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Original article

Biochemical Stress of Chromium in Tannery Effluents on the Fresh Water Fish, *Tilapia mossambica*(Pisces)

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ABSTRACT

Tannery effluent was brought from the Puliyanakannu Village, SIPCOT Industrial Estate of Ranipet, Tamil Nadu, India. The fresh water teleost, *Tilapia mossambica* was chosen as experimental animal. Chromium is the major constituent of tannery effluent, which is the heavy metal and highly toxic to aquatic fauna. The physico-chemical characteristic and oxygen consumption of tannery effluent were estimated. Histological changes in the liver and muscle of the experimental fish were observed. In the present investigation, the changes of histological structure of liver and muscle of *Tilapia mossambica* under chromium stress and toxic stress of tannery effluents on agriculture aquatic fauna were studied and the results are reported in this paper.

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1. Introduction

Aquatic pollution has become a global problem in recent years. Extensive industrialization has measurably influenced the quality of water of lakes, ponds and rivers all over the world. Natural waters contaminated by untreated wastes of industrial, technological and agricultural origin often contain various metallic compounds. Heavy metals due to the bioaccumulative and non-biodegradable properties constitute a core group of aquatic pollutants[1]. Water pollution is generally localized and confined, making it more severe. The pollutants undergo many reactions and can become hazardous 70% of India's fresh water is polluted, including several high altitude lakes. While water pollution is easier to study and manage, its control is highly complex and very costly. The effects of water pollution are not only devastating to people but also to animals, fish and birds. Polluted water is unsuitable for drinking, recreation, agriculture and industry. It diminishes the aesthetic quality of lakes and rivers. More seriously, contaminated water destroys aquatic life and reduces its reproductive ability. Eventually, it is a hazard to human health. Nobody can escape the effects of water pollution[2].

Very high organic loads accompany industrial effluents from sugar factories, distilleries, tanneries and paper industries. Byproducts of paper and pulp industry cause depletion of fish upto 40km downstream. The wastes from oil refineries and steel industries contain phenol, which imports a strong odour, apart from poisoning the water body. Fertilizer industry wastes contain ammonia, urea, phosphate and sulphate, which, in water, cause algal bloom and are toxic to aquatic fauna and flora. Alkaline industry wastes contain mercury that can kill human beings who consume mercurised fishes. Lead generated from battery, printing, petrol and paste-processing industries, trace and toxic elements such as zinc, copper, etc., and effluents from mining industries are injurious to aquatic organisms[3]. The leather industry is an important foreign exchange earner for India. The states of Tamil Nadu, West Bengal and Uttar Pradesh together have 88% of the tannery units of the country. Tanners use a large number of chemicals during the process, discharging toxic wastes into the rivers and degrading agriculture land. More than 1000 tanneries in Tamil Nadu are situated within a 25km distance from the centre of the town. Several of them have been in existence for thirty to forty years. Effluents from the tanneries are discharged into streams, which drain into ponds, thereby polluting the ground water sources and cultivable land.

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Manufacturing of leather, leather goods, leather boards and for produces numerous byproducts, solid wastes, high amount of wastewater containing different loads of pollutants and emissions into the air. The uncontrolled release of tannery effluents to natural water bodies increases health risks for human beings and environmental pollution. Effluents from raw hide processing tanneries which produce wet blue, crust leather or finished leather contain compounds of trivalent chromium and sulphides in most cases[4]. Chromium compounds are used for ferrochrome production, electroplating, pigment production and tanning. These industries, the burning of fossil fuels and waste incineration are sources of chromium in air and water. Most of the liquid effluent from the chromium industries is trapped and disposed of in land fills and sewage sludges, the chromium being in the form of the insoluble trivalent hydroxide[5]. The untreated effluent containing particulates of chromium from the tanneries are discharged into fresh water bodies and also affected the aquatic organisms. Chromium is one of the highly toxic heavy metal to aquatic fauna[6].

A variety of chemicals are used in the tanning industries, including lime, sodium chloride, sodium carbonate, ammonium chloride, sulphuric acid, tannins and dyes. All tanneries need a large amount of water for process leather and depend on ground water sources for their daily requirements. The discharged effluents from the processing units are stored in large lagoons and pollution occurs as the dissolved salts percolate into the surrounding soil. Histopathological studies on ichthyofauna are important in understanding the effect of heavy metals on the structural integrity of vital organs at cellular level. Information on the acute toxicity of chromium to fishes is mostly on the survival, growth, biochemical and physiological aspects. Hence, in the present investigation an attempt was made to study the histological structures of liver and muscle of *Tilapia mossambica* under chromium stress. Thus, the ground water sources are exploited to their fullest potential and polluted to a great extent. A state of severe pollution results from the cluster of tanneries in close proximity to each other. This study was carried out to assess the magnitude of the pollution problem. The aim was to analyze and understand the toxic effects of tannery effluents on agriculture aquatic fauna and the day-to-day life of people in Puliyanakannu Village, SIPCOT Industrial Estate of Ranipet.

2. Materials and Methods

2.1. Tannery Effluents

Tannery effluents were collected from the Puliyanakannu Village, SIPCOT (Small Scale Industrial Promotive Corporation of Tamil Nadu) Industrial Estate of Ranipet. It is on the Chennai-Bengaluru National Highway and functioning under the supervision of Ranipet Municipality. This industrial complex is one of the biggest exporting centers for leather and chemicals. The SIPCOT area harbours 77 leather industries, 35 engineering industries, 18 chemical industries, 5 oil industries and 5 miscellaneous industries.

2.2. Test Species

Test species selected for the present study is *Tilapia mossambica* is a euryhaline and eurythermal teleost belonging to family Cichlidae which includes fresh water species and it is commonly available in all fresh water ecosystem. *Tilapia mossambica* is capable of successfully invading a variety of

environmental conditions. It also possesses high reproductive potentiality, easy availability, wide tolerance, high adaptability and also of commercial value.

2.3. Acclimatization

The fishes were collected from local fresh water pond. Fishes with the length of 102cm and weight of 152gm were transferred to large aquaria (95 x 50 x 45cm) for a period of two weeks acclimatization as suggested by[7]. During the period of acclimatization, the fishes were fed with dried prawn powder mixed with cooked rice and libitum. Water was changed on alternative days and temperature was maintained at $30\pm 1^\circ\text{C}$.

2.4. Measurement of Oxygen Consumption

The species of *Tilapia mossambica* (Family: Cichlidae) was obtained from inland fishing centers and maintained under normal laboratory conditions. The fish were fed with groundnut cake and pellets. Fishes were divided into two groups of 3 fishes each. The first group was exposed to 100% effluent and the second was maintained as control in normal tap water without the metal toxicant for 30days. Both of the groups were aerated and maintained well. At the end of the time schedule for each group, the fishes were sacrificed and liver, muscle was immediately isolated and fixed in 10% formaldehyde for two days. The tissues were washed gently in tap water, dehydrated in successive grades of ethanol, cleared in xylene and embedded in paraffin. Sections of 4-6mm were cut and stained with haematoxylin. Histopathological technique was carried out. Oxygen consumption of control fishes and fishes exposed to different concentrations of industrial effluents was separately measured using a simple respiratory chamber by measuring the loss of oxygen content of the water in the respiratory chamber. Oxygen content of the water samples was estimated and it was expressed in ml/gm/hour. Muscle and liver were isolated from the fishes and kept in an ice-trough and used for histological studies.

2.5. Histology of Liver and Muscle

Morphology of the hepatocytes and muscle fibres were demonstrated. Thin slices of liver and muscle were taken; treated with Bouin's fluid and formaldehyde for initial fixation. The mixture is ripe in 48hours and may be used when a film with a metallic luster continues to form on the surface of the mixture that has stood in a coplin jar for 24hours. Treat sections with Bouin's fluid for 16-24hours. Wash sections thoroughly in tap water to remove picric acid. Treat sections for 1minute with an equal parts mixture of 0.3% potassium permanganate and 0.3% sulphuric acid. Decolorize with a 5% solution of sodium bisulphite. Wash well in running tap water. Stain in the chrome haematoxylin solution for 10-15minutes. Rinse in water and differentiate in 1% acid alcohol for 1minute to remove background staining. Wash in running tap water until the section is a clear red. Stain in 0.5% aqueous phloxine 5minutes. Rinse in water and treat with 5% phosphotungstic acid. Wash in running tap water for 5minutes when the section should regain its red colour.

3. Results

Oxygen consumption of control fishes and fishes which were exposed to industrial effluents is reported. The rate of oxygen consumption in control fishes is 3.264ml/gm/hour. 100% oxygen consumption shows significant decrease in the percentage oxygen consumption of fishes exposed to increasing percentage of

effluents. In the percent concentrations 1, 2, 3, 4, 5, 6, 7 and 7.5, the percent oxygen consumption varies from 86.060, 73.713, 60.416, 44.730, 35.049, 34.620, 17.340 and 12.469 in the decreasing order. Similarly when calculation was made for unit metabolism, the unit metabolism in controlled fish was found to be 0.2176ml/gm/hour and there was similar decrease with the increase in the percentage concentration of effluents.



3.1.Histology of Normal Liver

The liver of fish comprises of a continuous mass of hepatic cells arranged in cords. There is no clear division of hepatic cells into lobules. The hepatocytes are large in size and the nucleus is centrally situated. The pancreatic tissue is distinct with a well-developed cellular and a large number of blood sinusoids are found in hepatic mass.

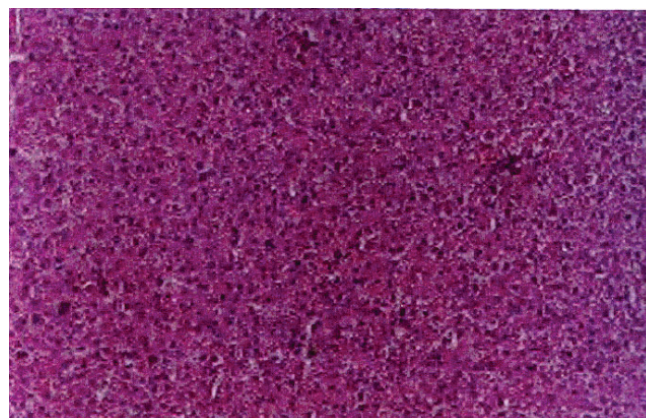


Table -1.Physico-Chemical Characteristics of Tannery Effluent

Parameter	Acceptable Limits	Values Recorded
Colour	----	Brown
Odour	Not desirable	Odour of Iron
pH	6-9 @	3.7
DO (cc/lit)	4-6 @	Nil

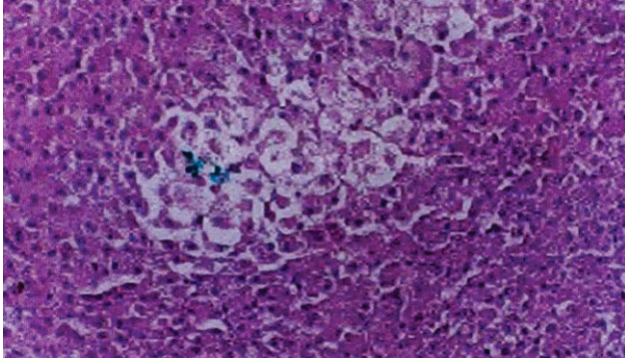
@ ISI Standards for disposal of industrial wastewater

Table 2. Oxygen Consumption of Control and Treated Fishes

Concentration of the Effluent	Oxygen Consumption	Unit Metabolism	Percent Oxygen Consumed	Percent Unit Metabolism
Control	3.264 ± 0.3	0.2176	-	-
1%	2.809 ± 0.4	0.1872	86.060 (-13.94)	86.029 (-13.970)
2%	2.406 ± 0.2	0.1604	73.713 (-26.287)	73.713 (-26.287)
3%	1.972 ± 0.5	0.1314	60.416 (-39.584)	60.385 (-39.613)
4%	1.460 ± 0.15	0.0973	44.730 (-55.27)	44.715 (-55.284)
5%	1.144 ± 0.7	0.0762	35.049 (-64.951)	35.045 (-64.954)
6%	1.130 ± 0.07	0.0753	34.620 (-65.38)	34.604 (-65.395)
7%	0.566 ± 0.03	0.0377	17.340 (-82.66)	17.325 (-82.674)
7.5%	0.407 ± 0.08	0.0271	12.469 (-87.531)	12.454 (-87.545)

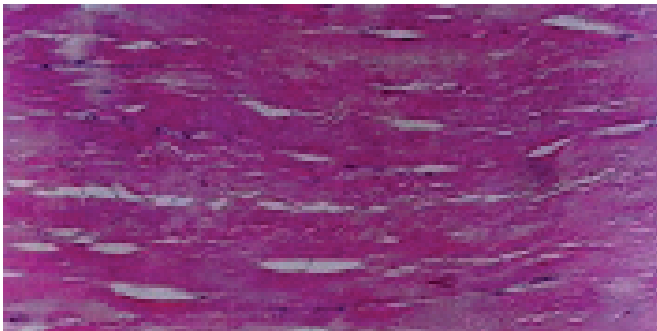
Oxygen consumption expressed in ml/gm /hour. Values are expressed in mean S.D. of six observations.

Tilapia mossambica when exposed to the tannery effluent water samples 30 days exposure, damage in cellular structure in the liver has observed. Focal vacuolar and diffused degeneration of hepatocytes were seen.



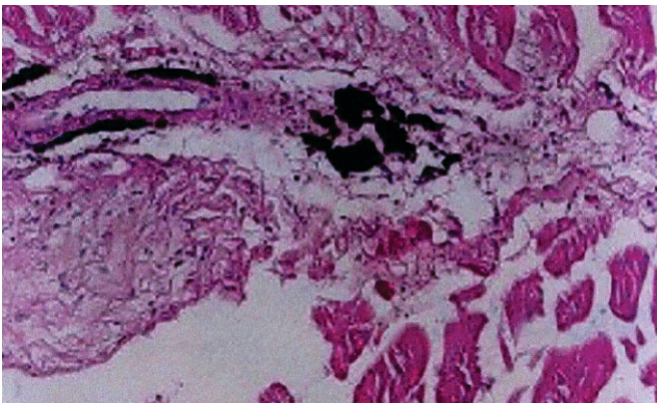
3.3. Histology of Normal Muscle

The muscle of fish is made up of muscle fibers. Layer of fibrils and connective tissue covers the muscle fibers and has a fluid portion called sarcoplasm, which has myofibrils and myofilaments. The myofilaments constitute the contractile elements of the muscle are innervated by nerves and blood vessels supply the blood to the muscle.



3.4. Histology of Experimental Muscle

The melanin deposition around the capillaries and arteries were noticed in the muscle tissue. The myofilaments were damaged.



4. Discussion

In the present study, the toxic contents of the tannery effluent affected the liver and muscle of *Tilapia mossambica* was impairing and functioning. Several investigators have pointed out histopathological changes in animals exposed to different pollutants [8, 9, 10]. Histopathological changes in different animals by chemical pollutant have been reported by several workers [11, 12]. The structural alteration induced by chromium in the gill of *Labeo rohita* was reported by [13]. The effect of distillery effluent on oxygen consumption of fresh water fish *Oreochromis mossambicus* [14]. The moderate histopathological alternation in gonads of *Channa punctatus* exposed to paper mill effluent was reported by [15]. [16] reported the effect of arsenite on certain aspects of protein metabolism in fresh water teleost, *Oreochromis mossambica*. Environmental pollutants disrupt the endocrine system of some wild fresh water fish were analyzed by [17].

5. Conclusion

Chromium compounds are used in ferrochrome, production, electroplating, pigment production and tanning. These industries, the burning of fossil fuels and waste incineration are sources of chromium in air and water. Most of the liquid effluent from the chromium industries is trapped and disposed of in landfills and sewage sludges the chromium being in the form of the insoluble trivalent hydroxide [18]. The toxicants enter the fish body mainly through the gill surface and hence they are the primary target organs to toxic stress. Similarly the structural alternations of liver and muscle were observed in the present study can be attributed to the sublethal effect of chromium from tannery effluents.

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