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EFFECT OF CADMIUM, COPPER AND LEAD CONTAMINATION ON GROWTH PERFORMANCE AND CHEMICAL COMPOSITION OF NILE TILAPIA (*Oreochromis niloticus*).

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ABSTRACT

Bolti (*Tilapia niloticus*) fish were grown in polluted water with one of the heavy metals namely cadmium, copper or lead. The concentrations of the tested metals in fresh water environment were 0.1 and 0.2 mg/l Cd, 0.25 and 0.50 mg/l Cu and 0.1 and 0.2 mg/l pb. An experiment of 56 days was performed to study the effect of the tested sublethal levels of each of cadmium, copper and lead on growth performance, body composition, residual concentration and muscular and abdominal area of fish body. A set of 140 fish with 10 g average initial body weight was divided into 14 similar groups in glass aquaria containing 75 liter of water in each. The treatments and the control were allocated to the experimental groups in doplicates. The fish were fed on a commercial diet containing about 25% crude protein at a rate of 3% of the body weight daily during the experimental period. The results showed that cadmium, copper and lead caused a significant reduction in the average weight gain (AWG), specific growth rate (SGR), survival rate (SR), feed conversion (FC), protein efficiency ratio (PER), and carcass dry matter and protein percentages. Residual concentration increased in fish tissues. Fish muscles area decreased at the two levels studied of each pollutant as compared with the control.

Keywords: Bolti-fish-Cu-Cd-Pb-performance-residues-muscular area.

INTRODUCTION

Environmental pollution represents a major problem in the world, specially in the less developed countries. Egypt is one of these countries which suffer from biosphers pollution (air/soil and water). Pollutants in water including heavy metals, pesticides, hydrocarbons and sewage wastes may accumulate in fish tissues and organs causing severe healthy problems to the consumers. Cadmium, copper and lead are toxic heavy metals, which widely used in many industries. Presence of cadmium and lead in water reduces growth, spawning activity and survival rate of different species of fish (Liloyed and Alabaster, 1980 ; Sastry and Subhodra, 1985 and Wang, 1993). Copper compounds are commonly used to treat some fish diseases in addition to its role for maintaining the normal biological activity of many enzymes. Yet, high levels of copper in water are toxic to fish. Sublethal dose of Cu is ranged from 0.02 to over 10 ppm depending on some factors such as water hardness, fish species, duration of exposure and stage of life (Draz *et al.*, 1993).

Sublethal levels of copper were found to depress feed intake and weight gain and decrease the concentration of ascorbic acid in the liver of rainbow trout (Stiff, 1977). The objective of this study was to investigate the effects of cadmium, copper and lead in fish-rearing water on growth performance, residues in different parts of fish body, chemical composition, and muscular and abdominal cavity area of *Tilapia niloticus*.

MATERIALS AND METHODS

This work was carried out in the Central Laboratory for Fish Research El-Abbassa, Sharkya Governorate. A total of 140 fingerlings of *Oreochromis niloticus* with 10 g initial body weight were used in this study. The fish were taken from the stock of El-Abbassa experimental earthern ponds. The fish were divided into 14 similar groups in glass aquaria containing 75 L of water with 10 fish in each. The groups were distributed into the experimental treatments in duplication. The fish in all groups were fed on a commercial diet (a roduct of Atmida Co. Dakahlya Governorate) at a rate of 3% of the live body weight daily for 8 weeks experimental period. Composition and proximate analysis of the fed diet are given in Table (1).

Table (1): Composition and chemical analysis of the diet used.

Items	%
Ingredients:	
Fish meal	12.3
Soybean meal	24.0
Yellow corn	40.0
Wheat bran	15.3
Sunflower oil	7.4
Vitamins & minerals mixture (1)	1.0
Chemical analysis (on DM basis):	
Dry matter	90.0
Crude protein	27.78
Ether extract	7.45
Ash	13.33
Crude fiber	3.89
Nitrogen free extract	47.55
ME (Kcal/kg)*	3513.05
P/E ratio (mg/kcal)**	79.08

* Metabolizable energy calculated using the value of 3.49, 8.1 and 4.5 kcal/g. for carbohydrate, fat and protein, respectively according to Pantha (1982).

** Protein/energy ratio (mg crude protein/k ME).

(1) Vitamin and mineral mixture (product of atmida Co. Dakahlya Governorate). Each kg contain: 6 million IU Vit. A ; 1.2 millon IU Vit. D_3; 6000 mg Vit. E. ; 1000 mg Vit. K_3; 400 mg Vit. B_1 ; 2000 mg Vit. B_2; 800 mg Vit. B_6 ; 4.8 mg Vit. B_{12}; 20 mg Biotin; 40 mg panthothenic acid ; 14000 mg Nicotinic acid ; 4800 mg Folic acid ; 2800 mg copper ; 160 mg Iodine ; 32000 mg Manganese ; 12000 mg Iron ; 20000 mg Zinc ; 60 mg Selenium and 50000 mg anti-oxidant.

On the basis of LC50 test which was conducted before the start of the experiment, two sublethal levels from each of cadmium, copper and lead were selected to study their effects on the growth performance, chemical composition and areas of muscullar and abdominal cavity of the fish. The two selected levels were 0.2 and 0.1 mg/l for cadmium, 0.5 and 0.25 mg/l for copper and 0.2 and 0.1 ng/ml for lead. Fish were weighed weekly and the amount of food quantity was adjusted accordingly. The daily ration was introduced at 2 equal meals at 8 a.m. and 2 p.m. Dechlorinated tap water was used to change one third of the water in each aquarium every day. Samples of water were taken weekly from each aquarium to determine water quality parameters (Abdelhamid, 1996) including temperature (via a thermometer), pH (using Orient Research Model 201-pH meter) and dissolved oxygen (means an Oxygen-Meter model 9070). Light was controlled by a timer to provide a 14-h light: 10 h dark as a daily photoperiod. Fish samples (4-5 fishes) from each group were obtained at the end of the experiment for chemical analysis and measuring areas of muscular and abdominal cavity. Chemical analysis for dry matter (DM), crude protein (CP), ether extract (EE) and ash in the diet used and in fish body (besides crude fiber, CF in the diet) were carried out according to A.O.A.O. (1980), while cadmium, copper and lead residues in different fish parts were estimated by atomic absorption spectrophotometry following the method of Capar (1977). The fish were examined also for infiltration / muscular areas using Echo Scan (Hs/s) Ultrasonic Diagnostic Instrument, Budapest Remeny Co.. The obtained numerical data were statistically analysed using SPSS (1997) for one-way analysis of variance. When F-test was significant, least significant difference was calculated (Duncan, 1955).

RESULTS AND DISCUSSION

1. Water quality parameters:

The most important physico-chemical parameters of tap water used in the experiment are shown in Table (2). Data in this table indicated that the values obtained lie in the acceptable range required for normal growth of tilapia (Abo-Salem *et al.*,1992).

Table (2): Some important measured physico-chemical parameters of water.

Parameter	Value
Water temperature (°C)	25.00-26.00
pH value	7.00-8.40
Salinity (%)	0.02-0.03
Dissolved oxygen (ppm)	5.00-5.70
Cadmium (ppb)	0.001
Copper (ppb)	0.003
Lead (ppb)	0.002

2. Growth performance:

Data concerning average weight gain (AWG), average daily gain (ADG), specific growth rate (SGR) and survival rate (SR) are presented in Table (3).

The results revealed that cadmium, copper and lead reduced growth significantly ($P<0.05$) at the tow levels studied. The negative effect on growth was more pronounced at the high level of either cadmium, copper or lead (Fig. 1). Exposure of *Nile Tilapia Orechromis niloticus* to cadmium at levels of 0.2 and 0.1 ppm decreased AWG by about 30.2 and 20.81%, respectively; while copper reduced AWG by about 27.1 and 16.6%, respectively; whereas lead reduced AWG by about 33.3 and 30.1%, respectively compared with the control. There were significant ($P<0.05$) differences in AWG between the control group and the other treated fish groups. However, the differences between the low levels of either cadmium or lead and the high levels were not significant ($P>0.05$) . Specific growth rate was also found to be the highest in the control group (Fig. 2) as in AWG and ADG and differ significantly ($P<0.05$) from all other treated fish groups. Survival rates were lower in all fish groups exposed to either cadmium, copper or lead than the control group (Fig. 3). The reduction in SR in treated groups ranged from 14.5 to 24.5% as compared to control. The differences in SR between the control group and the other treated groups were significant ($P<0.05$).

Table (3): Effect of different concentrations of cadmium copper and lead on growth and survival rate of *Tilapia niloticus* .

Treatment	AWG (g/fish)	ADG (g/fish/day)	SGR (%/day)	SR (%)
Cad. 1 (0.2 mg/l)	15.50^{c}	0.27^{c}	0.94^{a}	75.50^{a}
Cad. 2 (0.1 mg/l)	17.60^{b}	0.31^{b}	0.98^{a}	80.60^{a}
Cu 1 (0.5 mg/l)	16.20^{b}	0.28^{b}	0.95^{a}	78.70^{a}
Cu 2 (0.25 mg/l)	18.53^{b}	0.33^{b}	0.98^{a}	83.30^{a}
Pb 1 (0.2 mg/l)	14.81^{c}	0.26^{c}	0.91^{a}	78.80^{a}
Pb 2 (0.1 mg/l)	15.53^{bc}	0.27^{bc}	0.95^{a}	85.50^{a}
Control	22.23^{a}	0.39^{a}	1.20^{b}	100.0^{b}

a,b,c Means in the same column bearing the same letter do not differ significantly at 0.05 level.

1- Average weight gain = Final weight (g) – Initial weight (g)
2- Average daily gain = Final weight (g) – Initial weight (g)/Experimental period (d)
3- Specific growth rate = (Ln. Final weight – Ln. Initial weight/time (d) x 100
4- Survival rate % = (Total number of fish at the end of the experiment / total number of fish at the start of the experiment) x 100

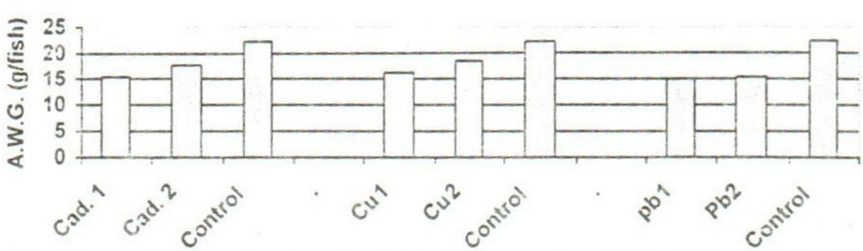


Fig. (1): Effect of cadmium, copper and lead on average weight gain of *O. niloticus*.

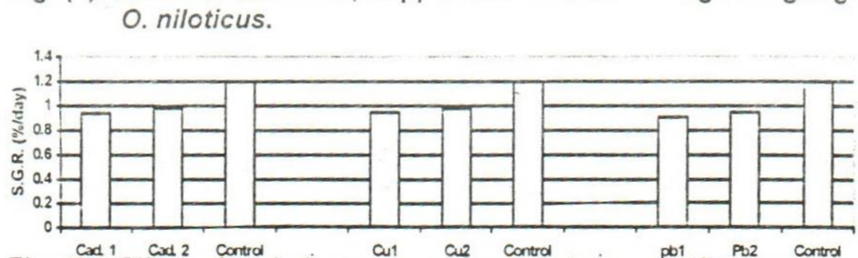


Fig. (2): Effect of cadmium, copper and lead on specific growth rate of *O. niloticus*.

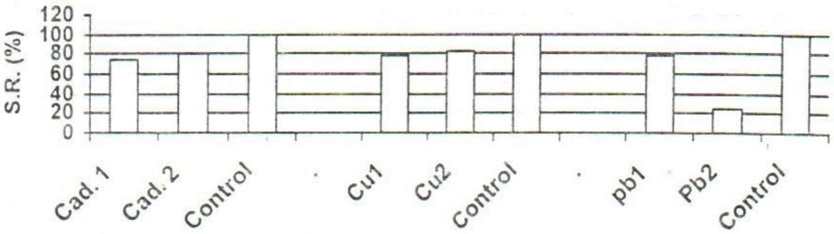


Fig. (3): **Effect of cadmium, copper and lead on survival rate of Nile tilapia *O. niloticus***

The present results were in agreement with those found by several workers. Mincken and Benoit (1971) reported a decrease in growth and survival rate of brook trout, when the fish treated with copper at level ranged from 1.9 to 32.5 ppb. Also, Stiff (1977) obtained an adverse effect of copper sulphate at 0.1 ppb on growth of rainbow trout. Moreover, Draz *et al.* (1993) recorded a significant decrease in final body weight and length of Nile Tilapia (*O. niloticus*) treated with 0.25 ppb copper sulphate. In addition, Hutchinson and Sprague (1985) reported a reduction in growth and reproduction of flag fish (*Jardanella floridae*) exposed to 29.5 ppb of copper chloride. Data in the literature obtained by many authors worked on different fish species revealed that cadmium and lead had also a negative effect on growth, survival and egg development of rainbow trout (Liloyed and Alabaster, 1980), on growth of catfish (Sastry and Subborhdra, 1985) and on body weight of tilapia (Wang, 1993).

On the contrary, the present results were in disagreement with the results obtained by some other investigators, who found that copper had no effect on body weight of sun fish (Causert, 1962), on growth performance of yellow perch (John and Frank, 1974) or on growth and reproductive of flathead minnows (Mount and Stephen, 1967). Eaton (1974) also did not observe any negative effect of cadmium chloride at 90 ppb on growth and survival of blue gill fish. The differences between the present results and those found by some workers on the effect of cadmium, copper and lead on growth may be due to the differences in the concentrations used from these metal compounds, the differences among fish species and environmental conditions. The negative effect of cadmium, copper and lead on growth of *Tilapia aureas* may be attributed to the effect of these heavy metals on the enzyme activities and metabolic processes. Several workers supported this assumption, since Bertolla *et al.* (1978) have shown an impairment of carbohydrate metabolism in *Clarias batrachus* and *Tilapia mossambicus*, when they exposed to cadmium. Belilies (1975) attributed the harmful effect of cadmium to its binding to the sulphhydryl enzymes, specially dehydrogenase. Other workers found that cadmium inhibited or decresaed the activity of some enzymes in the tissues of fresh water catfish. So, Sastry and Subborhodra (1985) reported that cadmium decreased the activity of alkaline phosphatase (in the liver, kidney and intestine), xanthine oxidase (in the liver and muscles) and glutamate dehydrogenase in the intestine and gills. They recorded also an inhibition of acid phosphatase activity and hexokinase in the liver, kidney, ovary and gills.

3. Feed efficiency and protein utilization:

Feed and protein utilization expressed as feed conversion ration (FCR) and protein efficiency ratio (PER) for *Tilapia niloticus* are given in Table 4.

Table (4): Effect of different concentrations of cadmium, copper and lead on efficiency of feed and protein utilizations by Nile tilapia.

Treatment	FCR(1)	PER(2)
Cad. 1 (0.2 mg/l)	2.10[b]	1.81[b]
Cad. 2 (0.1 mg/l)	2.21[b]	1.85[b]
Cu 1 (0.5 mg/l)	2.11[b]	1.89[b]
Cu 2 (0.25 mg/l)	2.30[b]	1.80[b]
Pb 1 (0.2 mg/l)	2.20[b]	1.77[b]
Pb 2 (0.1 mg/l)	2.20[b]	1.91[b]
Control	1.83[a]	2.08[a]

a,b,c Means in the same column bearing the same letter do not differ significantly at 0.05 level.
(1) Feed conversion ratio = Dry food consumed (g)/ Live weight gain.
(2) Protein efficiency ratio = Live weight gain (g)/Protein intake (g)

The data indicated that FCR and PER were poorer in fish groups grown in cadmium, copper and lead polluted water than in the control fish groups (Fig. 4&5). This could be explained by the poorer growth obtained from the treated fish groups as compared to the control fish group.

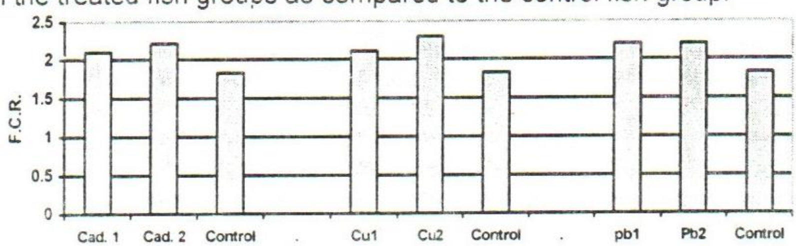


Fig. (4): Effect of cadmium, copper and lead on feed conversion ratio in Nile tilapia (*Creochromis niloticus*).

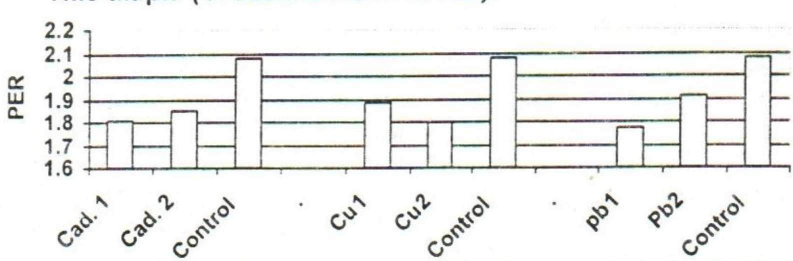


Fig. (5): Effect of cadmium, copper and lead on protein feed efficiency ratio in Nile tilapia (*Oreochromis niloticus*).

4. Body composition:

Values of dry matter (DM), *crude protein (CP), ether extract (EE) and* ash in body of Nile Tilapia are summarized in Table (5). The results showed that the control fish had the highest DM and CP values and the lowest EE and ash percentages. Within the two levels of each heavy metal, percentages of CP decreased as the level of the heavy metales increased, while the values of EE and ash increased with increasing the levels of these heavy metals.

Table (5): Effect of cadmium, copper and lead on the body composition of Nile Tilapia (on dry matter basis).

Treatment	DM, %	CP, %	EE, %	Ash, %
Cad.1 (0.2 mg/l)	22.11^{a}	58.85^{ab}	20.10^{b}	21.10^{b}
Cad. 2 (0.1 mg/l)	23.15^{b}	58.85^{ab}	19.90^{ab}	21.11^{b}
Cu 1 (0.5 mg/l)	24.40^{bc}	59.20^{b}	20.31^{b}	20.20^{b}
Cu 2 (0.25 mg/l)	21.51^{a}	60.20^{b}	20.20^{b}	20.00^{ab}
Pb 1 (0.2 mg/l)	22.13^{a}	60.10^{b}	20.20^{b}	19.80^{ab}
Pb 2 (0.1 mg/l)	25.31^{bc}	60.50^{b}	19.50^{b}	20.10^{b}
Control	26.10^{b}	65.22^{c}	16.50^{a}	18.10^{a}

a,b,c Means in the same column bearing the same letter do not differ significantly at 0.05 level.

The present results agree with those given by Abdelhamid and El-Ayouty (1991) who reported that lead pollution of fish rearing water increased ether extract and ash contents as well as lead residues in fish body, but it decreased the protein percentage in catfish.

These results were in agreement with those reported by Noaman *et al.* (1992), who found that CP content decresaed in *Tilapia nilotica* grown in water polluted with cadmium, copper and lead as compared with the untreated fish. Sastry and Gupta (1979) also found that muscular protein decresaed when the fish were exposed to cadmium chloride at 6.8 ppm, which was in agreement with the present results. The resultes obtained in this study dealt with ash content were similar to those reported by Noaman *et al.* (1992), who indicated that the higher content of heavy metal in water environment had led to the higher ash content in fish body parts. Data obtained here suggested that heavy metals might stimulate lipids deposition in the body of tilapia on the account of the fish protein. Some workers found a similar trend as mentioned previously, but others reported different results, where they found that the fish protein contents increased in heavy metal treated fish groups (Hassan, 1964 ; Merlini, 1978 and Khadre, 1990).

5. Heavy metal residues:

Table (6) showes the residues of the studied heavy metals in different parts of tilapia. Data indicated that the minimum concentrations of cadmium, copper and lead were obtained in the body of untreated fish groups (control). The highest metal residues were found in the head of fish groups exposed to the highest level of copper. Increasing the concentration of either cadmium, copper or lead caused a significant increase in the residues from these metals in all studies parts of *O. niloticus*. The lowest values of cadmium, copper and lead were observed in the muscles of the fish. There were significant differences in cadmium residues in fish parts between fish groups exposed to the high and low levels of cadmium, while the values of copper and lead residues between fish groups treated with the two levels of copper and lead were found to be no significantly different ($P<0.05$). Data in the literature showed generally that heavy metals concentrate in fish head, specially in gill tissue and/or in the viscera, specially in the liver and kidney (Schroeder, 1965 and Abo-Salem *et al.*, 1992). Abdelhamid *et al.* (2000) confirmed the present results, since they found that the lowest bioaccumulation factors of Cu and Pb were calculated in fish muscles than in the gills or the viscera. They added that pb residues were higher than those of Cu in the fish muscules.

Table (6): Residues (mg/g fresh weight) of cadmium, copper and lead in different parts of Nile tilapia.

Treatment	Head	Viscera	Muscles
Cad.1 (0.2 mg/l)	0.012	0.016	0.011
Cad. 2 (0.1 mg/l)	0.050	0.090	0.05
Cu 1 (0.5 mg/l)	0.015	0.018	0.07
Cu 2 (0.25 mg/l)	0.011	0.017	0.04
Pb 1 (0.2 mg/l)	0.013	0.019	0.06
Pb 2 (0.1 mg/l)	0.08	0.018	0.03
Control	0.001	0.003	0.001

On the other hand, Calamari and Marchett (1977) and Popoutsoglou (1991) reported that the highest residues of cadmium, copper and lead were found in the muscle tissues. These conflicting data may be due to several factors such as the level of heavy metals, used fish species, the period to which fish exposed and other environmental conditions.

6. Infiltration / muscular areas:

The fish were examined for infiltration muscular areas using Echoscan H5/s Ultrasonic Diagnostic Instrument (Budopest Remeny Co.). This test also presented the variations among the tested groups as illustrated in Figures (5 to 13), where Zm: the number of the actually zoom (zoom 2 to 8 cm), st: the number of the actually displayed memory is shown, PR: automatically identifies the type of connected probe (eg. L5 is Mhz liner prob), Ng: indicates the measure of actual near gain in a range of 1-16, FG: indicates the measure of the actual acaustic power, DP: active only with sector probe and indicates the measure of damping of piezo crystals of the probe, PA: picture averaging PP: post processing, DIR: direction of scanning, FR: fast or slow scanning, HI: high scanning rate, F: the various measurements can be selected, N: not active and ID: identifying characters.

This test presented the variations among the tested fish groups where Figs. 5, 8 and 11 presented the control with larger muscular (white) area than in Figs. 6 and 7 fish treated with cadmium, Figs. 9 and 10 for fish treated with copper and Figs. 12 and 13 for fish treated with lead. The increased abdominal (black) cavity area is proportional to the level of cadmium, copper and lead.

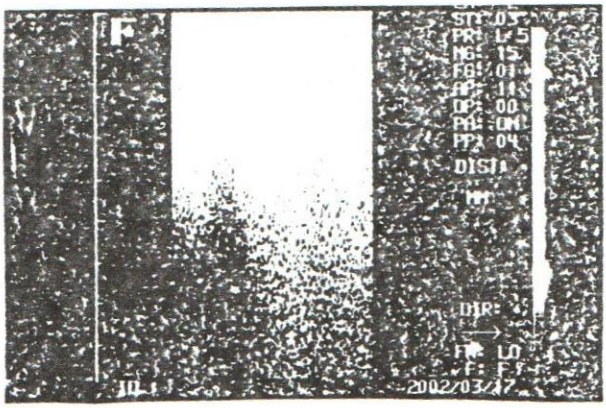


Fig. (5): Control

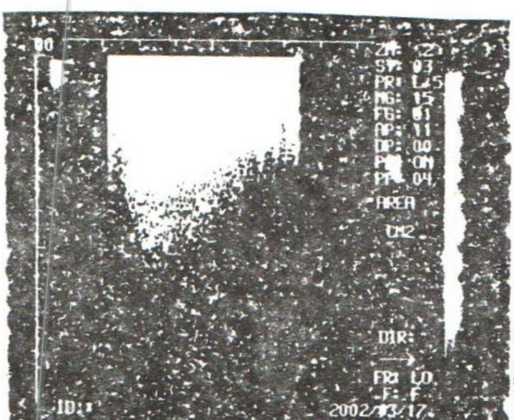


Fig. (6): Cd1

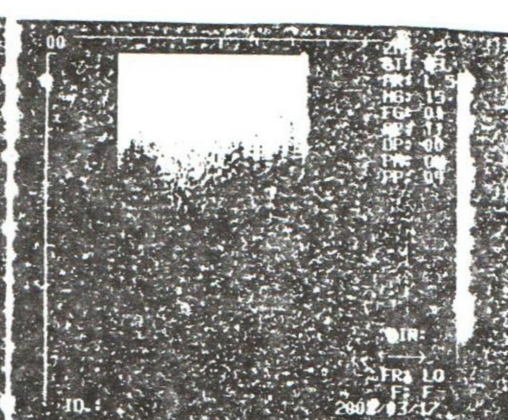


Fig. (7): Cd2

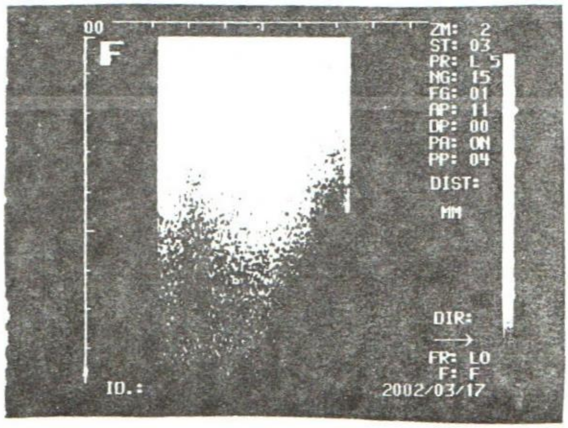


Fig. (8): Control

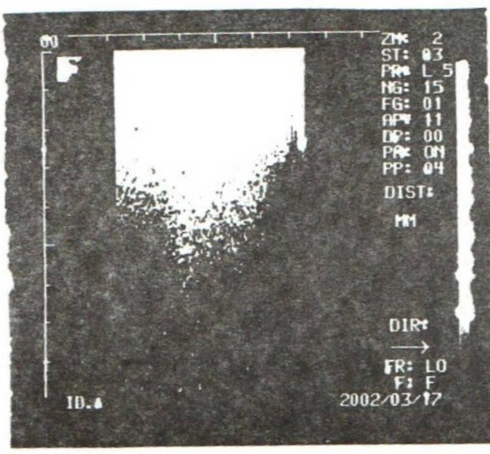


Fig. (9): Cu1

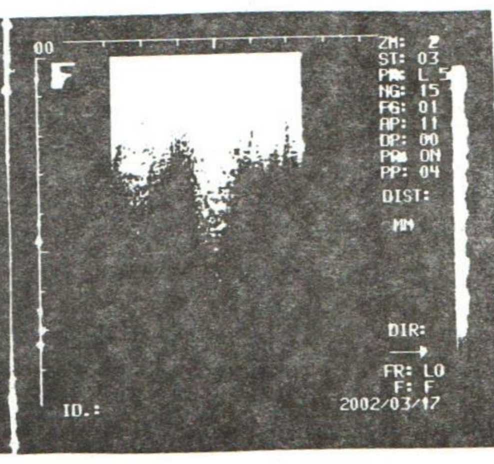


Fig. (10): Cu2

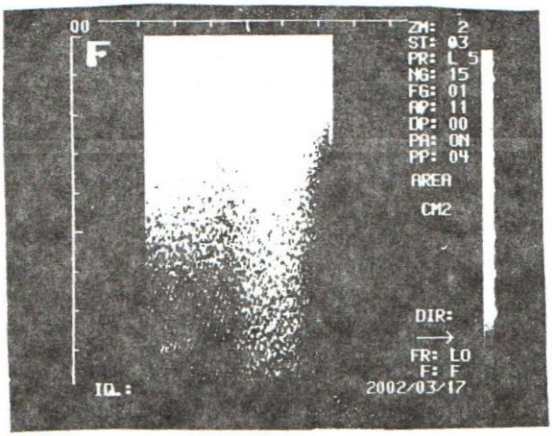
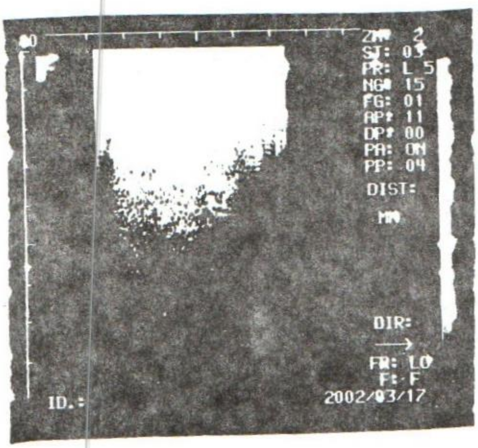


Fig. (11): Control



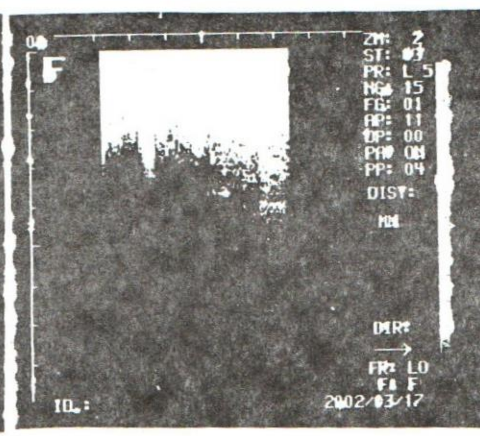


Fig. (12): Pb1

Fig. (13): Pb2

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تأثير كل من الكادميوم والنحاس والرصاص على النمو والتركيب الكيمـاوى فـى اسماك البلطى النيلى

محمود فؤاد اسماعيل سالم
معهد بحوث الانتاج الحيوانى – مركز البحوث الزراعية

أجريت تجربة استغرقت ٥٦ يوما لدراسة تأثير مستويين من كل من الكـادميوم (٠,٢-٠,١ مللجرام/لتر) ، النحاس (٠,٥ – ٠,٢٥ مللجرام/لتر) والرصاص (٠,٢-٠,١ مللجرام/لتر) فى الماء على النمو والتركيب الكيماوى لجسم الأسماك ، والمتبقى منها فى الأنسجة المختلفة للسـمك، ومساحة كل من العضلات والتجويف البطنى لأسماك البلطى النيلـى . اسـتخدمت ١٤٠ سـمكة بمتوسط وزن ابتدائى ١٠ جم ، وقسمت الأسماك الى ١٤ مجموعة متماثلة فى الوزن ، ووضـعت فى احواض زجاجية يحتوى كل منها على ٧٥ لترا من الماء ، وقد وزعت المعاملات والكنتـرول على المجموعات بحيث أن كل معاملة تكررت فى مجموعتين ، وأثناء التجربة غذيت الأسـماك على عليقة تحتوى على حوالى ٢٥% بروتين خام بمعدل ٣% من الوزن الحى يوميا.

أظهرت النتائج أن الكادميوم والنحاس والرصاص سببا انخفاضا معنويـا فـى متوسـط الزيادة فى الوزن ، ومعدل النمو النوعى ، ونسبة الأسماك الحية فى أسماك البلطى النيلـى عنـد المستويات التى درست بالمقارنة بالكنترول ، وكانت نسبة التحول الغذائى وكفاءة استخدام البروتين أسوأ فى المجموعات التى تعرضت لكل من الكادميوم والنحاس والرصاص.

وسبب كل من الكادميوم والنحاس والرصاص انخفاضا فى نسبة المادة الجافة والبروتين الخام فى جسم الأسماك ، بينما زادت نسبة المستخلص الاثيرى والرمـاد ، وكـان المتبقـى مـن الكادميوم والنحاس والرصاص فى المجموعات التى عوملت بأى من هذه العناصر أكبر عما فـى الكنترول ، وازداد هذا المتبقى بزيادة مستوى العناصر الثقيلة المختبرة ، وكان أعلى تركيـز مـن المتبقى فى رأس وأحشاء الأسماك التى عوملت بالمستويات العليا لكل مـن الكـادميوم والنحـاس والرصاص.

كما ظهر الاختلاف فى مساحات كل من العضلات والتجويف البطنـى ، حيـث قلـت مساحة العضلات فى الأسماك المعاملة بالعناصر الثقيلة بالمقارنة بالكنترول ، بالاضافة الى زيـادة مساحة التجويف البطنى وذلك على حساب مساحة العضلات والذى يعتبر الجـزء المـأكول فـى الأسماك.

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