International Journal Of

Recent Scientific Research

ISSN: 0976-3031

Volume: 7(1) January -2016

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THE OFFICIAL PUBLICATION OF
INTERNATIONAL JOURNAL OF RECENT SCIENTIFIC RESEARCH (IJRSR)
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International Journal of Recent Scientific Research
Vol. 7, Issue, 1, pp. 8142-8146, January, 2016

International Journal of Recent Scientific Research

ISSN: 0976-3031

RESEARCH ARTICLE

HISTOLOGICAL ALTERNATION IN SELECTED ORGANS OF *OREOCHROMIS NILOTICUS* FROM SELECTED LAKES AROUND COIMBATORE DISTRICT

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ARTICLE INFO

Article History:

Received 15th September, 2015
Received in revised form 21st November, 2015
Accepted 06th December, 2015
Published online 28st January, 2016

Key words:

Heavy metal, Pollution, Histopathology, *Oreochromis niloticus* , Muscle and gill.

ABSTRACT

Heavy metal contamination of aquatic ecosystems has been long recognized as a serious pollution problem. When fish are exposed to elevated levels of heavy metals in a polluted aquatic ecosystem, they tend to take these metals up from their direct environment. Heavy metal contamination may have devastating effects on the ecological balance of the recipient environment and a diversity of aquatic organism. In the present investigation Copper, lead, cadmium, zinc and Nickel concentrations were recorded in tissues of *Oreochromis niloticus* from selected study areas in 2014-2015. Histopathological alterations in fish tissues were also studied. Gill shows highest level of metals succeeded by muscle. The distribution of heavy metal in the fish organs analyzed were in the order of Zn > Cu > Pb > Ni and Cd. Heavy metal concentration was high in koolipalayam and orathupalayam lake. Several histopathological changes were noted in muscles and gills tissue attributable to heavy metals exposure.

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INTRODUCTION

In India urbanization and industrialization have boosted the economy of mankind through various means and ways in this modern world. But at the same time aquatic (Fresh water and Marine water) resource has become a huge challenge and a serious threat to different types of pollutants entering into the freshwater ecosystem affecting aquatic organisms, including fish indirectly *via* the food chain. Currently in India most of the aquatic water bodies receive million liters of industrial effluents, domestic waste, sewage and agricultural waste containing substances with high nutrient to toxic substances.

These pollutants may able to accumulate along the aquatic food chain which leads to severe risk for both animal and human health. The impact of pollutants cause decreases fertility and reproductive abnormalities in fishes, birds and mammals and at the same time alters immune function of aquatic organisms. Pollutants like heavy metals in the environment may have a natural origin but most of the heavy metals in water bodies come from anthropogenic activities leading to the deterioration of water quality causing main threat to aquatic fauna worldwide.

Among the pollutants heavy metals are considered to be major pollutants as they cause greatest threats to biota because of their persistence and possible bioaccumulation and bio magnification through food chains (Uysal *et al.*, 2009; Ricart *et al.*, 2010). Heavy metals pollutants contain toxic substances derived from sources like industrial waste discharges, sewage effluent, pesticides and agrochemicals etc which when discharged into water bodies will change the surface tension, thermal properties, conductivity, density and pH value along with biodegradable and non-biodegradable pollutants which will cause dermatological diseases, skin cancer and internal cancers (liver, kidney, lung and bladder).

The degree contamination in aquatic environment is frequently assessed by comparing containment concentration in associated biota. Prolonged exposure to water pollutants even in very low concentrations have been reported to induce morphological, histological and biochemical alterations in the tissues which may critically influence fish quality. Water analyses may be inefficient to identify the metal inputs to fluvial systems because of the inherent variability of flow and contaminant concentrations. With this regard, fish can be considered as one of the most significant indicators in aquatic ecosystems to analyze the impact of metal pollution because they occupy various tropic levels, and are the key species in tropic chains,

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and concentrate large amounts of metals (Barata *et al.*, 2010; Lasheen *et al.*, 2012). In addition, histopathological alterations can also be used as indicators for the effects of various anthropogenic pollutants on aquatic biota and are a reflection of the overall health of the entire population in the ecosystem (El-Bakary *et al.*, 2011). It is a cost effective tool to determine the health of organisms hence reflecting the health of an entire aquatic ecosystem. The exposure of fish to chemical contaminants is likely to induce a number of lesions in different organs as it provides a rapid method to detect the effects of irritants, especially chronic ones, in various tissues and organs.
Workers like Abdel-Warith *et al.*, 2011; Dhevakrishnan and Hussain Us Zaman (2012); Selvanathan *et al.*, 2013 stated that histopathological changes have been widely used as biomarkers in the evaluation of the health of fish exposed to contaminants. Gyaneswar *et al.*, (2014) Mohammad MN Authman *et al.*, 2015 gives a brief account of the toxic effects of heavy metals on fish and the use of fish as bio-indicator of the effects of heavy metals pollution. With this regards the present study was also carried out to investigate the heavy metal contamination of selected study areas Sulur lake, Koolipalayam lake and Orathupalayam lake and the histological alteration in the tissues of major polluted fresh water fish *Oreochromis niloticus* due to heavy metal contamination caused by the residues of these metals in their organs.

MATERIALS AND METHODS

Coimbatore is the second largest city in the Indian state of Tamilnadu. It is located at 11°00′58″N 76°58′16″E11.0161°N 76.971°E.Surrounded by the Western Ghats mountains range to the west and the North. Coimbatore the south Indian Manchester, is known for its textile factories, engineering firms, automobile parts manufacturers, health care facilities, educational institutions, pleasant weather, hospitality and for its kongu Tamil. The Noyyal River forms the southern boundary of the city. Noyyal's basin contains many lakes and ponds. They are the main water resource for the nearby villages mostly for various domestic activities. In the present study 3 lakes were selected they are namely sulur lake, koolipalayam lake and orathupalayam lake.

Demographical description of study area

***Sulur lake(L1): Location**:* Taluk – Coimbatore; **Village** – Sulur; **Latitude**: 11° 01’40″ N and **Longitude**: 77° 07’20″ E; **Size of Wetland**: Catchment area: 8.704 Sq.miles; Water Spread area – 0.332 Sq.km; Length of the Bund – 1450.00 M; **Capacity** – 17.94 M.cft; **Depth** – 8.50 Feet.

Koolipalayam lake (L2): Location: Taluk- Tirupur; village- Mannarai; **Latitude:** 11.134238° N and **Longitude:** 77.386521° E; **Size of Wetland**: Catchment area: 4.144 Sq. Km; **Lake bed area** – 243.35 acres; **Industries-** There are quite a few textile processing units and textile garmenting industries near to the lake.

***Orathupalayam lake (L3): Location**:* Taluk- Tirupur; village- Kangayam; **Latitude:** 11°06′31Z and **Longitude**: 77°32′23″E; **Size of Wetland**: Catchment area: 22.4555 sq.km; The maximum water level of 248 m and 250 m of top bund level. The capacity of the reservoir is 616 MCft . **Depth- 71** feet.

Collection of fish samples

The study species *Oreochromis niloticus* was collected from the selected study areas Sulur lake, Odatupalayam lake, Kolipalayam lake using gill net from the selected sites. From each sampling sites 10 fishes were collected as sample material with average weight ranging from 250 - 300 g, average length of 8-10cm and the target organs (gills and muscles) were excised for further analysis.

Heavy Metal Analysis from fish samples

Fish samples of 1.0 g dry weight were digested with 6ml of HNO_3 (65%) and 2ml of H_2O_2 (30%) at 280°C on a hot plate for 4hr. To the digested sample 2ml of 1N HNO_3 is add to the residue and the solution is evaporated again on the hot plate, till the sample gets completely digested. After cooling, the above sample 10ml of 1N HNO_3 was added. This solution gets diluted and filtered through a 0.45µ nitrocellulose membrane filter (Sharma *et al.*, 2000). The filtered solution is measured for metal concentrations using Atomic Absorption spectrometer. The entire triplicate sample was analysed in the Department of Chemistry, South India Textile Research Association (SITRA), Coimbatore.

Histopathological studies

Fishes were taken out, blotted dry with soft absorbent paper and dissected for the study of histopatological changes of the removed tissue samples like gills and muscles of *Oreochromis niloticus* fish. The removed organs were carefully preserved in labeled sample bottles containing 10% formalin, dehydrated in ascending grades of alcohol and cleared in xylene. The fixed tissues were embedded in paraffin wax and sections of 5 microns were cut, using Euromex Holland microtome. Sections were stained following Saad *et al.*, 2012 method with Hematoxylin and Eosin stain and were made for histological investigation under the microscope and their photos were taken by microscopic camera.

RESULT AND DISCUSSION

Heavy metal analysis in water samples of selected study area: The analysis of heavy metals in different organs of fish *Oreochromis niloticus* from the selected areas where shown in **Table No**: **2.** All the two organs of *Oreochromis niloticus* (muscle, gill) shows different values of heavy metals concentration. Gill shows highest level of metals succeeded by muscle. The distribution of heavy metal in the all fish organs analyzed were in the order of Zinc > Copper >Lead > Nickel and Cadmium. Among the metal analyzed the highest concentration level of Zinc and the minimum metal concentration is Cadmium. The permissible limits for heavy metals in water sample were represented in **Table No- 1**

Histopathology aberrations observed in experimental fishes: Histopathological alterations can be used as indicators for the effects of various anthropogenic pollutants on organisms and are a reflection of the overall health of the entire population in the ecosystem. These alterations are closely related to other biomarkers of stress since many pollutants have to undergo metabolic activation in order to provoke cellular change in the affected organism. Several studies reported that the exposure of fish to pollutants (agricultural, industrial and sewage) results in several histological alterations in different tissues of fish. These studies have been done for various reasons; many of them concerning food safety and public health interests where muscle tissues are generally the major edible portion of the fish Ashoka *et al*., (2011).

Histological changes in the muscle tissue of Oreochromis niloticus

Fish muscles are commonly analyzed to determine contaminant concentrations, and heavy metals have been quantified in muscle tissues from a variety of fish species Andreji *et al*., (2006); Soegianto and Hamami, (2007). The changes like abnormal arrangement of muscle bundles, focal disruption of muscle fibres, few congested blood vessels and mild lymphocytic infiltrate is seen. Aggregation of inflammatory cells and degradation of muscle fibers also observed in the muscle tissue of *Oreochromis niloticus* collected from Sulur lake (L1), Koolipalayam lake (L2) and Orathupalayam lake(L3).The histological changes were represented in **Plate No:1 a, b, c.** These changes are induced by untreated industrial effluents are discharged into the water bodies. All these changes indicate the fish under the highly stressful conditions due to the more polluted region receives the effluents from industrial complex. Similar changes have also been reported by Das and Mukherjee, (2000); Gbem *et al*., (2001); Bharat Bhusan Patnaik *et al.*, (2011) and Mano sathiyadevan *et al*., (2012) in various fish samples.

Histological changes in the gill tissue of Oreochromis niloticus

The gills are important organs in fish to perform respiration, osmoregulation, acid base balance and nitrogenous waste excretion Health, (1987). Fish gills are also vulnerable to pollutants in water because of their large surface area and external location. Gills apart from being the primary respiratory organ in fishes, are also responsible for ion regulation. For this reason, fish gills are considered to be the most appropriate indicators of water pollution levels.

Histological changes of gill tissue includes proliferation in the epithelial gill filaments, fusion and curling of secondary lamellae, aggregations of inflammatory cells , rupture of lamellar epithelium. Epithelial detachment ,degradation of gill tissues were also seen in gill tissue of L2) and Orathupalayam lake(L3). **Plate No-2 a, b, c.** The histopathological alterations attributed to the prolonged exposure to heavy metals resulted in respiratory, osmoregulatory and circulatory impairment. These findings were agreed by Fernandes *et al.*,(2008). Moreover, Alvarado *et al.* (2006) reported that, the dramatic increase of chloride cells in the gills that produces epithelial thickening of the filament epithelium enhances migration of chloride cells up to the edge of the secondary lamellae and provokes the hypertrophy and fusion of secondary lamellae.

External irritant are the most frequent causes of significant gill pathological changes and retard the respiratory function of the organ by reducing its surface area (Saad *et al.*, 2011). It is possible that the pathological alterations in the tissues of both studied fish could be a direct result of the heavy metals, pesticides, fertilizers, salts and sewage, which are entered to the lake with the drainage water.

Fish are important palatable pretentious food for mankind. Good quality foods are very important for the wellbeing of humanity and animal health. It has been used as an environment indicator of food source quality. Heavy Metals mainly tend to accumulate in fine grained sediments therefore large particles have a dilutive effect on metals concentration. Heavy metals contamination in sediment can affect the water quality and bioaccumulation of metals in the species resulting in potential long term implication on human health and the ecosystem. Heavy metal contamination in aquatic environment exerts an extra stress on fish, which tend to accumulate the heavy metals in metabolically active tissues and organs.

CONCLUSION

It could be concluded that the environmental contamination of selected study areas induced several histopathological alterations in the tissues of *Oreochromis niloticus*. Consequently, it is recommended to coordinate different efforts to rescue Lakes like fresh water bodies from the environmental pollution problems especially heavy metals which are highly toxic to biota. The overcoming of these problems can be possible by subjecting the drainage waters discharged into the lake to technical treatments that fulfill its safety. Hence, a scientific method of detoxification is essential to improve the health of these economic fish in any stressed environmental conditions.

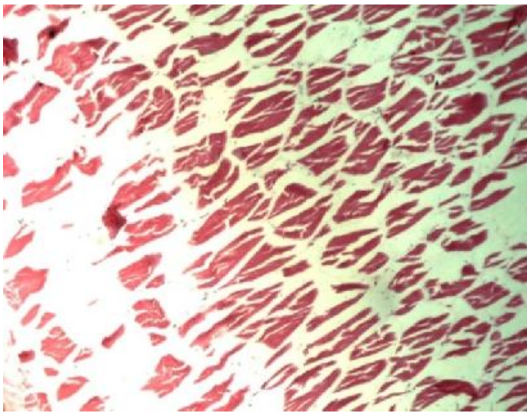
Table1 Standard Heavy metal values prescribed by WHO, FAO

Heavy metals(µg/g-1)	WHO	FAO
Cadmium (Cd)	**1.00**	**0.50**
Zinc (Zn)	**30.00**	**35.00**
Lead (Pb)	**0.50**	**0.50**
Nickel (Ni)	**15.00**	**10.00**
Copper (Cu)	**30.00**	**30.00**

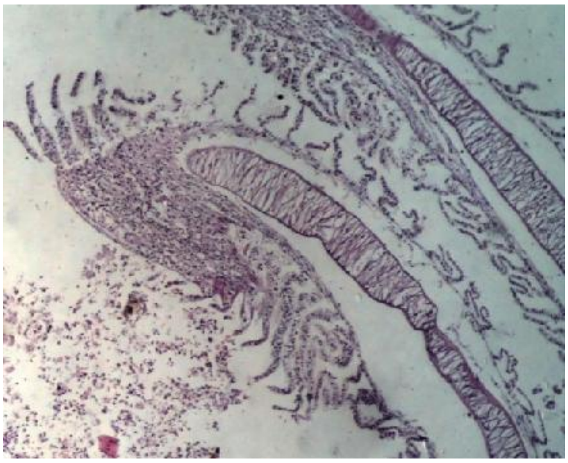
Table 2 Heavy metal concentration in muscle, gill tissues of *Oreochromis niloticus* from selected lakes in and around Coimbatore district

Metals (µg/Kg)	Samples	Sulur	Koolipalayam	Orathupalayam
Cadmium	Muscle	1.38±0.016^{c}	1.59±0.014^{a}	1.53±0.018^{b}
(Cd)	Gill	1.47±0.021^{c}	1.93±0.020^{a}	1.83±0.024^{b}
Zinc	Muscle	18.38±0.93^{c}	24.45±1.27^{b}	25.94±1.19^{a}
(Zn)	Gill	23.03±1.27^{c}	29.81±1.35^{b}	30.71±1.38^{a}
Lead	Muscle	5.91±0.872^{c}	8.64±0.747^{b}	9.69±0.822^{a}
(Pb)	Gill	8.35±0.793^{c}	10.31±0.798^{b}	12.69±0.868^{a}
Nickel	Muscle	4.17±0.173^{c}	5.43±0.154^{a}	5.37±0.167^{b}
(Ni)	Gill	5.86±0.176^{c}	6.74±0.179^{b}	6.93±0.183^{a}
Copper	Muscle	8.73±0.288^{c}	9.69±0.345^{a}	9.63±0.347^{b}
(Cu)	Gill	11.13±0.385^{c}	11.78±0.389^{a}	11.50±0.406^{b}

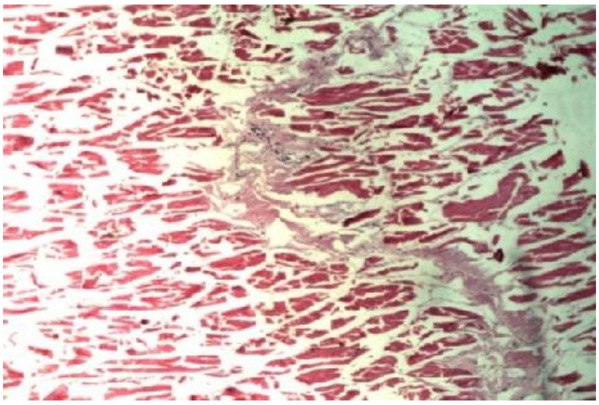
Values are mean ± SD of three samples in each group. $^{a\text{-}c}$ Row means followed by a common superscript are not significant at 5% level by DMRT



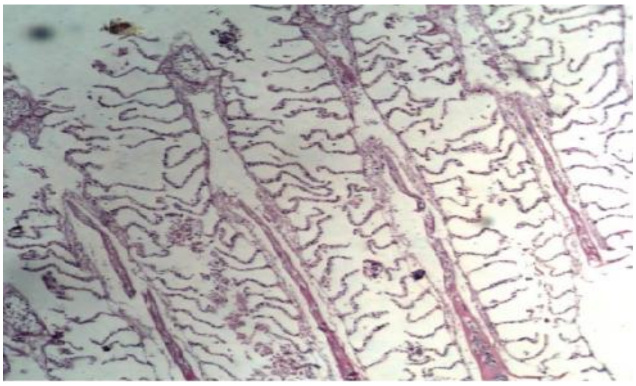
1a Focal disruption of muscle fibres



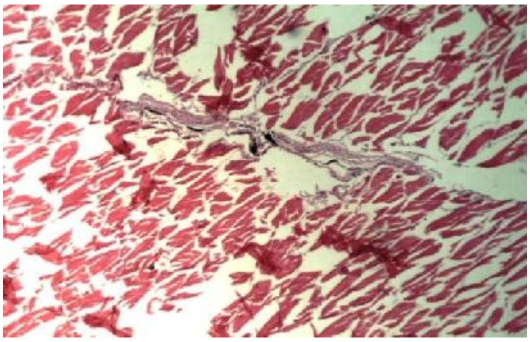
1b Rupture of lamellar epithelium



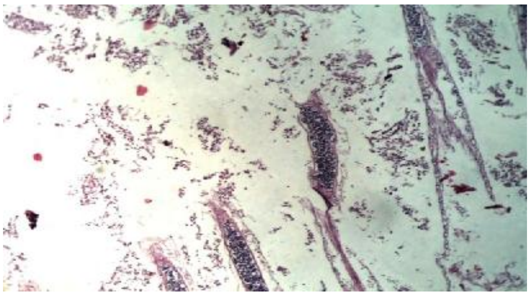
2a Lymphocytic infiltrate



2b Proliferation in the epithelial filament



3a Degradation of muscle fibers



3. b Degradation of gill tissues

1 a),2 a), 3 a) shows histological changes of muscle tissue of *Oreochromis niloticus* from selected study areas (L1,L2,L3). 1b), 2b), 3 b) shows histological changes of gill tissue of *Oreochromis niloticus* from selected study areas (L1,L2,L3).

Acknowledgement

The author is thankful to the Management Principal, Nirmala College For Women Coimbatore for providing necessary lab facilities and assistance. My heartfelt thanks to the faculty members of the Department of Zoology, Nirmala College For Women(Autonomous) Coimbatore for their comments and positive criticisms on the manuscript.

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How to cite this article:

Mercy M and Mary Fabiola SR.2016, Histological Alternation In Selected Organs of Oreochromis Niloticus From Selected Lakes Around Coimbatore District. *Int J Recent Sci Res.* 7(1), pp. 8142-8146.

ISSN 0976-3031