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Impact of water quality on biochemical and physiological parameters of *Oreochromis Niloticus* in different aquatic habitats

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ABSTRACT

Background: El-Wadi and El-Bats drainages considered the main reservoirs for agricultural and domestic non-treated drainage waters at El-Fayoum province. **Aims**: in the present study is to provide comparable data between El-Wadi, El-Bats drainages and Hawara channel (a branch of the Nile River) with using Nile tilapia (Oreochromis niloticus), as indicator of natural and anthropogenic impacts on aquatic ecosystem. **Material and method**: Heavy metals evaluated in our work represented in (Fe, Zn, Cd, Cu, and Mn). Moreover, the effect of pollutants and degraded water quality on the biochemical parameters which represented in evaluation of liver functions, kidney functions, protein profile, and lipid profile. . **Results** : The results showed that The lowest mean value of dissolved oxygen was observed in El-Bats drainage (5.5 mg/l), followed by El-Wadi drainage (5.93 mg/l), this could be attributed to agricultural, industrial, and waste municipal effluents which characterized by high load of organic wastes and the microbial activity that degraded the organic matter led to the oxygen consumption. While the highest mean value of dissolved oxygen (7.98 mg/l) was recorded in Hawara channel (a branch of Nile River). Also, mean value of Cd recorded the lowest concentrations between five heavy metals, and slightly exceeded permissible limit (0.01 ppm), while Zn recorded the highest concentrations in Hawara channel. However, in El-Bats and El-Wadi Drains, Cd showed the lowest concentrations between metals and exceeded permissible limit (0.01 ppm) and Fe showed the highest concentrations that exceeded permissible limit (1 ppm) in great concern according to WHO (1993). The results showed also significant increase of serum Ast, Alt, ALP, LDH activities, and the level of Urea, Uric acid, Creatinine, lipid profile parameters (Triglycerides, Total lipids, and Cholesterol), and protein profile parameters (Total protein, Albumin, and Globulin) concentration in Oreochromis niloticus collected from El-Bats and El-Wadi Drains compared with that collected from Hawara channel. From the present results, it can be concluded that deteriorated water quality represented in El-Wadi and El-Bats drainages by highly

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recorded heavy metals (Fe, Zn, Cd, Cu, and Mn) concentration affect negatively on biochemical parameters of Oreochromis niloticus and vice versa for Oreochromis niloticus collected from Hawara channel (a branch of the Nile River).

INTRODUCTION

The aquatic environment makes up the major part of our environment and resources. Therefore, its safety is directly related to human health **[1].** Pollution, loss of biodiversity, and habitat destruction are probably the main environmental threats for aquatic ecosystems. Moreover, the excessive contamination of aquatic ecosystems has evoked major environmental and health concerns worldwide **[2, 3]** as the aquatic environment is the ultimate recipient of pollutants produced by the natural influences and human activities **[4].**

The agriculture, industrial, and municipal effluents at El-Fayoum province represented mainly in two drainages, El-Wadi and El-Bats. El-Wadi drainage canal only passes about 226.3 million cubic meters which indicates its importance as one of the main drainage canals of this discharge system **[5]** .In addition, EL-Bats drainage canal passes about 100.84 million cubic meters which indicates its importance as the second drainage in our focus of this discharge system.

Water quality of the aquatic ecosystem is considered as the main factor controlling the state of health and disease [6,7] .Among the various toxic pollutants, heavy metals represent a very interesting group of elements due to their strong impact on stability of aquatic ecosystems, bioaccumulation in living organisms [8] , toxicity persistence and tendency to accumulate in water, sediments and fish organs [9] .

From the point of view of the biological importance of the trace heavy metals, all heavy metals are potentially harmful to most organisms at some level of exposure and absorption, although some elements have an essential function in the body of the organism, and have very important biological role as micronutrients in human diet [10] . At certain concentration above which it becomes very harmful for the organism and are highly toxic for aquatic organisms and their consumers including human when exceeding the recommended safety levels [11,12] .

Besides that, other elements are considered to have no biological function (non-essential) and are highly toxic at very low concentrations, e.g. mercury, lead, and cadmium, moreover, chronic exposure of fish to sub-lethal trace metal levels reduced swimming speed and growth. For the normal metabolism of fish, the essential metals must be taken up from water, food or sediment and suspended particulate material. However, similar to the route of essential metals, non-essential ones are also taken up by fish and accumulate in their tissues [13, 14, 15, 16, 17, 18].

Heavy metals contaminants of fish pose serious risks to biochemical parameters and enzymes activities. Blood parameters are considered biomonitors and therefore are important in diagnosing structural and functional status of fish exposed to contaminants [19].

Use of blood parameters provide an early warning of potentially damaging changes in stressed fish [20].

The effects of exposure to pollutants on fish physiology [21] have significant alterations in the plasma total proteins, albumin, globulin, total lipids, triglycerides, and Cholesterol, Urea, creatinine and uric acid, as well as, AST, ALT, ALP and LDH activities.

2. *Materials and Methods*.

The present field study was carried out on water, sediment and the Nile tilapia ; *Oreochromis niloticus* samples collected from the two main drains at El-Fayoum province, El-Wadi and El-Bats drains, in addition to Hawara channel, a branch of the Nile River.

Description of sampling Sites of collection:

Water, sediment and *Oreochromis niloticus* fish samples were collected from three sites at El-Fayoum province.

Site 1: Hawara channel, a branch of the Nile River.
Site 2: El-Wadi drainage canal, where agricultural, industrial and waste municipal effluents of El-Fayoum province discharged.
Site 3: El-Bats drainage canal, where agricultural and waste municipal effluents of El-Fayoum province discharged.

Sampling:

- Water, sediments and fish samples were collected at summer season of 2016 from each site for the following investigations.

I . Water sampling and analysis:

Water samples were collected approximately 20 cm below the surface [22] and kept in 500 ml sterile plastic containers. Samples were filtered in the field and acidified with 10% HNO3 for preservation, placed in an ice bath and brought to the laboratory.

A) Physicochemical analysis of water:

The water samples collected from different locations in the studied ecosystems were subjected to a number of physicochemical analyses as mentioned below :-

● **the pH** was measured by means of a pocket-pH meter (Micro Checkit ® pH+, Lovibond, England).
● **Dissolved oxygen (mg/l)** concentration was determined at the sampling site by means of Oxygen meter (model, YSI58).
●**Total hardness and total alkalinity (mg/l)** were measured by titration method according to the American Public Health Association standard methods [23].
● **Ammonia and nitrite (mg/l)** were measured according to the method described by the American Public Health Association standard methods [23] .

B). water analysis for heavy metals:

Heavy metal concentrations in water were determined by atomic absorption spectrophotometer (Perkin Elmer, 2280). The samples were prepared and analyzed sequentialy for zinc, copper, iron, cadmium, and manganese [23] . To 50 ml of water sample (in 500 ml Taylor flask), 0.50 ml of concentrated sulphuric acid was added, This was boiled down to obtain white fumes, cooled and 1.0 ml of 60% $HClO_3$ and 5.0 ml of concentrated HNO_3 were added, The resulting mixture was digested until a clear digest was obtained. This was cooled, filtered (No. 44 Whatman paper) into 500 ml volumetric flask, diluted to volume and mixed.
Heavy metal concentration (ppm) = reading of atomic absorption ×volume of diluted solution divided by Volume of water sample.

II. Sediment sampling and analysis for heavy metals:

Surface sediment samples from each of the three studied sites were collected and kept in plastic bags then placed in a cooler and transported to the laboratory. Sediment samples were dried at 105^o C (degrees Celsius) till constant weigh, and then disaggregated using a mortar and pestle. The material was then sieved through a 63-micron nylon mesh (the best sieve for heavy metals evaluation). Sample for metal analysis was prepared by treating 1g sample with 10 ml of concentrated nitric acid and 5 ml of 60% perchloric acid in 100 ml Kjeldahl flask. The mixture was heated with moderate heat using a hot plate for about 15 min until white fumes appear. The digest was cooled, then filtered (No. 44 whatman paper) into 50 ml volumetric flask with rinsing in de-ionized water and made up to mark with de-ionized water [24] .
Heavy metal concentration (µg/g) = reading of atomic absorption X volume of diluted solution divided by weight of sample (gm).

III. Fish sampling and analysis:

A total number of 90 adult fish of Nile tilapia; *Oreochromis niloticus* (30 fish/site), fishes were collected with the help of local fishermen.

A). Residual heavy metals in some selected organs of the studied Fishes:

Thirty fish from *Oreochromis niloticus* were collected at summer season of 2016 from each site for residual heavy metal analysis. The collected fish were dissected for its gills, liver, muscle, kidney and gonads tissues then , washed with de-ionized water, put in cleaned plastic bags and stored frozen until analysis was carried out. One gram of the prepared tissue sample (wet weight) was subjected to digestion by adding 10 ml of freshly prepared 1:1 concentrated HNO_3 : $HClO_3$ in beaker, covered with a watch glass till initial reaction subsided in about 1 hour and gently heated at 160 ºC in a sand bath on a hot plate till reduction of volume to 2-5 ml. The digests were allowed to cool and transferred to 25 ml volumetric flasks and made up to mark with de-ionized water. The digests were kept in plastic bottles and latter heavy metal concentrations were determined using an atomic absorption spectrophotometer.

Heavy metal concentration (μg/g) = reading of atomic absorption X volume of diluted solution divided by Weight of sample (g).

B). Blood sampling and biochemical analysis:

● **Blood sampling:** The fishes for blood analysis were brought to the laboratory alive in an aerated tank with a sandy bottom, in order to reduce capture stress. Blood samples were withdrawn from the arterial caudalies. The needle (heparinized glass pipette) was run quite deep through the middle line just behind the anal fin in a dorso-cranial direction. Moreover, serum was obtained by centrifugation of other blood sample. (At 5000 rpm for 12 minutes) and stored at -20 ºC for further analysis. The blood sample subjected to the following examinations.

● **Biochemical analysis (Serum analysis):**

●**Serum aminotransferase:** Serum Aspartate aminotransferase (AST) and Alanine aminotransferase (ALT) activities were estimated colorimetrically [25] .

● **ALP activity**[26] .

● **LDH activity** [27] .

●**Serum creatinine** [28].

● **Serum Uric acid and urea**[29] .

● **Serum total protein:** Serum total protein content was determined by Biuret test [30]

●**Serum albumin** [31] .

● **Serum globulin:** serum globulin was measured as the difference between plasma total protein and albumin.

●**Serum total lipid:** Serum total lipid concentration was determined colorimetrically by sulphovanillin reaction [32] .

● **Serum total cholesterol and triglycerides:** serum total cholesterol and triglycerides were determined colorimetrically [33] .

III). Statistical analysis: This study was carried out in summer season of 2016 and the results of the present study were statistically analyzed using analysis of variance (F-test) and Duncan's multiple range test to determine difference in means (among the three studied sites of collection for all studied parameters) using Statistical Analyses System, version 6.2 (SAS, 2000).

3. Results:

I . Water sampling and analysis:

Physicochemical analysis of water:

The quality indices of water collected from the three different studied sites of collection of Hawara channel, El-Wadi and El-Bats drainage canals are illustrated in table (1). The analysis of variance (F-test) showed significant differences in the values of pH, dissolved oxygen, total hardness, total alkalinity, ammonia and nitrite among the different studied sites (F-values = 123.71, 50.5, 62.79, 445.95, 104.43 and 132.67 respectively).

A).Analysis of heavy metals in water, sediment, and fish organs (residual heavy metals):

It is markedly observed that there were highly significant differences in (F-Value) for all heavy metal concentrations among water samples, sediment samples, and finally different organs; Gills, Liver, Muscles, Kidney, and Gonads for *O. niloticus* of studied sites of collection as illustrated in table (2), table (3), table (4), table (5), table (6), table (7), and table (8).

B). Biochemical analysis (Serum analysis):

It is markedly observed that there were highly significant differences in (F-Value) for all Serum constituents for *O. niloticus* of the three studied sites of collection as illustrated in table (9), and table (10).

4. Discussion

Water quality of the aquatic ecosystem is considered the main factor controlling the state of health and disease in both cultured and wild fishes. On the other hand, water quality state of a water body depends on a large number of physical, chemical, and biological indicators [34] . Degradation of the aquatic environment through industrial, waste municipal and agricultural effluents is a significant concern about the health of fish [35]. Fish could serve as biological indicators for pollutants and environmental degradation. Bioaccumulation of trace heavy metals can be harmful to aquatic organisms causing adverse effects for the aquatic [1] .

Deterioration of the natural water resources conditions by the action of effluents affect the quality and quantity of the fish [36] . So, the present study was conducted to provide data about the impact of water quality on biochemical and physiological parameters of *Oreochromis niloticus* along El-Wadi and El-Bats drainages that considered as reservoirs of the drainage water in El-Fayoum province, and Hawara channel that considered as a branch of the Nile River.

Water quality:

Water contamination is one of the most principal environmental and public health problems, which represented in our study in El-Wadi, and El-Bats drainages that act as reservoirs for agricultural, industrial and waste municipal effluents of El-Fayoum province.

The high concentrations of dissolved oxygen are very vital and important for aquatic organisms as it is required for the metabolism of aerobic organisms and organic matter decomposition [37] . The lowest mean value of dissolved oxygen was observed in El-Bats drainage (5.5 mg/l), followed by El-Wadi drainage (5.93 mg/l), this could be attributed to agricultural, industrial, and waste municipal effluents which characterized by high load of organic wastes and the microbial activity that degraded the organic matter led to the oxygen consumption. While the highest mean value of dissolved oxygen (7.98 mg/l) was recorded in Hawara channel (a branch of the Nile River).

The present data also showed high values of water ammonia and nitrite in water samples collected from El-Wadi and El-Bats drainages and the lowest values were recorded at Hawara channel. The total amount of effluents discharged from various urban activities must not be neglected. Heavy metal pollution in water is generally associated with agricultural, industrial, and municipal effluents [38] . Once metals are in the water column, they may be taken up by living organisms, deposited in the sediments or remain for some period in the water itself.

Heavy metals are widely distributed in aquatic system due to industrial development and the wide use of chemicals in agriculture. They may led to either an increase or decrease in growth of various fishes and within their permissible level, play an important role in biological systems [39] .

Heavy metals considered a class of pollutants with critically ecological concern. This is because they are not biodegradable and thus persist for a long time in aquatic environment. Their environmental concern is due to their build up along the food chain, which could led to bioaccumulation and biomagnification [40] .

Comparing the mean concentrations of heavy metals (Fe, Zn, Cd, Cu, and Mn) in water from the different studied locations, the results revealed the presence of Iron, Zinc, Cadmium and Manganese in the following order:

- **Iron:** El-Bats> El-Wadi>Hawara.
- **Zinc:** El-Bats > El-Wadi>Hawara.
- **Cadmium:** El-Wadi> El-Bats >Hawara**.**
- **Copper:** El-Wadi> El-Bats >Hawara.
- **Manganese:** El-Bats > El-Wadi>Hawara.

The results showed that Fe recorded the highest concentrations in El-Wadi and El-Bats drainages, which could be attributed to Fe liberation from sediments as sulphides [41] . Where Cd recorded the lowest concentrations however, the two heavy metals out of permissible limits (0.3 and 0.01 mg/l respectively) [42] .

The distribution of metals in sediments adjacent to settlement areas can provide researchers with evidence of the anthropogenic impact on ecosystems and, therefore, aid in assessing the risks associated with discharged human wastes. In aquatic ecosystems research, the role of the sediments in the cycling of chemical elements has often been underestimated and the exchange of elements (especially nutrients) between sediments and water is still a crucial topic [43] .

Comparing the average concentrations of heavy metals (Fe, Zn, Cd, Cu and Mn) in sediment samples collected from the three different sites. The results revealed the presence of these metals in the following order:

- **Iron:** El-Bats > El-Wadi>Hawara.
- **Zinc:** El-Bats > El-Wadi>Hawara.

- **Cadmium:** El-Bats > El-Wadi>Hawara.
- **Copper:** El-Bats > El-Wadi>Hawara.
- **Manganese:** El-Wadi> El-Bats >Hawara.

The present study revealed that metals are enriched in sediments as well as in fish, relative their levels in water. This indicates that relative to water, metals are adsorbed on sediment and bioaccumulated in fish. The deposition rate in sediment depends on metal concentrations in water and surface sediments [43, 44].

Residual Heavy Metals

In ecosystems, heavy metals such as Iron, zinc, cadmium, copper, and Manganese and others have received considerable attention due to their toxicity, long-term persistence, bioaccumulation, and biomagnification at various trophic levels [45]. When discharged into the water can enter the food chain, bioaccumulated in fish tissues and hence becomes a threat to man [43]. In the present study, bioaccumulation of metals in tissues varies from metal to metal and among different organs of the same organism [46]. Gills are in direct contact with the aquatic medium; therefore, metal concentrations in this organ reflect their concentrations in the external environment. In contrast, the concentrations in liver and kidney represent the rates of bioaccumulation and detoxification of pollutants, as a result concentrations of all studied trace heavy metals scored highly accumulation pattern, especially in these two organs. Also, in the present study, showed the lowest bioaccumulated heavy metals in muscles may be correlated with the fat-content in muscle tissues, low fat affinity to combine with heavy metals, and/or low metabolic activity of muscle [47]. Fe is an abundant and important element. The increase of Fe accumulation in the studied fish *(Oreochromis niloticus*) tissues along El-wadi, El-Bats drainages and Hawara channel was higher than the other metals possibly due to the increase of total dissolved Fe and consequently the free metal Fe concentration in El-Wadi, El-Bats, and Hawara channel, leading to an increase in metal uptake by different organs [48]. In the present study, heavy metals concentrations were higher in the gill than in the muscle tissue of the studied fish (*Oreochromis niloticus*). Metal concentration in the gill could be due to the element complexion with the mucus that is impossible to completely remove from the lamellae before tissue is prepared for analysis [49].

The low levels of the metals in the muscles may be due to the little blood supply to the muscular [50]. Our results revealed that the Mn is highly accumulated in gills than other organs of (*Oreochromis niloticus*) collected from El-Wadi and El-Bats drainages. These results [51].

Indicative biochemical Parameters

Total Serum proteins play an important role in the metabolism and regulation of water balance. The normal range of Serum total protein concentration in fish = 4.08±0.61g/dl [52]. So any disturbances make this reading up or down more useful in diagnosis of fish disease. The majority of plasma proteins which are synthesized in the liver are used as an indicator of liver impairment [53]. In the present study, there was elevation in plasma total protein (hyperproteinaemia), albumin and globulin in the studied fish (*Oreochromis niloticus*) from El-Wadi and El-Bats drainages when compared with fish collected from Hawara channel which were within range. This possibly due to activation of metabolic systems in response to pollutants exposure, degradation of the cellular material in the liver, several pathological conditions as damage of liver and kidney, as shown in the present study and relative changes in the mobilization of blood proteins, water loss in the plasma and/or induction of protein synthesis in liver [54]. The present findings are in agreement with previous reports of increased level of Serum proteins on exposure to heavy metals [55].

During chronic period of stress caused by heavy metals toxicity, Serum proteins are considered a source of energy: During stress condition, fish need more energy to detoxify the toxicant and to overcome stress. Since fish have fewer amount of carbohydrate, the next alternative source of energy is protein to meet the increased energy demand. The depletion of protein may have been due to their degradation and possible utilization of degraded products for metabolic purposes. [56] agree with this study observed that decrease in the protein content as observed in most of the fish tissues may be due to metabolic utilization of the ketoacids to gluconeogenesis pathway for the synthesis of glucose, or due to directing the free amino acids for the synthesis of proteins, or for the maintenance of osmo and ionic regulation. It could also be due to the production of heat shock proteins or destructive free radicals or could be a part of heavy metal induced apoptosis.

The level of total lipids, triglycerides and cholesterol in Serum and tissues of fish have been reported to be moderately sensitive to environmental pollutants but changes in these parameters seems to be attributed to many factors, such as the types of contaminant, the concentration, mode of its action, duration of exposure and fish species [49] . The increased value of Serum total lipids, triglycerides and total cholesterol indicated retardation of fat metabolism. The increase of total Serum lipids and cholesterol observed in the present study represented in El-Wadi and El-Bats drainages where agricultural, industrial and municipal effluents discharged from El-fayoum province may be due to one or more of the following reasons: release of cholesterol and other lipids constituents from damaged cell membranes, decrease hepatic excretion of cholesterol, increased production by the liver and other tissues by the effect of the pollutants (heavy metals), decreased activity of cytochrome P450 enzymes, thyroid dysfunction and finally blocked conversion of cholesterol to sex steroids as a result of gonad dysfunction [57, 58, 59] .

It is stated that the concentrations of cholesterol, an essential structural component of membranes , may increase due to liver and kidney failure causing the release of cholesterol into the blood [60] . This is repeated] in *Oreochromis niloticus* exposed to copper and zinc [59] .

It is evident that rise in Serum triglycerides represented in hypoactivity of lipoprotein lipase in blood vessels which breaks up triglycerides [59] .

Indicative Parameters of Liver Functions

Enzymes, such as aminotransferase (GOT and GPT) are important enzymes known to play a key role in mobilizing L-amino acids for gluconeogenesis and function as links between carbohydrate and protein metabolism during the fluctuating energy demands of the organism in various adaptive situations [61] . Alkaline phosphatase (ALP) distributed in tissue especially within cell membranes and would easily leak out of the cell in pollutants-induced tissue damage. LDH, the terminal enzyme in vertebrate anaerobic glycolysis, is one of the enzymes that have been employed for diagnosing liver, muscle and gill damages caused by pollutants in fish. Increased activity of LDH is a characteristic feature of a shift from aerobic to anaerobic metabolism leading to an elevated rate of pyruvate conversion into lactate, resulting in lactic acidosis [61] . In our study elevations in the activities of Serum GOT, GPT, ALP and LDH in *Oreochromis niloticus* From El-Wadi and El-Bats drainages reflect hepatic and myocardial impairment, leading to extensive liberation of the enzymes into the blood circulation. The elevation of serum GOT; GPT and ALP may be due to liver dysfunction [54] . In addition it is reported that the increase of serum GOT; GPT and ALP may be attributed to the hepatocellular damage or cellular degradation, perhaps in liver, heart or muscle [50] . There is another point of view most be taken in consideration discussed by (Yang and Chen, 2003) and reported that the increase of blood enzymatic activity may be due to increased enzymes synthesis. Increased serum activities of ALP have been explained by pathological processes such as liver impairment, kidney dysfunction and bone disease [53] . In the same aspect Serum GOT, GPT, ALP and LDH activities significantly increased in *Oreochromis niloticus* exposed to two metals (copper, lead) due to the release of enzymes from the destroyed tissues into the blood circulation [62] .Several histopathological alterations have been observed in the liver, kidney and muscle of the studied fish (*Oreochromis niloticus*) which support the observed increase in Serum GOT, GPT, ALP and LDH activities.

Indicative Parameters of Kidney Functions

It is well known that the renal failure is usually associated with decrease in urea, uric acid and creatinine excretion, thus leading to its increase in Serum. As a result, Serum urea, uric acid and creatinine were useful in diagnosis of renal function impairment, renal tubular necrosis, renal insufficient, impaired nitrogen metabolism, as well as muscle tissue damage [63] . In the present study, Serum urea, uric acid and creatinine showed highly significant increase in *Oreochromis niloticus* collected from El-Wadi and El-Bats drainages when compared to Hawara channel. This result supported that the environmental pollution caused by agricultural, industrial and municipal waste effluents of El-Fayoum province discharged in these drainages exerts harmful effects on the kidney tissues causing kidney dysfunction. It is recommended that the increased blood urea could occur at times of liver diseases, impaired kidney function and cardiac arrest [64] . Similar increase in Serum urea, uric acid and creatinine [65] .Urea in fish is synthesized by the liver and excreted primarily by the gills rather more the kidney. So in the present study the elevation of urea level may be attributed to gill dysfunction [66] . Other point of view concluded that kidney damage may result in reduced renal blood flow with reduction in glomerular filtration rate, resulting in azotemia characterized by increase in blood urea nitrogen, uric acid and creatinine [67] . The increased blood urea could occur at times of impaired kidney function, liver diseases and cardiac arrest [64] . Similar increase in Serum urea, uric acid and creatinine [65] . in the Nile Tilapia subjected to mercuric chloride. In the present study, several histopathological alterations have been observed in the liver, kidney and muscle of the studied fish (*Oreochromis niloticus*) which support the observed increase in Serum urea, uric acid and creatinine level.

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Table (1): (Means ± SD) of Physicochemical parameters of the three different studied sites

Studied sites of collection	PH	Dissolved Oxygen	Total Hardness	Total Alkalinity	NH3	NO2
Hawara channel (Branch of the Nile river)	7.48±0.16 **C**	7.98±0.124 A	85.33±11.67 B	106.33±11.5 B	0.153±0.02 B	0.11±.017 C
El-Wadi drainage canal	8.23±.05 **B**	5.93±0.22 B	576.7±66.6 A	350.7±10.5 A	0.68±.09 A	0.76±.09 A
El-Bats drainage canal	8.36±.08 **A**	**5.5±0.5 B**	**576±83.4 A**	**373±14.11 A**	**0.603±.08 A**	**0.636±.08 B**
***P*-values**	123.7**	50.5**	62.79**	445.95**	104.4**	132.7**

Data are represented as **means** of six samples **± S.D.** Means within the same column, with the same letter for each parameter are not significantly different, otherwise they do (SAS, 2000). ****** Highly significant difference at **P ≤ 0.01**.

Table (2): (Means ± SD) of heavy metals (Fe, Zn, Cd, Cu, and Mn) in water of the three different studied sites.

Studied sites of collection	Fe P.L 0.3 ppm	Zn P.L 5 ppm	Cd P.L 0.01 ppm	Cu P.L 1 ppm	Mn P.L 0.5 ppm
Hawara channel (Branch of the river Nile)	1.17±0.39 **C**	**15.5±0.97 C**	**0.216±0.036 C**	**1±0.128 C**	**0.286±0.135 C**
El-Wadi drainage canal	154.2±4.5 **B**	36.7±1.25 **B**	3.4±0.16 **A**	43.2±1.25 **A**	27.36±1.48 **B**
El-Bats drainage canal	185.4±10.5 **A**	44.3±2.6 A	3.05±0.098 B	37.36±1.66 B	29.5±1.35 A
F-values	1354.4**	425.86**	1489**	2161.1**	1182.5**

Data are represented as **means** of six samples **± S.D**. **P.l.** = Permissible level in water according to the Egyptian Organization for Standardization. Means within the same column, with the same letter for each parameter are not significantly different, otherwise they do (SAS, 2000). ****** Highly significant difference at **P ≤ 0.01**.

Table (3): (Means ± SD) of heavy metals (Fe, Zn, Cd, Cu, and Mn) in sediment of the three different studied sites.

Studied sites of collection	Fe ppm	Zn ppm	Cd ppm	Cu ppm	Mn ppm
Hawara channel (Branch of the river Nile)	**978.7±170.4 B**	**23.26±2.6 C**	**4.79±0.376 C**	**22.9±5.81 B**	**280±32.25 C**
El-Wadi drainage canal	3190±304.5 **A**	**65.9±5.56 B**	15.77±0.49 B	45.57±2.19 A	509±10.84 A

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El-Bats drainage canal	3370±383.6 A	88.6±3.67 A	16.56±0.239 A	48.37±2.01 A	471.33±10.5 B
F-values	118.72**	388.35**	1767.12**	82.27**	214.13**

Data are represented as **means** of six samples **± S.D.** Means within the same column, with the same letter for each parameter are not significantly different, otherwise they do (SAS, 2000). ** Highly significant difference at **P ≤ 0.01**.

Table (4): (Means ± SD) of Fe residual heavy metal concentration in organs (Gills, Liver, Kidney, Muscles and Gonads) of *Oreochromis niloticus* for the three different studied sites.

Studied sites of collection	Gills	Liver	Muscles	Kidney	Gonads
Hawara channel (Branch of the river Nile)	6.33±1.39 **C**	11.27±1.25 C	8.63±0.78 C	10.47±1.09 C	7.5±0.83 C
El-Wadi drainage canal	116±3.58 B	311.33±6.28 A	34.37±.66 B	126.3±2.25 B	64.7±3.68 A
El-Bats drainage canal	127±4.01 A	301.67±3.6 B	38.27±1.7 A	133.3±2.7 A	57.8±1.6 B
F-values	2541.8**	9672.5**	1185.6**	6243.5**	1041.9**

Data are represented as **means** of six samples **± S.D.** Means within the same column, with the same letter for each parameter are not significantly different, otherwise they do (SAS, 2000). ** Highly significant difference at **P ≤ 0.01**.

Table (5): (Means ± SD) of Zn residual heavy metal concentration in organs (Gills, Liver, Kidney, Muscles and Gonads) of *Oreochromis niloticus* for the three different studied sites.

Studied sites of collection	Gills	Liver	Muscles	Kidney	Gonads
Hawara channel (Branch of the river Nile)	11.73±1.7 **C**	13.63±2 B	12.47±.81 C	8.03±1.6 C	5.36±1.18 C
El-Wadi drainage canal	47.33±1.25 B	45.93±9.33 A	37.1±1.49 B	38.03±3.04 B	16.5±1.99 B
El-Bats drainage canal	52.37±2.42 A	47.63±8.37 A	41.1±1.67 A	46.73±1.19 A	21.2±2.28 A
F-values	854.42**	41**	761.62**	562.81**	112.6**

Data are represented as **means** of six samples **± S.D.** Means within the same column, with the same letter for each parameter are not significantly different, otherwise they do (SAS, 2000). ** Highly significant difference at **P ≤ 0.01**.

Table (6): (Means ± SD) of Cd residual heavy metal concentration in organs (Gills, Liver, Kidney, Muscles and Gonads) of *Oreochromis niloticus* for the three different studied sites.

Studied sites of collection	Gills	Liver	Muscles	Kidney	Gonads

Hawara channel (Branch of the river Nile)	0.74±0.13 **C**	0.6±0.17 C	0.39±.069 C	0.62±0.07 C	0.15±0.03 C
El-Wadi drainage canal	4.89±0.26 A	4.63±0.189 A	3.29±.125 A	4.06±.094 A	1.21±.023 A
El-Bats drainage canal	3.62±0.36 B	3.76±0.14 B	2.92±.107 B	3.68±.188 B	1.15±.027 B
F-values	365.36**	965**	1393.8**	1295.9**	2800.7**

Data are represented as **means** of six samples ± **S.D.** Means within the same column, with the same letter for each parameter are not significantly different, otherwise they do (SAS, 2000). ****** Highly significant difference at **P ≤ 0.01**.

Table (7): (Means ± SD) of Cu residual heavy metal concentration in organs (Gills, Liver, Kidney, Muscles and Gonads) of *Oreochromis niloticus* for the three different studied sites.

Studied sites of collection	Gills	Liver	Muscles	Kidney	Gonads
Hawara channel (Branch of the river Nile)	1.78±0.69 B	3.13±0.25 C	1.72±0.25 C	1.81±0.37 C	1.56±0.08 C
El-Wadi drainage canal	32.47±1.22 A	41.47±4.61 A	14.24±.52 A	20.73±1.99 A	8.85±0.21 A
El-Bats drainage canal	31.06±1.39 A	37.8±1.24 B	11.5±0.86 B	18.2±1.17 B	7.23±0.71 B
F-values	1384.7**	351.3**	720.7**	346.2**	474.14**

Data are represented as **means** of six samples ± **S.D.** Means within the same column, with the same letter for each parameter are not significantly different, otherwise they do (SAS, 2000). ****** Highly significant difference at **P ≤ 0.01**.

Table (8): (Means ± SD) of Mn residual heavy metal concentration in organs (Gills, Liver, Kidney, Muscles and Gonads) of *Oreochromis niloticus* for the three different studied sites.

Studied sites of collection	Gills	Liver	Muscles	Kidney	Gonads
Hawara channel (Branch of the river Nile)	1.63±0.41 **C**	1.73±0.16 B	1.06±0.06 C	1.36±0.2 C	0.74±0.11 C
El-Wadi drainage canal	16.3±0.4 B	6.66±1.02 A	3.38±.18 A	7±0.18 B	2.29±0.09 B
El-Bats drainage canal	18.23±1.5 **A**	7.29±0.8 A	2.46±0.15 B	8.06±0. 6 A	3.23±0.14 A
F-values	585.7**	96.15**	417.2**	521.3**	666.2**

Data are represented as **means** of six samples ± **S.D.** Means within the same column, with the same letter for each parameter are not significantly different, otherwise they do (SAS, 2000). ****** Highly significant difference at **P ≤ 0.01.**

Table (9): (Means ± SD) of protein and lipid profile parameters for *Oreochromis Niloticus* collected from the different studied sites.

Studied sites of collection	Total protein	Albumin	Globulin	Total lipid	Triglycerides	Cholesterol
Hawara channel (Branch of the river Nile)	4.02±0.132 **B**	1.54±0.06 B	2.48±0.19 B	968.3±17.18 B	155.35±21.02 B	154.5±18.98 A
El-Wadi drainage canal	7.3±0.173 A	3.2±0.438 A	4.1±0.284 A	1640.08±126.41 A	464.87±73.04 A	196.33±56.2 A
El-Bats drainage canal	7.43±0.41 A	3.47±0.59 A	3.95±0.186 A	1620.91±87.78 A	442.68±79.93 A	194.18±38.77 A
f.values	51.82**	6.75**	15.84**	109.79**	44.1**	1.99**

Data are represented as **means** of six samples ± **S.D.** Means within the same column, with the same letter for each parameter are not significantly different, otherwise they do (SAS, 2000). ****** Highly significant difference at **P ≤ 0.01.**

Table (10): (Means ± SD) of liver and kidney function parameters for *Oreochromis Niloticus* collected from the different studied sites.

Studied sites of collection	GPT	GOT	ALP	LDH	Creatinine	Uric acid	Urea
Hawara channel (Branch of the river Nile)	17.86±2.14 B	66.63±9.33 B	15.69±1.86 B	25.25±2.92 B	1.9±0.28 B	6.17±0.318 B	8.87±0.97 C
El-Wadi drainage canal	152.65±14.48 A	190.87±10.87 A	65.51±5.82 A	54.57±3.73 A	2.78±0.608 A	13.07±2.37 A	55.82±5.5 B
El-Bats drainage canal	150.85±20.85 A	180.78±19.27 A	70.15±10.06 A	54.90±5.54 A	3.12±0.219 A	11.37±1.75 A	64.02±4.89 A
F-values	165.69**	148.64**	118.4**	98.18**	14.31**	26.56**	289.04**

Data are represented as **means** of six samples ± **S.D.** Means within the same column, with the same letter for each parameter are not significantly different, otherwise they do (SAS, 2000). ****** Highly significant difference at **P ≤ 0.01.**