Study of Industrial Pollution and Its Effect on Nile Tilapia in Marryotia Canal

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Abstract

The present study was carried out to assay the bioaccumulation of ammonia ,nitrite ,nitrate in fresh water and the concentration of heavy metals (Lead, Cupper, Cadmium, Manganes and Zinc) in water and tissues of *Tilapia niloticus* selected from El-marryotia canal (one of the river Nile ramification in Al-Giza city) in Egypt, samples were collected during the period of July to October 2011 to determine water safety for human consumption and the hazardous effect of these metals on the histopathology of selected tissues and organs of fish.

Two samples were collected from two locations of canal, pH of water samples were measured and the concentration of ammonia, nitrate and nitrite were determined in these samples.

Twenty Tilapia were also collected from different locations of the canal, One gram of liver, abdominal and dorsal muscles tissue from each fish were taken for determination of lead, Cupper, Cadmium, manganese and Zinc in the selected tissues.

Specimens of gills, muscles, liver, kidney and intestine from each fish were fixed in 10% buffered neutral formalin, and processed for pathological study.

The results revealed that water samples polluted with ammonia, nitrite and nitrate respectively.,significant differences were recorded in concentration of Mn and Zn in both water samples. The mean concentration of Pb was 0.216±0.0108, 0.204±0.009 and Mn 0.901±0.041, 0.73±0.003 in both water samples which was above the World Health Organization safety standard, the results indicated that water were polluted with both two heavy metals.

More concentration of Pb1.362±0.0657 was noted in liver of fish , concentration of Pb (1.2±0.062), Cd (0.01.2±0.062) was the highest in abdominal muscles and lowest in the remaining tissues of fish , the mean concentration of Cu (0.6.2±0.032) was higher in abdominal muscle , Mn (0.54±0.02) and Zn (1,19.2±0.054) was more concentrated in dorsal muscles and all the heavy metals studied gave results with the permissible limits in fish tissue recorded by the standards of WHO.

The main histopathological changes observed were edema and sloughing of lamellar epithelia in gills of fish. In liver, it was noticed vacuolation of most hepatocytes with nuclear pyknosis . The findings in kidneys exhibited the presence of periglomerular edema and vacuolar degeneration of most of renal tubules .The fish muscle showed degeneration and edema with focal areas of necrosis of some muscle fibres.

keyword : Tilapia, Marryotia**,** bioaccumulation**,** heavy metals

Introduction

Contamination of fresh water with a wide range of pollutants that may be transformed into new potentially toxic compounds has become a matter of concern ones for the last few decades [1,2]

The natural aquatic systems may be extensively contaminated with heavy metals released from domestic,industrial and other man- made activities [3,4].

Some heavy metals are neuron toxic like Iron,Copper ,mercury , nickel , zinc, Cadmiums, chromium and manganese [5,6].The commonest source of heavy metal pollution are industrial and mining activities,petroleum exploration,processing and effluent management, atmospheric condensation and sewage disposal [7].

Fish are widely used to evaluate the health of aquatic ecosystems because pollutants build up in the food chain and are responsible for adverse effects and death in the aquatic systems [8]. Contamination by heavy metals in aquatic environment is a severe problem, it may alter the structure of the cell membrane of organisms [4].

Heavy metals cannot be destroyed through biological degradation, when exposed to higher concentrations, organs of aquatic animals may accumulate heavy metals [9,10,11].

Several studies carried out on various fishes have shown that heavy metals may alter the physiological activities , biochemical parameters both in tissues,blood and histopathological alterations in fish tissues [12,13]

Material and Methods

The present study was carried out to assay the bioaccumulation of heavy metals (lead, Copper, Cadmium and mercury) in fresh water, tissues of *Tilapia niloticus* from Marryotia canal between July to October 2011 samples wee included :

1-Water samples: two samples were taken from two location of_Marryotia canal using clean containers at depth about 30 cm from the water surface to avoid floating materials.The collected water samples were filtered through whatman No. 42 to get rid of suspended material, pH of water samples were measured at site of taking water samples ,concentration of ammonia ,nitrate, nitrite and heavy metals were determined in these water samples by atomic absorption spectrophotometric technique

2-Fish samples: 20 fishes were collected from different locations of the canal of approximately 300-400 gm body weight and length of 20-25 cm.

Biochemical analysis:

One gram of liver, abdominal and dorsal muscles tissue from each fish were taken and burned in oven and used for determination of Pb, Cu, Cd, Zn in the above selected tissue using spectrophotometer.

Pathological examination:

Specimens of gills, muscles, liver, kidney and intestine from each fish were fixed in 10% buffered neutral formalin, and processed then paraffin sections were obtained of 4-5 micron and stained with Haematoxylin and Eosin stain [14] and examined under light microscope.

Results

The present study showed variable concentration of heavy metals in water and selected fish tissues samples used in this study which illustrated in table 1,2 and 3

As noticed in table(1) the mean concentration of pH water in the two location of all samples studied was 6.8±0.18 6.6±0.22 which was with the permissible level of WHO safety reference standard [14] , the results revealed that the concentration of ammonia, (.13±0.07, 0.21±0.09 nitrite (0.09±0.01, 0.05±0.004) and nitrate (2.1±0.17,2±0.19) respectively was above the permissible level based on WHO safety [15] .

Table(2) showed the mean concentration of heavy metals(Pb Cd,Cu,Mn,Zn) in water samples from the two locations was mentioned in table(1) ,significant differences were recorded in concentration of Mn(0.901±0.041) and Zn (0.3165±0.014**)** in both water samples at p ≤0.01 using t- student test .The mean concentration of Pb was 0.216±0.0108, 0.204±0.009 and Mn 0.901±0.041, 0.73±0.003 in both water samples were above the WHO safety standard , the result indicated that water was polluted with both two heavy metals.

The mean concentration of heavy metals (Pb, Cd,Cu,Mn,Zn) in liver, abdominal and dorsal muscles were illustrated in table 3, significant differences were recorded in the concentration of these heavy metals in fish samples using ANOVA test.. More concentration of Pb1.362±0.0657 was noted in liver of fish .

The concentration of Pb (1.2±0.062), Cd (0.01.2±0.062) was the highest in abdominal muscles and lowest in the remaining tissues of fish , the mean concentration of Cu (0.6.2±0.032) was also higher in abdominal muscle,Mn (0.54±0.02) and Zn (1,19.2±0.054) was more concentrated in dorsal muscles and all the heavy metals studied gave results with the permissible limits in fish tissue recorded by the standards of WHO standard [15,16]

Histopathological findings:

The histopathological examination of different fish tissues revealed that several pathological changes, gills showed congestion of the central vein sinus of the primary lamellae (Fig. 1) . Edema with focal areas of degeneration and necrosis of the lining epithelium of the secondary lamellae (Fig. 2) . Desquamation of the lining epithelium of the secondary lamellae with mononuclear cells aggregation (Fig. 3). Some areas showed epithelial hyperplasia and fusion where both the secondary lamellae and primary filaments were clumped together in a mass of tissue with mononuclear cells aggregation (Fig. 4).

Examination of the fish muscular tissues revealed focal areas of degeneration of muscle bundles and necrosis (Fig. 5). Interstitial edema with fragmentation of some muscle fibres (Fig. 6).

Liver showed severe vacuolar degeneration of the hepatocytes with nuclear pyknosis of the majority of hepatic cells (Fig. 7). Congestion of the hepatic blood vessels with haemorrhages and intravascular haemolysis of some red blood cells (Fig. 8 a&b) as well as presence of inflammatory cells within the blood vessels (Fig. 9) .haemosidrosis and activation of Kupffer cells were also seen (Fig. 8b).

The main pathological findings of intestine were degenerative and necrotic changes of some lining mucosal epithelium (Fig.12).Other areas showed hypertrophy with fusion of some villi and sloughing of the lining epithelium of the tip of some villi. Edema and mononuclear cells aggregation in the submucosa and within the blood vessels (Fig. 13)

Discussion

This study documented that water of canal studied were polluted with ammonia, nitrite and nitrate, relative to [15] safety values. Ammonia is very toxic to fish and aquatic life. Ammonia concentration of 0.06 mg/l can cause gill damage in fish. Water naturally contains less than 1 milligram of nitrate-nitrogen per liter and is not a major source of exposure. Higher levels indicate that the water has been contaminated.[2].Common sources of nitrate contamination include fertilizers, animals wastes, septic tanks, municipal sewage treatment systems, and decaying plant debris [17] Nitrites can produce a serious condition in fish called "brown blood disease". react directly with hemoglobin in human blood and other warm-blooded animals to produce methemoglobin [17] . Manganese is neurotoxic and manifests in the form of impulsive and aggressive behavior in some cases euphoria and sexual stimulation. [3]. Several histopathological changes were noted in gills, abdominal and dorsal muscles, liver, kidneys and intestine which attributable to heavy metals exposure .

The toxicity of these elements is due to the ability to cause oxidative damage to living tissues. Damages include enhanced lipid per- oxidation , DNA damage, enzyme inactivation and the oxidation of protein sulfydryl groups [18].

In our study, examination of gills revealed severe pathological alterations due to entry of this metals and provokes lesions and damage [19] .Gills also showed edema of the primary lamellae, several previous studies reported that edema of the gill epithelium is one of the main structural changes caused by the exposure to heavy metals and sometimes referred as a first sign of pathology [20 ,21,22]. Cellular degeneration in the gill epithelium is also observed in fish exposed to different pollutant as described by [21 ,22] . Swelling and hyperplasia of the gills epithelium could serve as a defence function , as these alterations increase the distance a cross which water borne irritants must diffuse to reach the blood stream. [23].

The fish muscle is one of the ultimate parts for heavy metal accumulation.The heavy metals were uniformly spread over the body muscles. The present study revealed various degenerative and necrotic changes of most of the muscular tissues . The present finding was supported by the other finding of [24, 25].

The liver showed vacuolar degeneration and nuclear pyknosis in the majority of hepatic cells. These findings were apparent as the liver is considered the organ of detoxification, excretions and binding proteins [2]. The liver of fish is sensitive to environmental contaminants because many contaminants tend to accumulate in the liver and exposing it to a much higher levels than in other organs [26].The presence of haemosiderin pigments as a result of internal bleeding and hemolysis in the hepatic tissues.

Kidneys showed hydropic swelling of most of the lining renal tubules with pyknotic nuclei. Also, periglomerular edema and atrophy of the glomerular tuft capillaries with few inflammatory cells infiltration were seen[25,26]. Congestion of most of the renal blood vessels, interstitial haemorrhage and presence of macrophages laden with hemosidrin pigments were also observed. Similar alterations in kidneys of Tilapia were observed in several species of fish exposed to heavy metals and these alterations were described by[27] and [28]who found that kidney is the gate way for many heavy metals detoxification in body and considerable amount of heavy metals accumulated in the kidney tissue[29].

The intestine showed various alterations in the present study , similar alterations in the intestine of *Liza parsia* exposed to Hgcl2 were described by [30]

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Table (1): The mean concentration of pH ,ammonia , nitrite and nitrate in water samples from two locations of Elmarriyotia canal

Parameter	Value N=3 , Mean±SD		WHO Unite
	Sample 1	Sample 2	
PH	6.8±0.18	6.6±0.22	
Ammonia(NH3)	0.13±0.07	0.21±0.09	0.05 mg/L
Nitrite	0.09±0.01	0.05±0.004	0.06 mg/L
Nitrate	2.1±0.17	2±0.19	1 mg/L

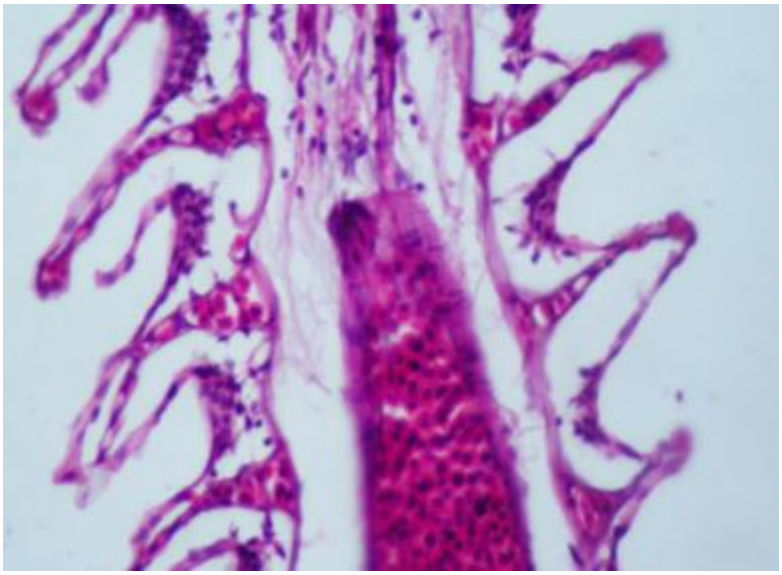
Table (2): the mean concentration of heavy metals (Pb, Cd , cu , Mn and Zn) in water sample from two locations of Nile canal (N=2 , Mean±SD)

Water Sample	Pb(mg/L)	Cd(mg/L)	Cu(mg/L)	Mn(mg/L)	Zn(mg/L)
Sample 1	0.216± 0.0108b	0± 0a	0.0203 0.0009a	0.901± 0.041c	0.3165± 0.014a
Sample 2	0.204± 0.009b	0.0± 0.0a	0.0181 0.0008a	0.73± 0.003b	0.2419± 0.010a
WHO unit	0.01	0.005	0.03	0.5	3.0

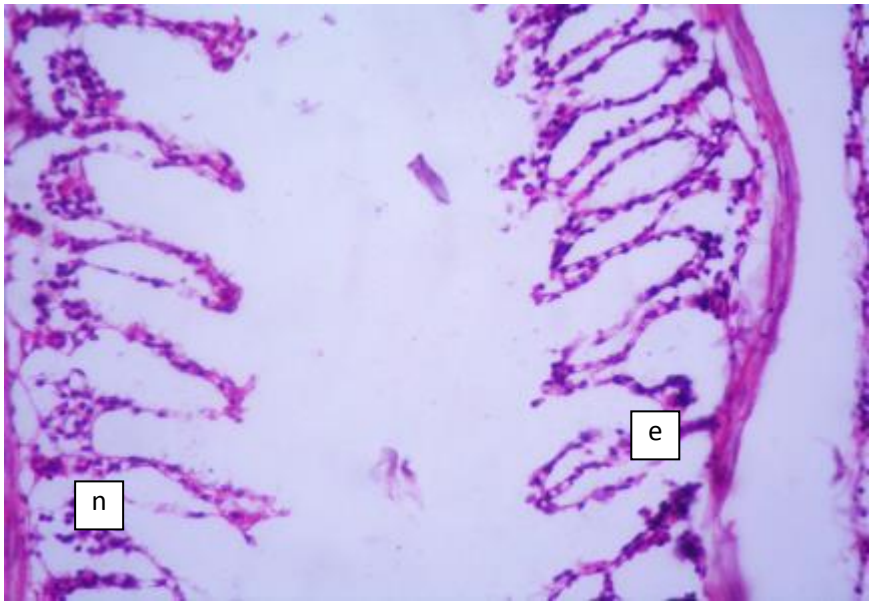
Table (3): Bioaccumulation of heavy metals(Pb Cd,cu,Mn,Zn) in liver abdominal and dorsal muscle of fish tissue included in this study (n=20, Mean ±SD)

Group	Pb(mg/dl)	Cd(mg/dl)	Cu(mg/dl)	Mn(mg/dl)	Zn(mg/dl)
Abdominal .muscle N=20	1.25± 0.0625a	0.014± 0.0006a	0.676± 0.032a	0.73± 033b	1.40± 0.063b
Liver N=20	1.362± 0.0657a	0.049± 0.0024b	1.765± 0.084c	1.409± 0.068c	1.624± 0.075b
Dorsal muscle N=20	1.254± 0.062a	0.01± 0.0004a	1.131± 0.055b	0.45± 0.025a	1.19± 0.054a
F-calculated	2.112	11.654#	14.115#	19.542#	7.332#
WHO unit	1.5	0.2	1.5	2.5	150

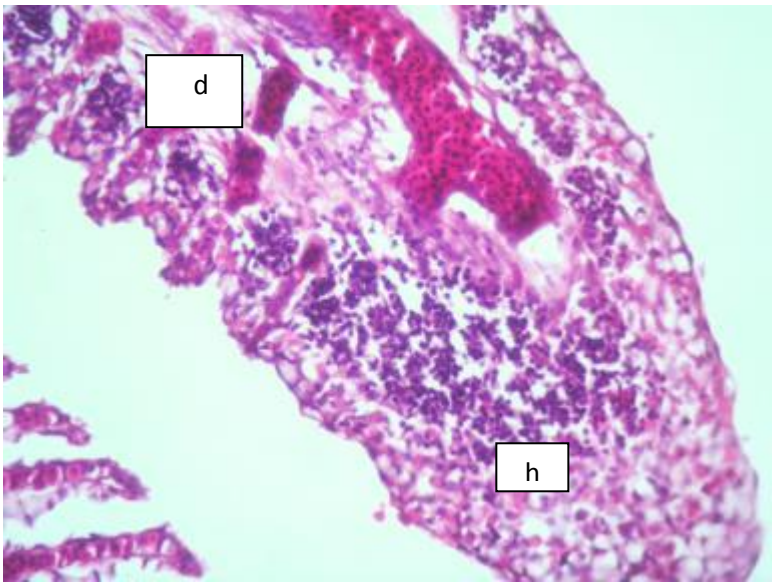
Significant at p <0.05 using ANOVA test



Fig(1): Gills section showing congestion of the central vein sinus of the primary lamellae. H&E (x 20)



Fig(2): Gills section showing edema with focal areas of degeneration and necrosis of the lining epithelium of the secondary lamellae (H&E X 200). e: edema n: necrosis



Fig(3): Gills section showing of the lining epithelial separation of with mononuclear cells aggregation (H&E X 200)d:degeneration h : hemorrhage

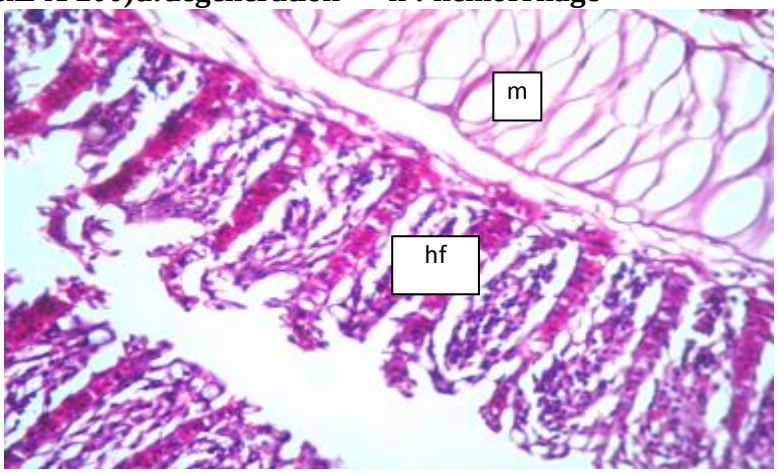
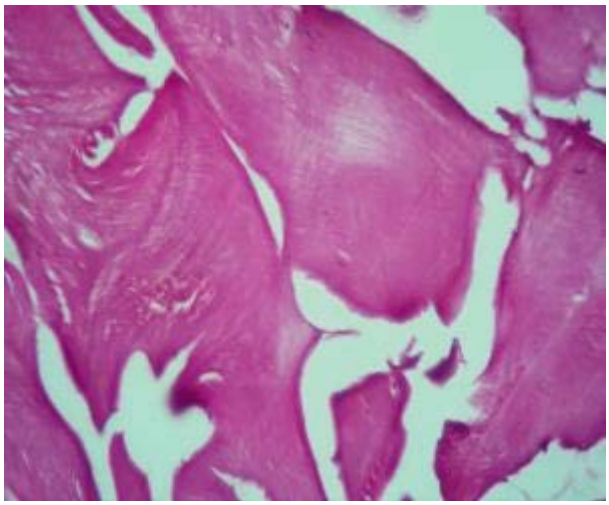
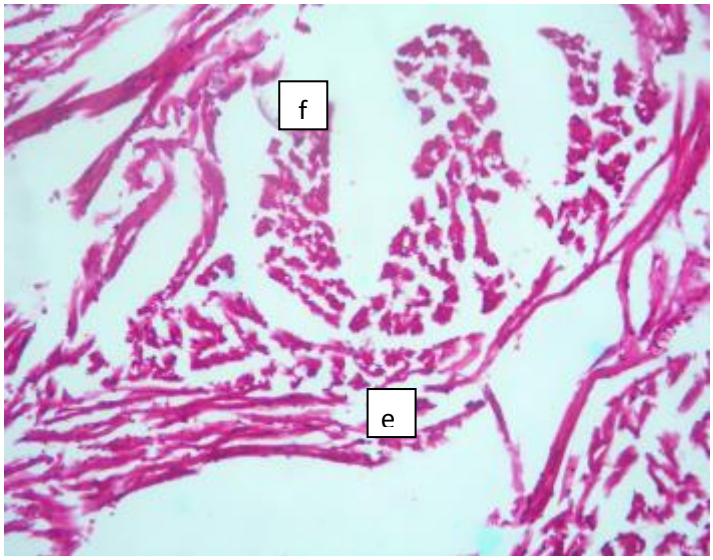


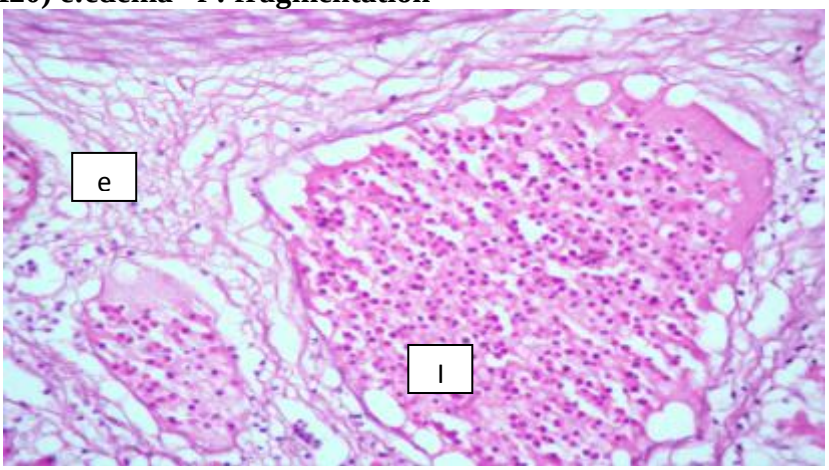
Fig (4): Gills revealing epithelial hyperplasia and fusion of both secondary and primary lamellae with mononuclear cells aggregation(H&E X 200). hf : hyperplasia m: mononuclear cells



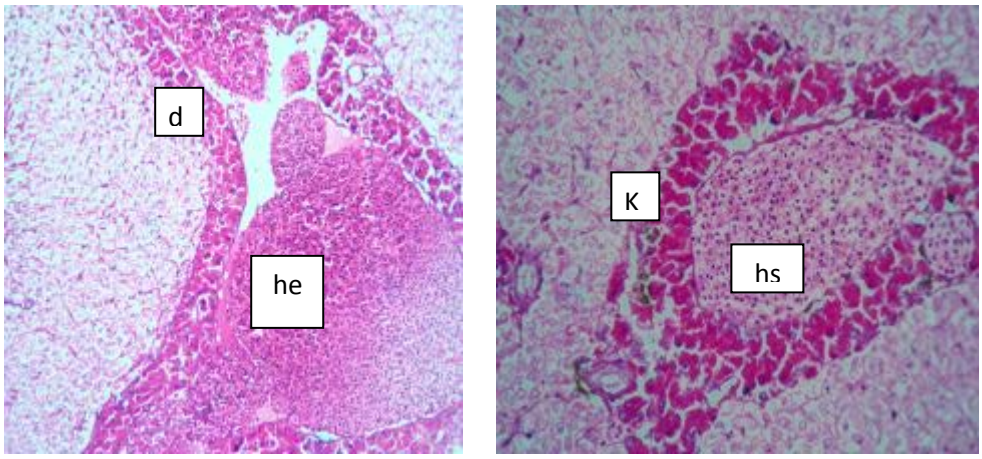
Fig(5): flesh showing focal areas of degeneration and necrosis of the muscle bundles (H&E X 40) .



Fig(6): Muscular tissues revealing, interstitial edema with fragmentation of some muscle fibres (H&E X20) e:edema f : fragmentation



Fig(7): Intestine of fish revealing accumulation of inflammatory exudate within the dilated blood vessels and presence of edema in the submucosa (H&E X 40). I : inflammatory cells e:edema



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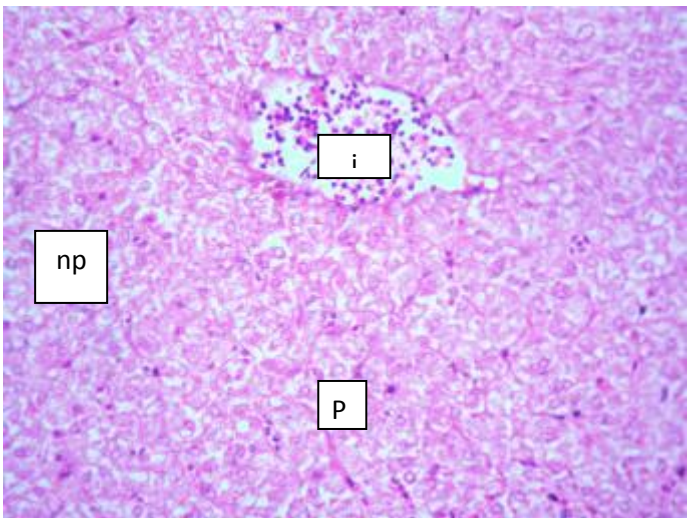
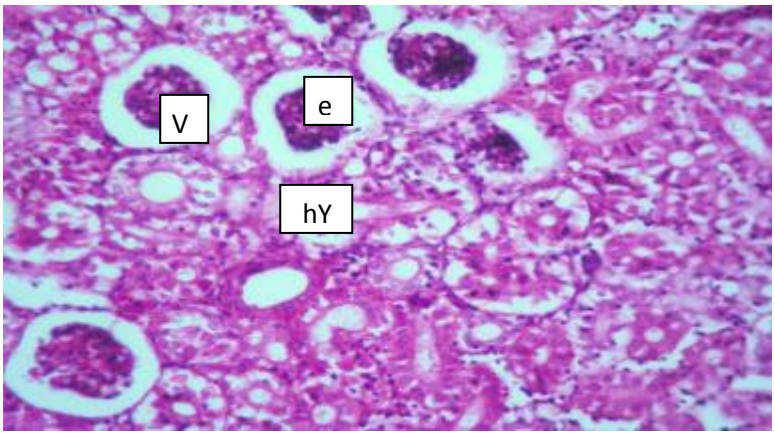
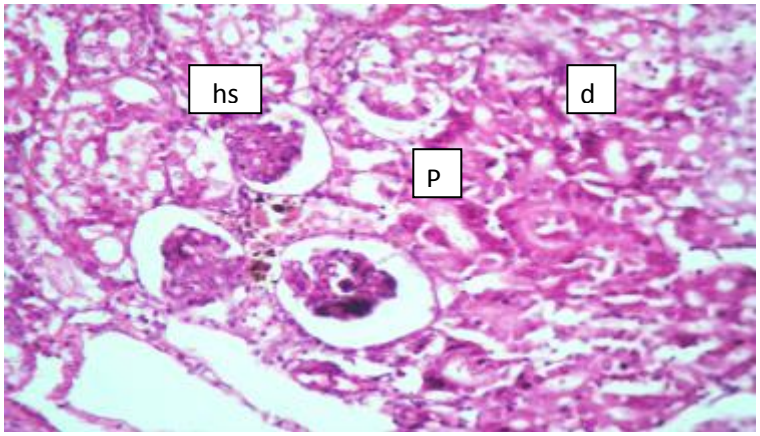
Fig(8a&b): a- Liver showing dilatation of the hepatic vessel and engorged with blood as well as intravascular hemolysis (H&E X 400). d : dilatation of vessel he : hemolysis b- Liver showing hemosidrosis and activation of Kupffer cells (H&E X 400). hs:hemosidrosis k : kupffer cells



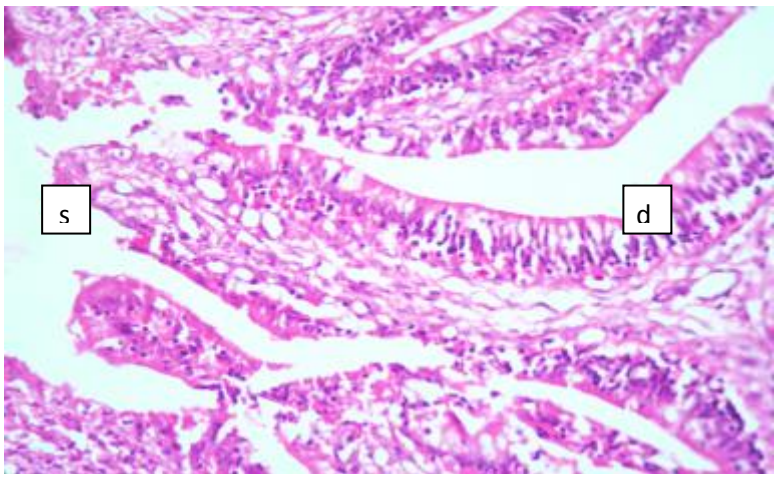
Fig(9): Section in liver revealing presence of necrobiotic changes in the hepatic cells with nuclear pyknosis (H&E X 40). Np: necrobiotic changes p : pyknosis i:inflammatory cells



Fig(10):Kidney revealing vacuolar and hyaline degeneration of most of the lining renal tubules with pyknotic nuclei. Periglomerular edema with atrophy of the tuft capillaries (H&E X 40). v : vacuolar degeneration hY: hyaline degeneratione : edema



Fig(11):Cross section in kidney showing presence of macrophages laden with hemosidrin pigment, periglomerular edema and various degenerative changes of the renal tubules (H&E X 40). hs : hemosidrin pigment p : periglomerular edema d : degenerative change



Fig(12): Intestine exhibiting degenerative changes with sloughing of some lining intestinal epithelium (H&E. X 400). d:degeneration s : slouphing.

دراسة التلوث الصناعي وتاثيراته على اسماك البلطي النيلي لمياه قناه المريوطية

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استلم البحث في:31 كانون الثاني 2012 ، قبل البحث في: 17 حزيران 2012

الخلاصة

صممت الدراسة الحالية للتقصي عن التراكم الحيوي للامونيا والنترات والنتريت في المياه العذبة والمعادن الثقيلة مثل (الحديد- النحاس- الكادميوم -المنغنيز- الزنك) في المياه وانسجة اسماك البلطي النيلي في قناة المريوطية (احد التفرعات الناتجة عن نهر النيل _ مدينة الجيزة)في جمهورية مصر العربية للمدة مابين تموز ولغاية تشرين الاول 2011 لتحديد مدي صلاحية تلك المياه للاستهلاك البشري ودراسة تاثير تلك المعادن الخطرة في امراضية الانسجة والاعضاء المختارة للدراسة . .

جمعت لهذه الدراسة عينتان ماخوذة من مكانين مختلفين من القناة، اذ قيس الاس الهيدروجيني لها وتحديد تركيز كل من الامونيا –النتيريت والنترات والمعادن الثقيلة في عينات المياه . جمعت عشرون سمكة بلطي نيلي من اماكن مختلفة من القناة وتم تشريحها واخذ عينات من الغلاصم والامعاء والكبد والانسجة العضلية البطنية الظهرية للدراسات المرضية النسجية اضافة الى انه تم جمع غرام واحد من الكبد والانسجة العضلية الظهرية والبطنية لتحديد تركيز كل من الحديد والنحاس والكادميوم والمنغنيز والزنك في الانسجة المختارة لهذه الدراسة.

كشفت نتائج الدراسة عن ان عينات المياه كانت ملوثة بالامونيا والنترات والنتريت على التوالي وسجل وجود فروق معنوية في تركيز كل من المنغنيز والزنك في كل عينات المياه موضوع الدراسة وتركيز النحاس0.676±0.032 اعلى في الانسجة العضلية البطنية و اظهر كل من المنغنيز والزنك تركيزا على في الانسجة العضلية الظهرية(0.45±.025، 1.1.19±0.05)على التوالي منه في بقية الانسجة ولقد اعطت جميع العناصر نسبا طبيعية وضمن الحدود المسموح بها لقياسات منظمة الصحة العالمية.

اما بالنسبة الى التغييرات النسجية المرضية التي لوحظت في هذه الدراسة فلقد تمثلت بوجوداستسقاء وانسلاخ في اما الدراسة التشريحية المرضية للكبد فلقد اظهرت تكون ,الانسجة الطلائية لصفائح خياشيم الاسماك موضوع الدراسة فجوي في اغلب الخلايا الكبدية مع وجود تغلظ نووي فضلا عن وجود صبغة الهيموسدرين في عينات المياه التي كانت اعلى من النسب المسموح بها لمنظمة الصحة العالمية مما اشار الى تلوث العينات بهذان العنصرين .

اما بالنسبة الى الموجودات المرضية في كليات الاسماك فلقد لوحظ ان هناك استسقاء بين الكبيبات الكلوية وتحلل فجوي في معظم النبيبات البولية كما تبين وجود تحلل واستسقاءمع مناطق محورية تنخرية في بعض الالياف العضلية الخاصة بالاسماك موضوع الدراسة.

الكلمات المفتاحية : البلطي، المريوطية، التراكم الحيوي، المعادن الثقيلة.