

# **SUMMARY**

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With the extensive growth of industrial development the use of metals have grown enormously. The industries these days discharge large amounts of heavy metals in the aquatic environment through their effluent. Among the various environmental pollutants heavy metals and their salts constitute the most widely distributed group of highly toxic and long retained substances.

In the present thesis, Chapter-I deals with the introduction of the problem taken up for investigations. With the enormous growth of the industries, the load of the heavy metals in the aquatic environment has grown considerably, affecting the aquatic flora and fauna. The fish *Cirrhinus mrigala* (Ham.) and *Heteropneustes fossilis* (Bloch) are very common fish inhabiting ponds and rivers such as Yamuna and Betwa passing through district Jalaun. These fish form a major bulk of the edible variety of the fish locally. Hence, in the present study the effect of heavy metals, cadmium, mercury, and nickel on these fish has been undertaken.

Although much of work on the effect of heavy metals on fresh water and marine fish has been done, yet the literature on the effect of these metals on these fish is scanty. Chapter-II deals with the historical account of the research work, which has been undertaken by different workers on the effect of different metals and other pollutants on fish. The

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outbreak of Minamata disaster and Itai-itai disease in Japan due to consumption of mercury affected fish and cadmium contaminated rice had drawn the attention of the scientists worldwide to standardize the permissible limits of the metals in the aquatic systems. A voluminous account of the work on the effect of different heavy metals on fish has been done, which has been presented in this chapter.

In Chapter-III, the experimental programme has been discussed, wherein material and methods used for the present investigations have been given. Three heavy metals cadmium, mercury and nickel have been used in the form of cadmium sulphate, mercuric chloride and nickel sulphate. Two fresh water edible varieties of fish, *Heteropneustes fossilis* (Bloch) and *Cirrhinus mrigala* (Ham.) have been used to determine the toxicity of these metallic pollutants.

In the present investigation, the fish *H. fossilis* (Bloch) has been subjected to cadmium sulphate and nickel sulphate while *C. mrigala* (Ham.) has been treated with mercuric chloride. Short term and long term bioassay tests had been arranged. Observations were made for acute toxicity, causing mortalities among the fish used for tests. Median lethal concentration values (LC50 values) for different exposure durations, 24 hrs, 48 hrs, 72 hrs. and 96 hrs were ascertained. Lethal threshold concentration for each metal and fish were recorded. Behavioural changes due to metal treatment were noted. Histopathological changes were studied. The changes in growth in terms of changes in weight and length were studied in different

concentrations of the metallic pollutants for definite period of time. The accumulation of these metals in the vital organs was also observed. The methods for conducting these experiments followed EIFAC (1983)<sup>96</sup> and APHA (1985)<sup>3</sup>.

Chapter-IV describes the following results observed in the experiments and Chapter-V deals with the discussion in details. The studies on the toxicity of these heavy metals revealed that mercury is comparatively more toxic and the median lethal concentrations (LC50) were calculated by probit analysis and were observed for different durations to be 680 µg/l, 533 µg/l, 516 µg/l and 428 µg/l for 24 hrs, 48 hrs, 72 hrs and 96 hrs for *Cirrhinus mrigala* (Ham). The lethal threshold concentration, which is the minimum concentration of the metal, sufficient enough to cause first mortality within 96 hrs, was observed to be 50 µg/l and the minimum concentration at which 100% mortality was observed in 30 days exposure was recorded as 100 µg/l. The toxicity of cadmium sulphate was studied on **Heteropneustes fossilis** (Bloch). The lethal threshold concentration of cadmium sulphate was found to be 0.5 mg/l in which the first mortality was recorded in 48 hrs, Median lethal concentration (LC50) for cadmium sulphate were recorded as 4.6 mg/l, 2.85 mg/l, 2.62 mg/l and 2.31 mg/l for 24 hrs, 48 hrs, 72 hrs and 96 hrs durations. 6.0 mg/l concentration was found to be highly toxic which caused 100% mortality in 48 hrs.

The acute toxicity of nickel sulphate was studied on **Heteropneustes fossilis** (Bloch) which was found to be minimum of

three metals used. The lethal threshold concentration was recorded as 1.0 mg/l in which first mortality was observed in 96 hrs. The LC 50 values were calculated to be 11.36 mg/l, 8.8 mg/l, 7.25 mg/l and 6.6mg/l for 24 hrs, 48 hrs, 72 hrs and 96 hrs. At 12.0 mg/l concentration, all the test fish were recorded to be dead in 96 hrs.

The rate of opercular movement were observed to study the change in the respiratory frequency due to stress caused by the metallic pollutants. The fish showed the higher rate of opercular movement as first response to the effect of metals which declined after 24 hrs. or 48 hrs in different fishes and concentrations of the metallic pollutants used for the treatment to the fish.

The change in the swimming patterns and behaviour of the fish in the aquarium was studied due to the action of these metals. The change in the active swimming pattern was recorded which slowed down and the fish showed even sluggish movements in higher concentrations of the metals (the avoiding reaction of the fish declined to minimum).

The studies on the growth in length and weight of the fish were made. The maximum growth in length of *Cirrhinus mrigala* subjected to mercuric chloride was recorded to be 3.07 percent in 200 µg/l concentration while minimum growth (length) of 1.33 percent was observed in 300 µg/l concentration. In weight the maximum growth was recorded in 50 µg/l mercuric chloride while loss of weight by 1.6 percent was observed in 300 µg/l concentration. In the growth studies in *Heteropneustes fossilis* (Bloch) subjected to cadmium sulphate, the fish showed 3.17 and 2.98 percent growth in length in 0.5mg/l and 1.0mg/l while in other concentrations the growth was not observed.

In weight, the maximum growth of 3.49 percent was observed in 0.5 mg/l concentration. A loss in weight was registered in 2.0 and 2.5 mg/l concentrations. The effect of nickel sulphate was studied on *Heteropneustes fossilis* (Bloch) where maximum increase in length was found to be 0.47 percent in 1.0 mg/l and maximum gain in weight was recorded to be 2.96 percent in 1.0 mg/l nickel sulphate, while loss of weight, which was measured by 1.46 percent and 1.14 percent in 4.0 and 6.0 mg/l concentrations.

The histopathological studies were made to observe the effect of these metals on different vital organs of fish. These histopathological changes were observed in gills, liver and kidney of the fish. Due to the effect of metals, these organs showed lesions and different degrees of degenerative changes depending upon the dose i.e. concentration of the metals and the duration of the exposure.

The first symptom which was seen in gills, was the excessive secretion of the mucous, the first immediate protective response by a fish. Degenerative changes in gills included, vacuolar degeneration of the cytoplasm of the epithelial cells, necrosis of epithelial cells, eccentric displacement of nuclei, pycnosis, haemorrhage, hyperplasia and hypertrophy. Histopathological changes observed in liver are the degeneration of cytoplasm of hepatocytes, necrosis, hyperaemia, haemorrhage and engorgement of blood cells in blood vessels. Tubulonecrosis, degenerative changes in the epithelial cells of the tubules, haemorrhage and oedematous conditions of Bowman's capsule were the histopathological changes observed in the kidney of the fish.

These histopathological lesions were found to be severe and more prominent in higher concentrations of the metals and also in the fish, which were given a longer exposure to the metals. These histopathological lesions were found to be more severe and prominent in *Cirrhinus mrigala* subjected to mercuric chloride, lesser effect was observed in *Heteropneustes fossilis* subjected to cadmium sulphate and comparatively the least effect was observed in the fish subjected to nickel sulphate.

The accumulation of the metals, mercury, cadmium and nickel was also studied in the vital organs, gills, liver and kidney. The estimation of cadmium and mercury was done by dithizone method, APHA (1985)<sup>3</sup> and nickel was estimated by heptoxime method, APHA (1985)<sup>3</sup>. The gills, which form about 60 percent area, the largest surface of the fish to encounter the surrounding aquatic medium containing metal is found to register the maximum accumulation. The accumulation in gills, liver and kidney has been found to increase with concentration of the metal used and the duration of the exposure. The gills showed the minimum accumulation of 3.6 percent in 48 hrs, in 0.5 mg/l concentration of cadmium sulphate while maximum of 12.8 percent was found to accumulate in 5 days only. the liver registered the minimum and maximum accumulation of 1.2 and 4.25 percent in the same concentration of 0.5 mg/l and 5.0 mg/l of cadmium sulphate. The kidney accumulated 0.4 percent and 2.3 percent, the minimum and maximum values.

For mercury, the fish accumulated 3.1, 5.2, 5.6, 5.7 and 6.0 percent in gill in 24 hrs in 50, 100, 200, 400 and 500 $\mu$ g/l

mercuric chloride concentrations. The maximum accumulations in the gills were 25.0, 28.4, 31.0, 30.2 and 29.1 percent in 30, 30, 25, 20 and 20 days respectively. 1.3 percent mercury was found to accumulate in liver in 24 hrs while 10.4 percent metal accumulated in 30 days in 50  $\mu\text{g}/\text{l}$ . In 100 $\mu\text{g}/\text{l}$  concentration these values for the same durations were 3.4 and 11.9 percent. The maximum accumulation of the metal in the liver was recorded to be 13.2 percent in 400  $\mu\text{g}/\text{l}$  concentration in 20 days exposure. The kidney is the other important target organ for metals. The minimum accumulation in kidney was 1.1 percent while the maximum accumulation was 7.6 percent in 50  $\mu\text{g}/\text{l}$  and 400  $\mu\text{g}/\text{l}$  concentration in 24 hrs and 20 days respectively.

For nickel sulphate, the gill registered the minimum accumulation of 1.2 percent in 24 hrs. in 1.0 mg/l concentration while 16.5 percent was the maximum accumulation in 8.0 mg/l in 30 days exposure. These values for the liver were 0.5 percent and 4.5 percent in 1.0mg/l and 8.0 mg/l in 24 hrs and 30 days respectively. In kidney of the fish affected by nickel sulphate the accumulation was not of the order of other two metals. The minimum of 0.2 percent in 6.0 mg/l in 24 hrs exposure was accumulated in kidney while maximum of 2.6 percent accumulated in kidney after 30 days of exposure in 8.0 mg/l concentration of nickel sulphate.

The reason for accumulation of these metals in these vital organs has been attributed to the fact that these metals form complexes with ligands forming macromolecule which in the cells are unable to cross the smaller pores.

In the present investigations it is clearly indicated by the observations and the results that of the three metals taken to study the effect of fish, mercury is highly toxic as compared to cadmium followed by nickel. The standards formed by the Indian standards Institution be reviewed because it has been established that the LC50 value of different metals vary with other environmental conditions and their individual toxic effect differs when other metals interact simultaneously in the aquatic medium.