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PHYSIOLOGICAL STUDIES ON TWO POPULAR TELEOSTS, *OREOCHROMIS NILOTICUS AND BAGRUS BAYAD,* INHABITING El-SAHEL CANAL AND DAMIETTA BRANCH AT DAKAHLIA PROVNCE.

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ABSTRACT

In the present study. two popular fish for human consumption (*Oreochromis nitoticus & Bagarus bayad*) inhabiting El-Sahel canal and Damietta branch at Dakahlia province were used for evaluating the toxicological effects of heavy metals (Cr, Ni, Zn and Cu) exposure through period of one year. Present results revealed an increase in the studied metals bioaccumulation in the muscles of fish samples from El-Sahel canal. Decreased total protein and glycogen contents in both liver and muscle tissues with inhibition of the brain acetylcholine esterase (AchE) were also recorded. However, the reverse was observed by lactic dehydrogenase (LDH) activity in both liver and muscle and the antioxidants, reduced glutathione (GSH) and glutathione-S-transferase (GST) in the liver tissue of fish inhabiting heavy metals polluted water, in comparison to those from the less polluted site. At the same time, it was also noted that all tested biomarkers in the two examined fish exhibited seasonal variations through the entire study period that reached lowest values during winter and highest during summer. In all, these results revealed the adverse effects of the aquatic heavy metals pollution on two of the most locally eaten fish that seemed to be seasonally depended.

INTRODUCTION

As a result of the human activities, the aquatic environment has been increasingly contaminated by heavy metals ***(Skjelkvale et al., 2001).*** This causes adverse effects on the health status of aquatic organisms, especially fish ***(Gbem et al., 2001). Zyadah (1995) and Zhou et al. (1998)*** reported that fish exposure to heavy metals results in their accumulation in target organs, with concomitant physiological alterations that ultimately used as bioindicators for pollution. These include changes in the metabolic activities ***(Zaghloul et al., 2002),*** as well as in the antioxidant defense system ***(Dautremepuits et al., 2003).***

Regarding the above explanations, it was of interest to study the impact of aquatic environmental pollution on two fish species (*Oreochromis niloticus* & *Bagrus bayad*) of economic importance that inhabiting two water regions along Dakahlia province. This was done by: (1) investigating the concentration of accumulated heavy metals (Ni, Zn, Cr & Cu) in the muscles of the two selected fish species and the surrounding water during a study period of one year. (2) evaluating a number of biomarkers (total protein, glycogen content, acetylcholinesterase activity, as well as antioxidant defense components; reduced glutathione and glutathione-S-transferase) in tissues of the examined fish, taking into consideration the effect of seasonal variations during the study period.

MATERIALS AND METHODS

Study area:

Fish samples were collected from two water localities at Dakahlia province that considered as natural sources for fishery. The first locality is El-Sahel canal, commonly called Khadrawia canal at an area parallel to Talkha Fertilizer Factory that lies east to the canal, while the second locality is the Damietta branch of the river Nile running in Mansoura city at an area known as Gizzerhat El-Ward that lies west to Mansoura city. The first locality was selected as polluted area; where it receives several sources of pollution, as industrial and agricultural wastes, while the second one was selected as relatively less polluted area (reference site).

Collection of fish samples:

Samples of two freshwater teleosts (*Oreochromis niloticus* and *Bagrus bayad*) that considered the most abundant fish consumed by the local population, were collected seasonally through period of one year from January 2002 to December 2002. Fish were brought to the laboratory on the same day for investigation. Also, samples of surface water were collected seasonally from the two study sites during the study period for heavy metals analysis.

Preparation of tissue homogenate:

Fish were dissected to remove the epiaxial muscle on the dorsal surface, the entire liver, and brain from each one. Samples from such organs were weighed, homogenized and centrifuged. The supernatent was then collected for further biochemical analysis. Another samples from both liver and muscle tissues were removed for determination of glycogen content. A third sample of the muscle tissue was removed, weighed and dried, then digested by conc. nitric acid for analysis of heavy metals. At the same time, the collected water samples from the two studied sites were filtered and prepared for analysis. The values of tested metals (Cr, Ni, Zn & Cu) in both muscles and water samples were measured using an Atomic Absorption Spectrophotometer (Perkin Elmer 3110).

Biochemical analysis:

Glycogen content was determined as described by Nicolas *et. al.*, (1956). Reduced glutathione (GSH) content and glutathione-S-transferase (GST) activity were assessed following the methods of Prins and Loose (1969) and Habig *et. al.*, (1974), respectively. Moreover, total protein, lactic dehydrogenase (LDH) activity and acetylcholine esterase (AchE) activity were measured, using kits from Diamond Company, Egypt.

Statistical analysis:

The impact of aquatic environmental pollution was determined using *t*- test analysis ($p< 0.05$), by comparing fish species collected from the two sampling sites, where fish from Nile less polluted locality was considered as control samples. Statistical

analysis were performed using the SPSS statistical soft ware package.

RESULTS

In the present study, differences between fish samples collected from the two studied sites were assessed seasonally, through period of one year in order to determine the influence of aquatic pollution on the examined fish.

Obtained data indicated significant increases in the concentrations of all tested metals (Cr, Ni, Zn & Cu) in the muscles of fish samples collected from the polluted canal locality if compared to those from Nile locality. A finding that goes in parallel with the increased concentrations of tested metals in surface water of El-Sahel canal, relative to Nile water (table 1).

On comparing the biochemical measures assessed in both liver (table 2) and muscle (table 3) of the tested fish from the two studied localities, both fish species collected from El-Sahel canal showed significant decreases for all tested biomarkers through the entire study year, except for LDH activity in both liver and muscle and the antioxidants (GSH & GST), in the liver tissue that showed higher values in comparison with that collected from the River Nile branch (table 2 & 3).

Meanwhile, brain AchE activity showed significant decreases for all fish samples collected from El-Sahel canal, when compared to those from Nile branch water (table 4).

Beside the above recorded changes in response to site pollution, the two examined fish species from the two selected sites showed seasonal changes in all tested parameters that attained lowest values during winter, but seemed to increase at the rest of the year, reaching the highest values during summer, with the exption of LDH activity that showed the reverse behavior.

Table (1): Heavy metal concentrations in the surface water (µg/l) and muscles (µg/g d.t.) of the two examined fish species collected from two different water resources along Dakahlia province.

Seasons		Nile				Canal			
		Cr	Ni	Zn	Cu	Cr	Ni	Zn	Cu
Winter	Water	0.76 ±0.09	0.88 ±0.04	0.10 ±0.02	0.19 ±0.01	1.12 ±0.22	0.99 ±0.17	0.15 ±0.06	0.28 [a] ±0.03
	O. niloticus	28.47 ±1.92	21.64 ±2.02	23.46 ±0.67	23.88 ±0.77	42.36 [b] ±2.92	38.64 [b] ±1.53	36.69 [b] ±2.59	37.47 [b] ±1.51
	B. bayad	18.12 ±0.98	17.29 ±1.36	13.30 ±2.60	13.93 ±0.84	38.88 [c] ±1.47	36.80 [c] ±2.97	23.65 [c] ±0.51	26.52 [c] ±0.51
Spring	Water	0.62 ±0.08	0.85 ±0.33	0.07 ±0.00	0.25 ±0.03	1.04 [a] ±0.14	0.93 ±0.26	0.10 ±0.01	0.39 [a] ±0.04
	O. niloticus	37.97 ±2.05	36.46 ±2.16	26.44 ±1.82	30.21 ±2.05	67.05 [b] ±2.93	50.87 [b] ±2.52	46.82 [b] ±1.69	53.80 [b] ±0.60
	B. bayad	30.78 ±1.29	27.41 ±1.00	18.69 ±0.99	20.17 ±1.22	45.67 [c] ±1.60	37.31 [c] ±1.93	24.64 [c] ±1.05	29.81 [c] ±1.43
Summer	Water	0.18 ±0.03	0.45 ±0.12	0.03 ±0.00	0.28 ±0.02	0.92 ±0.12	0.61 ±0.14	0.08 ±0.01	0.33 [a] ±0.00
	O. niloticus	63.65 ±1.11	42.47 ±2.56	31.92 ±0.31	38.35 ±2.65	79.92 [b] ±2.66	76.21 [b] ±1.93	48.15 [b] ±1.07	54.46 [b] ±1.31
	B. bayad	67.69 ±2.28	57.53 ±1.60	20.89 ±2.84	32.53 ±1.81	74.72 ±2.72	66.84 [c] ±0.38	29.60 [c] ±1.37	38.21 ±1.78
Autumn	Water	0.94 ±0.003	0.55 ±0.15	0.05 ±0.00	0.39 ±0.04	1.05 ±0.09	0.67 ±0.10	0.06 ±0.00	0.44 ±0.04
	O. niloticus	49.78 ±2.22	39.94 ±0.58	24.68 ±1.42	34.79 ±1.67	70.89 [b] ±2.54	66.29 [b] ±1.85	47.14 [b] ±1.44	50.00 [b] ±1.85
	B. bayad	37.88 ±2.04	30.98 ±1.76	16.23 ±1.68	20.49 ±2.09	47.93 [c] ±2.68	43.08 [c] ±2.04	25.19 [c] ±1.52	36.93 [c] ±1.37

Values are means ± S.E , n = (4-5) & d.t = dry tissue.

a= significant at $p < 0.05$, compared to the surface water collected from river Nile.

b= significant at $p < 0.05$, compared to fish (*O. niloticus*) collected from river Nile.

c= = significant at $p < 0.05$, compared to fish (*B. bayad*) collected from river Nile.

Table (2): Total protein (g/100g w.t), glycogen content (mg/100g w.t), LDH (U/g w.t) and antioxidant components, GSH (mg/100g w.t)& GST(U/mg w.t) in the liver of the two examined fish species collected from two different water resources along Dakahlia province.

Seasons		Nile					Canal				
		T. Protein	Glycogen	LDH	GSH	GST	T. Protein	Glycogen	LDH	GSH	GST
Winter	*O. niloticus*	35.34 ±1.19	12.69 ±0.56	651.54 ±31.94	1.81 ±0.07	1.59 ±0.06	21.80 [b] ±0.88	8.10 [b] ±0.22	840.53 [b] ±36.27	2.15 [b] ±0.09	1.87 [b] ±0.10
	B. bayad	26.17 ±1.04	12.46 ±0.73	1121.4 9 ±36.60	1.51 ±0.06	1.28 ±0.05	18.75 [c] ±0.93	7.61 [c] ±0.75	1432.28 [c] ±30.03	1.88 [c] ±0.05	1.56 [c] ±0.12
Spring	*O. niloticus*	36.56 ±1.34	14.57 ±0.72	582.35 ±28.25	1.99 ±0.08	2.02 ±0.06	23.93 [b] ±0.57	8.50 [b] ±0.42	755.13 [b] ±37.72	2.26 [b] ±0.06	2.33 [b] ±0.10
	B. bayad	29.80 ±1.74	14.20 ±0.84	840.48 ±33.67	1.77 ±0.07	1.72 ±0.07	18.79 [c] ±0.85	8.22 [c] ±0.69	1376.77 [c] ±28.17	2.04 [c] ±0.07	2.09 [c] ±0.09
Summer	*O. niloticus*	44.60 ±2.24	16.89 ±1.02	404.89 ±28.58	2.28 ±0.09	2.25 ±0.10	24.23 [b] ±0.97	9.41 [b] ±0.63	657.17 [b] ±37.77	2.87 [b] ±0.07	2.56 [b] ±0.08
	B. bayad	33.73 ±1.59	18.76 ±1.01	715.02 ±36.71	1.98 ±0.09	1.86 ±0.06	19.94 [c] ±0.85	8.76 [c] ±0.35	1308.12 [c] ±32.24	2.64 [c] ±0.09	2.17 [c] ±0.08
Autumn	*O. niloticus*	33.52 ±1.58	16.21 ±1.09	523.41 ±28.88	2.20 ±0.09	2.13 ±0.08	24.01 [b] ±1.18	8.68 [b] ±0.51	724.12 [b] ±29.92	2.58 [b] ±0.08	2.23 ±0.09
	B. bayad	31.89 ±1.35	16.93 ±0.45	818.13 ±37.01	1.78 ±0.07	1.67 ±0.04	19.18 [c] ±0.56	8.45 [c] ±0.36	1363.18 [c] ±36.97	2.10 [c] ±0.06	1.77 ±0.05

Values are means ± S.E , n = (10-12) & w.t = wet tissue.

b= significant at $p < 0.05$, compared to fish (*O. niloticus*) collected from river Nile.

c= = significant at $p < 0.05$, compared to fish (*B. bayad*) collected from river Nile.

Table (3): Total protein (g/100g w.t), glycogen content (mg/100g w.t), LDH (U/g w.t) in the muscle of the two examined fish species collected from two different water resources along Dakahlia province.

Seasons		Nile			Canal		
		T. Protein	Glycogen	LDH	T. Protein	Glycogen	LDH
Winter	*O. niloticus*	32.03 ±1.10	0.78 ±.009	1704.26 ±35.99	19.00 [b] ±1.02	0.49 [b] ±0.03	2133.47 [b] ±34.22
	B. bayad	27.04 ±1.08	0.87 ±0.02	1779.66 ±35.52	18.14 [c] ±0.86	0.43 [c] ±0.03	2332.00 [c] ±32.10
Spring	*O. niloticus*	34.81 ±1.37	0.90 ±0.03	1522.34 ±31.54	21.59 [b] ±1.00	0.56 [b] ±0.02	1997.33 [b] ±33.01
	B. bayad	30.54 ±1.07	0.96 ±0.06	1298.05 ±34.17	20.58 [c] ±1.01	0.45 [c] ±0.06	1727.22 [c] ±29.91
Summer	*O. niloticus*	40.90 ±2.27	1.56 ±0.05	1395.21 ±32.31	22.00 [b] ±1.48	0.67 [b] ±0.03	1874.33 [b] ±26.38
	B. bayad	33.19 ±1.47	1.49 ±0.08	1067.61 ±32.09	21.72 [c] ±0.80	0.61 [c] ±0.03	1608.91 [c] ±29.10
Autumn	*O. niloticus*	34.46 ±1.34	0.94 ±0.06	1423.18 ±28.40	21.62 [b] ±1.11	0.57 [b] ±0.04	1940.39 [b] ±27.24
	B. bayad	31.22 ±1.57	0.99 ±0.02	1175.65 ±26.44	21.00 [c] ±0.96	0.55 [c] ±0.02	1670.92 [c] ±34.99

Values are means ± S.E , n = (10-12) & w.t = wet tissue.

b= significant at $p < 0.05$, compared to fish (*O. niloticus*) collected from river Nile.

c= = significant at $p < 0.05$, compared to fish (*B. bayad*) collected from river Nile.

Table (4) : AchE activity (U/g w.t) in brain of the two examined fish species collected from two different water resources along Dakahlia province.

Seasons		Nile	Canal
Winter	*O. niloticus*	770.68 ±21.12	631.85 [b] ±23.99
	B. bayad	1115.69 ±24.30	769.42 [c] ±21.79
Spring	*O. niloticus*	1280.00 ±26.21	1046.08 [b] ±29.77 [c]
	B. bayad	2112.85 ±35.74	1310.83 ±37.45 [c]
Summer	*O. niloticus*	1603.36 ±49.63	1307.82 [b] ±44.54
	B. bayad	2489.52 ±51.74	1418.92 [c] ±40.30
Autumn	*O. niloticus*	1471.12 ±25.03	1181.86 [b] ±18.99
	B. bayad	1602.16 ±32.91	1351.02 [c] ±39.88

Values are means ± S.E , n = (10-12) & w.t = wet tissue.
b= significant at $p < 0.05$, compared to fish (*O. niloticus*) collected from river Nile.
c = significant at $p < 0.05$, compared to fish (*B. bayad*) collected from river Nile.

DISCUSSION

1. Trace metals bioaccumulation in fish:

As a result of industrial and agricultural activities, the aquatic systems have been increasingly contaminated by heavy metals ***(Gbem et al., 2001).*** Investigations of metal concentrations in fish have indicated their toxicity and tendency to bioaccumulate in fish tissues ***(Abd El-Nasser et al., 1996).*** In fish, there are two ways for metal bioaccumulation. The first is from water via gills and skin and the second way is via uptake of contaminated food ***(Barron, 1995 and Wong et al., 2001).***

In this field of study, other investigators added that the extent of metal bioaccumulation by fish tissues seemed to be governed by various environmental factors, such as seasonal variations ***(Hamed, 1998).*** In support to this, the present data described that the level of the detected metals (Cr, Ni, Zn, Cu) in the muscles of the tested fish (*B. bayad & O. niloticus*) tended to vary from season to another, where the minimal values were recorded during winter and the maximal ones were registered during summer. These observations are probably attributed to the seasonal variations in water temperature which affect the occurrence and behavior of most aquatic organisms and consequently their metabolic activities that are sensitive to water temperature ***(Haggag et al., 1999).*** Thus, any change in the temperature would affect the metabolism with subsequent influence on the rate of detoxification and accumulation of toxicants. It follows that the metabolic acceleration due to heat may accelerate metal accumulation in the fish tissues, while the decline in the metabolic rates of fishes when the environment becomes colder, may reduce the rate of incorporation of metals ***(Roennberg et al., 1990).*** Also, other studies suggested that the seasonal differences in fat accumulation could have considerable influence on metal bioaccumulation in fish, as lipids are known to have great affinity to combine with heavy metals ***(Cunningham and Tripp, 1973).*** From this study and the others mentioned above, the effect of season is proved as a factor in the metals accumulation in fish.

Other field studies indicated that fish accumulate metal in proportion to their levels in the aquatic environment and the extent of accumulated metals in fish tissues is higher than their

concentrations in the surrounding water ***(Miller et al., 1993).*** These findings goes in harmony with the present notion that the concentrations of the detected metals (Cr, Ni, Zn& Cu) in the muscles of the tested fish were much higher than that found in the water of the sampling sites (Nile & canal). In association, the canal fish tended to accumulate metals at higher extent than those of the Nile fish. This is mostly related to the fact that the canal fish exposed to higher metal (Cr, Ni, Zn& Cu) concentrations under natural field condition than the Nile fish, as a result of the increase in metal discharge from industrial and agricultural activities in the vicinity of El-Sahel canal.

2. Total protein content:

In various fish species, proteins are of importance as structural compounds, biocatalysts and hormones for control of growth and differentiations ***(Begum and Vijayaraghavan, 1996).*** So variation in fish proteins could be used as bioindicator for monitoring physiological status of the tested fish.

In the present study, examined fish at the two studied localities exhibited lowered protein values in both liver and muscle tissues during winter, followed by general increase during the rest of the year, particularly in summer. On studying this seasonal pattern of protein changes, it was suggested that protein decline during winter is probably a metabolic adaptation to the food shortage in the environment ***(White et al. 1986).*** During this period of inadequate food supply (starvation period), energy required for metabolic maintenance may be provided from utilization of protein reserves, which mainly accumulate in the muscle tissue ***(Haggag et al., 1999).*** Utilization of tissue proteins at winter may also related to the fish reproductive cycle, during which the development of gonads for spawning may proceed, causing reduction in the tissue protein reserves to overcome the condition of food shortage ***(White et al., 1986).*** In contrast to this, the increased protein reserves during summer, as shown in the present study and in other ones ***(Weatherly and Gill, 1987)*** seemed to be caused by the availability of food and abundance of phytoplankton with feeding increase during summer ***(Larsson, 1985).***

Beside the above mentioned seasonal influences, the present study investigated other environmental variables that directly affect fish protein reserves. Present data indicated marked depletion of total protein content in both liver and muscle of the fish samples collected from EL-Sahel canal when compared to those from the river Nile. This protein depletion could be attributed to the increased pollutants in the canal water, including hydrocarbons found in sewage wastes ***(Reinfelder et al., 1998)*** and heavy metals, found in the industrial and agricultural effluents ***(Zagloul et al., 2002).*** Bioaccumulation of pollutants, particularly metals, as (Cr, Ni, Cu & Zn) in fish may critically influence tissue protein level and fish quality ***(Reader et al., 1989).*** This may be explained as follows: exposure to metals as (Cu and Zn) may lead to high accumulation in the gills that may cause damage in their structure and a reduction in the rate of oxygen consumption, causing sharp reduction in the metabolic rate of fish and consequently decreased protein content in tissues. Moreover, the decreased tissue protein in fish living in polluted environment may be a result of the decrease in insulin level caused by metal toxicity, as in case of Cu and Zn ***(Zagloul et al., 2002).*** Insulin is known to have profound effects on the proteogenic pathways in fish ***(Ablett et al., 1981).*** It stimulates the inward cellular transport of amino acids, particularly in muscles, leading to interacellular accumulation of amino acids with subsequent increase in the protein content ***(Reda et al., 2002).***

3. Glycogen content and lactic dehydrogenase (LDH) activity:

In the present study, fish collected from the two studied sites tended to exhibit marked variations in the liver and muscle glycogen contents that seemed to occur inversely with the lactic dehydrogenase (LDH) activity, throughout the study period. Obtained variations were as follows: decreased glycogen content, but increased LDH activity in both liver and muscle of the studied fish during winter. This was accompanied by an increase in glycogen content with a decrease in LDH activity, during the rest of the year (particularly at summer). For explanation, it was suggested that: (1) The depletion of glycogen content with an elevation in the activity of LDH (the terminal enzyme in vertebrate anaerobic glycolysis) may be related to the fact that fish at winter

(starvation period) generally exhibit diminished Kreb's cycle enzyme activities with inhibition of the oxidative metabolism, that is compensated by stimulation of glycolysis, as reflected by the enhanced LDH activity ***(Seddon and Prosser, 1997).*** (2) The increased glycogen content with decreased LDH activity, particularly during summer, could be related to the increased feeding of fish species during summer. Thus, glycogen could be stored in the liver and muscle tissues ***(Haggag et al., 1999).*** These findings in all, agree with other authors ***(Sheridan and Mommsen, 1991)*** who mentioned that the glycogen stores can be modulated by natural environmental factors (as seasonal variations and food availability).

Next to this, other investigators ***(Almeida et al., 2001)*** have demonstrated that environmental pollution could be another factor determining tissue glycogen in fish. Of the important consideration in this regard, is the effect of metal pollution ***(Reingfelder et al., 1998).*** This has been considered by several authors ***(Reda et al., 2002)***, who reported that metal pollution produce a stress response due to their toxic action, with subsequent changes in carbohydrate metabolism, including glycogen depletion ***(Wenderlaar-Bonga, 1997).*** This agrees with other studies indicating that depletion of glycogen is considered as a biomarker for pollution by trace metals including, Cu ***(Zaghloul et al., 2002)***, Zn ***(Hilmy et al., 1987)*** and Cr ***(Alakal and Shamsi, 1986).***

In support to these findings, the present data revealed that fish collected from EL-Sahel canal showed depleted glycogen content with elevated LDH activity in both liver and muscle tissues compared to the river Nile fish. Thus, indicating enhanced glycogen breakdown as a stress response of fish to metal toxicity. Considering the biological consequences, glycogen breakdown indicates metabolic changes towards the formation of lactate which may have adverse effects on the fish. Accumulation of lactate may lead to metabolic acidosis and subsequent muscle fatigue with implications on escape capacities in fish ***(Gagnon, 2002).***

4. Acetylcholine esterase (AchE) activity:

In the present investigation, the recorded differences in the brain AchE activity of the tested fish at the selected sites and seasons was taken as a biomarker for the effect of the environmental variables on the studied fish. Obtained data indicated marked inhibition in the brain AchE activity of the fish collected from the polluted canal, as compared to those from the river Nile. Meanwhile, the mentioned enzyme showed lowered activity during winter, followed by higher enzyme activities during the rest part of the year, specially for fish at summer period.

Concerning the seasonal influence on the brain AchE activity, it is probably related to the fact that fish during search of food displayed sustained swimming pattern, as consequence of relatively high AchE activity ***(Szabó et al., 1991).*** Another point to be taken is the variation in fish behavioural habits (as feeding and swimming pattern) from season to another ***(Ferenczy et al., 1997).*** So, the variations in fish habits with seasons may induce differences in the enzyme activity.

On the other side, the inhibition in the brain AchE activity of the fish samples collected from the polluted area may be caused due to fish exposure to various pollutants, as agricultural pesticides and herbicides, as well as metals.

In this context, it has accepted that pesticides demonstrated acute toxicity in fish through inhibition of brain AchE activity ***(Lionetto et al., 2003).*** The same toxic effect was observed with herbicides in the work of ***(Hassanein, 2002)*** who reported that fish respond to herbicide stress by a reduction of brain AchE activity.

With regard to the effect of metal pollution, several studies indicated that heavy metals such as (zinc & mercury) can be considered as environmental inhibitors for AchE activity in fish ***(Lionetto et al., 2003).*** Inhibition of AchE causes accumulation of the neurotransmitter, acetylcholine, at the synapse and blocking the neurotransmission in the respiratory centers of the brain which leads to death ***(Soliman et al., 1995).*** In addition, fish with reduced AchE will be unable to maintain a normal upright position in the water, resulting in uncontrolled drifting, inability to protect themselves from predation or other hazards ***(Hassanein, 2002).***

5. The antioxidant defense components:

Fish exposure to certain organic and metal pollutants is thought to generate free radicals specially reactive oxygen species (ROS) with subsequent alterations in fish antioxidant defenses, such as glutathione ***(Varanka et al., 2001).*** Reduced glutathione (GSH; L-δ-glutamyl-L-cysteinyl-glycine) is probably the most abundant cellular thiol, that is detected virtually in all tissues but show, in general, high concentration in the liver (as a major organ for antioxidant defenses in fish ***(Otto et al., 1999).*** Apparently, GSH is important in protecting against deleterious effects of cell exposure to ROS by reacting with them to form glutathione disulphide (G-S-S-G). This antioxidant effect occurs spontaneously through GSH or may also catalyzed by glutathione transferase (GST; an enzyme for which GSH is utilized as substrate), ***(EL-Wakf, 1998).***

Recently, it was also demonstrated that fish antioxidant components (as GSH & GST) can be modulated by the natural environmental factors, as food availability and seasonal variations ***(Wilhelm et al., 2001).*** In support to this seasonal influence, the present data indicated marked reduction in the hepatic antioxidants (GSH and GST) of the two studied fish species during winter for each locality, but elevation in the same parameters was recorded during the rest of the year, particularly during summer. Such seasonal pattern; i.e. enhancement of antioxidants coinciding with high temperature period at summer was confirmed by ***Wilhelm et al., (1993)*** who provided explanation that warm temperature could increase oxygen consumption and aerobic metabolism by fish, which involve increased ROS production. Thus, representing a situation of oxidative stress associated with enhanced antioxidant capacity in fish.

Another factor to be mentioned concerning the antioxidant defenses is the effect of pollution, as evidenced in the present work by the increased hepatic antioxidants (GSH & GST) in the fish samples collected from EL-Sahel canal, as compared to those from the river Nile. These findings are in general agreement with a previous study by ***EL-Shenawy, (2002)*** who reported that the increased hepatic GSH could probably linked to fish exposure to various pollutants, particularly metals as (Cr, Ni, Zn & Cu) which

stimulate the antioxidant GSH, aiming at restricting pollutants toxicity ***(Wilhelm et al., 1997).*** Moreover, the increased GST activity could be related to the enzyme sensitivity to a large variety of pollutants, either organic or inorganic, as identified by several authors ***(Lemaire et al., 1994).*** It has also suggested that the increased GST activity in response to pollution could be related to fish adaptation to the continuous exposure to pollutants, even if their concentrations in the environment still meet acceptable sanitary conditions ***(Lopes et al., 2001).***

CONCLUSION

Data of the present study demonestrated that the investigated water locality at El-Sahel canal is more metal (Cr, Ni, Zn & Cu) polluted than Gizzerrhat El-ward area at the river Nile. Consequently, the tested fishes (*O. niloticus & B. bayad*) inhabiting El-Sahel canal locality showed worse physiological status than those from the Nile locality. This was indicated by the increased metal (Cr, Ni, Zn & Cu) accumulation in their muscles and the depleted total protein and glycogen contents in both the liver and muscle tissues. Inhibition of the brain AchE was also recorded, with elevations in the liver antioxidant defense agents (GSH and GST) to overcome the increased metal pollution in the canal locality. Obtained data also indicated that seasonal variations during the study period greatly affected the tested physiological parameters in the two fish species from the two water localities, reaching maximal during summer and minimal during winter.

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الملخص العربى

دراسات فسيولوجية على إثنين من الأسماك العظمية الشائعة، البلطى النيلى و البياض، المستوطنة لقناة الساحل و فرع دمياط من نهر النيل فى محافظة الدقهلية، مصر.

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فى هذه الدراسة تم اختيار نوعين من الأسماك العظمية الشائعة " البلطى النيلى و البياض" المستوطنة لمياه مختلفة الجودة (لقناة الساحل و فرع دمياط من نهر النيل) فى محافظة الدقهلية و ذلك لتقدير الأثار الناجمة عن التعرض للتلوث البيئى.

و قد أظهرت النتائج زيادة تراكم العناصر الثقيلة (الكروم، النيكل، الزنك و النحاس) فى أنسجة العضلات لأسماك لقناة الساحل و فرع دمياط من نهر النيل. و كذلك نقص مستوى البروتين و النشا الحيوانى فى أنسجة العضلات والكبد و نشاط الأسيتيل كولين إستريز فى أنسجة المخ . و على العكس من ذلك أظهرت النتائج زيادة فى نشاط إنزيم اللاكتك ديهيدروجينيز فى أنسجة العضلات و الكبد و كذلك مستوى مضادات الأكسدة (الجلوتاثيون و الجلوتاثيون ترانسفريز) فى أنسجة الكبد لأسماك المياه الملوثة بالمقارنة بأسماك نهر النيل (فرع دمياط).

و فى نفس الوقت أظهرت النتائج حدوث تغيرات موسمية فى جميع المعايير المقيسة فى أنسجة الأسماك محل الدراسة خلال مدة البحث و كانت هذه التغيرات أقل ما تكون خلال فصل الشتاء و أعلاها خلال فصل الصيف.

و قد أوضحت النتائج التأثير الضار لتلوث البيئة المائية بالمعادن الثقيلة على أسماك البلطى والبياض التى تؤكل محليا. و قد بدى هذا التأثير معتمدا على التغيرات الموسمية.