailia Canal, Egypt

Parameter	Abu Z'abaal	BELBIES	Abbassa	El-Tal Al-Kepeir
ALT (U/I)	24.12 \pm 3.18	21.45 \pm 2.96	30.71 \pm 4.52	36.61 \pm 4.88
AST (U/I)	25.41 \pm 3.39	28.83 \pm 4.34	33.65 \pm 4.14	45.31 \pm 6.29
Cholesterol (mg/dl)	222.3 \pm 11.62	167.9 \pm 7.81	255.63 \pm 14.4	283.71 \pm 9.8
Glucose (mg/dl)	30.31 \pm 3.91	44.71 \pm 6.28	52 \pm 4.82	59.64 \pm 4.2
Creatinine (mg/dl)	1.43 \pm 0.44	1.75 \pm 0.61	1.78 \pm 0.52	2.06 \pm 0.18
Urea (mg/dl)	12.45 \pm 3.2	19.19 \pm 5.5	22.81 \pm 5.12	26.71 \pm 4.19
Uric acid (mg/dl)	2.70 \pm 0.66	3.21 \pm 0.52	3.97 \pm 0.82	3.41 \pm 0.44

DISCUSSION

In Egypt and other developing countries, where environmental protection laws have not been enforced, industrial and domestic wastes are dumped indiscriminately into water bodies. These wastes have been reported to contain toxic and hazardous substances including metals. The contamination of water resources by trace metals is of important concern because of their toxicity, persistence and bio-accumulative nature (Ikem *et al.*, 2003). Heavy metals may enter an aquatic ecosystem from different natural and anthropogenic sources, including industrial or domestic sewage, storm runoff, leaching from landfills, shipping and harbor activities and atmospheric deposits (Rajeshkumar, & Munuswamy, 2011). From the analysis of heavy metals for samples of water and fish muscles in the four sites under study, the last site, which is the city of El-Tal Al-Kepeir city, showed higher concentrations of zinc, copper and cadmium compared to the other three sites. Moreover, the concentration of both iron and lead was higher in the Abbassa area than the other sites. This is due to the reception of all pollutants from the canal from its inception in addition to the sources of pollution present in these two stations. These results coincide with the findings of some studies conducted on the River Nile and some other water surfaces in Egypt (Khallaf *et al.*, 1998; Al Yousuf *et al.*, 2000; Kraemer, 2005; Authman, 2008; Authman *et al.*, 2008; Bahnasawy *et al.*, 2011). The levels of heavy metals had seasonal differences in the living or dead organic and inorganic matter in the water column. The reason is due to human activity from industrial and urban activity, petroleum contamination and sewage disposal causing water pollution (Edwards *et al.*, 2001; Santos *et al.*, 2005). Generally, the increase in heavy metals concentrations in the Nile water including its branches, such as Ismailia Canal might be attributed to the direct inputs from different sources (industrial wastes and atmospheric inflow of dust containing car exhaust). In addition, the increase in density of boats and ships, which discharges its effluent directly to the Nile contains a high amount of Pb in both the dissolved and particular phases (Ibrahim & Tayel, 2005; Osman, *et al.*, 2009).

The present results showed that the metal concentrations in fish muscle of *Oreochromis niloticus* are closely associated with the metal content of water; heavy metals were detected in the following order: Fe > Zn > Cu > Pb > Cd. This may be attributed to the abundance of these metals in water. A remarkable relationship between heavy metals concentrations in aquatic organisms was also observed (Ibrahim *et al.*, 2000; Ibrahim & El-Naggar, 2006). The analysis of heavy metal levels in examined muscle tissue provides

information on potential risk to the fish themselves and the consumers of these fish. Pollution by heavy metals in aquatic ecosystem is growing at an alarming rate and has become an important worldwide problem (Malik et al., 2010).

Studies were carried out to determine the hematological and biochemical values related to sex, age, size, environmental and physiological conditions and comparative studies on fish blood parameters to determine the systematic relationship between some species. MCH and MCHC values increased in the blood of Nile tilapia collected from Abu Z'abaa City and Abbassa region, compared to Belbies City and El-Tal Al-Kapeir City. Adeyemo (2007) found a significant increase in MCH and MCHC values of the African catfish exposed to lead. The increase in MCH with a decrease in RBC, Hb and MCV values (observed here) was previously recorded in the study of Mukherjee and Shiha (1993) for the major carp exposed to cadmium. The perturbation in these blood indices may be attributed to a defense reaction against toxicity. Since hematological and blood biochemical parameters are influenced by a variety of environmental stressors, they have the potential to be used as biomarkers of the detected Nile River pollution. Their evaluation in fish has become an important means of understanding the toxicological impacts of exposure hazards (Borges et al., 2007; Sudova et al., 2009; Li et al., 2011).

In the present study, significant reductions in the values of RBCs, Hb, Hct and MCV were observed in the blood of the Nile tilapia and the African catfish collected from downstream sites (polluted sites), compared to those collected from less polluted upstream areas. The present results find supports from Summarwar (2012) who recorded remarkable reduction in the levels of RBCs, Hb, Hct and MCV in the blood of *Clarias batrachus* inhabiting polluted sites, compared to those caught from clean site. Numerous studies showed that the peripheral RBCs, Hb, Hct and MCV of fish displayed a decreased incidence after exposure to different pollutants under field and lab conditions (Zutshi et al., 2010; Mekkawy et al., 2011; Gupta & Chandra, 2013). The observed reduction in RBCs, Hb, Hct and MCV values may be attributed to one or more of the following factors: (i) Hem-dilution of blood due to damage and later bleeding in the gills. (ii) The destruction of mature RBCs and the inhibition of erythrocyte production. (iii) Hemolytic crisis that results in severe anemia in fish exposed to heavy metals (Desai & Parikh, 2012; Musa et al., 2013).

CONCLUSION

This study provides valuable information on heavy metals in water and fish muscles (edible tissues) in the four regions under study in this important branch of the Nile. It showed that the average concentration of iron in the Abbassa region is within the permissible limit, while that of Pb surpassed the permissible limit in all regions regarding the water samples. In addition, the concentration of heavy metals measured in the muscles of the fish studied was generally lower than the permissible levels issued by the World Health Organization. The present study revealed that haematological and biochemical parameters are recognized as an important tool for monitoring fish health and help in detecting any variation in water quality. The study indicated that there was an alteration in the blood of *O. niloticus* that could lead to dysfunction in this species. In addition, the results provided evidence that parameters can be a sensitive indicator of water pollution.

REFERENCES

- Adeyemo, O. K. (2007).** Hematological Profile of *Clarias gariepinus* (Burchell, 1822) Exposed to Lead. Turkish J. Fisheries and Aquatic Sci., 7: 163-169.
- Al-Rawi, S. M. (2005).** Contribution of man-made activities to the pollution of the Tigris within Mosul area/ Iraq. Inter. J. Environ. Res. & Public Health, 2(2): 245-250.
- Al-Yousuf, M. H.; El-Shahawi, M.S. and Al-Ghais, S.M. (2000).** Trace metals in liver, skin and muscle of *Lethrinus lentjan* fish species in relation to body length and sex. Sci. Tot. Environ., 256: 87-94.
- Authman, M. M. N. (2008).** *Oreochromis niloticus* as a biomonitor of heavy metal pollution with emphasis on potential risk and relation to some biological aspects. Global Veterenaria, 2 (3): 104-109.
- Authman, M.M.N.; Bayoumy, E.M. and Kenawy, A.M. (2008).** Heavy metal concentrations and liver histopathology of *Oreochromis niloticus* in relation to aquatic pollution. Global Veterenaria, 2(3): 110-116.
- Bahnasawy, M.; Khidr, A.A. and Dheina, N. (2011).** Assessment of heavy metal concentrations in water, plankton, and fish of Lake Manzala, Egypt. Turkish Journal of Zoology, 35 (2): 271-280.
- Begum, A.; Mustafa, A.I. ; Amin, N.; Chowdhury, T.R.; Quraishi, S.B. and Banu, N. (2013).** Levels of heavy metals in tissues of shingi fish (*Heteropneustes fossilis*) from Buriganga River, Bangladesh. Environ. Monit. Assess., 185: 5461-5469.
- Bitten-Court, L.J.G.; Kreutz, L.C.; De Souza, C.; Rodrigues, L.B. and Fioreze, I. (2003).** Hematological changes in jundia (*Rhamdies quelen*) after acute and chronic stress caused by usual aquacultural management, with emphasis on immune suppressive effects. Aquaculture, 237: 229-236.
- Borges, A. ; Scotti, L.V. ; Siqueira, D.R. ; Zanini, R. and Do Amaral, F. (2007).** Changes in hematological and serum biochemical values in jundia, *Rhamdia quelen* due to sublethal toxicity of cypermethrin. Chemosphere, 69: 920-926.
- Chen, Y.Q.; Edwards, J.J. ; Kridel, S.J. ; Thornburg, T. and Berquin, I. M. (2007).** Dietary fat gene interactions in cancer. Cancer. Metastasis. Rev., 26: 535-551.
- Dacie, J.V. and Lewis, S.M. (2001).** Practical Haematology. In: Lewis, S.M., Bain, B.J. and Bates, I. Eds., Practical Heamatology, 9th Edition, Churchill Livingstone, Harcourt Publishers Limited, London, PP 444-451.
- Desai, B. and Parikh, P. (2012).** Impact of curzate (fungicide) on hematological parameters of *Oreochromis mossambicus*. International J. Scientific Engineering Res., 3(7): 1-6.
- Din, J.N. ; Harding, S.A. ; Valerio, C.J. ; Sarma, J. ; Lyall, K. ; Riemersma, R.A. ; Newby, D. E. and Flapan, A.D. (2008).** Dietary intervention with oil rich fish reduces platelet monocyte aggregation in man. Atherosclerosis, 197(1):290-306.

- ECS (Egyptian Chemical Standards) (1994).** Protection of the Nile River and water stream from pollution, Ministry of Irrigation, Cairo, Egypt, Law No 4.
- Edwards, J. W. ; Edyvane, K .S. ; Boxall, V.A. ; Hamann, M. and Soole K. L . (2001).** Metal levels in seston and marine fish flesh near industrial and metropolitan centers in South Australia. *Marine Pollution Bulletin*, 42: 389-396.
- EOS (1993).** Egyptian Organization for Standardization. Maximum levels for heavy metal concentrations in food. ES 2360-1993, UDC:546.19:815, Egypt.
- Gabriel, O. M. ; Rita, O. ; Clifford, A. and Kennedy, O. (2006).** Heavy metal pollution of fish of Qua. Iboe River Estuary: Possible implications for neurotoxicity. *The internet J. of Toxicol.*, 3(1): 1-6.
- Geriesh, H.M. ; Balke, Klaus, D and ElRayes, A. E. (2008).** Problems of drinking water treatment along Ismailia Canal Province, Egypt. *J. Zhejiang University Sci.*, 9 (3): 232–242.
- Gupta, A. and Chandra, R. (2013).** Impact of effluent of River Ramganga on the hematology of fresh water fish (*Heteropneustes fossilis*). *International J. Environmental Sci.*, 4: 19-22.
- Heier, L. S.; Teien, H. C.; Oughton, D.; Tollefsen, K.E. and Olsvik, P.A.(2013).** Sublethal effects in Atlantic salmon (*Salmo salar*) exposed to mixtures of copper, aluminium and gamma radiation. *J Environment Radioactivity*, 121: 33-42.
- Ibrahim S. and Tayel, I. (2005).** “Effect of heavy metals on gills of *Tilapia zilli* inhabiting the River Nile water (Damietta Branch and El-Rahaway Drain),” *Egypt Journal of Aquatic Biology & Fish.*, 9(2): 111- 128.
- Ibrahim, A. M., M. H. Bahnasawy, S. E. Mansy and R.I. El-Fayomy. (2000).** On some heavy metal levels in water, sediment and marine organisms from the Mediterranean coast of Lake Manzalah. *Egypt. J. Aqua. Biol. & Fish.*, 4 (4): 61-81.
- Ibrahim, H.; Ibrahim, M. and Samhan, F. (2009).** Distribution and bacterial bioavailability of selected metals in sediments of Ismailia Canal, Egypt. *J. Hazardous Materials*, 168: 1012-1016.
- Ibrahim, N. A. and G. O. El-Naggar. (2006).** Assessment of heavy metals levels in water, sediment and fish at cage fish culture at Damietta Branch of the river Nile. *J. Egypt. Acad. Environ. Develop.*, 7 (1): 93-1114.
- Ikem, A.; Egiebor, N. and Nyavor, K. (2003).** Trace elements in water, fish and sediment from Tuskegee Lake, Southeastern USA. *Water Air Soil Pollut.*, 147: 79–107.
- Kalay, M. and Canli, M. (2000).** Elimination of essential (Cu and Zn) and non-essential (Cd and Pb) metals from tissues of a fresh water fish, *Tilapia zillii*. *Trop. J. Zool.*, 24: 429-436.
- Khallaf, E.A. ; Galal, M. and Authman, M. (1998).** Assessment of heavy metals pollution and their effects on *Oreochromis niloticus* in aquatic drainage water. *Journal of the Egyptian German Society of Zoology*, 26 (B): 39–74.

- Kraemer, L.D.; Campbell, P. G. and Hare, L. (2005).** Dynamics of Cd, Cu and Zn accumulation in organs and sub-cellular fractions in field transplanted juvenile yellow perch (*Perca flavescens*). Environ. Pollut., 138: 324–337.
- Li, Z.H.; Velisek, J.; Grabic, R.; Li, P. and Kolarova, J. (2011).** Use of hematological and plasma biochemical parameters to assess the chronic effects of a fungicide propiconazole on a freshwater teleost. Chemosphere, 83: 572-578.
- Malik, N.; Biswas, A.K.; Qureshi, T.A.; Borana, K. and Virha, R. (2010).** Bioaccumulation of heavy metals in fish tissues of a freshwater lake of Bhopal. Environmental Monitoring and Assessment, 160: 267-276.
- Marcovecchio, J. E. (2004).** The use of *Micropogonias furnieri* and *Mugil liza* as bioindicators of heavy metals pollution in La Plata River estuary, Argentina. Sci. Tot. Environ., 323: 219–226.
- Mekkwaw, I.A.; Mahmoud, U.M. and Sayed, A.E.D.H. (2011).** Effects of 4-nonylphenol on blood cells of the African catfish *Clarias gariepinus* (Burchell, 1822). Tissue and Cell, 43: 223-229.
- Mohammed, A. S. (2008).** Microbiological studies on Ismailia Canal from River Nile, Egypt. M.Sc. Thesis, Fac. Sci. Al-Azhar Univ., Cairo, Egypt.
- Mukherjee, J.R. and Shiha, G.M. (1993).** Cadmium toxicity on hematological and biochemical aspects in an Indian freshwater major carp, *Labeo rohita* (Hamilton). J Freshwater Biol., 5: 245-251.
- Musa, S.; Aura, C.; Ogello, E.; Omondi, R. and Charo-Karisa, H., (2013).** Hematological response of African catfish, *Clarias gariepinus*, (Burchell 1822) fingerlings exposed to different concentrations of tobacco (*Nicotiana tabacum*) leaf dust. Zoology, 2013:1-7.
- Osman, A.G.; Abouel Fadl, K. Y.; Abd El Baset, M.; Mahmoud, U.M.; Kloas, W. and Moustafa, M.A. (2018).** Blood biomarkers in Nile tilapia *Oreochromis niloticus* and African catfish *Clarias gariepinus* to evaluate water quality of the River Nile. Journal of Fisheries Sciences, 12(1): 1-15.
- Osman, H.A.M.; Ibrahim, T.B.; Ali, A.T. and Derwa, H.I.M. (2009).** Field application of humic acid against the effect of cadmium pollution on cultured tilapia *Oreochromis niloticus*. World Applied Sci. J., 6: 1569-1575.
- Rajeshkumar, S. and Munuswamy, N. (2011).** Impact of metals on histopathology and expression of HSP 70 in different tissues of Milk fish (*Chanos chanos*) of Kaattuppalli Island, South East Coast, India. Chemosphere, 83: 415-421.
- Ralph, S. and Ramadan, A.B. (2008).** Environmental studies on water quality of the Ismailia Canal, Egypt. Wissenschaftliche Berichte Fzka, 7427:1-47.
- Santos, I.R.; Silva-Filho, E.V.; Schaefer, C.E.; Albuquerque-Filho, M.R. and Campos, L.S. (2005).** Heavy metal contamination in coastal sediments and soils near the Brazilian Antarctic Station, King George Island. Marine Pollution Bulletin, 50: 85-194.

-
- Sayed, A.H.; Mekkawy, I.A. and Mahmoud, U.M. (2011).** Effects of 4-nonylphenol on metabolic enzymes, some ions and biochemical blood parameters of the African catfish *Clarias gariepinus* (Burchell, 1822) African J. Biochem. Res., 5: 287-297.
- Seriani, R.; Abessa, D.M.S.; Kirschbaum, A.A. ; Pereira, C.D.S. and Romano, P. (2011).** Relationship between water toxicity and hematological changes on *Oreochromis niloticus*. Brazilian J. Aquatic Sci. Technol., 15(2): 47-53.
- Singh, R.K.; Chavan, S.L. and Sapkale, P.H. (2007).** Heavy metal concentrations in water, sediments and body tissues of red worm (*Tubifex* spp.) collected from natural habitats habitats in Mumbai, India', Environmental Monitoring and Assessment, Vol. 129, pp.471–481.
- Sudova, E.; Piackova, V.; Kroupova, H.; Pijacek, M. and Svobodova, Z. (2009).** The effect of praziquantel applied per os on selected hematological and biochemical indices in common carp (*Cyprinus carpio* L.). Fish Physiol. Biochem., 35: 599–605.
- Summarwar, S. (2012).** Comparative hematological studies of *Clarias batrachus* in Bisalpur Reservoir and Pushkar Lake. Indian J. Fundamental Applied Life Sci., 2: 230-233.
- World Health Organization (WHO) (1999).** Summary and Conclusions. Proceedings of the 53rd Meeting of the Joint FAO/WHO Expert Committee on Food Additives, Rome, 1-10 June 1999
- Zutshi, B.; Prasad, S.G.R. and Nagaraja, R. (2010).** Alteration in hematology of *Labeo rohita* under stress of pollution from Lakes of Bangalore, Karnataka, India. Environ Monit Assess, 168:11–19.