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A Study on Acute Toxic Effect in Labeo Rohita of Untreated Sewage Water

Akash Diwakar[1]* Archana Singh[2]

[1] Research Scholars, Department of Zoology, Sai Meer Degree College, Uttar Pradesh

[2] Supervisor, Department of Zoology, Sai Meer Degree College, Uttar Pradesh

Abstract – This research is intended to examine acute toxicity of untreated water in Labeo rohita. A 96-hour acute toxicity test to measure the LC50 of 5 concentrations of untreated sewage water was undertaken, namely, 10%, 25%, 50%, 70% and 100%. Results of the research demonstrate that in less than 24 hours, 100 percent of the fingerlings exposed to a concentration of untreated sewage water had a mortality rate. The exposure of the fingerlings to 10%, 25% and 50% UT likewise exhibited substantial ($p < 0.05$) development in the 96-hour timeframe. The research therefore found that untreated sewage water in Labeo rohita may have acute harmful effects, which need the treatment of waste water before it is released into water bodies.

Keywords – Toxicity, Toxic Effect, Acute Toxic Effect, Fish, Labeo Rohita, Sewage Water

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INTRODUCTION

Pollution of the aquatic environment is a severe and developing concern all over the globe these days. The increasing quantity and volume of industrial, agricultural, and commercial chemicals released into the aquatic environment has resulted in a variety of negative consequences on aquatic creatures, including fish. The pollution of aquatic systems with heavy metals has piqued the interest of various researchers in both developed and developing nations throughout the globe.

Fish consumption has risen dramatically in recent years as a result of its nutritional and therapeutic advantages. Heavy metal concentrations in fish tissues reflect previous or current exposure, with heavy metal intake mostly occurring via the gills, skin, or diet. Because metals are known to influence core metabolic pathways, they may have a significant negative influence on both human and animal life. As a result, there is increasing worry that metals deposited in fish tissues may pose a health danger, particularly to those who consume fish (1-3).

Toxicity refers to a substance's ability to have a negative or damaging impact on a living creature. Acute toxicity tests are used to see whether certain quantities of test material or effluent have a detrimental impact on a set of test organisms during a regulated short-term exposure. In India, the fast growth of the human population, along with fast urbanization, industrialization, and agricultural expansion, has had a significant influence on both the quality and quantity of water. Eutrophication and water quality degradation are caused by anthropogenic activities such as oxygen depletion, increased BOD & COD loads, changes in transparency, pH, phosphate, and nitrate levels. Pollutants in the environment have been shown to harm aquatic creatures. Aquatic communities, such as plankton, fish, and invertebrates, are under severe stress as a result of ecological exploitation. In this case, fish bioassays may be highly valuable for evaluating the quality of water that has been contaminated with complicated mixes of harmful compounds. Using fish bioassays, growth retardation and a range of pathological outcomes have been discovered in contaminated river waters. Bioassay studies may be used to test for both non-specific effects and specific effects. In order to effectively address the challenge of survival in a changing environment, an organism's behavior permits it to adapt to external and internal inputs. As a result, behavior is a selected reaction that is continually altering as a result of direct contact with the environment's physical, chemical, social, and physiological components. The health and fitness of aquatic organisms may be harmed by behavioral and morphological changes caused by sublethal toxicity. Contaminants typically alter locomotor activity, and fish swimming patterns are a highly structured species-specific response. Because behavioural markers of toxicity connect physiological function with ecological processes, they seem to be excellent for evaluating the impacts of aquatic pollution on fish populace. Importantly, studies are starting to link physiological alterations to behavioural disturbance, giving physiological toxicity measurements ecological importance (4).

Metals accumulate in fish at far greater quantities than in water and soil because they are at the top of the aquatic food chain. Heavy metals may be absorbed by fish via the epithelium or mucosal surface of their skin, their gills, and their gastrointestinal system.

Blood is a sign of an animal's physiological status, according to Zutshi, B., et al., 2010. As a result, a field research was carried out to look at the hematological parameters of the wild rohu population, Labeo rohita (Ham). Rohu fish of varied sizes and weights were collected for this study from Hebbal and Chowkalli lake. Hemoglobin content, RBC and WBC count, and PCV and MCHC values all showed significant variances. A substantial drop in RBC count (p 0.5), hemoglobin content, PCV and MCHC values, as well as an increase in leukocyte count and MCV values, were identified in fish from Chowkalli Lake, indicating severe anemia. Lake B fish exhibited less RBC and lower serum protein and lipid concentrations. In comparison to lake A, fish from lake B had greater glucose serum concentrations at first, subsequently lower concentrations (900-1,500 g). The change in parameter values may be related to fish exposure to various kinds of pollutants, which are mostly prevalent in the Chowkalli lake, which gets heavy metals, synthetic detergents, petroleum products, and other acid and alkali chemicals from surrounding local enterprises. Other characteristics of these fish are their black bodies and aggressive temperament (5).

MATERIALS AND METHODS

Collection and analysis of municipal sewage water

Municipal sewage waste water was collected from the Sewage Water Treatment (STP) facility in the hamlet of Bhattian in the Punjab state of India. This collecting location is situated inside the municipal borders of Ludhiana at 30o5757N and 75o4954E, 10.7 kilometers from Punjab Agricultural University in Ludhiana, Punjab, India. Two STPs are situated in Bhattian, each measuring 36.42 hectares and spanning 5.7 kilometers, and treating untreated sewage wastewater using two separate technologies, namely the Sequencing Batch Reactor (SBR) and the Upflow Anaerobic Sludge Digestor (UASB). The fingerlings were raised in water acquired from the facility utilizing SBR technology, which has a capability of processing 50 million litres per day (MLD) with a 90% effluent removal efficiency. Physico-chemical parameters such as pH, temperature, Biochemical oxygen demand (BOD), Dissolved oxygen (DO), and free carbon-dioxide (CO2) were measured in the control, untreated, and treated water using standardized procedures provided by the American Public Health Association (APHA).

Fingerlings collection and acclimatization

The Guru Angad Dev Veterinary and Animal Sciences University in Ludhiana, Punjab, India provided fingerlings of Labeo rohita (7.620.25 cm in length, 8.250.32 g in weight). The fingerlings were acclimatized to laboratory conditions in tanks (35 liters capacity) containing chlorine-free tap water that met acceptable temperature, conductivity, dissolved oxygen, and biochemical oxygen requirements for ten days. A steady photoperiod (12-hour light:dark cycle) was maintained through the acclimatization phase and studies.

During the acclimation stage, fingerlings were given commercial fish food ad libitum. One day before the trial, the feeding was stopped. Electrically controlled aerators (2 aerators/35 ltrs. tub) and filters (2 filters/tub) were used to keep the tubs aerated at all times. The experimental approach adhered to the Organization for Economic Cooperation and Development's guidelines (OECD).

Acute toxicity test

For 24, 48, 72, and 96 hours, two batches of seven healthy fingerlings (n=7) were exposed to dechlorinated tap water (control group), treated sewage water, and different concentrations of untreated sewage water (ten percent, twenty-five percent, fifty-five percent, seven-hundred percent, and one hundred percent). After 24, 48, 72, and 96 hours of exposure, the percent mortality rate of the fingerlings was observed in the exposed groups. In control and treated water, none of the fingerlings died; nevertheless, mortality was detected in fingerlings exposed to various quantities of untreated sewage water. When the fish did not react to a glass rod probe, it was assumed to be dead. The deceased specimens were removed from the tubs as soon as they were discovered. Robertson et al. [25] provided POLO software to determine the fatal concentration (LC50) of untreated sewage water to Labeo rohita over 96 hours at various concentrations of untreated sewage water.

Statistical analysis

CPCS I software was used to do a multifactor analysis of variance (ANOVA) to find significant differences between groups.

RESULTS AND DISCUSSION:

The fingerlings were exposed to control, treated, and various quantities of untreated sewage water (UT) for 96 hours in this investigation. The percent mortality rate and percent growth rate in fingerlings were assessed at 24, 48, 72, and 96 hours. There was no mortality in the fingerlings exposed to control, treatment, and 10% UT for 24, 48, 72, and 96 hours. However, two fingerlings died after 96 hours of exposure to a 25% UT concentration, four fingerlings died after 24 hours of exposure to a 50% UT concentration, and six fingerlings died after 48 hours of exposure to a 50% UT concentration, but none died after 72 hours. Six fingerlings died after 24 hours of exposure to a 75 percent UT concentration, and eight fingerlings died after 48 hours of exposure. All of the fingerlings exposed to water at a concentration of 100 percent UT died in less than 24 hours. The percent mortality in cases of control, 10%, 25%, 50%, 75%, and 100% was found to be 0, 0, 14.28, 71.42, 100, and 100%, respectively (Table 1).

Table 1: Mortality rate of Labeo rohita fingerlings exposed to control, treated and different concentrations of untreated sewage water (UT) at varied time intervals

Duration Groups	No. of test fishes	24 hr	48 hr	72 hr	96 hr	Total no. of dead fishes	Per cent mortality
Control	14	0	0	0	0	0	0
Treated	14	0	0	0	0	0	0
10% UT	14	0	0	0	0	0	0
25%UT	14	0	0	0	2	2	14.28
50%UT	14	4	6	0	0	10	71.42
75%UT	14	6	8	-	-	14	100
100%UT	14	14	-	-	-	14	100

The lethal concentration (LC50) of UT was estimated using POLO software after the examination of the mortality rate in fingerlings. The LC50 of UT was determined to be 51.70 mg/ltrs.

Table 2: The value of lethal concentration (LC50) of Untreated sewage water (UT) against *Labeo rohita* for cumulative experimental period of 96 hours

Parameter	Concentration of UT(mg/L)	Slope	Heterogeneity	Degree of freedom
LC50	51.70	51.64±2.45	0.00	3

During the acute toxicity test, the fingerlings' percent growth rate was also monitored every 24 hours. In contrast to the control and treatment groups, fingerlings subjected to 10%, 25%, and 50% concentrations of UT exhibited a significant reduction (p0.05) in percent growth rate after exposure. At 24, 48, 72, and 96 hours of exposure, the least growth rate was recorded in fingerlings subjected to a 50 percent concentration of UT, i.e. 0.550.08, 0.600.02, 0.710.21, and 0.980.46 (Table 3).

Table 3: Per cent growth rate of Labeo rohita fingerlings after exposure to control, treated and untreated sewage water (UT) at different time intervals

Groups Duration	24 hr	48 hr	72 hr	96 hr
Control	2.13 ± 0.02^{a1}	2.69 ± 0.01^{a1}	2.73 ± 0.18^{a1}	3.50 ± 0.41^{a1}
Treated	2.33 ± 0.12^{a1}	2.45 ± 0.09^{a1}	2.49 ± 0.16^{a1}	3.41 ± 0.09^{a1}
10% UT	0.79 ± 0.04^{a2}	0.82 ± 0.03^{a2}	1.60 ± 0.05^{b2}	1.63 ± 0.06^{b2}
25%UT	0.75 ± 0.02^{a2}	0.80 ± 0.09^{a2}	1.24 ± 0.13^{b3}	1.98 ± 0.12^{b3}
50%UT	0.55 ± 0.08^{a3}	0.60 ± 0.02^{b3}	0.71 ± 0.21^{c4}	0.98 ± 0.46^{d4}
75%UT	-	-	-	-
100%UT	-	-	-	-

Values are Mean±S.E

CD (5%): A (Groups) = 0.034561, B (Duration) = 0.024561, AB (Interaction) = 0.045361

Values in rows with distinct numeric superscripts (1-4) are substantially different (p0.05).

The values in columns with distinct alphabetic superscripts (a-d) vary considerably (p0.05).

The findings of this research demonstrated that when fingerlings were exposed to various concentrations of UT for 96 hours, they died and grew at a slower pace. The measured value of LC50 of UT indicated that some harmful impurities/substances were present in untreated sewage water, causing death and a reduction in fingerling growth rate. The value of LC50 calculated after exposing Labeo rohita fingerlings to waste water collected from different sites of Tung Dhab Drain was 72.45 percent for Tung Dhab drain near a paper mill outlet and 83.20 percent for another site of Tung Dhab drain near village Mahal, according to a study conducted. During this acute toxicity test, the mortality rate in Labeo rohita fingerlings was shown to rise proportionately with an increase in the concentration of wastewater taken from Tung Dhab drain, Amritsar, after a 24 hour interval for 96 hours. At 100 percent concentration of wastewater recovered from both locations, the percent mortality was measured as 100 percent (6). Similarly, using Nile tilapia, Oreochromis niloticus, as the test organism, an experiment was undertaken to determine the acute toxicity level of effluents from the inlet and outflow of the biological lagoons of the Hawassa Textile waste treatment facility. The data was collected for 24, 48, 72, and 96 hours in order to investigate the effects of effluent toxicants on Oreochromis niloticus behavioural responses and survival rate. The fish supplied at lower effluent concentrations exhibited normal swimming behavior, but the fish placed at higher effluent concentrations exhibited irregular swimming, gasping, and frequent surfacing behavioural reactions. There was no fish mortality in the control group, and the outlet effluent concentration was 10% (v/v). At 100 percent (v/v) inlet effluent concentration, the greatest percentage mortality was found, followed by 100 percent (v/v) outlet and 40 percent (v/v) inlet effluent concentrations. Inlet and outlet wastewater had 96-hour LC50 and acute toxicity unit (ATU) values of 30.5 percent (v/v), 3.279, 71.5 percent (v/v), and 1.399, respectively. The treatment plant's overall efficiency level for removing toxicants was found to be 57.33 percent (v/v) in this investigation. However, the treatment plant's efficiency need be enhanced in order to utilize the water for irrigation and other household uses; otherwise, using wastewater in its current state is dangerous (7).

The LC50 values for wastewater revealed in this investigation are consistent with the values reported by different employees to Labeo rohita when exposed to municipal wastewater from various sources. After exposing Channa striatus to this wastewater for 96 hours, Yadav et al. found an LC50 value of 70.0 percent of industrial wastewater acquired from a fertilizer company discharge point at Phulphur, Allahabad. Similarly, Lopamudra conducted an LC50 - 96 hr test to evaluate the safe sewage water concentration level for Labeo rohita survival and development. They gathered sewage water from a sewage canal in East Kolkata Corporation's metropolitan metropolis, and used several sub-lethal concentrations to compute the LC50 of the sewage water, such as 25%, 50%, 75%, and 100%. (8).

Using a fish bioassay on Lebistus reticulate, the toxicity of the effluent was also determined. The wastewater's LC50 value was found to be 6%, indicating that the effluent was hazardous in nature. The presence of excessive organochlorine pesticides (OCs) in the wastewater (1.719 mg/l), as well as sulphide concentrations of 17.60 mg/l, which were both greater than the mandated level of 2.0 mg/l, generated stench in the surrounding area. It also included up to 80 mg/l of oil/grease. The dissolved oxygen level of the water was lowered by a layer of floating oil on the surface. Toxicity was transferred to the fish by all of these factors. This research concluded that prior to release of raw wastewater into surface water bodies, toxicity testing using fish bioassays should be performed to determine a dilution factor (9).

Patro discovered that after being exposed to industrial wastewater effluent for 7, 14, 21, and 28 days, the growth rate of the freshwater fish Oreochromis mossambicus decreased considerably, with an additional 28 days of recovery. In contrast to the control, the percent drop in body weight was 7.2 percent, 14.96 percent, 25.06 percent, and 31.89 percent at the 7th, 14th, 21st, and 28th days of exposure, respectively. In the control fish, there were no evidence of poisoning. Recovery was also shown to be non-significant in the exposed group, with a 47.96 percent drop on the 28th day of recovery instead of any recovery. Under stress, restlessness, and agitated movements, a reduction in feeding activity, loss of body fluids, and loss of ions from the body may result in a drop in body weight and percent growth rate of fish (10-12).

CONCLUSION:

According to the findings, the lethal concentration (LC50) of untreated sewage water confirms the existence of poisonous organisms in the water. According to the findings, untreated sewage has the ability to cause acute toxic effects in Labeo rohita fingerlings, and the toxic effects in fingerlings were shown to grow as the time of exposure increased.

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Corresponding Author

Akash Diwakar*

Research Scholars, Department of Zoology, Sai Meer Degree College, Uttar Pradesh