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Toxicity bioassay of a textile industry effluent using the teleost fish *Poecilia reticulata*

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ABSTRACT

Textile effluent is one of the major sources of water pollution and significantly affects the aquatic ecosystem in general and fish in particular. Histopathological changes have been widely used as biomarkers in evaluating the health of aquatic species exposed to contaminants. A variety of molecular, biochemical, and histological responses in fish have been employed as biomarkers of various environmental stresses. Fishes serve as valuable models for assessing the effect of various types of pollutants. Short term acute toxicity test was performed for a period of 96 hours using different concentrations of textile industrial wastewater with the aim of evaluating the acute toxicity of the effluent on the fish *Poecilia reticulata.* LC_{50} and safe concentration values indicated that the textile effluent is highly toxic to the fish. The study essentially stresses the importance of proper treatment of the textile effluent before discharge and the necessity to take control methods to safeguard the aquatic environment.

Key words: *Textile effluent, Wastewater, Toxicity,* LC_{50}*, Poecilia reticulata.*

Introduction

Industrial discharge is the significant source that contaminates the aquatic ecosystem. Textile industries consume large volumes of water and become a cause for the pollution of the aquatic environment by the release of untreated or partially treated effluents (Verma *et al*., 1978). Aquatic organism's uptake effluents through the food chain at various trophic levels and are susceptible to various physiological disorders (Karthikeyan *et al*., 2006). Biological changes in fish due to exposure of environmental toxicants are called bio-markers and are widely used for environmental risk assessment (Van-der-oos *et al*., 2003). Fishes are at higher levels of food chain and accumulation of contaminants in fish biomagnifies the toxicants found in water and hence used to evaluate health of the aquatic ecosystem (Kock *et al*., 1996). Fishes are exposed to aquatic toxicants via the delicate respiratory surface of the gills extends over half of the body's surface area and make intimate contact with the surrounding water (Wendelaar, 1997). Toxicity study is an integral part of ecotoxicological studies which help to find out toxicant limit and its safe concentration, as well as fate and effects of chemical contaminants. Fishes are considered as an ideal test organism for ecological studies to examine both acute and chronic toxicity of pollutants. It is proven that fishes are sentinel organisms for monitoring genotoxic pollutants in the aquatic environment because they play an important role in the food web. They are bio-concentrators and are responsive to pollutants even at low concentrations. Low concentrations of the pollutant may not result in fish mortality, but they can cause sublethal effects on them. Fish forms an appropriate model because of their sensitivity towards the alterations in the medium in which they live. In the present study, *Poecilia reticulata* were exposed to lethal concentration of textile effluent for a short term and their general behavior, rate of survival, and mortality were studied.

Materials and Methods

Collection and maintenance of experimental fish

Guppy, *Poecilia reticulata* is a freshwater ornamental fish which satisfies the qualities of an experimental fish model (OECD, 2019). Fishes were collected from a private fish farm in Nagercoil. They were disinfected with 0.1% solution of Potassium permanganate for 5 minutes, Active and healthy fishes of uniform size (1.5 to 2 cm) were selected for acclimatization for 2 weeks during which they were fed with standard pelleted fish feed.

Collection of textile effluent

Textile effluent was collected from the inflow of an effluent treatment plant (ETP) in Tiruppur of Tamil Nadu. Preliminary screening of the effluent was carried out to determine the appropriate concentration range of various toxic chemicals in the textile effluent for test animals.

Static Bioassay

Healthy fishes with active movements were selected for the toxicity testing. Acute toxicity testing was done by time-dependent (96 hours) static renewal in conformity with standard guidelines APHA, 1998). The effluent was diluted to obtain different concentrations. Ten fishes were used per concentration and the experiment was conducted in triplicates. The control group was without the textile effluent keeping all other conditions (pH 7.4 and temperature 37^{o}C). Mortality of fish was recorded at 6, 12, 24, 48, 72, and 96 hours. The percentage of corrected mortality was calculated using Abbott's formula.

Corrected mortality percentage=Percentage living in control- Percentage living in treatment/ Percentageliving in control × 100

Determination of LC_{50}

The concentrations at which 50% of test animals which can survive (96 Hour LC_{50}) were calculated by subjecting the data to probit analysis. The lethal concentration, lethal dose and safe concentration were calculated by bioassay test value using the weighted regression method of probit analysis (Finney, 1971; Robert and Boyce, 1972).

Lethal Dose (LD)= LC_{50} value × Exposure time

Safe Concentration = 48 hr LC_{50} × 0.3/ S^2

Where, S= 24 hr LC_{50} / 48 hr LC_{50}

Results

Table 1. Mortality of *P. reticulata* at different concentrations of the effluent after exposure to the textile effluent of 96 hours

Hours of exposure	LCL	UCL	LC_{50} in % concentration
6	0.313	0.336	129.331
12	0.309	0.326	119.339
24	0.299	0.315	107.177
48	0.286	0.30	92.430
72	0.281	0.297	89.252
96	0.230	0.259	57.323

The fishes showed behavioural changes including signs of weakness, irregular swimming, restlessness, jerky movements, revolving, convulsions and extension of fin rays. The results of mortality analysis of the effluent against *P. reticulata* yielded the derived toxicity indices (LC_{50}) ranging from 0.5% (v/v) at 96 hours to 1.3% (v/v) at 24 hours. No mortality was observed in the control group. The percentage of mortality was found to be increased with the subsequent increase in the effluent concentration.

The lethal concentrations (LC_{50}) were inversely proportional to the duration of exposure. 48 hours LC_{50} value was 0.9 with a LD_{50} value of 43.2, likewise, the 24 hours LC_{50} value was 1.1 with a LD_{50} value of 26.4. The safe concentration of the effluent was calculated as 0.22 ppb (Table 1). nhr LC 50 and confidence for *P. reticulata* exposed to textile effluent are represented in Table 2. The LCL and UCL for 48 hours were 0.286 and 0.30 respectively. For 96 hours, 0.230 and 0.259 were recorded as the respective LCL and UCL.

Discussion

The textile wastewater released into nearby water bodies result in serious problems directly and indirectly to the aquatic biota. Hence it becomes essential to find the lethal concentrations of these effluents to safeguard them for the devastating effects. Acute toxicity tests have an important role in assessing the effect of toxicants on animals and they have wide applicability in evaluating the toxicity of various types and mixture of pollutants on fish and other animals (Craddock, 1977). For textile effluent, chemical assay is not feasible. Some toxicants in the effluent have not yet been identified. Consequently, they cannot be analyzed chemically. The toxicity of a compound or effluent to any animal depends upon many factors such as size, sex, weight, age and strain. It is also influenced by environmental factors such as change in pH, DO, temperature, hardness and relative humidity.

Behavioral parameters can serve as good biomarkers of pollution even without sacrificing the animal (Shukla and Tripahi, 2010). In control group of fishes, normal behavior such as constant opercular movement, steady balance, normal surfacing and non-aggressive movement seem to be significant. During the toxicity study, the fishes showed altered behavior including signs of weakness, irregular swimming, restlessness, jerky movements, revolving, convulsions, extensions of fin rays and thick mucus covering over the whole-body surface. During the exposure, some noticeable damage or injuries were shown among fishes especially those exposed to higher concentrations. The injuries probably weaken the fish's resistance to toxin and consequently result in mortality. This could be due to the stress and cumulative impact of heavy metals present in the effluent.

A fall in fish respiration was noted in the study by counting the gill cover movement. Similar to the study, fall in respiration on exposure to naphthalene and respiratory distress in hatchlings of *L.rohita and C.mrigala* was reported (Dange and Masurekar, 1984). Respiratory distress in rainbow trout, *Oncorhynchus mykis* was observed when exposed to malachite green (Ross *et al*., 1985). Due to the resulting stress, hatchlings secrete mucus that cause blockage of gills resulting in precipitation of mucus leading to a decrease in respiratory surface area which may be the reason for the observed mortality (Verma, 2011). It shows the sign of asphyxiation which is an indication of gasping that leads to death (Kalavathy *et al*., 2001). Such alterations are also reported in different fishes exposed to pesticides (Venugopal *et al*., Vineet *et al*., 2008). Mucus covering on the body and gills reduce oxygen supply and causing death in *Anabas testudineus* (Bloch) when treated with monocrotopus (Santhakumar and Balaji, 2000).Fish suffered from anoxemia and die due to depression of gill function when exposed to methylene blue and rose bengal (Srivastava *et al*., 2000; Tonogai *et al*., 1980). Erratic behavioral responses of the test organisms were caused by high concentration of toxicants found in industrial effluent (Adewoye *et al*., 2005; Soni *et al*., 2006).

The lethal concentrations (LC_{50}) were inversely proportional to duration of exposure, concentration and toxicity factor. Similar observations were reported in catfish where-in no mortality was observed in the control group (Gabriel and Okey, 2009). The LC_{50} value of diazinon 60 EC to *A. testudineus, C. punctatus* and *B. gonionotus*were 6.55, 3.09 and 2.27 ppm for 96 hrs exposure (Rahman *et al*., 2002). The LC_{50} value of the heavy metal Cadmium chloride for fish *L. rohita*was 687.38, 653.99, 632.78 and 589.76 mg/l and for Lead nitrate 796.57, 740.17, 694.39 and 681.68 mg/l for 24, 48, 72 and 96 hours respectively. The LC_{50} values were 0.127% for tannery effluent, 1.1376% for a vegetable oil factory effluent and 0.137% for a paper mill effluent.

The acute toxicity of a raw effluent from a battery manufacturing plant to fresh water fish *O. niloticus* for 96 hrs had been reported (Monkiedje *et al*., 2004). The 48 h LC_{50} and 48 h LC_{90} are 20.8% and 26.6% (v/

Table 2. nhr LC50 and confidence intervals for P. reticulata at exposed to textile effluent after 96 hours exposure to the textile effluent

S.No.	Exposure in Hours	LC_{50} (ppb)	No. of fish exposed	% of mortality	Regression Equation Y= a+bx	Lethal Dose (LD) (ppb)	Safe Concentration (ppb)
1	6	1.3	10	50	Y= 7.821+(-11.51) X	7.8	0.22
2	12	1.2	10	50	Y= 8.398+(-12.44) X	14.4	
3	24	1.1	10	50	Y= 11.095+(-17.52) X	26.4	
4	48	0.9	10	50	Y= 12.273+(-19.12) X	43.2	
5	72	0.8	10	50	Y= 10.371+(-15.23) X	57.6	
6	96	0.5	10	50	Y= 6.590+(-6.58) X	48	

v) of effluent, whereas 96 h LC_{50} and LC_{90} values are 16 and 20.7% (v/v). The LC_{50} were 6.784, 5.784, 5.257 and 4.845 % at 24, 48, 72 and 96 hrs LC_{50} for *Barytelphus aguerini* exposed to sugar mill industrial effluents (Matkar and Gangotri, 2003). For *L. rohita* hatchlings, 100 % mortality was found in 90 mg L^{-1} concentration of dye after 96 hours of exposure. In all concentrations above 90 mg l^{-1}, all the hatchlings of *L. rohita* died within 72 hours of exposure (Patial and Kaur, 2013).

Mortality analysis of chrome plating industry effluent against *P. reticulata* recorded the 96-hour LC_{50} value of 1.67 ml/l (Angelin *et al*., 2015). LC_{50} value of 37.9 µg/l was recorded at 96 hours when the fish was exposed to copper sulphate treated water (Shuhaimi-Othman *et al*., 2010). LC_{50} value of 58.84 mg/l for *O. niloticus* treated with copper sulphate solution at 96 h of exposure was observed (Ezeonyejiaku *et al*., 2011). The 96 h LC_{50} was found to be 71.5% (v/v) and 1.399 as ATU for outlet and the safe concentration of effluent for outlet wastewater was 7.15% (v/v) (Workagegn, 2010).

The LC_{50} values obtained in the study are in agreement with a previous work where-in 35.518 % 96 h LC_{50} was recorded for *Clarias gariepinus* and 2.5 ATU for synthetic resin effluent (Dhanusi and Oranusi, 2012). A higher value of 96-hour LC_{50} 57.323 was recorded which varies from a previous related study wherein 96-hour LC_{50} was 39.726 (Selvaraj *et al*., 2015). The results of mortality analysis of the untreated effluents against *O. mossambicus* yielded the derived toxicity indices values ranging from 0.9% at 24 h to 0.16% at 96 h (Amte and Mhaskar, 2012). The safe dischargeable concentration was 0.22 ppb which was much lesser than 3.60 % (v/v) for the effluent as observed in *C. carpio* (Roopadevi *et al*., 2012).It is observed that the textile effluent impart toxicity in *P. reticulata* and therefore the present level of treatment of effluent prior to discharge appears insufficient.

Conflict of Interest- None

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