

SOME STUDIES ON THE EFFECT OF PREVAILING HEAVY METALS POLLUTION ON *ORECHROMIS NILOTICUS*

By

EI-Banna, S. and Samia M. EI-Hoshy*

Dept. of Aquatic Anim. Med. Agric. Res. Center, Anim. Health Re.s. Inst. Alex. Lab.

**Dept. of Food Hygiene Agric. Res. Center, Anim. Health Re.s. Inst. Alex. Lab.*

ABSTRACT

This study was carried out in 2 private fish farms at Janaklies, Alexandria Governorate to investigate the residual effect of heavy metals pollution on *Oreochromis niloticus* fish. 40 water samples and 40 *O. niloticus* fish samples ranging 150-200 were collected. Heavy metals were estimated in water and fish. The results were evaluated according to the international standards of WHO and FAO. The results showed a positive correlation between the concentration of heavy metals in fish and its concentration in water containing organic fertilizers. Some blood parameters, serum constituents and liver metabolites were not significantly decreased except serum glucose level and hepatosomatic index.

INTRODUCTION

Fish production has been considered one of the main sources of animal protein in developing countries including Egypt. Environmental pollution of water, soil and food becomes a threat to the existence of many plants and animals so it threatens the survival of human.

Heavy metals were recognized as toxic substances due to their low rate of elimination from the consumer body either human or animal. Fish absorb heavy metals from water through gills, skin and digestive tract. Industrial and agricultural discharges, such as coal, oil combustions, phosphate fertilizers, plastics and pesticides were considered the main sources of heavy metal pollutants of water **WHO (1992); Marcel Galab (1997) and Wafa Eleraky et al., (2003).**

The aquatic environment including streams, rivers and lakes act as a reservoir for foreign organic chemicals which may be toxic to man and fish (**Bend et al., 1980**).

This study was done to discuss the problem of water pollution with heavy metals and its residual effect on fish and on blood profile.

MATERIAL AND METHODS

Water and Fish samples were collected from two private farms during the period of fish growing, 1st farm received artificial feeding plus organic fertilizers and inorganic fertilizers, while, the second farm not received artificial feeding, organic or inorganic fertilizers, both two fish farms were stocked with *Oreochromis niloticus* fish and supplied with fresh water.

1-Determination of heavy metals:

a-Water samples:

Water samples were preserved by addition of 1 ml concentrated nitric acid per liter (1:1) until time of analysis. Water samples were analyzed according to method described by **APHA (1989)**. Water samples were filtered through 0.45 μ membrane filter. the filtrate was collected for measurement of lead, cadmium, Copper and zinc, in water using air/acetylene flame atomic absorption spectrophotometer (Unic CAM 696 AA spectrometer)

b-Fish samples:

Twenty fish samples of *Oreochromis niloticus* were obtained from each farm, the fish body weight ranged from 150 - 200 gram, each fish sample was represented by one gram offish flesh and digested according to the method described by **Finerty et al., (1990)**. The obtained solutions were analyzed by using Air/Acetylene fame atomic absorption spectrophotometer (Uni CAM 696 AA Spectrometer).

2- Determination of some blood and serum parameters:

Serum and blood samples were collected from 20 fish from the examined cultured *O.niloticus*. and 20 fish from control aquarium that received dechlorinated tap water. The blood were taken over a heparinized vials by severing the caudal peduncle of fish (**Dabrowska et al., 1989**).

The erthrocyte counts were determined by a double hacmocytoimeter using the method of **Wintrobe (1934)**.

Haemoglobin content was estimated by the method of **Van Kampen and Zifstra (1981)**, Serum protein was determined according to **Josphon and Gyllensward (1975)**, Serum glucose was estimated according to **Trinder (1969)**, Serum lipids were determined according to **Zollner and Kirch**

(1962), Liver glycogen was estimated according to **Handel (1965)**, while total lipids in liver were determined according to **Blight and Dyer (1959)**.

RESULTS AND DISCUSSION

Fish had been regarded as a desirable nutritional source of high quality protein, minerals and vitamins.

Recently, a great attention had been paid extensively to the possible danger of environmental pollution offish farms.

Lead is unknown to be essential for the maintenance of biological systems and the general view is to decrease exposure to lead as possible, lead retained in the organs for a long time, being a dangerous cumulative poison (**Allcroft. 1951**).

Permissible limit of lead in water according to **WHO (1984)** is 0.05 mg/L (Table. 1). accordingly, the level of lead in waier obtained from 1st farm and 2nd farm exceeded this limit. lead levels in the examined water samples are recorded in Table (2).

Water pollution with lead resulted in accumulation of lead in fish in concentrations exceeded the permissible limit intended by **Egyptian Organization for Standardization and Quality Control "E. O. S. Q. C." (1993)** (Table. 1). The highest concentrations of lead were recorded in fish collected from 1st farm followed by 2nd Farm. These figures are parallel to the levels of lead in the examined water samples.

The high lead findings in the present study may be due to these farms subjected to water from motor cars (high gasoline combustion), industrial discharges or agricultural discharges as super phosphate fertilizers, which considered one of the primary sources of lead poisoning in fish in Ugypt. These findings coincide with those reported by **Nabawi et al., (1987)**.

Cadmium is a accumulative poison. It has found in air. food. soil. plants and water (**Commission of the European Communities, 1978**).

Our results revealed that, the mean concentration of cadmium in water of the 2 farms, exceeded the permissible limit of **WHO (1984)** (0.001 mg/L). Similarly, the mean tissue cadmium levels in fish exceeded the permissible limits indicated by **FAO / WHO (1992)** (0.05 mg/L).

A correlation between cadmium levels in water and tissue residues in fish were observed, where the highest concentrations of cadmium in water and in fish were existed in 1st farm that received organic fertilizers followed by 2nd farm. The high cadmium level in water and fish should be considered with a great care because many studies revealed that cadmium had a

significant role in the incidence of some diseases e.g . diabetes mellitus (**Merali and Sigal, 1977**).

Copper is an essential element, having a role in formation of erythrocytes, release of tissue iron and development of bone, CNS and connective tissue. **WHO (1984)** set a guide line value for Copper in whaler to be 1mg/ L. Accordingly Copper level in examined water samples obtained from 1sl farm exceeded tins limit, while the copper not detected in 21KI farm. The high water Chopper level in 1sl farm was parallel to a high fish copper level. At the same time this elevated level of 'copper is still under the permissible limit as shown in Table (1).

The estimated levels of zinc in water of 2 farms (Table. 2) were under the permissible limits which intended by **WHO (1984)** (Table. 1) also (table. 2) indicated that, tissue zinc residue in fish is also under the accreted limit (50 ppm) indicated in Table (I). However there, was a correlation between zinc concentration in water and its residues in fish.

The obtained results of Chopper and zinc in Table (2) revealed that copper and zinc accumulation in fish w'-ere not so remarkable. These findings coincided with **Heth et al., (1966)** and **Clark (1989)** who stated that although fish from areas known to be contaminated, contain higher concentrations of Chopper and zinc than those from uncontaminated areas. Copper and zinc do not generally accumulate in food chains and do not make any hazard toxicity to man.

Table (3) cleared that, the erythrocyte counts, haematocrit value haemoglobin content in blood .

Niloticus fish reared in polluted water was non significantly decreased in comparing with fish reared in fresh water, this may be attributed to haemolysis of red blood cells caused by water compounds (**Mazher et al., 1987**).

Also Table (3) showed that the serum protein and lipids of polluted O.N. fish were non significantly decreased. Our results nearly similar to **Kandil (1987)**.

Glucose concentration was significantly increased in polluted O.N. fish. This may be attributed to the high activation of glycolysis process (conversion of liver glycogen to blood glucose) and tins raises the concentration of glucose in the blood. Similar observation were also found by **Hilmy et al., (1987)**.

Table (3) showed that the values of protein, lipid, glycogen and water content in the liver of O.N. fish reared in polluted water were non significantly decreased. The same observations were also found by **Kandil (1987)**. While hepatosomatic index was increased significantly, this may be attributed to increasing size and weight of the liver due to concentrated of heavy metals.

In the present study the elevated values of some heavy metals in water and muscular tissues of *O. niloticus* resulted from environmental pollution. Therefore the preventive measures intended for minimizing the pollution of water and raw Fish with such metals should be undertaken including sanitary protection of surface water from these pollutants, minimizing the use of phosphates for land fertilization as possible and regular analysis of water and raw fish for these pollutants and the results should be evaluated according to the international guide lines as a fruitful advice to delay pollution of water.

REFERENCES

- Alleroft, R. (1951):** Vet. Rec. 63 - 583. (C.F. Clake & Clake. Vet. Toxicology 4th Ed.).
- APHA, American Public Health Association (1989):** Standard methods for the examination of water and wastewater, 17th Ed. APHA. Washington. D. C.20005.
- Bend, J. R.; James, M. O. and Dritchard, J. B. (1980):** Aquatic toxicology in introduction to environmental toxicology. Edited by Guthrie, E. I7, and Perry, J.J. First Edition, Elsevier, New York. (172 - 185).
- Bligh, E.G. and Dyer, W.T. (1959):** A rapid methods of total lipid extraction and purification. Can. J, Biochem. Physiol, 37: 911:917.
- Clark, R. B. (1989):** marine pollution. Oxford Science Publications, 2nd Ed. Larendom Press. Oxford.
- Commission of the European Communities (1978):** Criteria (dose/effects relationships) for cadmium. Xford. Pergmon Press.
- Dabrowaska, II.; Burgdorf, K. and Gunther, K.(1989):** Interaction between dietary protein and magnesium level in tilapia (*Oreochromis niloticus*). Aquaeult., 76:277-291.
- Egyptian Organization for Standardization and Quality Control "E. O. S. Q. C. " (1993):** Maximum residue limits for heavy metals in food. Ministry of Industry. No. 2360 / 1993. p.p. 5. Cairo Egypt.
- El-Nabawi. A.; Heinzow, B. and Kruse, II. (1987):** As, Cd, Cu. Pb, Hg and Zn in fish Iron} the Alexandria region, Egypt. Bull. Environ. Contam. Toxicol 39: 889-897.
- FAO/WHO (1992):** Codex alimentarius Commission, Standard programme Codex Committee oil food additives and contaminates 2411 Session, Hague. 23 - 28., March. 1992.
- Finerty, M. W.; Madden, J.D.; Feagly, S. E. and Grodner, R. M. (1990):** Effect of environs and seasonably on metal residues in tissues of wild and pond raised Cray fish in Southern Louisiana. Arch Environ. Contain. Toxicol. 19: 49.
- Handel, E.V. (1965):** Estimation of glycogen in small amount of tissue. Ana.

Biochem., H: 156-265.

Lletli, D. A.; Beeker, W. M. and Iloekstar, W. G. (1966): Effect of calcium, phosphorus and zinc on zinc 65 absorption and turnover in rats fed semi-purified diets. *J. of Nutr.*, 88(3): 331 - 337.

Hilmy, A.M.m El-Domiatty, N.A.; Mousa, E.I. and Abu-Samra. W.E. (1987): Variation in the blood of juvenile *Clarius lazera* subsequent to acute and subacute copper exposure. *Bull. Inst. Ocean. And Fish.* 13: 65-77.

Kandil, A.E. (1987): Physiological studies on the grass carp (*Cenopharyngdon idelia*) under environmental pollution. Ph.D., Thesis. Cairo Univ., 1 86pp.

Mazhar, E.M.; Ashry, A.M. and Khadry, S.M. (1987): Effects of environmental pollution by mercury on blood parameters of the Nile catfish (*Clarius laizera*). *Proc. Zoo. Soc. A.R. Egypt*, 13: 247-258.

Marcel, F. Galab. (1997): Clinicopathological studies on fish exposed to some environmental pollution in Manzala lake. Ph.D. Thesis, Fac. Vet., Suez Canal University.

Merali, Z. and singhal, R. L. (1977): Long-term effects of orally administered cadmium on neonatal rats. *Proc. Of the 1st Intern. Congress on Toxicology. Held March. 30 - April 2, hi Toronto, Canada.*

Saleh, H.H. (1982): Fish liver as indicator of aquatic environmental pollution. *Bull. Inst. Ocean. And Fish.* 8: 69-79.

Steel, R. G. D. and Torrie, J. H. (1980): Principles and procedures of statistics. McGraw Hill Book Co., New York.

Trinder, P. (1969): Determination of glucose in blood serum or plasma by enzymatic colorimetric method. *Ann. Clin. Biochem.*, 6: 24pp.

Van Kampen, E.J., and Zijfstra, W.C. (1981): Recommendation for haemoglobinometry in human blood. *Clin. Chem. Acta.* 6: 338pp.

Wafaa Eleraky; Saleh, G. and Dief, M. (2003): Some studies on heavy metal pollution in water and fishes. *Egypt. J. basic Appl. Physiol.* 2 (2): 1 85 - 200.

Wintrobe, M.M. (1934): Variation in the size and haemoglobin content of erythrocytes in the blood of variation vertebrate. *Folia. Haematologica.* 51: 32-49.

World Health Organization (1984): Guidelines for drinking water. WHO, Geneva.

World Health Organization (1992): Environmental health criteria. No. 134. Cadmium. WHO. Geneva.

Zollner, N. and Kirsh, K. (1962): Uber die quantitative besliming von h'poiden (Mikromethod) mitles der vielen naturlichen lipoden (alien bekannten plasma h'poiden) gemeinsamen sullbphosovanillin-reaktion. *Z. ges. Exp. Mod.*, 135: 545-561.

Table (I): Recommended international levels of heavy metals in water and fishes.

Metal	Permissible Limits in water	References	Permissible Limits in fish	Country and References
Lead (Pb)	0.050 p.p.m	WHO (1984)	0.1 mg/Kg	Egypt: Egyptian Organization for Standardization and Quality Control 'E. O. S. O. C. ' (1993).
			0.5 p.p.m	FAO/WHO (1992)
			2.0 mg/Kg	England: (MAAFF.1979)
			4.0 µg/g	NewZeland. In : Julshamn
Cadmium (Cd)	0.005 p.p.m	WHO (1984)	0.05 p.p.m	FAO / WHO (1992)
			0.1 mg/g	Egypt: 'E. O. S. O. C. ' (1993).
			1.0 µ/g	Spain: Boletin Oficial del Estado (1991). In: Schuhmacher and Domingo (1996).
Copper (Cu)	1.000 p.p.m	WHO (1984)	20.0 p.p.m	Food Stuffs, Cosmetics and Disinfectants (1972).
			20.0 µg/g	Spain: Boletin Oficial del Estado (1991). In: Schuhmacher and Domingo (1996).
Zinc (Zn)	5.000	WHO (1984)	50.0 p.p.m	Food Stuffs, Cosmetics and Disinfectants (1972).
			50.0 mg/Kg	England: Food Standard Committee "FSC." In: Eromosele et al. (1995)

Table (2): Heavy metals in water and *O. niloticus* Fish (Mean ± S.E)

Sources of samples	Lead		cadmium		Copper		Zinc	
	water	Fish	water	Fish	water	Fish	water	Fish
	p.p.m		p.p.m		p.p.m		p.p.m	
Fish farm received fertilizers	0.71 [^] ± 0.20	2.14 [^] ± 0.35	0.56 [^] ± 0.06	0.484 [^] ± 0.04	1.3 [^] ± 0.06	2.6 [*] ± 0.3	3.5 [*] ± 0.3	7.55 [*] ± 1.2
Fish farm Not received fertilizers	0.56 [^] ± 0.3	1.30 [^] ± 0.06	0.03 [^] ± 0.07	0.38 [^] ± 0.04	ND	0.85 ± 0.13	0.23 [*] ± 0.15	0.70 [*] ± 0.20

ND : Not determined

Increasing

* : Decreasing

Table (3): Blood parameters, serum analysis and liver metabolites of *O. niloticus* fish reared in polluted water.

Item	Fresh water (Control)	Polluted water
Bood parameters:-		
Erythrocyte counts million/m ³	1.692±0.115	1.299±0.130
Haematocrit value %	37.99±1.39	29.86±1.90
Haemoglobin content gm/100ml	6.91 ±0.33	4.33±0.79
Serum analysis:-		3.33±0.76
Protein (gm/100ml serum)	4.99±0.88	2.66±0.36
Lipid (gm/100ml serum)	3.21±1-0.66	95.50±<8.99
Glucose (mg/100 ml serum)	81.66±6.33	
Liver metabolites:-		1.8.66±1,40
Protein (gm/100gm tissue)	19.3.1 ±1.30	2.33±0.44
Lipid (gm/100 gm tissue)	3.99±0.33	2.66±0.55
Glycogen (gm/100 gm tissue)	3.33±0.18	51.13±2.10
Water content (gm/100 gm tissue)	56.19±1.9	
Hepatosomatic index	1.30±0.31	2.88±3.36

الملخص العربي

دراسة تأثير تلوث المياه بالمعادن الثقيلة على اسماك مزارع البلطي النيلي

صلاح البنا
سامية محمد الحوشى
مركز البحوث الزراعية - معهد بحوث صحة الحيوان - قسم أمراض الأسماك - معمل الإسكندرية
مركز البحوث الزراعية - معهد بحوث صحة الحيوان - معمل جمرک الإسكندرية

أجريت هذه الدراسة في مزرعتين تابعتين للقطاع الخاص في منطقة جناكليس

التابعة لمحافظة الإسكندرية لدراسة مشكلة تلوث المياه بالمعادن الثقيلة و مدى تأثير هذه الملوثات على أنسجة تلك الأسماك.

تم تجميع 40 عينة من الماء من الزرعتين و في نفس الوقت تم تجميع 40 عينة

من اسماك البلطي النيلي عددها 40 سمكة يتراوح وزن كل منها بين 150 - 200 جرام من تلك المزارع.

بعد تحضير العينات الخاصة بالمعادن الثقيلة و تجهيزها تم اختبار تلك الملوثات

في المياه و الأسماك.

نتائج هذه الدراسة تم تقييمها على حسب المعدلات العالمية الموضوعه و المحددة

بمنظمة الصحة العالمية و منظمة الأغذية و الزراعة.

أوضحت نتائج هذه الدراسة وجود ارتباط إيجابي بين معدل ترسيب المعادن الثقيلة

في عضلات البلطي النيلي و معدل تركيزها في المياه و أيضا أوضحت نتائج هذه الدراسة

زيادة معدل تركيز المعادن الثقيلة في الأسماك و الماء المزود بالمخصبات الطبيعية.