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EFFECT OF ALUMINIUM SULPHATE ON BIOCHEMICAL AND HEMATOLOGY CHANGES IN *CATLA CATLA* FINGERLINGS-A TOXICITY STUDY

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ABSTRACT

In the present study was to assessing the Aluminium sulphate toxicity in freshwater fish as *Catla catla*. Fish samples were exposed to toxicants as aluminium for 72h and their cumulative mortality was calculated in 12 hours intervals. Results were were analyzed by SPSS 20 to obtain number of cumulative mortality and lethal concentrations. LC_{50} of *Catla catla* at 72hr was 50mg/L for Aluminium. The mortality at any fixed time increased with the increase in concentration and for a particular concentration mortality increased with the increase in exposure time due to accumulation of toxicants to a dangerous level leading to death. Overall it can be concluded that aluminium sulphate has significant toxicity to *Catla catla*. This study provides new evidence, using biochemical, oxidative stress markers, hematological, liver markers and histological parameters that exposure to levels of aluminium may be an important factor on fish in the acidified soft waters. It is concluded that the fishes can effectively used as monitors of water quality with respect to metals. Also, it can conclude that biochemical and histological parameters could be ranked as possible biomarkers of pollution.

KEYWORDS: *Catla catla*, Aluminium sulphate, Biochemical and histological parameters.

INTRODUCTION

Aquatic animals provide useful models for toxicological evaluations that bridge the gap between real world and laboratory problems. Select aquatic organisms are adaptable to laboratory experimentation in areas such as acute toxicity testing and chronic sublethal risks

evaluation, including such phenomena as carcinogenesis, mutagenesis anti teratogenesis. The metal which has a relatively high density and toxic at low quantity is referred as 'heavy metal', The excess quantities of metals are detrimental as these destabilize the ecosystems because of their bioaccumulation in organisms, and elicit toxic effects on biota and even death in most living organisms (Gupta *et al*., 2013).

Aluminium (Al) is the most widely distributed metal in the environment (Exley and House, 2011) occurring naturally in the trivalent state (Al^{+3}) as silicates, oxides and hydroxides, but may combine with other elements such as chlorine, sulphur, fluorine, as well as form complexes with organic matter (Martin, 1992). Environmental media may be contaminated by Al from anthropogenic sources and through the weathering of rocks and minerals. Weathering processes on rocks release more Al to the environment than human-related activities (Lantzy and MacKenzie, 1979). Exposures to Al occur in occupations associated with mining and processing of ore, scrap metal recycling, deployment and use of Al-containing compounds and products, and during engagement in Al metal cutting, sawing, filing and welding. Animals and humans living in environments contaminated by industrial wastes may also be exposed to high levels of Al (Boran *et al.*, 2013). In the present study to investigate the biochemical, oxidative stress and histological studies of *Catla catla* on exposure to aluminum sulpahte.

MATERIAL AND METHODS

Collection and acclimation of experimental fishes

Fingerlings of *Catla catla* (average weight 6.45 g) were procured from Fish farm, Thitta, Thanjavur District, Tamil Nadu, India, using cast net and maintained in the laboratory in a glass aquarium tank and acclimated in aerated tap water with continuous aeration for two weeks prior to experimentation. During this period, fishes were fed with a known amount of fish food.

Experimental design

In this experiment, fishes of uniform size (length 7.50cm) and weight (6.45 gm) were segregated from the stock and acclimated for 3 days to the lab conditions, temperature (28±2°C), pH 7.5–7.8, and an almost normal photoperiod (12:12-h L/D). The fishes were divided into two groups (one control and one experimental groups) of ten individuals each and introduced into the trough containing dechlorinated tap water. All the experimental fishes were administered orally with the respective material.

Group I : Control fish with 0.09% saline,

Group II : With a single dose of Aluminium sulphate (50mg/L)

Aluminium sulphate was first dissolved in water at 70°C and then cooled to 37°C before administration. The fishes were killed at 24 h by decapitation. Tissue samples from the muscles were extracted and prepared for biochemical analysis. A dup licate was run simultaneously and the pooled samples were analysed.

BIOCHEMICAL ESTIMATIONS

Preparation of homogenate

The 1g tissue (Liver and muscle) was weighed and homogenized using a Teflon homogenizer. Tissue homogenate was prepared in 0.1 M Tris Hcl buffer (pH 7.4) and used for the estimation of various biochemical parameters. Protein was estimated by the method of Lowry *et al.* (1951). Total lipids in tissues was estimated by the method of Folch *et al.*, (1957). Carbohydrate present in fish tissues was quantified using Anthrone method. Malondialdehyde was estimated by the thiobarbituric acid assay method of Beuge and Aust (1978). Reduced glutathione was estimated by method of Moron *et al* (1979). The activity of mitochondrial glutathione peroxidase was assayed by the method of Rotruck *et al* (1973). Copper-zinc superoxide dismutase activity was determined by the procedure of Kakkar *et al.* (1984) in plasma. The activity of catalase was assayed by the method of Beers and Sizer (1952). The SGOT and SGPT were estimated by the method of Reitman and Frankel (1957). Hematological profile was estimated by Cyanmethaemoglobin method (Dacie and Lewis, 1968) (Beacon Diagnostic Kit).

Histopathological studies

Histological studies of liver, muscle and gill tissues were carried out by the method of (Ochei and Kolhatkar, 2000). The various steps involved in the preparation of tissues for histopathological studies were fixation, dehydration, clearing, impregnation, embedding, section cutting, staining and mounting.

RESULTS

The results of toxic effects of Aluminium sulphate on *Catla catla* fingerlings, in terms of proximate composition, oxidative stress markers, liver function analysis, hematological changes and histological alterations were given in the following (Table 1 to 5 and Plate 1).

Bioassay toxicity tests

Acute toxicity of 72 hour 50mg/L of Aluminium sulphate exposed on *Catla catla* fingerlings were depicted in figure 1. The toxicity tests were carried out with the 50mg/L of Aluminium sulphate based on the 72 hour 60% of mortality rate were observed (table 1). On the basis of toxicity study, 50mg/L of Aluminium sulphate used further studies

Table 1: Aluminium sulphate against the *Catla catla* fingerlings (Total exposed fish 10).

Hours (50mg/L)	Fish mortality (Per 10 fish)	% of Fish mortality
12	0	0
24	2	20
48	4	40
72	6	60

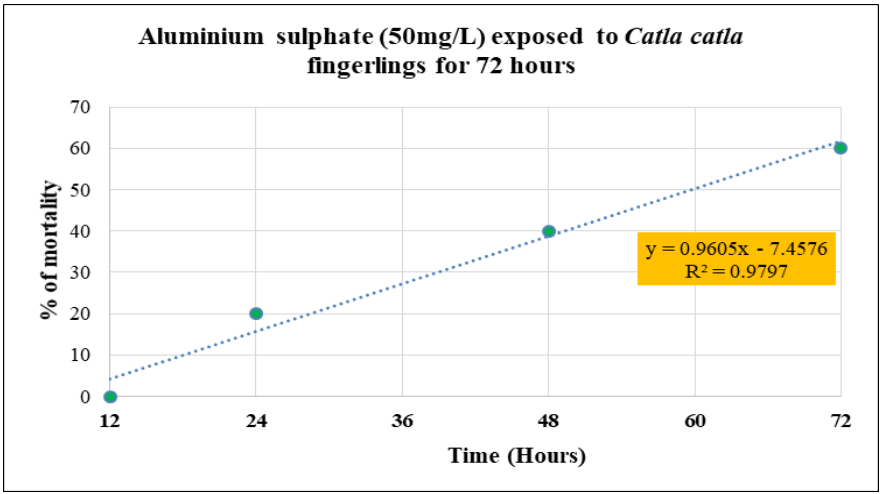


Figure 1: Aluminium sulphate against the *Catla catla* fingerlings

Effect of Aluminium sulphate on proximate composition of *Catla catla* fingerlings

Aluminium sulphate toxicity on fish was observed for the alteration in proximate composition viz. Protein, carbohydrate and lipid of muscle tissues when exposed to 50mg/L of Aluminium sulphate for 72 hours (table 2).

Table 2: Effect of Aluminium sulphate on proximate composition in *Catla catla* fish muscles.

Parameters	Group I (Control fish)	Group II (Experimental fish)	*P* value
Protein (mg/gm)	85.47±2.19	73.95±2.33	**P*<0.05
Carbohydrate (mg/gm)	31.61±1.57	23.18±1.44	**P*<0.05
Lipids (mg/gm)	12.84±0.65	7.92±0.71	**P*<0.05

Values are expressed as Mean ± standard deviation for 3 experiments. Data was calculated by student t-Test (Independent sample, P value two tail) using MS-excel ver. 2013. Statistically significant level 0.05. *P<0.05 statistically significant differences and NS Non- significant.

In 50mg/L of Aluminium sulphate expose to *Catla catla* fingerlings, the protein of muscle in group I (85.47±2.19 mg/gm tissues) and group II (73.95±2.33 mg/gm tissues) were observed. The exposed (group II) fish also significant decrease compare with control (group I). In 50mg/L of Aluminium sulphate expose to *Catla catla* fingerlings, the carbohydrate of muscle in group I (31.61±1.57 mg/gm tissues) and group II (23.18±1.44 mg/gm tissues) were observed. The exposed (group II) fish also significant decrease compare with control (group I). In 50mg/L Aluminium sulphate expose to *Catla catla* fingerlings, the lipids of muscle in group I (12.84±0.65 mg/gm tissues) and group II (7.92±0.71 mg/gm tissues) were observed. The exposed (group II) fish also significant decrease compare with control (group I).

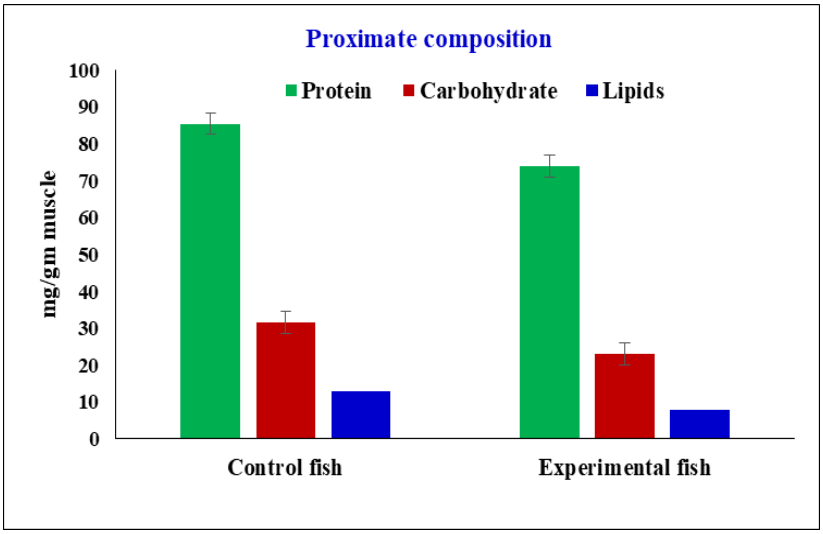


Figure 2: Effect of Aluminium sulphate on proximate composition in *Catla catla* fish muscles.

Effect of Aluminium sulphate on oxidative stress markers in *Catla catla* fingerlings

Toxicity of Aluminium sulphate on oxidative stress markers viz. lipid peroxidation, reduced glutathione, superoxide dismutase, glutathione peroxidase, and catalase in liver tissues of and *Catla catla* were studied (table 3).

Table 3: Effect of Aluminium sulphate on oxidative stress markers in *Catla catla* liver.

Parameters	Group I (Control fish)	Group II (Experimental fish)	*P* value
MDA (nmol/L)	1.63±0.43	4.96±0.74	**P*<0.05
Reduced glutathione (GSH) U/ml	26.41±2.37	35.84±2.79	**P*<0.05
Superoxide dismutase (SOD) U/ml	5.53±1.25	9.12±1.72	**P*<0.05
Catalase (Cat) U/ml	0.95±0.07	3.76±0.15	**P*<0.05
Glutathione peroxidase (GPx) U/ml	3.14±0.23	6.59±0.42	**P*<0.05

Values are expressed as Mean ± standard deviation for 3 experiments. Data was calculated by student t-Test (Independent sample, *P* value two tail) using MS-excel ver. 2013. Statistically significant level 0.05. **P*<0.05 statistically significant differences and NS Non- significant. In 50mg/L Aluminium sulphate expose to *Catla catla* fingerlings, the MDA of liver in group I (1.63±0.43 nmol/L) and group II (4.96±0.74 nmol/L) were observed. The exposed (group II) fish also significant increase compare with control (group I). In 50mg/L Aluminium sulphate expose to *Catla catla* fingerlings, the Reduced glutathione (GSH) of liver in group I (26.41±2.37 U/ml) and group II (35.84±2.79 U/ml) were observed. The exposed (group II) fish also significant increase compare with control (group I). In 50mg/L Aluminium sulphate expose to *Catla catla* fingerlings, the Superoxide dismutase (SOD) of liver in group I (5.53±1.25 U/ml) and group II (9.12±1.72 U/ml) were observed. The exposed (group II) fish also significant increase compare with control (group I). In 50mg/L Aluminium sulphate expose to *Catla catla* fingerlings, the Catalase (Cat) of liver in group I (0.95±0.07 U/ml) and group II (3.76±0.15 U/ml) were observed. The exposed (group II) fish also significant increase compare with control (group I). In 50mg/L Aluminium sulphate expose to *Catla catla* fingerlings, the Catalase (Cat) of liver in group I (3.14±0.23 U/ml) and group II (6.59±0.42 U/ml) were observed. The exposed (group II) fish also significant increase compare with control (group I).

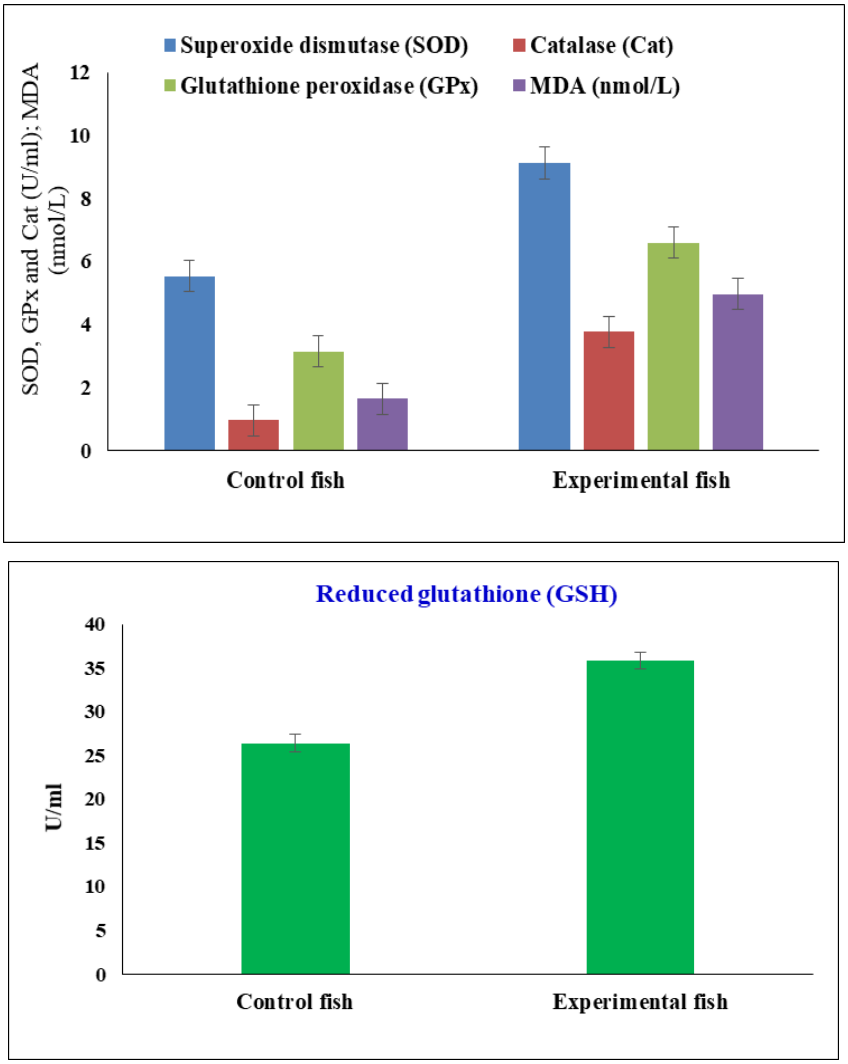


Figure 3: Effect of Aluminium sulphate on oxidative stress markers in *Catla catla* liver.

Effect of Aluminium sulphate on liver function in *Catla catla*

Catla catla treated with Aluminium sulphate were investigated for plasma enzyme activities such as Aspartate transaminase (AST) and Alanine transaminase (ALT) to analyse the liver function. The exposed (group II) fish also significant increase compare with control (group I) in AST and ALT (table 4).

Table 4: Effect of Aluminium sulphate on liver function in *Catla catla.*

Parameters	Group I (Control fish)	Group II (Experimental fish)	*P* value
AST (IU/l)	19.04±0.94	25.73±0.87	**P*<0.05
ALT (IU/l)	15.31±0.56	19.46±0.73	**P*<0.05

Values are expressed as Mean ± standard deviation for 3 experiments. Data was calculated by student t-Test (Independent sample, P value two tail) using MS-excel ver. 2013. Statistically significant level 0.05. **P*<0.05 statistically significant differences and NS Non- significant.

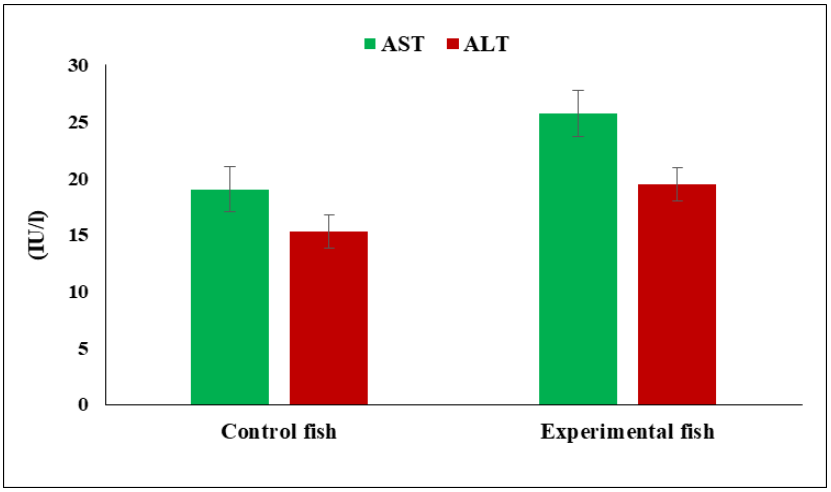


Figure 4: Effect of Aluminium sulphate on liver function in *Catla catla.*

Effect of Aluminium sulphate on haematological changes in *Catla catla* fingerlings

Blood is a sensitive marker which is affected with the environmental changes. Hence hematologic evaluation can be used in monitoring the health status of fish. Biochemical changes in blood values are particularly important to diagnose disease and toxicity in fish. A number of blood parameters such as total RBC count, total WBC count and haemoglobin (Hb) content have been used as indicators of metal pollution in the aquatic environment. In the present study haematological profile such as RBC, WBC and haemoglobin concentration were examined in *Catla catla* exposed to Aluminium sulphate (table 5).

Table 5: Effect of Aluminium sulphate on haematological changes in *Catla catla.*

Parameters	Group I (Control fish)	Group II (Experimental fish)	*P* value
Hb (g/dl)	5.32±0.35	3.18±0.41	**P*<0.05
RBC (Million/cu.mm)	1.76±0.05	1.04±0.09	**P*<0.05
WBC (Cu. mm)	6080.00±216.94	7400.00±209.71	**P*<0.05

Values are expressed as Mean ± standard deviation for 3 experiments. Data was calculated by student t-Test (Independent sample, P value two tail) using MS-excel ver. 2013. Statistically significant level 0.05. **P*<0.05 statistically significant differences and NS Non- significant. In 50mg/L Aluminium sulphate expose to *Catla catla* fingerlings, the Hb and RBC of blood in group I (5.32±0.35 g/dl and 1.76±0.05 Million/cu.mm) and group II (3.18±0.41g/dl and 1.76±0.05 1.04±0.09) were observed. The exposed (group II) fish also significant decrease compare with control (group I). In 50mg/L Aluminium sulphate expose to *Catla catla* fingerlings, the WBC of blood in group I (6080.00±216.94 Cu. mm) and group II

(7400.00±209.71 Cu. mm) were observed. The exposed (group II) fish also significant increase compare with control (group I).

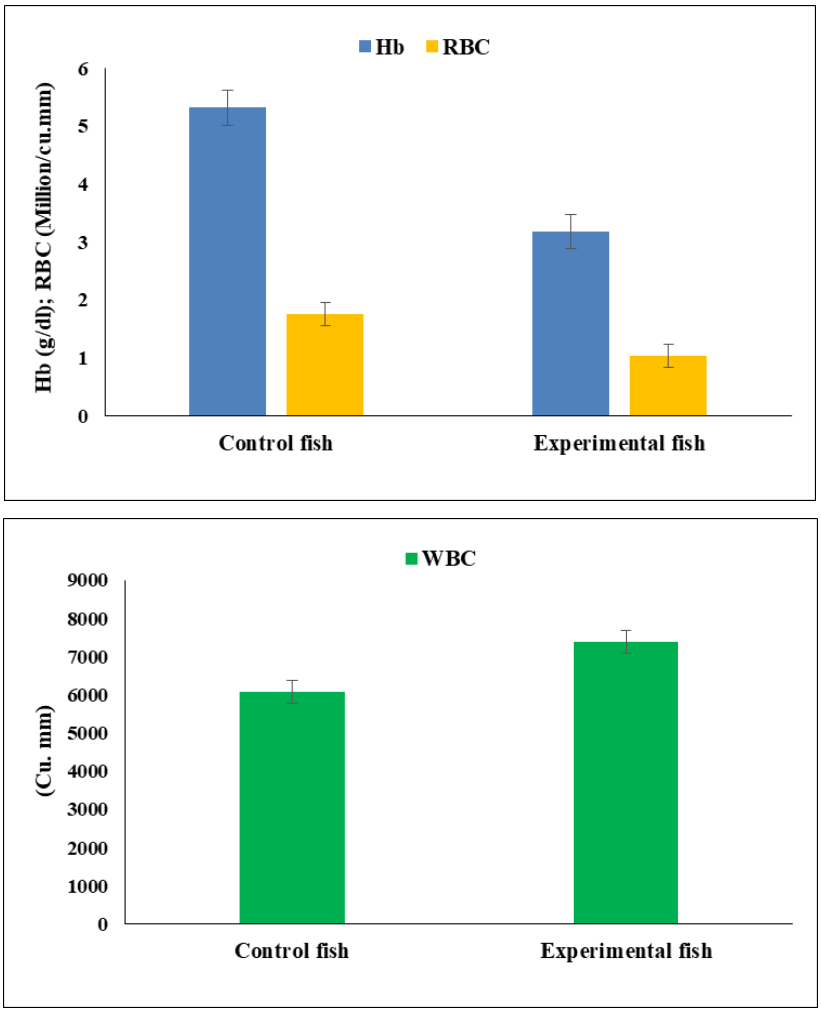
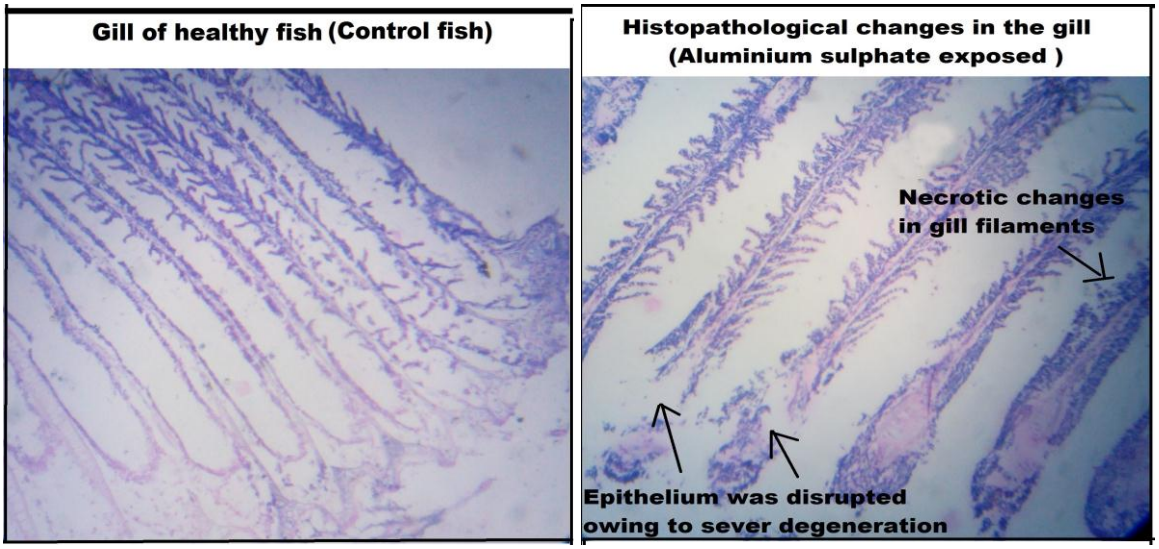


Figure 5: Effect of Aluminium sulphate on haematological changes in *Catla catla.*

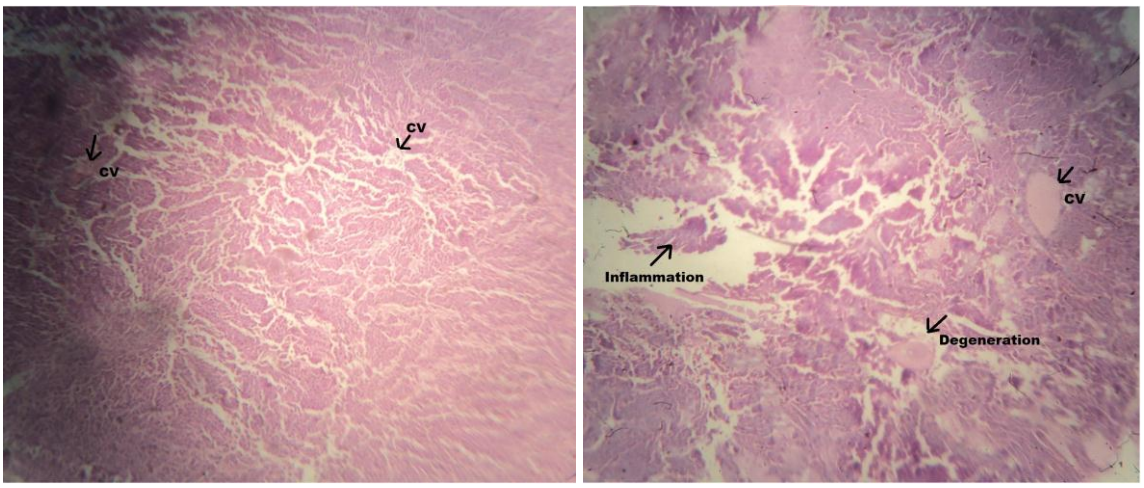
Histological observation of Aluminium sulphate exposed *Catla catla* fingerlings in gills and liver tissue

Histopathological alterations of gills and liver of *Catla catla* exposed to 50mg/L of Aluminium sulphate for 3 days were analyzed. Histological changes observed in gills and liver of *C. catla* on aluminium exposure (Pale 2). Gill observed the necrotic changes and sever degeneration as compared to control gill. Liver showed that sever inflammation and degeneration as compared to control liver.



Group I **Group II**

Histopathological observation of Gills



Group I **Group II**

Histopathological observation of Liver

Plate 2: Histopathological observation of Gills and Liver in *Catla catla* treated with Aluminium sulphate (CV – Central vein).

In control (group I) fish of *Catla catla* the secondary gill lamellae (SGL) appeared as finger like structures. The SGL were thin, slender and attached on either side of the primary gill lamellae (PGL). The secondary gill lamellae were highly vascularised and surrounded by a thin layer of epithelial cells. The overall observation of present investigation indicated that marked (group II) histopathological changes were found in the gills of *C. catla* 3 days exposure of Aluminium sulphate 50mg/L. In control (group I) fish of *Catla catla* hepatic cells were polygonal in shape with distinct nuclei. Large number of blood sinusoids and normal

veins were also seen around the hepatocytes. Present investigation observed marked (Group II) histopathological changes in the liver of *C. catla* exposed to Aluminium sulphate 50mg/L for 3 days.

DISCUSSION

Acute toxicity is referred as the harmful effects on organisms after a short period of exposure to a toxicant. It is helpful to understand the quantity of a toxicant below which it may be considered safe for the toxicant in the environment (Ay *et al*., 1999). These toxicity studies are the very useful in evaluating the water quality parameters required for fish's survival. In aquatic toxicology, mortality is the important factor since it is easy to evaluate and has obvious biological and ecological significance.

Thus, an attempt was made in the current study to evaluate the 96 hours LC_{50} values of Aluminium sulphate on marine *Anadara rhombea.* Aluminium and its compounds have not part take any role in biological functions. The presence of Aluminium in any form in living organisms cause cytochemical and histopathological effects. The organic Aluminium compounds affects central nervous system. Whereas, Aluminium sulphate damage liver, digestive tract and kidney. Fish may absorb Aluminium directly from contaminated water or indirectly from feeding on organisms living in the contaminated water (Javed, 2003). Although several studies have been conducted to assess the toxicity of heavy metals to algae, the number of studies dealing with the toxic effect of heavy metal on aquatic animals is limited (Harmon *et al.,* 2005). In the present investigation, were analyzed by SPSS 20 to obtain number of cumulative mortality and lethal concentrations. LC_{50} of *Catla catla* at 72hr was 50mg/L for Aluminium. The mortality at any fixed time increased with the increase in concentration and for a particular concentration mortality increased with the increase in exposure time due to accumulation of toxicants to a dangerous level leading to death.

Measurement of biochemical parameters is a commonly used diagnostic tool in aquatic toxicology and bio monitoring. Biochemical biomarkers have been used in order to prevent irreversible damage in whole organisms, communities and ecosystems. The impact of a number of contaminants on aquatic ecosystems can be assessed by the measurement of their external levels in the surrounding water or sediments, or by determining some biochemical parameters in bivalves and other organisms that respond specially to the degree and type of contamination (Nicholson and Lam, 2005).

Biochemical approach has been advocated to provide an early warning of potentially harmful changes in stressed bivalves. Analysis of chemical substances in tissues and body fluids, toxic metabolites, enzymes activities and other biochemical variables have frequently been used in documenting the toxin interaction with biological systems. Tissues reflect the physiologic state of an animal because they are the products of intermediate metabolism (Artacho *et al.*, 2007).

The decreased content of protein due to stress under exposure to heavy metal toxicity. During stress conditions bivalves need more energy to detoxify the toxicant and to overcome stress. Since bivalves have fewer amounts of carbohydrates so next alternative source of energy is protein to meet the increased energy demand (Singh *et al.*, 2010). Our result agrees with the earlier report (Rajkumar and John Milton, 2011).

The decreased content of carbohydrates due to stress under exposure to heavy metal toxicity. During stress conditions bivalves need more energy to detoxify the toxicant and to overcome stress. Bivalves have high amounts of carbohydrates used as a source of energy to meet the increased energy demand under stress. A similar decrease in carbohydrate has been reported in bivalves exposed to copper sulphate (Padewar *et al*., 2011). The storage or mobilization of metabolic substrates such as glucose, glycogen, lactate, lipid, and protein are disrupted by exposure to several trace metals, including aluminium, manganese, nickel and metal mixtures in a polluted habitat (Levesque *et al.*, 2002).

Lipids form reserve food in the body of the organism and used as a third major source of energy during stress (Shandilya *et al*., 2010). In the present study investigated the protein content in different body tissues of aluminium treated *Catla catla*. Exposure of Aluminium chloride, protein content in *Catla catla* was significantly decreased in 72hrs exposure. The decreased content of lipids due to stress under exposure to heavy metal toxicity. Heavy metals are recognized as a strong biotoxicants, because of their persistent nature and cumulative action to the aquatic flora and fauna (Sharma and Agrawal, 2005). Phospholipids, also called structural lipids, are playing an important role in the cell membranes formation. Shaikh, (2011) has also studied the lipid alterations in various animals after exposure to toxicants.Over all, protein, carbohydrate and lipid content in different body tissues of aluminium treated *Catla catla* were investigated. Exposure of $AlCl_2$, protein, carbohydrate and lipid content in *Catla catla* were significantly.

All aerobic organisms produce free radicals as a side product during the reduction of molecular oxygen by mitochondria. Free radicals play an essential role in maintaining the physiological condition of the body. Oxidative stress is induced by excess accumulation of the reactive oxygen and nitrogen species (RONS) that can be able to damage basic components for cell function and survival. Oxidative stress is defined as the condition occurring when the physiological balance between oxidants and antioxidants is disrupted in favor of the former with potential damage for the organism (Vignini 2011).

Malondialdehyde (MDA) is the major aldehyde resulting from the peroxidation of biological membrane polyunsaturated fatty acid. MDA, a secondary product of lipid peroxidation is used as an indicator of tissue damage by series of chain reactions (Ray and Husain, 2002). In the present study, the MDA content in body tissues of aluminium treated *Catla catla* investigated. Exposure of Aluminium chloride, MDA content in *Catla catla* was significantly increased in 72hrs exposure. In the present study, the SOD, catalase, Gpx activities and GSH content in tissues of aluminium treated *Catla catla* investigated. Exposure of Aluminium chloride, SOD, catalase, Gpx activities and GSH content in *Catla catla* was significantly decreased in 72hrs exposure.

SOD catalyzes the dismutation of two superoxide anions ($O^{\bullet}_2$ − and $^{\bullet}OH$) to molecular oxygen and hydrogen peroxide (Fridovich, 1998). GPx and CAT catalyze the conversion of hydrogen peroxide to water. MDA is regarded as a useful biomarker for measuring the level of oxidative stress (Nesto *et al.,* 2007). There is a close relationship between environmental stress in bivalves and the rate of cellular reactive oxygen species (ROS) generation. When the rate of ROS production exceeds the rate of its decomposition by antioxidant defenses and repair systems, oxidative stress can be established by ROS leading to several toxic processes, including DNA damage, chemical carcinogenesis, lipid peroxidation activation and enzymatic inactivation, especially CAT, GPx and SOD. The activities of these antioxidant enzymes serve as protective responses to eliminate reactive free radicals. MDA and these antioxidative enzymes have been detected in a number of bivalve species (Charissou *et al*., 2004).

Present study observations show that changes in the activities of antioxidant enzymes reflect the time course of oxidative stress in the fish caused by Al, and could be used as potential biomarkers for ecotoxicological bioassays of heavy metals. Enzymes catalyze specific biochemical reactions in the body. Changes in their levels and properties alter the functional

ability of an organism. The diagnosis of organ disease/damage is aided by measurement of a number of non-functional plasma enzymes characteristic of that tissue or organ. The amount of enzyme released depends on the degree of cellular damage, the intracellular concentration of the enzymes and the mass of affected tissue. The concentration of the enzymes released reflects the severity of the damage. ALT and AST are enzymes normally present in the liver, heart, muscles and blood cells. They are basically located within hepatocytes. So when liver cells are damaged or die transaminases are released into blood stream, where they can be measured they are therefore of index of liver injury (Reitman and Frankel, 1957). Mild inflammatory conditions are also likely to release cytoplamic enzymes (Vasudha et al., 2006). In the present study, the ALT and AST activities in tissues of aluminium treated *Catla catla* investigated. Exposure of Aluminium chloride, ALT and AST activities in *Catla catla* was significantly decreased in 72hrs exposure.

Haematological profile of blood can provide important information about the internal environment of the organism. Hematological studies on fishes have assumed greater significance due to the increasing emphasis on pisciculture and greater awareness towards the anthropological pollution of marine water resources. Such studies have generally been used as an effective and sensitive index to monitor physiological and pathological changes in. Hematological indices like hemoglobin (Hb) content, total red blood cell count (RBC) and total white blood cell (WBC) may be altered in after exposure to heavy metals (Adhikari *et al*., 2004).

Blood characteristics are very sensitive to environmental pollutant and their response to undesirable materials is very fast, so in this study we detected different levels of blood cells and hematological parameters. Hematological profile as Hb, RBC and WBC in aluminium treated *Catla catla*. Exposure of $AlCl_2$, hematological profile in *Catla catla* was significantly altered i. They also reported that that the size of these bivalves had an influence on their haematological parameters. According to Babatunde *et al.*, (1992), any changes in the constituent component of blood sample when compared to the blood profile could be used to interpret the metabolic and health status of the animal. Gabriel *et al.*, (2011). Based on the results obtained from this study it has been concluded that aluminium exposed *Catla catla* exhibited alterations in various hematological parameters. So these parameters could be a useful indicator to understand the physiological state of which is under stress when exposed to aluminium toxicants.

Assessment of histological alterations is an important method adopted to assess the impacts of pollution. In general, organs were in direct contact with the surrounding environment, and the liver, kidney and muscles are the major target organs for toxicants (Oliva *et al*., 2009). In the present study, observation of metal accumulation in *Catla catla* tissues of gill and liver. Tissue changes in test organism exposed to a sublethal concentration of a toxicant is a functional response of an organism that provides information on the nature of the toxicant. The toxic effects of heavy metals with respect to histological changes have been reviewed (Waqar, 2006). Hence there is a need to study the histopathological changes in gills and liver of *Catla catla* in response to pollutants. Moderate alterations were observed in gill followed by liver of *Catla catla* due to gill was the first body tissue to exposure to toxicants. Histological approach is a useful tool for evaluating the toxicants action at tissue level providing data concerning tissue damage, structural and functional changes (Sprague, 1973) in tissues and organs. Histological analysis appreciates the pathological conditions by diagnosing the abnormalities or damages of the tissues of animals exposed to heavy metals.

CONCLUSION

Overall it can be concluded that aluminium sulphate has significant toxicity to *Catla catla*. This study provides new evidence, using biochemical, oxidative stress markers, hematological, liver markers and histological parameters that exposure to levels of aluminium may be an important factor on fish in the acidified soft waters. It is concluded that the fishes can effectively used as monitors of water quality with respect to metals. Also, it can conclude that biochemical and histological parameters could be ranked as possible biomarkers of pollution.

REFERENCES

1. Adhikari, S., Sarkar, B., Chatterjee, A., Mahapatra, C. T., & Ayyappan, S. Effects of Cypermethrin and Carbofuran on Haematological Parameters and Prediction of their Recovery in a Freshwater Teleost, *Labeo rohita* (Hamilton), *Ecotoxicology and Environmental Safety*, 2004; *58:* 220-226.
2. Artacho, P., Soto-Gamboa, M., Verdugo, C., & Nespolo, R. F. Blood biochemistry reveals malnutrition in black-necked swans (*Cygnus melanocoryphus*) living in a conservation priority area. *Comp. Biochem. Physiol.*, 2007; *146:* 283-290.

3. Ay, Ö. *et al*., Copper and lead accumulation in tissues of a freshwater fish Tilapia zillii and its effects on the branchial Na, K-ATPase activity. *Bulletin of environmental contamination and toxicology*, 1999; *62*(2): 160-168.
4. Babatunde, G. M., Fajimi, A. O., Oyejide, A. O. Rubber seed oil versus palm oil in broiler chicken diet. Effect on performance nutrient, digestibility, haematology and carcass characteristics. Animal Feed Science Technology, 1992; 35: 133-146.
5. Beers R and Sizer I. A spectrophotometric method for measuring the breakdown of hydrogen peroxide by catalase. Journal of Biological Chemistry, 1952; 195: 133.
6. Beuge JA and Aust SD. The thiobarbituric acid assay. *Methods in Enzymology*, 1978; 52: 306-307.
7. Boran AM, Al-Khatib AJ, Alanazi BS, Massadeh AM. Investigation of aluminum toxicity among workers in aluminum industry sector. *Eur Scient J.*, 2013; *9*: 440–451.
8. Charissou, A. M., Cossu-Leguille, C., & Vasseur, P. Relationship between two oxidative stress biomarkers, malondialdehyde and 8-oxo-7, 8-dihydro-2′-deoxyguanosine, in the freshwater bivalve Unio tumidus. *Science of the total environment*, 2004; *322*(1-3): 109-122.
9. Dacie J.V., and Lewis S.M., 1968. Practical Hematology, 4th edition J and A, *Churchill, UK.*, 37: 3-6.
10. Exley C, House E. Aluminium in the human brain. *Monatshefte fur Chemie*, 2011; 142: 357–363.
11. Folch J, Less, Stanley GHS. A simple method for the isolation and purification of total lipids from animal tissues. J. Biol. Chem., 1957; 226: 497-509.
12. Fridovich, I. Oxygen toxicity: a radical explanation. *J. Exp. Biol*, 1998; 210: 1 203-1 209.
13. Gabriel, U. U., Ezeri, G. N. O. & Opabunmi, O. O. Influence of sex, source, Health status and acclimation on the Haematology of Clarias gariepinus African Journal Biotechnology, 2011; 3: 463-437.
14. Gupta N, Kushwaha AK, Chattopadhyaya MC Adsorption studies of cationic dyes onto Ashoka (Saraca asoca) leaf powder. J Taiwan Inst Chem Eng, 2013; 43: 604–613.
15. Javed, M. Relationships among water, sediments and plankton for the uptake and accumulation of metals in the river Ravi. *Indus J. Plant Sci.*, 2003; *2*(4): 326- 331.
16. Kakkar P, Das B and Viswanathan PN. A modified spectrophotometric assay of SOD. *Ind J Biochem Biophy*, 1984; 21: 130-132.

17. Lantzy RJ, MacKenzie FT. Atmospheric trace metals: global cycles and assessment of man's impact. *Geochim Cosmochim Acta,* 1979; *43*: 511–525.
18. Levesque, H. M., Moon, T. W., Campbell, P. G. C., & Hontela, A. Seasonal variation in carbohydrate and lipid metabolism of yellow perch (Perm - Iavesc-ens) chronically exposed to metals in the field. *Aquat. Toxicol.,* 2002; *60:* 257-267.
19. Lowry O.H, Rosenbrough N.J, Farr A.L and Randall R.J., Protein measurement with the Folin's reagent. Journal of Biological Chemistry., 1951; 193: 265-276.
20. Martin RB. Aluminum speciation in biology. *Ciba Found Symp,* 1992; *16*: 95–125.
21. Moron M.S., DsePierre J.W and Manerwik K.B., Levels of glutathione, glutathione reductase and glutathione-s-transferase activities in rat lung and liver Acta of Biochemica, biophysica, 1979; 582: 67-68.
22. Nesto, N., Bertoldo, M., Nasci, C., & Da Ros, L. Spatial and temporal variation of biomarkers in mussel (Mytilus galloprovincialis) from the Lagoon of Venice, Italy. Mar. Environ. Res., 2007; 58: 287–291.
23. Nicholson, S., & Lam, P. K. S. Pollution monitoring in Southeast Asia using biomarkers in the mytilid mussel Perna viridis (Mytilidae: Bivalvia). *Environment International*, 2005; *31*(1): 121-132.
24. Oliva. M., Garrido, M. C., Sales, M. D., & Gonzalez de Canales, M. L. Sublethal and lethal toxicity in juvenile Senegal sole (*Solea senegalensis*) exposed to copper: *A preliminary toxicity range-finding test. Exp. Toxicol. Pathol*, 2009; 61: 113–121.
25. Padewar, S. K., A. B. Harkal, Jagtap, A. R., & Mudkhede, L. M. Study of carbohydrate metabolism in selected tissues of fresh water bivalves, (*Lamellidens marginalis*) under copper sulphate toxicity. *Asian Journal of Bio Science,* 2011; *6*(2): 246-247.
26. Ray, G., & Husain, S. A. Oxidants, antioxidants and carcinogenesis, 2002.
27. Reitman S and Frankel S. A colorimetric method for the determination of serum glutamic oxaloacetic and glutamic pyruvic transaminase. Am J Clin Path, 1957; 25: 56.
28. Rotruck JT, Pope AL, Ganther HE, Swanson AB, Hafeman DG and Hoekstra WG. Selenium: biochemical roles as component of glutathione peroxidase. Sci., 1973; 179: 588-590.
29. Shaikh, L. M. Treatment of acute otitis media in children under 2 years of age. *New England Journal of Medicine*, 2011; *364*(2): 105-115.
30. Shandilya, S. M., Nalam, M. N., Nalivaika, E. A., Gross, P. J., Valesano, J. C., Shindo, K., & Kono, T. Crystal structure of the APOBEC3G catalytic domain reveals potential oligomerization interfaces. *Structure*, 2010; *18*(1): 28-38.

31. Sprague, J. D. The ABC of Pollution Assays using Fish, Biological Methods for Assessment of Water Quality, *A.S.T.M. STP,* 1973; *528:* 6-30.
32. Vignini, A. Stroke and oxidative stress. In: Gadoth N, Go ¨bel HH (eds) Oxidative stress and free radical damage in neurology. Oxidative stress in applied basic research and clinical practice. Humana Press, Totowa, 2011; 137–152.
33. Waqar, A. Levels of selected heavy metals in Tuna fish. *Arab. J. Sci. Eng.,* 2006; *31:* 89-92.