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Oxidative stress, hematological, biochemical, and hormonal alterations induced by exposure tilapia fish of Edko Lake, Egypt, to organochlorine pesticide residues in Edko Lake water.

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Abstract

Background: Organochlorine (OCl) pesticide residues, including gamma-HCH, beta-HCH, heptachlor epoxide, p, p DDE, Dieldrin, endrin aldehyde, endosulfan sulfate, and endrin ketone, were found in the water of Egypt's Edko Lake in the Beheira Governorate. Therefore, the present study evaluated the effect of these OCl pesticide residues on some hematological, biochemical, and hormonal indices of lake tilapia fish.

Results: The impact on hematological parameters indicated that these OCl residues cause a significant increase in WBC counts, hematocrit (HCT) values in male and female fish and a considerable rise in HGB and lymphocytes (LYM) in male fish. Liver function biomarkers, such as aspartate amino transaminase (AST) and alanine amino transaminase (ALT), increased significantly because of these OCl residues. In contrast, alkaline phosphatase (ALP) went down a lot. In addition, they decreased the level of glucose compared to the control. Levels of biomarkers related to kidney functions, i.e., uric acid and creatinine, increased significantly. Also, antioxidant enzymes like catalase, glutathione peroxidase (GPx), and glutathione reductase (GR) increased. However, the activity of GST decreased a lot in both male and female fish, and the level of superoxide dismutase (SOD) was reduced in male fish. Results revealed an alteration in thyroid hormone levels (thyroxine (T4) and triiodothyronine (T3)) in fish plasma compared with the control. Also, a significant decrease in folic stimulation hormone (FSH) in female fish and a substantial reduction in progesterone and testosterone hormones in male fish.

Conclusions: The OCl pesticide residues found in Edko Lake water directly induced significant adverse effects on hematological components and biomarkers of liver and kidney functions and may have caused hormonal disturbances that harm fish health.

Keywords: OCl pesticide; Nile Tilapia fish; Hematocrit (HCT); Superoxide dismutase (SOD); Thyroxine (T4); Triiodothyronine (T3)

1. Introduction

Contamination of water by pesticides, either directly or indirectly, can kill fish, reduce fish productivity, or elevate concentrations of undesirable chemicals in edible fish tissue, which can affect the health of humans consuming these fish. Organochlorine pesticides are highly stable under different environmental conditions, have a persistent nature, and have chronic adverse effects on wildlife and humans. Studies have been primarily restricted to the direct effects of individual compounds in assessing the hazards of pesticides to fish. However, simultaneous exposure to pesticide

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mixtures could modulate metabolism and toxicity under field conditions. The high pesticide concentrations in the water and sediments indicate serious pesticide residue issues in the fish tissues. Ntow [1] showed that fish samples could be considered one of the most significant indicators in freshwater systems for estimating pesticide pollution levels. Burke *et al.* [2] suggested that, as with mammals, it had been shown that, after functional damage to tissues and organs of fish, some specific cellular enzymes leak into the blood plasma, where they have been detected. Therefore, knowledge of the hematological characteristics is an essential tool that can be used as an effective and sensitive index to monitor physiological and pathological changes in fish, and exposure to chemical pollutants can induce either an increase or decrease in the hematological level and the lymphocytes, the most abundant type of leucocyte in the peripheral blood of *C. gariepinus* exposed to paraquat herbicide. Neutrophils followed this. In contrast, the least significant leucocyte was the eosinophil [3&4].

Environmental contaminants such as herbicides, heavy metals, and insecticides are known to modulate antioxidant defense systems and cause oxidative damage in aquatic organisms by ROS production. Thus, the activity of antioxidant enzymes and the occurrence of oxidative damage have been proposed as indicators of pollutant-mediated oxidative stress [5]. Oxidative stress is one of the significant factors in the pathogenesis of liver disease. Fish have antioxidant defense pathways that neutralize ROS. These pathways include antioxidant enzymes like superoxide dismutase (SOD), catalase (CAT), and glutathione reductase (GR), as well as non-enzymatic antioxidants like reduced glutathione (GSH) [6]. Nowadays, the culture of Nile tilapia, *O. niloticus,* dramatically increases toward intensification, and this species is also widely distributed in many parts of the world. Edko Lake is one of Egypt's most economically significant north Egyptian lakes. These lakes had been exposed to environmental deterioration, whose negative result was reflected in their fisheries and, consequently, their fish production (quality and quantity). Analysis of water and fish samples collected from Edko Lake water revealed the presence of Heptachlor epoxide, p,p-DDE, Dieldrin, p, p-DDD, and Endrin ketone [7&8]. Therefore, the present study determines the adverse effects of organochlorine pesticide (OCl) residues in Edko Lake water on hematological, biochemical, and hormonal biomarkers in male and female Nile Tilapia fish (*Oreochromis niloticus*. Linnaeus, 1758) of Edko Lake.

2. Materials and Methods

2.1. Pesticide standards

A mixture of certified reference standard pesticides containing 18 organochlorine pesticides (OCPs) at a concentration (2000ug ± 0.5%) /ml for each of heptachlor epoxide, p, p- DDE, Dieldrin, p, p- DDD and Endrin Ketone were obtained from SUPLECO company (Bellefonte, PA, USA).

2.2. Study area and samples collection

Surface water was collected from Edko Lake, El-Behera Governorate, Egypt (Figure 1), and laboratory tap water (blank water).

2.3. Fish samples

Fish samples were collected from different sites of Edko Lake (figure 1), caught by fishermen, and placed in a tank containing oxygen. The collected fish samples were transferred immediately to the laboratory in an icebox, and their weights were recorded and later dissected to remove the tissue, liver, and gills of fish samples; then, samples were ground using a mini chopper, stored in aluminum wrappers, freeze-dried at -80 °C, and lyophilized to study the toxic effect of OCl residues on some hematological, biochemical, and hormonal biomarkers in the blood, serum, and plasma of tested fish.

2.4. Toxicological Experiments

Nile tilapia fish were collected from Edko Lake water to study the effect of OCl pesticide residues found in lake water on hematological, biochemical, and hormonal parameters of male and female fish and compare them with those of control fish. Fish in the control group were divided into two groups, males and females. Fish in each grade received tap water after staying for 24 hours. All experiments used eight fish from both Edko Lake and laboratory fish, males and females. Blood samples were collected from the caudal vein of each fish by syringe from the arteries as described by [10]. The blood samples were collected in heparinized tubes and serum separation tubes. The serum and plasma of the blood samples were separated as follows: samples were left for 30 min at room temperature and then centrifuged for 10 min at 10000 rpm. The supernatant serum and plasma were kept at -20 C until analysis.

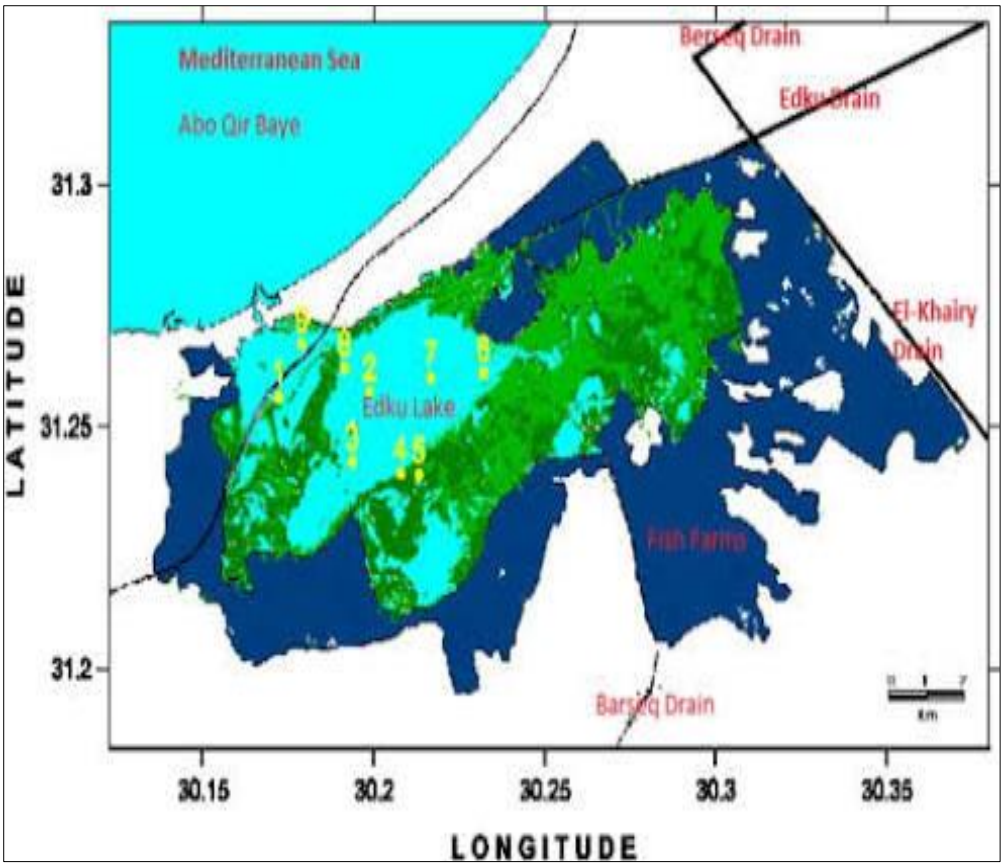

Figure 1 Map of Edko Lake

2.5. Hematological parameters

- **Hematocrit value** (packed cell volume) was determined according to [11&12] using the micro hematocrit centrifuge Model SH120. Wintrobe hematocrit heparinized tubes were packed with blood samples at 2/3 of their volume and stopped at one of their ends. Then, hematocrit tubes containing blood samples were centrifuged at 12000 r.p.m. for seven minutes. The hematocrit value was obtained by reading the packed cell volume on a unique graduated hematocrit measurement. The obtained data were expressed as percentages of hematocrit value to the total blood volume.
- **Total Hemoglobin was determined** according to the Wintrobe method [13].
- **The counting of red blood corpuscles (RBCs):** Hemocytometer, microscope Novex, and red cell count pipette was used to achieve the goal of this experiment. The Hemocytometer slide was focused under the microscope, and RBCs were counted according to the method of [14&15]. The hemocytometer slide is divided into 25 squares, each of which is divided into 16 smaller squares. The RBC count was done for five out of the 25 squares, and the number of RBCs was multiplied by the factor of 104 to obtain the RBC count for/ cm3.
- **White blood cell counts (WBCs)** were achieved according to the methods of [14&15]. As mentioned in RBC counting, the microscope and the hemocytometer were used. However, in the case of WBC counting, a specific area in the hemocytometer slide divided into four larger squares was used. The counts were multiplied by a factor of 50 to obtain the number of WBCs per cm3.

2.6. Measurement of kidney function biomarkers

- **Measurement of uric acid** concentrations was carried out according to the method reported by [16&17] using Boehringer Mannheim Diagnostic Kits.
- **The creatinine measurement in the fish serum** was done using the method of [18] Boehringer Mannheim Gmbh Diagnostics Kits. The color complex results from an alkaline solution's picrate and creatinine reaction.

2.7. Measurement of liver function biomarkers

- **The determination of serum AST** activity was determined according to the method described by [19] using Boehringer Mannheim GMBH Diagnostics Kits. This method uses photometry to determine how much oxaloacetate hydrazone is made when 2,4-dinitrophenylhydrazine is added.
- **Serum ALT activity** was determined using the method [19] described using Boehringer Mannheim Gmbh Diagnostics Kits. The method depends on a photometric estimation of pyruvate by monitoring the concentration of pyruvate hydrazone formed with 2, and 4-dinitrophenyl hydrazine.
- **Measurement of alkaline phosphatase (ALP)** was measured according to the method [20] reported using Diamond Diagnostics Kits.
- **Total protein was determined according to the method [21]** using Boehringer Mannheim Gmbh Diagnostics Kits.
- **Glucose content was measured** according to the method [22] described using Boehringer Mannheim Gmbh Diagnostics Kits.
- **Determination of Lactate dehydrogenase (LDH)** was carried out according to the method [23] using Diamino Diagnostics Kits.
- **Plasma Superoxide dismutase (SOD) was determined** according to [24] using Boehringer Mannheim Gmbh Diagnostics kits.
- **Plasma Catalase was determined** according to [25&17] using Boehringer Mannheim Gmbh Diagnostic kits.
- **Plasma Glutathione peroxidase (GPx) was determined** according to [26] using Boehringer Mannheim Gmbh Diagnostics kits.
- **Glutathione Reductase (GR) was determined** according to [27] using Boehringer Mannheim Gmbh Diagnostics kits.
- **Determination of plasma glutathione-S-Transferase (GST)** was carried out according to the method reported by [28] using Kits. The Bio diagnostic Glutathione -S-transferase assay kit measures total GST activity (cytosolic and microsomal) by measuring the conjugation of 1-chloro- 2, 4- dinitrobenzene (CDNB) with reduced glutathione. The conjugation is accompanied by an increase in absorbance at 340 nm. The rate of increase is directly proportional to the GST activity in the sample.
- **Quantitative Determination of Follicle- Stimulation Hormone (FSH)** was carried out according to the method reported by [29, 30, 31,&32]using International Immuno Diagnostics Kits
- **Testosterone hormone was determined in fish serum** according to the method reported by [33, 34&35] using International Immuno Diagnostics Kits.
- **Determination of progesterone** hormone concentration in fish serum according to the method reported by [36&37].
- **Triiodothyronine (T3) hormone concentration was determined in fish serum** according to the methods reported by [38&39] using International Immuno Diagnostics Kits.
- **Determination of L-Thyroxine (T4)** hormone concentration in fish Serum was carried out according to the methods reported by [40] using International Immuno Diagnostics Kits.

2.8. Statistical analysis

Statistical analyses were conducted using a computer program, "SAS"; the statistical design was a factorial CRD (Complete Randomized Design).

3. Results

Analysis of water samples from Edko Lake revealed the presence of Heptachlor epoxide, p, p-DDE, Dieldrin, p, p-DDD, and Endrin ketone with concentrations of 0.2309, 1.3524, 0.4104, 1.2622, and 0.1087 µg/l, respectively [8&9]. In addition, the adverse effects of the mixture of OCl residues (3.3646 µg/l, found in Edko Lake water) on hematological, biochemical, and hormonal biomarkers in Edko Lake fish were determined.

3.1. Effects on hematological parameters

Results in Table 1 show a significant increase in WBC counts in female fish with a value of 79.097 ± 1.746 * 109/l, relative to the control (38.79 ± 0.293 * 109/l). Also, a significant increase of HGB in male Edko Lake fish was 18.873 ± 1.598 g/dl relative to the control (15.67 ± 0.565 g/dl). No substantial changes in RBC counts were observed in male and female Edko Lake fish.

Results in Table 2 revealed a significant increase in hematocrit (HCT) in males and females of Edko Lake fish and a significant increase in lymphocytes (LYM) in male fish relative to control. Thus, the above results revealed substantial differences between hematological parameters in the fish collected from Edko Lake and those of control fish, indicating the harmful effects of OCl residues on lake fish.

Table 1 Effect of OCl pesticide residues in Edko Lake water on the RBC, WBC, and HGB count in the blood of treated Edko Lake fish

Treatments	Hematological Parameters					
	WBC (* 10^9/l) Mean ± SD* Male	WBC (* 10^9/l) Mean ± SD* Female	RBC (* 10^{12}/l) Mean ± SD* Male	RBC (* 10^{12}/l) Mean ± SD* Female	HGB (g/dl) Mean ± SD* Male	HGB (g/dl) Mean ± SD* Female
Control	26.283^{f} ± 1.090	38.79^{e} ± 0.293	2.283^{a} ± 0.234	1.503^{b} ± 0.116	15.67^{dc} ± 0.565	13.203^{d} ± 0.308
Lake fish	48.99^{d} ± 1.633	79.097^{c} ± 1.746	2.42^{a} ± 0.400	1.483^{b} ± 0.196	18.873^{ba} ± 1.598	14.307^{d} ± 1.013

*Data shown with the same symbols in the vertical column did not differ at 0.05 statistical levels.

Table 2 Effect of OCl pesticide residues in Edko Lake water on the HCT and LYM percentages in treated Edko Lake fish blood

Treatments	HCT		LYM	
	Mean ± SD* (%) Male	Mean± SD* (%) Female	Mean ± SD* (%) Male	Mean ± SD* (%) Female
Control	22.3^{cb} ± 3.890	18.643^{c} ± 0.878	31.513^{d} ± 1.440	21.407^{d} ± 0.987
Lake fish	38.487^{a} ± 3.248	25.65^{b} ± 2.660	48.393^{c} ± 2.021	34.51^{d} ± 0.876

*Data shown with the same symbols in the vertical column did not differ from each other at 0.05 statistical levels

3.2. Effects on biochemical biomarkers in the serum of Edko Lake fish

Table 3 shows how OCl pesticide residues in the water of Edko Lake affected the activity of aspartate aminotransaminase (AST), alanine aminotransaminase (ALT), and alkaline phosphatase (ALP) in the blood serum of Nile Tilapia fish from Edko Lake. Results show a significant increase in AST and ALT activity and a significant decrease in ALP activity in male and female fish.

Table 4 presents the results of the effect of OCl pesticide residues on the activity of lactate dehydrogenase (LDH) and creatinine and uric acid levels in the Edko Lake fish. These results show a significant increase in the activity of LDH in female fish, a significant increase in creatinine in male fish, and a substantial increase in uric acid in both male and female fish compared with control fish.

Table 3 Effect of OCl pesticide residues in Edko Lake water on the activity of AST, ALT, and ALP enzymes in the blood of treated Edko Lake fish

Treatments	AST activity (U/l)		ALT activity (U/l)		ALP activity (U/l)	
	Mean ± SD* Male	Mean ± SD* Female	Mean ± SD* Male	Mean ± SD* Female	Mean ± SD* Male	Mean± SD* Female
Control	2.910^{cd} ±1.164	0.976^{d}± 0.682	3.298^{d} ± 2.929	0.582^{d} ± 0.000	42.688^{b} ± 15.204	71.864^{a} ± 16.610
Lake fish	45.784^{a} ± 10.036	15.714^{cb} ± 6.559	37.967^{a} ± 7.060	17.533^{c} ± 3.356	10.671^{c} ± 2.256	19.284^{cb} ± 7.218

*Data shown with the same symbols in the vertical column did not differ from each other at 0.05 statistical levels

Table 4 Effect of OCl pesticide residues in Edko Lake water on the activity of LDH and levels of Creatinine and Uric acid in the blood of treated Edko Lake fish.

Treatments	LDH activity (U/l)		Creatinine (g/dl)		Uric acid (mg/dl) finish	
	Mean ± SD* Male	Mean± SD* Female	Mean SD* Male	Mean ± SD* Female	Mean ± SD* Male	Mean ± SD* Female
Control	280.627^{b} ± 33.810	399.353^{b} ± 21.075	0.978^{b} ± 0.539	1.733^{ba} ± 1.222	6.634^{d} ± 0.247	5.033^{d} ± 0.300
Lake fish	179.894^{b} ± 85.175	1798.042^{a} ± 484.636	2.204^{a} ± 0.425	0.62^{ba} ± 0.475	66.4^{b} ± 30.128	28.4^{cd} ± 9.934

*Data shown with the same symbols in the vertical column did not differ from each other at 0.05 statistical level

Results in Table 5 show the effect of OCl pesticide residues in Edko Lake water on the activity of catalase and the levels of glucose and total protein in Edko Lake fish. Statistical data show a significant decrease in glucose concentration in male and female fish (157.752 ± 3.697 and 138.760 ± 7.262 mg/dl, respectively) as compared with control fish (195.730 ± 38.703 and 197.796 ± 18.802 mg/dl, respectively). Also, OCl residues caused a slight decrease and increase in total protein in male and female fish relative to control fish. Catalase activity in the same table revealed a significant rise in Edko Lake tilapia fish.

Table 5 Effect of OCl pesticide residues in Edko Lake water on the activity of Catalase and Glucose Total protein levels in the blood of treated Edko Lake fish

Treatments	Glucose		Catalase activity		Total protein	
	Mean ± SD* (mg/dl) Male	Mean± SD* (mg/dl) Female	Mean ± SD* Male	Mean ± SD* Female	Mean ± SD* (g/dl) Male	Mean ± SD* (g/dl) Female
Control	195.730^{a} ± 38.703	197.796^{a} ± 18.802	274.348^{b} ± 57.072	349.794^{ba} ± 325.103	6.275^{b} ± 1.068	5.029^{b}± 0.156
Lake fish	157.752^{ba} ± 3.697	138.760^{b} ± 7.262	537.723^{ba} ±58.537	732.510^{a} ± 59.351	4.704^{b}± 0.444	6.430^{b} ± 1.289

*Data shown with the same symbols in the vertical column did not differ from each other at 0.05 statistical level

Results in Table 6 show a significant decrease in the activity of GST in males and females of Edko Lake fish compared to the control. Conversely, data in the same table revealed a significant increase in glutathione peroxidase activity in females of Lake fish.

Table 6 Effect of OCl pesticide residues in Edko Lake water on the activity of Glutathione – S –Transferase and Glutathione peroxidase enzymes in the blood of treated Edko Lake fish**.**

Treatments	**Glutathione – S-Transferase activity**		**Glutathione peroxidase activity**	
	Mean ± SD* (mU/ml) Male	Mean ± SD* (mU/ml) Female	Mean ± SD* (mU/ml) Male	Mean ± SD* (mU/ml) Female
Control	761.115^{cb} ± 18.723	1489.423^{a} ± 26.726	81.056^{c} ± 5.616	372.856^{bac}± 136.636
Lake fish	681.441^{c}± 121.090	1009.508^{b} ± 159.493	84.298^{c} ± 71.697	437.701^{ba} ±160.121

*Data shown with the same symbols in the vertical column did not differ from each other at 0.05 statistical levels

Results in Table 7 show the effect of OCl pesticide residues on the activities of glutathione reductase (GR) and superoxide dismutase (SOD) in male and female Edko lake fish. These results revealed a significant increase in the activity of GR and SOD in male and female Lake fish relative to control fish.

Table 7 Effects of OCl pesticide residues in Edko Lake water on the activity of Glutathione Reductase and Superoxide Dismutase enzymes in the blood of treated Edko Lake fish.

Treatments	**Glutathione Reductase activity**		**Superoxide Dismutase activity**	
	Mean ± SD* (mU/ml) Male	Mean± SD* (mU/ml) Female	Mean ± SD* (mU/ml) Male	Mean ± SD* (mU/ml) Female
Control	6.422^{cb} ± 0.278	2.844^{c} ± 0.168	147.727^{ba} ± 19.682	215.909^{a} ± 70.966
Lake fish	13.332^{a} ± 1.386	8.577^{b} ± 0.880	45.455^{b} ± 19.682	181.818^{a} ± 52.075

*Data shown with the same symbols in the vertical column did not differ from each other at 0.05 statistical levels

Growth and thyroid hormone systems have important implications for fish behavior. Results of the effect of OCl residues on growth and thyroid hormones in lake fish show a slight alteration in the levels of thyroxine hormone (T4) in both male and female Edko lake fish and a significant decrease in the levels of triiodo thyroxine hormone (T3) in Edko Lake fish as shown in (Table 8).

Table 8 Effect of OCl pesticide residues in Edko Lake water on the levels of Triiodo thyroxine (T3) and Thyroxine (T4) hormones in the blood of treated Edko Lake fish.

Treatments	**Triiodo thyroxine (T3)**		**Thyroxine (T4)**	
	Mean ± SD* (ng/ml) Male	Mean± SD* (ng/ml) Female	Mean ± SD* (Mg/ml) Male	Mean ± SD* (Mg/ml)Female
Control	0.536^{b} ± 0.165	2.386^{a} ± 1.325	26.195^{a} ± 6.367	26.018^{a} ± 0.444
Lake fish	0.222^{b} ± 0.114	0.481^{b} ± 0.097	20.45^{a} ± 5.124	26.710^{a} ± 0.940

*Data shown with the same symbols in the vertical column did not differ from each other at 0.05 statical levels

The effect of these OCl residues in Edko Lake water caused a significant decrease in follicular-stimulating hormone (FSH) in female fish and a non-significant reduction in male fish relative to control. Also, progesterone and testosterone hormones had decreased significantly in the male fish and had slightly decreased in females of Edko Lake fish, as shown in (Table 9).

Table 9 Effects of OCl pesticide residues in Edko Lake water on Folic stimulate hormone (FSH), Progesterone, and Testosterone hormone levels in treated Edko Lake fish.

Treatments	FSH		Progesterone		Testosterone	
	Mean ± SD* (U/l) Male	Mean± SD* (U/l) Female	Mean ± SD* (g/dl) Male	Mean ± SD* (g/dl)Female	Mean ± SD* (mg/dl)Male	Mean ± SD* (mg/dl)Female
Control	8.047^{ba} ± 2.307	7.78^{ba} ± 1.701	67.744^{a} ± 0.521	54.878^{ba} ± 0.460	13.09^{a} ± 1.962	12.504^{a} ± 0.229
Lake fish	6.913^{ba} ± 4.100	3.491^{b} ± 0.983	55.671^{ba} ± 9.348	55.528^{ba} ± 6.311	11.235^{a} ± 0.080	11.862^{a} ± 0.459

*Data shown with the same symbols in the vertical column did not differ from each other at 0.05 statical levels

4. Discussion

Various sources of pesticides (including herbicides, fungicides, and insecticides) frequently impact aquatic environments. Fish species are described as suitable monitors for the effects of toxic compounds because of their ecological and economic relevance. The adverse impact of OCl pesticide residues, 3.3646 µg/l, found in Edko Lake water by [8, 9] on hematological, biochemical, growth, and hormonal biomarkers in tilapia lake fish was determined. Knowledge of hematological characteristics is an essential tool that can be used as an effective and sensitive index to monitor physiological and pathological changes in fish. Exposure to chemical pollutants can induce either an increase or decrease in the hematological level of eosinophils [3, 4]. Results of the present study revealed that these OCl pesticide residues caused a significant increase in WBC counts, hematocrit (HCT), slight changes in RBC counts in male and female fish, and a substantial increase in lymphocytes (LYM) in male fish relative to control. These results disagree with [41, 42, and 43]. When these OCl residues are used to treat fish, the fish's immune system may be activated, or the fish may be stressed, which is shown by a change in the WBC count that turns into leukocytosis. Leucocytes regulate immunological functions and the protective response to stress in fish [6, 7].

The liver in fish is an organ that performs various functions associated with the metabolism of xenobiotics [44]. Our results show a significant increase in the activity of AST and ALT and a considerable decrease in the activity of ALP in both male and female fish. These results agree with those of [45, 46] and disagree with those of [47]. Alterations in serum AST, ALT, and ALP levels are conventional indicators of liver injury. The cytoplasmic enzyme lactate dehydrogenase (LDH) is widely used as a marker of organ or tissue lesions in toxicology and clinical chemistry. Therefore, it has been used for demonstrating tissue damage in fish. LDH is essential during glycolysis and directly affects fish development [48]. The data from the present study showed a significant increase in the activity of LDH in female fish compared with control fish. This result agrees with [45, 49] and disagrees with [50]. Elevated levels of this enzyme in the plasma indicate transient damage to either muscle fibers (cardiac) or other tissues. Increased activity of LDH can be explained as a consequence of pathological changes in hepatic tissue. Catalytic activities of plasma enzymes (i.e., LDH, ALT, and AST) may indicate a stress reaction, as the increased values indicate stress-based tissue impairment.

Concerning the effect of OCl residues on kidney function, our results revealed a significant increase in uric acid and creatinine, a considerable decrease in glucose concentration, and a slight reduction in total protein in males and females of Edko Lake fish. These results disagree with those of [51] and agree with those of [52].

Under normal physiological conditions, slight oxidative stress can trigger the antioxidant defense systems, including SOD, CAT, and GST, as a compensatory response. Thus, the reactive oxygen species (ROS) can be removed to protect the organisms from oxidative damage [53]. The activity of antioxidant enzymes may be increased or inhibited under chemical stress depending on the intensity and duration of pressure applied and the susceptibility of exposure species. The liver in fish is an organ that performs various functions associated with the metabolism of xenobiotics {44]. Hepatocytes, like other cells, depend on antioxidant enzymes for protection against reactive oxygen species produced during the biotransformation of xenobiotics [54]. Among the enzymes that comprise this defense system are SOD and CAT. Superoxide dismutase (SOD) is one of the enzymes responsible for removing hydrogen peroxide, which is metabolized to oxygen and water [55]. Our results revealed that OCl residues caused a significant increase in the activities of catalase (CAT) and glutathione peroxidase in males and females of Edko Lake fish. These results agree with [56, 57]. Also, our results show a significant decrease in glutathione-S-transferase activity (GST) in lake fish, consistent with those of [58, 59]. Concerning how these OCl residues affect the levels of superoxide dismutase (SOD) and

glutathione reductase (GR) in fish from Edko Lake, the results showed that the activity of SOD and GR decreased significantly in both male and female fish. These results agree with those of [60, 61].

Growth and thyroid hormone systems affect fish behavior [62]. Exposure to numerous pesticides has altered both thyroxine (T4) and triiodothyronine (T3) levels in the plasma of several fish species after both acute and chronic exposure [63]. Results show a non-significant alteration in the content of T4 in both male and female fish and a significant decrease in T3 levels in male fish. Follicular-stimulating hormone (FSH) is released from the gonadotrophs in the pituitary gland [64]. Pesticides interfere with the normal function of FSH, and abamectin and indoxacarb significantly reduce the content of the FSH hormone. Our results indicated a significant decrease in the level of FSH in female lake fish and a substantial reduction in progesterone and testosterone hormones in male Edko Lake fish. These findings are nearly identical to those of [65], who mentioned that malathion interferes with the aromatase enzyme, leading to reduced estradiol (E2) and T hormone in eels (*Monopterus albus).* Span et al. [66] came to similar conclusions. They found that male Carassius auratus treated with atrazine had lower levels of testosterone and plasma estradiol since atrazine stimulates the aromatase enzyme, which turns testosterone into estradiol. However, our results disagree with [67]. Many authors, including [68, 69], explained how the sex hormone changed and suggested that this change might result from pesticide interference with the production of free cholesterol, the sex hormone precursor to steroid production.

5. Conclusions

Overall, the results suggest that the OCl pesticide residues found in the water of Edko Lake directly caused changes in hematological parameters and enzymatic activities. In addition, these findings indicate that the OCl pesticide residues caused metabolic disorders, cell damage in specific organs (like the liver, kidney, and oxidative system), and disruptions of growth and hormone systems that may have harmed fish health.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors confirm no known conflicts of interest with this publication.

Statement of ethical approval

The experimental work on rats was performed with the approval of the Animal Care and Experimental Committee, Faculty of Agriculture, Damanhur University, Egypt, and according to the Guide for Care and Use of Laboratory Animals (NRC, Acad. Press; Washington, DC, USA,1996

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Authors' contributions

All authors contributed to the study's conception and design. They performed material preparation, data collection, and analysis. M.A. Abbassy conceived and designed the experiments, wrote the paper, and submitted it for publication on behalf of the co-authors. ; Eman Nour Eldin performed the experiments and analyzed the data; M.A. Khalifa and Omar A. Omar contributed reagents/materials/analysis tools.

Consent for publication

The authors consent to the publication of identifiable details, including images and other details within the text to be published in the journal.

Availability of data and materials

All data generated or analyzed during this study are included in this article. Also, the related datasets are available from the corresponding author upon reasonable request.

References

[1] Ntow W. Joseph (2005). Pesticide residues in Volta Lake, Ghana. Lakes& Reservoirs: Policy and Management for Sustainable Use, Vol. 10, Issue 4 /p. 243-248

[2] Burke, C. W., and C. J. Eastman(1974). Thyroid hormones. Brit. Med. Bull., 30:93-99.

[3] Alohan et al. (2014). Hematological indices in fish plasma indicate exposure to a commonly used herbicide. Australian Journal of Basic and Applied Sciences 8(6):245-250

[4] Nwani, C.D., Nagpure, N.S., Kumar, R., Kushwaha, B., Kumar, P. and Lakra, W.S. (2010). Lethal concentration and toxicity stress of carbosulfan, Glyphosate, and Atrazine to freshwater air-breathing fish Channa punctatus (Bloch.) Int. Aquat. Res., 2: 105-111.

[5] Monteiro, D.A., Alves de Almeida, J., Rantin, A.F.T., Kalinin, L., (2006). Oxidative stress biomarkers in the freshwater characid fish, Brycon cephalus, exposed to organophosphorus insecticide Folisuper 600 (methyl parathion). Comp. Biochem. Physiol. 143, 141–149.

[6] Modesto, K.,A. and Martinez, C, B, (2010). Effects of Roundup Transorb on fish: Hematology, antioxidant defenses, and acetylcholinesterase activity. Chemosphere 81(6)781-7

[7] Velisek, J., and A. Stara (2018). Effect of thiacloprid on early life stages of common carp (Cyprinus carpio). Chemosphere, Vol. 194, 481-487.

[8] Abbassy M.A., Khalifa M.A., Nassar A.K., Omar A.O., Salim Y.M. and Nour-Eldin E.E. (2020). Comparative study of organochlorine pesticide residues in the water of both Edko Lake and fish farm in El-Beheira Governorate, Egypt. Austin Environ Sci. Vol 5(1):1-6.

[9] Abbassy, M.A., M. Khalifa, A.K. Nassar, E. Nour El-Deen (2021). Analysis of organochlorine pesticides (OCPS) residues in fish from Edko lake (North of Egypt) using eco-friendly methods and their health implications for humans. Toxicological Research, 37(4), 495-503.

[10] Congleton,J. and W.J. LaVoie (2011). Comparison of blood chemistry values for samples collected from juvenile chinook salmon by three methods. Journal of aquatic Animal Health. 13 (2): 168-172

[11] Bull, B.S. ; J.A. Koepke ; E. Simson ; O.W. van Assendelft (2000). Procedure for determining packed cell volume by the microhematocrit method; approved standard-third edition. NCCLS Document H7-A3. 20(18).

[12] Bull, B.S. ; O.W. van Assendelft ; K. Fujimoto; et al (2001). (International Council for Standardization in Hematology Expert Panel on Cytometry).Recommendations for reference method for the packed cell volume (ICSH Standard 2001). Lab Hematology; 7:148-170.

[13] Wintrobe, M. M. (1965). Clinical hematology. 4th edition lea and Febiger, Philadelphia.

[14] Britton, C. J. (1963). Disorders of the blood, 9th Edition J. A. Churchill Ltd.: London.

[15] Seiverd, E. C. (1964). Hematology for Medical Technology, 3rd Edition, lea and Feiger: Philadelphia.

[16] Barham, D. and P. Trinder. (1972). Analyst, 97:142-145.

[17] Fossati, P. ; L. Prencipe and G. Berti (1980). Clin. Chem., 2612: 227-273.

[18] Henry, R. J. (1974). Clinical Chemistry, principles and technics,2nd Edition, Harper and Row, p.525.

[19] Reitman, S. and Frankel, S. (1957). A colorimetric method for determination of serum glutamic oxaloacetic and glutamic pyruvic transaminase. Amer. J. Clin. Pathol. 28:56

[20] Rec, GSCC (DGKC). (1972). Determination of enzyme activities. J. Clin Chem. Clin. Biochem., 10:192. C.F. Randox manual procedures,

[21] Henry, R. J. (1964). Clinical Chemistry Harper and Row publishers, New York. P.181.

[22] Trinder, P. (1969). Enzymatic determination of glucose in blood serum. Annals of Clinical Biochemistry, 6, 24.

[23] Rec, GSCC (DGKC). (1970). Determination of enzyme activities. J. Clin Chem. Clin. Biochem., 8: p 658. C.F. Randox manual procedures, Edition III. No. LD 420. Antrim, UK. AP. 309.

[24] Nikinmaa M, Huestis W (1984). Adrenergic swelling of nucleated erythrocytes: cellular mechanisms in a bird, domestic goose and two teleost's, striped bass, and rainbow trout. J. Exp. Biol., 113: 215-222

[25] Aebi, H. (1984). Catalase in Vitro. In Packer, L.,(Ed.), Methods in Enzymology, Academic Press, 105, 121-126.

[26] Pagila, D.E. and Valentine, W. N. (1967). Studies on the quantitative and qualitative characterization of Erythrocyte peroxidase. Journal of Laboratory and Clinical Medicine, 70: 158-169.

[27] Goldberg, D.M. and R. J. Spooner (1983). Glutathione Reductase , "In H.U. Bergmeyer, J. Bergmeyer and M. GraBI, Eds., Methods of Enzymatic Analysis, 3rd Ed. Verlag Chemie,Weinheim, 1983, pp. 258-265.

[28] Habig,W.H., M.J. Pabst and W.B. Jakoby(1974). Glutathione -S-transferases. The first enzymatic step in mercapturic acid formation. The J. Biol. Chem.Vol. 249,22, 7130-7139.

[29] Marshall, J. C. (1975). Clinic in Endocrinol Metab. 4: 545.

[30] Cohen, K. L. (1977). Metabolism. 26: 1165.

[31] Abranham, G. E. (1981). Radio assay systems in Clinic. Endocrinol. Marcel Dekker, Inc., New York

[32] Rebar, R. W.; G. F. Erickson and S. S. C. Yen (1982). Fertil. Steril. 37: 35.

[33] Chen, A.; J. J. Bookstein and D. R. Meldrum (1991). Diagnosis of a testosterone-secreting adenoma by selective venous catheterzation. Fertil. Steril. 55(6): 1202- 1203

[34] Heinonen, P. K. (1991). Androgen production by epithelial ovarian tumors in postmenopausal women. Maturitas., 13(2): 117-133

[35] Tietz, N. W. (1995).Clinical guide to laboratory tests, third edition. Saunders Company, Philadelphia, pp. 578- 580

[36] Cameron, E .D. H. and J. J. Scarisbrik (1973). Radioimmuno assay of plasma progesterone. Clin. Chem. 19: 1403-1408.

[37] Kakabakos, S. E. and M. J. Khosravi. (1992). Direct time resolved florescence immunoassay of progesterone in serum involving the biotin streptavidin system and the immobilized antibody approach. Clin. Chem. 38: 725-730

[38] Spector, D. A.(1976). Thyroid function and metabolic state in chronic renal failure. Ann. Int. Med., 85:724-730.

[39] Cavalieri, R. R. and B. Rapoport (1977). Impaired peripheral conversion of thyroxine to triiodothyronine. Ann. Rev. Med., 28:57-65.

[40] Schuurs, A. and Van Weemen (1977). Enzyme-immunoassay. Clinica Chimica Acta 81(1), 1-40.

[41] Adeyemo OK (2007). Hematological profile of Clariasgariepinus (Burchell, 1822) exposed to lead. Turkish J. Fish. Aquat. Sci. 7: 163-169

[42] Banaee M, Mirvaghefi AR, Rafei GR, MajaziAmiri B (2008). Effects of sub-lethal diazinon concentrations on blood plasma biochemistry of common carp. Int. J. Environ. Res., 2: 189-198

[43] Banaee M, Sureda A, Mirvaghefi AR, Ahmadi K (2011). Effects of diazinon on biochemical parameters of blood in rainbow trout (Onchorhynchus mykiss). Pest. Biochem. Physiol., 99: 1-6

[44] Jiminez, B. D. and Stegeman, J. J. (1990). Detoxification enzymes as indicators of environmental stress on fish. Am. Fish. Soc. Symp. 8: 67-79.

[45] Tiwari S, Singh A (2004) Piscicidal activity of alcoholic extract of Nerium indicum leaf and their biochemical stress response on fish metabolism. Afri. J. Trad. CAM. 1:15-29.

[46] Abd-Algadir, M.I., M.K. Sabah Elkhier and O.F. Idris (2011). Changes of the fish liver (Tilapia nilotica) made by herbicide (Pendimethalin). Journal of Applied Biosciences 43: 2942-2946

[47] Ayalogu OE, Igbon NM, Dede EB (2001). Biochemical changes in the serum and liver of albino rats exposed to petroleum samples (gasoline, kerosene, and crude petroleum). J. Appl. Sci. Environ. Mgt. 5(1):97-100

[48] Gronczewska,J.and E.F.Skorkowski (2020). Lactate dehydrogenase in fish spermatozoa and its role in sperm cell bioenergetics. Indian Journal of Experimental Biology,Vol.5: 7-13.

[49] Abalaka SE, Esievo KAN, Shoyinka SVO (2011). Evaluation of biochemical changes in Clarias gariepinus adults exposed to aqueous and ethanolic extracts of Parkiabiglobosa pods. Afr. J. Biotechnology., 10: 234-240

[50] Hedayati A, Safahieh A, Savari A, Marammazi JG (2010). Assessment of aminotransferase enzymes in yellowfin sea bream (Acanthopagrus latus) under the experimental condition as biomarkers of mercury pollution. World J. Fish Mar Sci.,2:186-192

[51] Pickering, A.D. (1981). Stress and Fish. Academic Press New York, 367 pp.

[52] Al-Ghanim, K. A. (2012). African Journal of Agricultural Research Vol. 7(4): 561-567

[53] Livingstoone, D. R. (2001). Contaminants stimulated reactive oxygen species production and oxidative damage in aquatic organisms. Mar. Pollution Bull., 42:656-666.

[54] Londis, W. G. and Yu, M. H. (1995). Introduction to Environmental Toxicology: Impacts of Chemicals upon Ecological Systems. Lewis Publishers, Boca Raton.

[55] van der Oost, Beyer, J. and Vermeulen, N.P.E. (2003). Fish bioaccumulation and biomarkers in environmental risk: Environ. Toxicology and Pharmacology,13(2): 57-149.

[56] Gad, N. S. and Yacoub, A. M. (2009). Antioxidant defense agents and physiological responses of fish to pollution of Rosetta Branch of the River Nile, Egypt. Egypt. J. Aquat. Biol. & Fish., 13(4):109-128.

[57] Stara, A., Pagano, M., Albano M. and Savoca, S. (2021). Effects of long-term exposure of Mytillus galloprovincialis to thiacloprid: A multi biomarker approach. Environmental Pollution 289(3): 117892 DOI: 10. 1016/j.envpol.2021. 117892.

[58] Simonata, J.; Guedes, C. and Martinez, C. (2008). Biochemical, physiological and histological changes in neotropical fish, Prochildus lineatus exposed to diesel oil. Ecotoxicol. Environ. Saf ., 69:112-120 .

[59] Nahragan, J.; Camus, L.; Gonzalez, P.; Jonsson, M.; Chritiansen, S. and Hop, H. (2010). Biomarker response in polar cod exposed to dietary crude oil. Aquatic Toxicol., 96(1):77-83.

[60] Matos, P.; A. Fontaı ´nhas-Fernandes, F. Peixoto, J. Carrola, E. Rocha (2007). Pesticide Biochemistry and Physiology 89: 73–80.

[61] Peixoto,F.; D. Simo˜ es, D. Santos, A. Fontaı ´nhas-Fernandes, (2006). Toxicological effects of oxyfluorfen on oxidative stress enzymes in tilapia Oreochromis niloticus. Pest. Biochem. Physiol. 85: 91–96.

[62] Devlin, R.H., Biagi, C.A., Yesaki, T.Y., Smailus, D.E., Byatt, J.C., (2001). Growth of domesticated transgenic fish. Nature 409,781–782

[63] Leemans, M., S. Couderg, B. Demeneix, and J-B. Fini, (2019). Pesticides with potential thyroid hormone-disrupting effects: A Review of recent data. Front. Endocrinol. (Lausanne) 10: 743.

[64] Puri, D. (2011). Textbook of Medical Biochemistry, 3rd Edition. Elsevier, A division of Reed Elsevier India Private Limited. ISBN 978-81-312-2312-3.

[65] Singh, H. (1993). Effect of malathion on steroidogenesis and sex reversal in Monopterus albus. Mar Environ Res 35: 159-164.

[66] Spano, l.; Tyler, C.R.; Van Aerle, R.; Devos, P.; Mandiki, S.N.M.; Silvestre, F.; Thome, J.P. and Kestemont, P. (2004). Effects of atrazine on sex steroid dynamics, plasma vitellogenin concentration and gonad development in adult goldfish (Carassius auratus). Aquat. Toxicol. 66:369-379.

[67] Singh, S. and Singh, T.P. (1987). Impact of malathion and hexachlorocyclohexane on plasma profile on three sex hormones during different phases of the reproductive cycle in Clarias batarachus. Pesticide Biochem. Physiol. 27, 301-308.

[68] Garcia-Reyero, N.; Barber, D.S.; Gross, T.S.; Johnson, K.G.; Sepulveda, M.S.; Szabo; N.J. and Denslow, N.D. (2006). Dietary exposure of largemouth bass to OCPs changes expression of genes important for reproduction. Aquatic Toxicology. 78:358-369.

[69] Lal, B and Singh, T.P. (1987). The effect of malathion and γ-BHC on the lipid metabolism in relation to reproduction in the tropical teleost, Clarias batarachus. Environ Poll, 48:37-47.