

Studies on the Effect of Electroplating Effluent on Biochemical Components of Fish *Cyprinus carpio*

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Abstract: Industrial effluents contributing to aquatic pollution contain a vast array of toxic substances, which include heavy metals. It leads to alteration in physical, chemical and biochemical properties of water bodies as well as that of environment. Electroplating effluent induces deleterious effects on non-target aquatic organisms resulting in imbalance of an ecosystem. The fishes *Cyprinus carpio* were treated with the sub lethal concentration of 5% Electroplating effluent for 24, 48, 72 and 96 hours. Another group was maintained as control. At the end of each exposure period, fishes were sacrificed and tissues such as liver, gill, muscle and kidney were dissected and removed. Samples were tested for carbohydrate, protein and lipid content in the various tissues of *Cyprinus carpio* under effluent stress were found to be decreased when compared with control. The use of biochemical approach has been advocated to provide an early warning to potentially damaging changes in stressed organisms. The health of any organism is influenced by the physiological activities taking place in body studies on the toxic effects of electroplating effluent on fish have shown varied responses according to the type of species and dosages.

Key words: *Cyprinus carpio* • Electroplating effluent • Protein • Carbohydrate • Lipid

INTRODUCTION

Water pollution [1] is the biggest threat of urbanization, industrialization and modern agricultural practices. Discharge of toxic elements from industrial process, mining and agriculture developments may have detrimental effects on aquatic animals [2]. The chemicals present in the industrial effluent affect the normal life of animal [3]. Among industries, electroplating industries plays an important role in creating heavy metal pollution in water bodies through direct discharge of effluent in water bodies. The continuous input of metals into the aquatic environment directly through waste disposal or indirectly via settling of waterborne particles necessitates a continued assessment of their effects on the ecosystem [4].

Biochemical parameters can show the impact of water pollution on fish [5]. *Cyprinus carpio* is an economically important freshwater fish and commonly cultured in many parts of the world). It appears to possess the same biochemical pathway to do with the toxic effects of

endogenous and exogenous agent as do mammalian species. [6]. Gills are the first organ to get affected by pollutants [7].

Biochemical profiles in fish and other aquatic organisms under heavy metal stress serve as important bio-indicators for monitoring of aquatic environment [8-10]. Metals ions are stable and are known to be persistent as environmental contaminants since they cannot be degraded or destroyed. They are harmful to aquatic life and water contaminated by them remains a serious public health problem to human. Electroplating industry is one of the industries where a various heavy metals are used during cleaning technique as a result of which the excess of metals released into the effluents. Pollution of heavy metals is realized due to their non biodegradability under such a situation, it becomes important to remove even the traces of contaminated metal in our ecosystem. The research is needed for efficient and cost-effective remedies. The use of biological materials for removing and recovering heavy metals from contaminated industrial effluents has emerged as a

potential alternative method to conventional techniques, which may be expensive or ineffective. The present study was to observe the effect of electroplating effluent on the biochemical component in gill, liver, kidney and muscle of fresh water fish *Catla catla*. The effluent from the electroplating industries is the cause of serious ground water and soil contamination in vicinity area which pose significant threat to human health and ecology.

MATERIALS AND METHODS

Test Organisms: Bulk of sample fishes, a *Cyprinus carpio* ranging in weight from 4-5 g and measuring 5-7 cm in length were procured from Aliyar Dam. Fishes were acclimatized to laboratory conditions for 2 weeks in a large Syntax tank. The water was changed twice in a day to maintain the oxygen content and to remove the excreta of fishes. The fishes were fed regularly with conventional diet rice bran and oil cake 1:1 ratio. Feeding was stopped two days prior to the experiment in order to keep the animal more or less in the same state of metabolic requirement. Fish were not fed during the toxicity tests. Fishes about the same size irrespective of sexes were selected for the experiment

Acute Toxicity Tests: The undiluted electroplating industrial wastewater collected, was considered as 100% solution. From this, the selected concentrations of the waste water for the experiments were obtained by diluting it with clean non-chlorinated ground water. In acute bioassay studies, the determination of LC50, the lethal concentration at which 50% of fish dies, is of prime importance to derive the sub-lethal concentrations so as to treat the organisms. The LC50 values for 24, 48,

72 and 96h exposure to electroplating industrial wastewater were obtained using Probit Analysis [11]. Fish were exposed to 5% electroplating industrial waste water in 96 hour were taken as the sub lethal concentrations.

Sampling: At the end of experimental period, the surviving fish were sacrificed by decapitation, dissected and tissues (liver muscle, gills and kidney) were isolated from control as well as the experimental fish. The tissues were homogenized with 80% methanol, centrifuged at 3500 rpm for 15 minutes and the clear supernatant was used for analysis of different parameters.

Biochemical Analyses: The total protein was estimated following the method [12]. Carbohydrate was estimated using the method [13] and the cholesterol was estimated using the method [14]. The results were expressed as mg/g wet weight of the tissue.

Statistical Analysis: All measurements were performed in average of three replicates. Data obtained was analyzed using the SPSS/PC+ Statistical package (ver.11.5). Significant difference between control and experimental groups were determined using Duncan’s test for multiple range comparisons.

RESULT AND DISCUSSION

Total Carbohydrate: In the present study the maximum reduction of carbohydrate content was observed upon 96 hours exposure (Table 2). In the tissues, the trend of decrease in carbohydrate content was liver > kidney> muscle> gill. Table shows that the change in

Table 1: Changes in Protein Content in the Tissues of *Cyprinus Carpio* Exposed to Electroplating Effluent For Different Exposure Periods

Tissues mg/g	Exposure Periods				
	Control	24 Hours	48 Hours	72 Hours	96 Hours
Gill	2.47±0.03	1.92±0.01	1.71±0.04	1.50±0.04	1.30±0.03
t-value		12.28**	15.58**	19.44**	23.84**
% change		22.26	30.76	39.27	47.37
Liver	2.75±0.11	2.23±0.04	1.89±0.04	1.68±0.21	1.42±0.03
t-value		4.03**	2.98*	9.18**	11.24**
% change		18.90	31.27	38.90	48.36
Kidney	3.10±0.07	2.70±0.06	2.18±0.03	1.78±0.04	1.52±0.03
t-value		9.98**	19.75**	22.95**	32.34**
% change		12.90	29.67	42.58	50.96
Muscle	4.11±0.04	3.64±0.03	2.74±0.03	2.01±0.03	1.98±0.03
t value		5.15**	12.30**	23.14**	18.28**
% change		11.43	33.33	51.09	51.82

Results are mean (±SD) of 5 observations % = Parenthesis denotes percentage increase/decrease over control. ** - Significant at 1% level, NS- Non Significant, *- Significant at 5% level

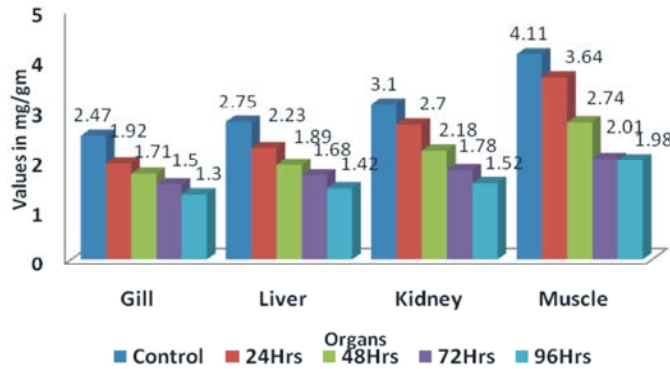


Fig. 1: Changes in Protein Content in the Tissues Of *Cyprinus Carpio* Exposed to Electroplating Effluent for Different Exposure Periods

carbohydrates was dependent significantly on the concentration of effluent as well as period of exposure for most of the tissues. Liver tissues showed 29.54, 27.24, 24.11 and 22.58 mg/g of protein in 5% of electroplating effluent and 31.10 mg/g of protein in control after 24, 48,

72 and 96 hours exposures. Table 1 shows the decreased value of protein content in kidney as 23.50, 19.10, 16.38 and 14.52 mg/g in 5% of electroplating effluent exposure and 28.10 mg/g in control after 24, 48, 72 and 96 hours exposures. In muscle tissues 20.68, 18.62, 16.50 and

Table 2: Changes In Carbohydrate Content In The Tissues of *Cyprinus Carpio* Exposed To Electroplating Effluent For Different Exposure Periods

Tissues mg/g	Exposure Periods				
	Control	24 Hours	48 Hours	72 Hours	96 Hours
Gill	23.89±0.03	20.32±0.04	18.78±0.03	15.34±0.01	13.29±0.03
t value		34.03**	53.28**	76.18**	94.24**
% change		14.94	21.39	35.78	44.37
Liver	31.10±0.05	29.54±0.04	27.24±0.03	24.11±0.04	22.58±0.01
t value		20.15**	65.31**	93.54**	102.61**
% change		5.01	12.41	22.47	27.39
Kidney	28.10±0.06	23.50±0.07	19.10±0.04	16.38±0.04	14.52±0.03
t value		16.18**	21.75**	42.95**	62.34**
% change		16.37	32.02	41.70	48.32
Muscle	26.47±0.03	20.68±0.01	18.62±0.04	16.50±0.04	14.30±0.03
t value		39.28**	52.58**	69.44**	73.84**
% change		21.87	29.65	37.66	45.97

Results are mean (±SD) of 5 observations % = Parenthesis denotes percentage increase/decrease over control.

** - Significant at 1% level, NS- Non Significant, *- Significant at 5% level

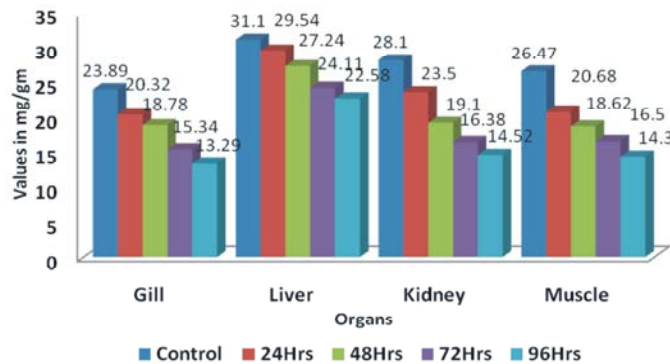


Fig. 2: Changes in carbohydrate content in the tissues of *cyprinus carpio* exposed to electroplating effluent for different exposure periods

14.30 mg/g of protein in 5% of electroplating effluent exposure and 26.47 mg/g in control after 24, 48, 72 and 96 hours respectively. The protein level in gill is also reduced. In control the protein level is 23.89 mg/g. It is decreased to 20.32, 18.78, 15.34 and 13.29 mg/g in 5% of electroplating effluent exposures for 24, 48, 72 and 96 hours

Protein: The level of protein was found to be decreased in all tissues compared to control. The decrease in protein content was muscle > kidney> liver> gill on 96 hours exposure (Table 1). Table 1 shows the maximum decreased value of protein content in gill as 1.92, 1.71, 1.50 and 1.30 mg/g in 5% of electroplating effluent exposure and 2.47 mg/g in control after 24, 48, 72 and 96 hours exposures. Muscle tissues showed 3.64, 2.74, 2.01 and 1.98 mg/g of protein in 5% of electroplating effluent and 4.11 mg/g of protein in control after 24, 48, 72 and 96 hours exposures. In kidney tissues 2.70, 2.18, 1.78 and 1.52 mg/g of protein

in 5% of electroplating effluent exposure and 3.10 mg/g in control after 24, 48, 72 and 96 hours respectively. The protein level in liver is also reduced. In control the protein level is 2.75 mg/g. It is decreased to 2.23, 1.89, 1.68 and 1.42 mg/g in 5% of electroplating effluent exposures for 24, 48, 72 and 96 hours.

Lipid: The present observation indicated that the extent of decrease in cholesterol was more in high concentration upon 96 hrs exposure (Table 3). In the tissues, the trend of decrease in lipid content was gill> kidney> muscle> liver. In the gill 39.43, 37.94, 33.19 and 28.18 mg/g of lipid in 5% of electroplating effluent exposure and 41.00 mg/g in control after 24, 48, 72 and 96 hours respectively. The lipid level in kidney is also reduced. In control the lipid level is 34.10 mg/g. It is decreased to 32.60, 30.24, 25.28 and 22.72 mg/g in 5% of electroplating effluent exposures for 24, 48, 72 and 96 hours. Table 1 shows the decreased value of lipid content in muscle as 30.20, 27.75, 24.50 and

Table 3: Changes In Lipid Content In The Tissues Of *Cyprinus Carpio* Exposed to electroplating Effluent For Different Exposure Periods

	Exposure Periods				
	24 Hours	48 Hours	72 Hours	96 Hours	
Control					
Gill	41.00±0.04	39.43±0.03	37.94±0.03	33.19±0.03	28.18±0.03
t value		30.10**	55.26**	90.14**	112.20**
% change		3.83	7.46	19.04	31.26
Liver	29.89±0.05	26.10±0.03	23.59±0.03	19.60±0.2	16.33±0.04
t value		30.03**	50.28**	86.28**	92.14**
% change		12.67	21.07	34.42	45.37
Kidney	34.10±0.04	32.60±0.05	30.24±0.02	25.28±0.02	22.72±0.03
t value		9.98**	49.75**	52.95**	82.34**
% change		4.39	11.32	25.86	33.37
Muscle	31.47±0.03	30.20±0.01	27.75±0.03	24.50±0.03	20.10±0.04
t value		29.28**	32.58**	49.44**	63.84**
% change		4.03	11.82	22.15	36.13

Results are mean (±SD) of 5 observations % = Parenthesis denotes percentage increase /decrease over control,

** - Significant at 1% level, NS- Non Significant, *- Significant at 5% level

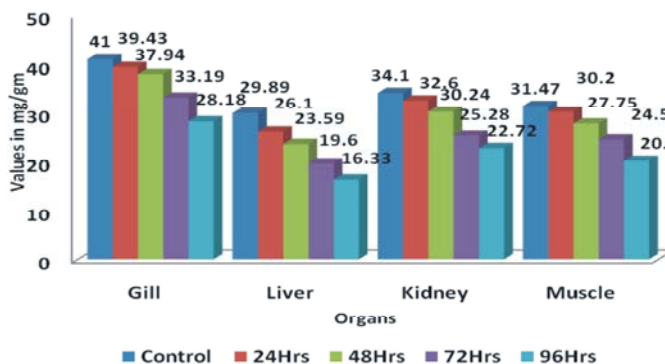


Fig. 3: Changes in Lipid Content in the Tissues Of *Cyprinus Carpio* Exposed to Electroplating Effluent for Different Exposure Periods.

20.10 mg/g in 5% of electroplating effluent exposure and 31.47 mg/g in control after 24, 48, 72 and 96 hours exposures. Liver tissues showed 26.10, 23.59, 19.60 and 16.33 mg/g of lipid in 5% of electroplating effluent and 29.89 mg/g of lipid in control after 24, 48, 72 and 96 hours exposures. The liver showed the highest percentage decrease (99.90) in cholesterol content). Lipids serve as energy source for fish metabolism and hence reveal their importance during stress condition. Decrease in muscle lipid indicates that lipid hydrolysis might be accelerated to derive energy to overcome toxic stress.

Environmental stress invokes compensatory metabolic activity in the organs of an animal through modification and modulation of the quantity and quality of problems. The changes in protein, carbohydrate and lipid levels in different tissues of fish after the treatment with electroplating effluent are presented in Tables 1 and 2 and 3 while analyzing the changes in the protein, lipid and carbohydrate it became clear that they fluctuated during different intervals of treatment.

In fishes, generally the carbohydrate reserves may be rapidly utilized under unfavorable conditions and the great variations found in the tissues indicate that the level of mobilizable carbohydrate reserves may fluctuate widely and rapidly in response to fluctuations in the nutritional state of the animal. Carbohydrates which supply the major portion of the metabolites for the energy requirements in a normal individual is oxidized for the energy requisites. Carbohydrates may be converted to glycogen or shunted in the metabolic pathway to supply the carbon chain for amino acids or converted in to fat [15]. At sublethal concentration, when the liver carbohydrate content decreased the blood sugar level increased which suggests the breakdown of liver glycogen (glycogenolysis). The mobilization of glucose from the liver to the blood and its availability for utilization by the needy tissues for ensuring normal metabolic processes in the body appears inevitable when the fish is exposed to toxic medium. Depletion of the glycogen content in the liver and muscle was also observed in fish *Mystus cavasius* exposed to electroplating industrial effluent. Depletion of glycogen in the liver and kidney suggests that these tissues do not contribute much anoxia resulting from resulting from pollution stress, since anoxia and hypoxia are known to increase carbohydrate consumption or may be due to generalized disturbances in carbohydrate consumption [16].

Proteins are important organic substance required by organisms in tissue building. The amino acid is the building blocks of proteins which are synthesized in the

body must be supplemented through diet. Since, the food value of fish is directly dependent on their protein content, the contamination by the toxic substance will reduce their nutritive value [17]. The alteration in protein value may also be related to some structural changes in the liver, the arrangement of hepatic words leading to the alteration of liver metabolism. The decrease in liver protein is also attributed to the inhibition of protein synthesis.

The decrease in protein content suggests an increase in proteolytic activity and possible utilization of its products for metabolic purpose. The fall in protein level during exposure may be due to increased catabolism and decreased anabolism of proteins [18]. A significant reduction in the levels of proteins and glycogen [19]. The gradual accumulation of toxicants present in the industrial effluent has been shown to cause drastic reduction in the protein content progressively in the fish tissues [20]. Proteins are mainly involved in the architecture of the cell. During chronic period of stress, they are also a source of energy [21]. The depletion of protein content in liver, muscle, kidney and gill tissues may have been due to their degradation and possible utilization of degraded products for metabolic purposes. In this work the protein content of *Cyprinus carpio* at different sub lethal concentrations of electroplating effluent decreased in all exposure periods. The depletion in tissue protein indicated rapid utilization of energy stores to meet the energy demands warranted by the environment.

Lipid is an important constituents of animal tissue, which plays a prime role in energy metabolism. Lipids are also important in cellular and sub-cellular membranes. A gradual decrease in lipid content in various tissues of *Cyprinus carpio* after treatments of electroplating effluent of various periods of exposure. Earlier researchers [22] also suggested that the decrease in lipid content in *Cyprinus carpio* may be either due to the uptake of lipid by the tissue for utilization at cellular levels or due to increased lipolysis or mitochondrial injury, which affect the fatty acid oxidation mechanism [23, 24]. The considerable decrease in total lipid in tissues might be due to drastic decrease in glycogen content in the same tissue which is an intermediate source of energy during toxic stress conditions [25]. After glycogen, lipid content may be used for energy production to overcome toxic stress. Some workers support these results in which lipid content decreased in animals after exposure to pollutants. Significant decrease in lipid of *Labeo rohita* when exposed to heavy metal cadmium [26]. The effect of dairy effluent on *O. mossambicus* and

reported that lipid content was decreased [27]. Reduction in protein, glycogen and lipid in tissues of freshwater fish *Labeo rohita* induced by heavy metals from electroplating industry [28]. Biochemical changes in freshwater fish, *Catla catla* on exposure to heavy metal toxicant cadmium chloride [29]. Changes in protein levels of certain tissues of freshwater fish *Heteropneustes fossilis* induced by copper [30].

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CONCLUSION

The present study reveals that electroplating effluent, is highly toxic to fish, leading to effect the nutritive value of the fish and all the metabolites studied are found to be sensitive which reflect changes in the normal activities of various functional systems. Thus biochemical alterations in fish can be considered as biomarkers to access the health status of the fishes as well as aquatic bodies polluted by toxicants. Thus environmental protection is the major requirement of the society.

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