

Behavioral and morphological changes in a freshwater fish, *Labeo rohita* exposed to tannery industry effluent



Zoology

KEYWORDS : Acute toxicity, behavioral responses, morphological changes, tannery industry effluent, *Labeo rohita*

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ABSTRACT

Acute toxicity of the tannery industrial effluent was determined for 96h period against Labeo rohita. The LC50 value of effluent was calculated by Finney method (1971). Three sublethal concentrations 3.53%, 1.76% and 0.88% were selected on basis of (1/2, 1/4, 1/8) of LC50 value. Various behavioral and morphological changes were observed in the three sublethal concentrations for 96h. Behavioral changes include erratic swimming, gulping of air, opercular movement, loss of equilibrium, restlessness and sluggishness. Morphological changes like loosening of scales, sinking of eyeball, redness of eye, profuse mucous secretion, bleeding from gills and haemorrhages were recorded. Concentration 3.53% proved to be highly toxic whereas 1.76% and 0.88% concentrations cannot be ignored because they also induced changes in behaviour and morphology of fishes. Control fishes were also continuously monitored and compared with the changes caused by effluent in each concentration.

2. Introduction:

Pollution of aquatic ecosystem is recognized globally as a potential threat to both human and animal populations which interact with aquatic environment (Biney *et al.*, 1987). Water resources are polluted due to the discharge of industrial effluents, agricultural run-off having insecticides, pesticides, heavy metals, fertilizers, chemicals, sewage and other domestic wastes. In Punjab due to rapid industrialization and population explosion the conditions of freshwater bodies are deteriorating which poses potential risk to the health of aquatic organisms. Fishes store, concentrate, metabolize toxicants and exhibit behavioral and morphological changes. They also act as bio-indicator to monitor water quality.

Behaviour allows an organism to adjust to external and internal stimuli in order to meet the challenges of survival in a changing environment while morphological changes are external changes occur due to the changing environment. Study of behavioral and morphological changes due to the effect of toxicants in fishes is the best suitable method to check the pollution in water resources. Thus, it is a promising tool in ecotoxicology and act as diagnostic endpoint for screening, differentiating the chemical effluents and their effect on aquatic organism.

Most of the work pertaining behavioral and morphological changes in fishes due to the effect of pesticides, herbicides or heavy metals dissolved in water had been reported. So far, effect of industrial effluent on fishes has not been done. In the present study, tannery industry effluent has been taken.

Tannery industry is one of red industry (PPCB, 1999) whose effluent is highly toxic to aquatic system. In Punjab, there are about 30 tannery industries in Jalandhar, dumping their wastes in Kala Sanghia drain which flows into chitti bein and then joins river Satluj. Family cyprinidae is one of the largest families of freshwater fishes having 2400 species belonging to 220 genera. These fishes are abundant in rivers of Punjab. Thus, they are ideal to monitor genotoxic effect of tannery effluent. *Labeo rohita* is selected because is easily available and highly consumed by people of Punjab. The main objectives of the study are: a) To calculate 96h LC₅₀ value of the effluent b) To prepare three sublethal concentrations c) To record various behavioral responses and morphological changes in *Labeo rohita* due to the effluent.

3. Materials and methods:

Freshwater fish, *Labeo rohita* of about 6-8 cm in length and

32-58 gms in weight were collected from government fish seed farm, Patiala. They were treated with 0.1% of KMnO₄ solution for 30 minutes to remove any external infections and Fishes were fed with pelted feed and feeding was stopped 24h prior to acute toxicity test. Each experiment was performed in triplicate.

Effluent of tannery industry was taken directly from the waste outlet of an industrial unit based in Jalandhar for the conduct of genotoxicity test against the fish. Tannery industry effluent contains several complex organic and inorganic components like sulphides (sodium sulphide, sodium hydrosulphite and calcium hydrosulphide), sodium chloride, cyanides, dimethyl amines, chromium sulphate salts, oil, grease, alum salts and suspended solids (EPR, 2010).

The 96h LC₅₀ value of tannery industry effluent was calculated by following the method given by Finney (1971). For this, 10 fishes each were exposed to 50L of normal water (control) and five concentrations (2%, 4%, 6%, 8% and 10%) of tannery industry effluent. Mortality of fishes was recorded in each concentration for 96h. The values of mortality were converted into Probits and concentrations into log values and graph was plotted to determine 96h LC₅₀ value. Control group was also monitored.

Behavioral responses and morphological changes in three sublethal concentrations (3.53%, 1.76% and 0.88%) of LC₅₀ value of effluent were recorded after 24h, 48h, 72h and 96h durations of exposure. Control group fishes were also monitored to assess the normal behavior.

4. Results:

The results indicated different mortality rate of fishes which increased with a corresponding increase in the concentration of tannery industry effluent. The 96h LC₅₀ comes out to be 7.07 (L/L). Various behavioral responses and morphological changes in fishes of both control and treated groups in all the sublethal concentrations after all the exposure periods were recorded (Table).

a) Behavioral responses:

Control fishes: Fishes were very active and showed well synchronized movement. They mostly settled at base of experimental tub, while sometimes came on the surface of water and actively responded to slight disturbances. No mortality was recorded in the control.

Treated fishes: Treated fishes showed erratic and rapid swimming in all the three sublethal concentrations. The effect was more pronounced at 24h while less at 96h. In 3.53% it was 25 min at 24h, 18min at 48h, 16min at 72h and 10min at 96h, in 1.76% it was 15min at 24h, 12min at 48h and 10min at 72h and 96h and in 0.88% it was 8min at 24h and 5min for 48h, 72h and 96h. Gulping air at surface or jumping out of water was also observed it was maximum in 3.53% and 1.76% concentrations and increased with the duration of exposure (24h, 48h, 72h and 96h) while it was minimum in 0.88% for at all exposure periods. Opercular movements were fast at 24h, normal at 48h and 72h and slow at 96h in all the three sublethal concentrations. Loss of equilibrium in fishes was rarely seen. It was shown only in 3.53% after 72h and 96h of exposure periods while absent in other concentration and exposure duration. Fishes were hitting against wall in 3.53% for all-time intervals, in 1.76% only at 72h and it was absent in 0.88%. Fishes showed restlessness in 1.76% and 3.53% concentrations at 24h and 48h of exposure periods. Fishes became sluggish after 72h and 96h time intervals in 1.76% and 3.53% concentrations. Two fishes in 3.53% and one fish in 1.76% were died after 96h of exposure. Before death, they jumped out of water, gulped air and then lied at surface of water with jerky movements. Lastly, dead fishes lied at the surface of water with belly upward and open mouth.

b) Morphological changes:

Control fishes:

Morphologically, *Labeo rohita* is laterally compressed, fusiform, blackish grey coloured fish with large bulging eyes, prominent snout and fringed lips. Body is covered with overlapping cycloids scales (Fig. 1).

Treated fishes:

Treated fishes subjected to three sub-lethal concentrations at various time intervals exhibited morphological changes. Loosening of scales was observed in 3.53% and 1.76% concentrations after 72h and 96h of exposure while it was absent in 0.88% (Fig. 2). Sinking of eyeball was examined in all the concentrations after 72h and 96h of exposure (Fig. 3). Redness of eyes was

recorded in 3.53% after 72h and of all the concentrations after 96h for time intervals (Fig. 4). Profuse mucous secretions were recorded in all concentrations (3.53%, 1.76% and 0.88%) after 24h, 48h, 72h and 96h exposure, while maximum secretion was shown by the fishes in 3.53% concentration (Fig. 5). Bleeding from gills was observed in 1.76% and 3.53% concentrations only after 96h of exposure (Fig. 6). Hemorrhages near lips were recorded only in 3.53% concentration after 72h and 96h of time intervals (Fig. 7).

After 96h, survived fishes were transferred to normal freshwater for recovery. It was observed that morphological changes once occurred remained and did not became normal.

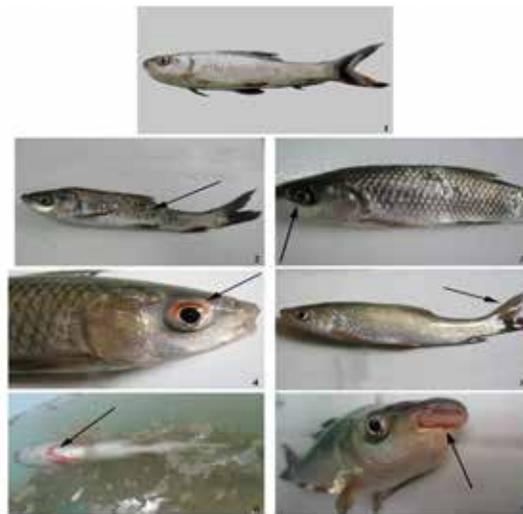


Fig. 1: *Labeo rohita* (Control) Morphological changes: Fig. 2: Loosening of scales, Fig. 3: Sinking of eye ball, Fig. 4: Redness in eye, Fig. 5: Profuse mucous secretion, Fig. 6: Bleeding from gills, Fig. 7: Haemorrhages at mouth

Table. Behavioral responses and morphological changes in *Labeo rohita* after treatment with tannery industry effluent.

EXPOSURE PERIOD AND CONCENTRATION																
A. BEHAVIORAL RESPONSES	24h				48h				72h				96h			
	C	C1	C2	C3												
1. Erractic Swimming	A	L	M	M	A	L	M	M	A	L	M	M	A	L	M	M
2. Gulping air at surface	A	L	M	M	A	L	M	M	A	L	M	M	A	L	M	M
3. Opercular movements	A	F	F	F	A	N	N	N	A	N	N	S	A	N	S	S
4. Loss of equilibrium	N	A	A	A	A	A	A	A	A	A	A	P	A	A	A	P
4. Hitting against wall	A	A	A	P	A	A	A	A	A	A	P	P	A	A	A	P
5. Restlessness	A	A	P	P	A	A	P	P	A	A	A	A	A	A	A	A
6. Sluggishness	A	A	A	A	A	A	A	A	A	A	P	P	A	A	P	P
7. Fish lied at surface	A	A	A	A	A	A	A	A	A	A	A	A	A	A	P	P
B. MORPHOLOGICAL CHANGES																
1. Loosening of scales	A	A	A	A	A	A	A	A	A	A	P	P	A	A	P	P
2. Shrinking of eye balls	A	A	A	A	A	A	A	A	A	P	P	P	A	P	P	P
3. Redness in eyes	A	A	A	A	A	A	A	A	A	A	A	P	A	P	P	P
4. Profuse mucous secretion	A	P	P	P	A	P	P	P	A	P	P	P	A	P	P	P
5. Bleeding from gills	A	A	A	A	A	A	A	A	A	A	A	A	A	A	P	P
6. Haemorrhages at mouth	A	A	A	A	A	A	A	A	A	A	A	P	A	A	P	P

Less= L, More= M, Fast= F, Normal= N, Slow= S, Present= P, Absent= A, Control= C, 0.88%= C1, 1.76%= C2, 3.53%= C3.

5. Discussion:

Industrial development has contributed significantly in uplifting everyone's living standard but at the same time, brought pollution danger. Fishes act as bio-indicators because they are very sensitive to the changes occur in the aquatic environment and play an important role in monitoring the water pollution. Behavioral study gives direct response of the animals to the pollutants. Warner *et al.* (1966) commented that "The behavioral activity of an organism represents the final integrated results of a diversified biochemical and physiological processes". Thus, a single behavioral parameter is generally more comprehensive than a physiological or biochemical parameter. Thus, behavior is a selective response that constantly adapting through direct interaction with physical, chemicals, social and physiological aspects of environment. The present study is an attempt to monitor the behavioral and morphological changes induced in *Labeo rohita* by tannery industry effluent.

Control fishes released in well aerated water were found to be alert and respond actively to slight disturbances. They behaved naturally by showing movements in well coordinated manner. Fishes treated with three sublethal concentrations of tannery industry effluent for different time interval, showed behavioral responses and morphological changes due to the effect of effluent. Behavioral responses *viz.* erratic and rapid swimming, gulping of air, opercular movement, loss of equilibrium, hitting against the wall, restlessness and sluggishness were examined. Morphological changes like loosening of scales, redness in eyes, sinking of eyeball, profuse mucous secretions, bleeding from gills and haemorrhages around mouth were also recorded. Behavioral and morphological changes are increased with the increase in concentration of the effluent.

It has been suggested that restlessness, erratic and jerky swimming, loss of equilibrium, hitting against the wall and hyperac-

tivity in fishes might occur due to the inactivation of acetylcholinesterase (AChE), leading to accumulation of acetylcholine at synaptic junctions (Fulton and Key, 2001) Surfacing or gulping of air might be due to the demand of high oxygen level after exposure (Katja *et al.*, 2005). Bisht and Agarwal (2007) also suggested that profuse mucous secretion is considered to be a defense mechanism to neutralize the effect of toxicants by coagulation and to prevent the entry of toxicants in the body.

Loss of equilibrium, erratic swimming and restlessness are common behavioral responses in several fishes exposed to a variety of toxicants as recorded in *Labeo rohita*, *Catla catla* and *Cirrhinus mrigala* exposed to fenvalerate (Susan and Sobha, 2010), in *Oreochromis niloticus* and *Clarias batrachus* exposed to copper sulphate (Ezeonyejiaku, 2011) and in *Oreochromis niloticus* exposed to cypermethrin (Yaji *et al.*, 2011). Hitting against the wall was noticed in *Labeo rohita*, *Catla catla* and *Cirrhinus mrigala* exposed to fenvalerate (Susan and Sobha, 2010).

Results of present study clearly indicated that behavioral and morphological changes were maximum at higher sublethal concentration (3.53%) and longer exposure period of the tannery industry effluent, thus it is highly toxic. Impact of other low concentrations (0.88% and 1.76%) cannot be ignored. However, prolonged dumping of this amount of toxicant can also cause behavioral changes and morphological responses in fishes. *Labeo rohita* is highly consumed by the people of Punjab. So, fishes captured from these water bodies are indirectly affecting the human health and also responsible for various diseases like cancer, intestinal infections and skin disorders. It is therefore, suggested that both small scale as well as large scale industries to treat their waste water in treatment plant before dumping. Healthy aquatic environment should be maintained to save fish fauna and human health.

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