



Studies on Toxicity of Detergents to *Mystusmontanus* and Change in behaviour of Fish

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Abstract

The acute toxicity of two detergents to *Mystusmontanus* was investigated using static bioassay's. The 96h LC50 values was determined by the Litchfield and Wilcoxon graphical method. These were for Det-I 20.0mg/litre and Det-II 23.5mg/litre. During exposure period, the test fishes exhibited several behavioural changes before death such as restlessness, rapid swimming, and loss of balance, respiratory distress and haemorrhaging of gill filaments amongst others. Opercula ventilation rate as well as visual examination of dead fish indicates lethal effects of the detergent on the fish.

Keywords: Toxicity, detergents, *Mystusmontanus*, static bioassay's, behavioural changes.

Introduction

The ecosystem plays an important role in the lifetime of living organisms due to its physico-chemical properties and formation of food value. The activities like urbanization, industrialization, agricultural and other activities. These activities has exploited the environment and disturbed the delicate ecological balance between living and non-living components of the biosphere. The unfavourable conditions created by man, not only threatened the survival of humans but also the other living organisms. The number of species with time is likely to become rare.

A serious concern has grown up over the years around the world about the rivers turning murky, fish rotting on sea shores, tree withering, cities choking up with foul air, toxic chemicals being cycled into food stuffs and epidemic diseases appearing so frequently.

Nowadays, the major rivers and aquatic bodies of world were heavily polluted due to domestic, industrial discharge and detergents. A huge amount of various brands of detergent powders, flakes and various shampoos and toilet soaps. The surface-active property of detergents has been rendered them to be the most powerful cleaning agents. Because of this quality, are being used on a large scale for the past three to four decades. These detergents causes foam in the water bodies, reduce the air-water interaction leading to deficiency of oxygen to the aquatic animals under the water surface. The detergents and surfactants are non-degradable pollutant accumulated in water body. These detergents enter the food web through uptake by vegetation, planktons, fishes and zooplanktons.

Large number of brands of detergent powders and flakes are available in the market; the composition of a typical packaged detergent¹ and two detergent samples is given in table-1.

Table-1
Contents of packed detergents and two detergent samples consider for toxicity test

Content of Detergent	Content of a Typical packaged Detergent	Det-I	Det-II
Surfactant	20-30 %	66.5 %	67.4 %
Polyphosphate	28 %	18.0 %	23.0 %
Sodium Silicate	6.0 %	8.0 %	3.0 %
Sodium Sulphate	35 %	4.1 %	3.5 %
Carboxymethyl cellulose	1 %	2.5 %	2.1 %
Bleach	< 1 %	0.8 %	0.8 %
Colour	< 1 %	Traces	Traces
Perfume	<1 %	0.06 %	0.01 %

Detergents affect both chemical and biological characteristics of receiving water bodies. Surfactants of various detergents having benzene ring, branched alkyl chain or ethoxylate chain are resistant to anaerobic biodegradation. In the course of biodegradation H₂, H₂S, CO₂ and CH₄ were produced. The final products of biodegradation are CO₂ and CH₄².

The surfactants are one of the major contents of detergents, commercial surfactants like linear benzene sulphonate (LAS), alkyl sulphates (AS), alkyl ethoxysulphates (AES), ethoxylates (AE), alkyl phenol ethoxylates (APE), Cetyltrimethyl Ammonium Bromide (CTAB) etc are synthetic chemicals, which are used in large amounts in detergents, soap, shaving creams, fabric softeners, additives for food, paint, leather and textile items, pesticides, defoliants, antiseptics, disinfectants³.

Excessive amount of phosphate in detergents are responsible for eutrophication of natural waters and brunt the diversity of plankton⁴. The toxic and median lethal levels of the detergents were 6 and 3 % respectively. They suggested that the detergent affects the feeding; growth metabolic rate and conversion efficiency were decreased with increasing concentrations of the detergent in the fish *Macropodus cupanus*. These detergents were found to interfere with the food utilization, growth and conversion of fishes⁵. Detergents also bring reduction in the biochemical contents in the tissues of aquatic animals. The 24hrs LC₅₀ value of detergent to *Macrobrachium lamarrei* was 0.5%, also investigated the reduction in glycogen, protein and lipid contents of tissues of prawn with increase in concentration and time of exposure⁶. But studies pertaining to the acute toxicity of detergent to fishes are scanty.

Hence an attempt is made in the present investigation to determine the acute toxicity of two household detergents to the freshwater fish *Mystus monatus*.

LC₅₀ is a concentration in which 50% of the experimental animals survive. Estimation of LC₅₀ by interpolation involving plotting of data in a graph with concentration on X-axis, while percentage on Y-axis. A straight line is drawn between maximum points representing survival at maximum successive concentrations that were lethal to more and less than of the total number of test animals exposed to the toxicant. The concentration at which this crosses the 50% survival line is the LC₅₀ value⁷.

Material and Methods

The freshwater fish *Mystus montanus* were collected by the fisherman from Mula river before Mahalunge village and were used for bioassay studies. The fishes were brought into the laboratory for the acclimation by providing sufficient aeration. The fish *Mystus montanus* were selected irrespective of sex for experiments. The size or length ranged from 12.3cm to 14.5cm and weight ranging from 18.72gm to 23.86gm. Fishes were acclimated in glass tank in the laboratory for seven days as per the method in APHA⁸. They were divided into different groups each containing ten fishes for the experiment. Dead fish number were also recorded simultaneously and removed immediately. During experimentation, the methods recommended⁸. Detergents like Det-I and Det-II are weighed accurately as per requirement and dissolved in water before the transfer of fishes into the aquarium, simultaneously control set was arranged.

Static renewal bioassay tests were conducted in order to evaluate the acute toxicity of Det-I and Det-II, dechlorinated tap water was used for acclimation of fish as well as experiment and control. Fishes were fed with soaked dry prawn once in day. The moderate size fishes were selected for experiment and placed in different concentrations of Det-I and Det-II to determine LC₅₀ values after 96 hours. Static bioassays, without

aeration and with the toxicant added to the test medium at the beginning of the test were used to determine the toxicity of detergents Det-I and Det-II. The bioassays were conducted in a glass aquarium containing twenty litres of dechlorinated water with preliminary testing.

After appropriate toxicity range of the test, solutions were determined by preliminary testing, 16 concentrations of Det-I ranging from 5.0mg/litre to 50mg/litre for *Mystus montanus* and 15 concentrations of Det-II ranging from 5.0mg/litre to 55.0mg/litre to *Mystus montanus*. Observations on survival were made after 24, 48, 72 and 96 hours. LC₅₀ (concentration required for 50% mortality) values are calculated by graphical method. Control group of animals was maintained simultaneously.

Table-2
Physico-chemical characteristics of the diluent's water used in acute toxicity or 96 hours LC₅₀

Factors	Diluents water used for toxicity of Detergents	
	Det-I	Det-II
PH	7.08 –7.24	7.2-7.45
Temperature	25- 28°C	26- 28°C
Suspended Solids	00	00
Total Alkalinity	172-174 mg/litre	170-173 mg/litre
Total acidity	20-23mg/litre	19-21mg/litre
Dissolved Oxygen	6.62-6.95 mg/litre	6.7-7.02mg/litre
Biological Oxygen Demand	00	00
Chemical Oxygen Demand	00	00
Nitrate	0.17-0.19 mg/litre	0.16-0.19mg/litre
Phosphate	0.1-0.23 mg/litre	0.05-0.21mg/litre

All the tests are carried out, using standard procedure as per BIS (Bureau of Indian Standard) and APHA.

Results and Discussion

To study the changes in body colour, behaviour pattern, swimming movements and opercular movements were observed.

Acute toxicity with Det-I and Det-II: Toxicity studies: During the period of acute toxicity tests no mortality was observed in control group. The LC₅₀ values of both Det-I and Det-II studied for period of 96 hours were found. Among these two detergents studies the LC₅₀ value of Det-I for exposure period of 96 hour was found minimum (20.0mg/litre) and LC₅₀ value of Det-II for an exposure period of 96 hours was found maximum (23.5mg/litre).

Table-3
Acute toxicity data of Det-I in *Mystusmontanus*.

Sub group No.	No. of Fishes Exposed	Detergent Conc. --mg/lit	Mortality of fishes noted after time intervals of									% Mortality within 96 hours
			30 min	60 min	2 hrs	6 hrs	12 hrs	24 hrs	48 hrs	72 hrs	96 hrs	
1	10	50mg/lit	1	2	1	1	2	3	00	00	00	100
2	10	45mg/lit	2	1	1	1	1	2	2	00	00	100
3	10	40mg/lit	1	1	1	1	2	2	2	00	00	100
4	10	35mg/lit	1	1	2	1	1	2	2	00	00	100
5	10	30mg/lit	1	1	00	1	1	2	1	1	2	100
6	10	28mg/lit	00	1	00	1	1	2	2	1	1	90
7	10	25mg/lit	1	00	00	1	1	1	2	1	00	70
8	10	24mg/lit	00	1	1	00	1	1	2	1	00	70
9	10	23mg/lit	00	00	00	00	2	1	1	1	1	60
10	10	22mg/lit	00	00	1	00	2	1	00	1	1	60
11	10	21mg/lit	00	00	00	1	1	2	00	1	1	60
12	10	20mg/lit	00	1	1	00	00	2	00	00	1	50
13	10	19mg/lit	1	00	00	1	00	00	1	00	1	40
14	10	18mg/lit	00	00	1	00	1	00	1	00	1	40
15	10	15 mg/lit	00	1	00	00	00	1	00	00	1	30
16	10	10 mg/lit	00	00	00	00	00	00	00	1	1	20
17	10	5.0 mg/lit	00	00	00	00	00	1	00	00	00	10
18	10	Control	00	00	00	00	00	00	00	00	00	00

Table-4
Acute toxicity data of Det-II in *Mystusmontanus*

Sub group No.	No. of Fishes Exposed	Detergent Conc. --mg/lit	Mortality of fishes noted after time intervals of									% Mortality within 96 hours
			30 min	60 min	2 hrs	6 hrs	12 hrs	24 hrs	48 hrs	72 hrs	96 hrs	
1	10	55mg/lit	1	1	1	1	3	2	1	00	00	100
2	10	50 mg/lit	1	1	2	1	1	3	1	00	00	100
3	10	45mg/lit	1	1	1	1	1	2	2	1	00	100
4	10	40mg/lit	1	1	00	1	2	2	1	2	00	100
5	10	35mg/lit	00	1	1	1	1	1	00	2	3	100
6	10	30mg/lit	1	00	00	2	1	00	1	1	1	70
7	10	28mg/lit	00	00	00	1	2	1	1	1	00	60
8	10	25mg/lit	00	00	1	1	1	1	1	1	00	60
9	10	24mg/lit	00	00	1	00	1	2	00	2	00	60
10	10	23.5mg/lit	00	00	1	00	1	2	00	00	1	50
11	10	23mg/lit	00	1	00	00	00	1	1	00	1	40
12	10	22mg/lit	00	00	00	00	1	1	2	00	00	40
13	10	21mg/lit	00	00	00	00	00	1	1	1	1	40
14	10	20mg/lit	00	00	1	00	00	1	00	1	1	40
15	10	15 mg/lit	00	00	00	00	1	00	1	1	00	30
16	10	10 mg/lit	00	00	00	00	00	1	00	00	00	10
17	10	5 mg/lit	00	00	00	00	00	00	1	00		10
18	10	Control	00	00	00	00	00	00	00	00	00	00

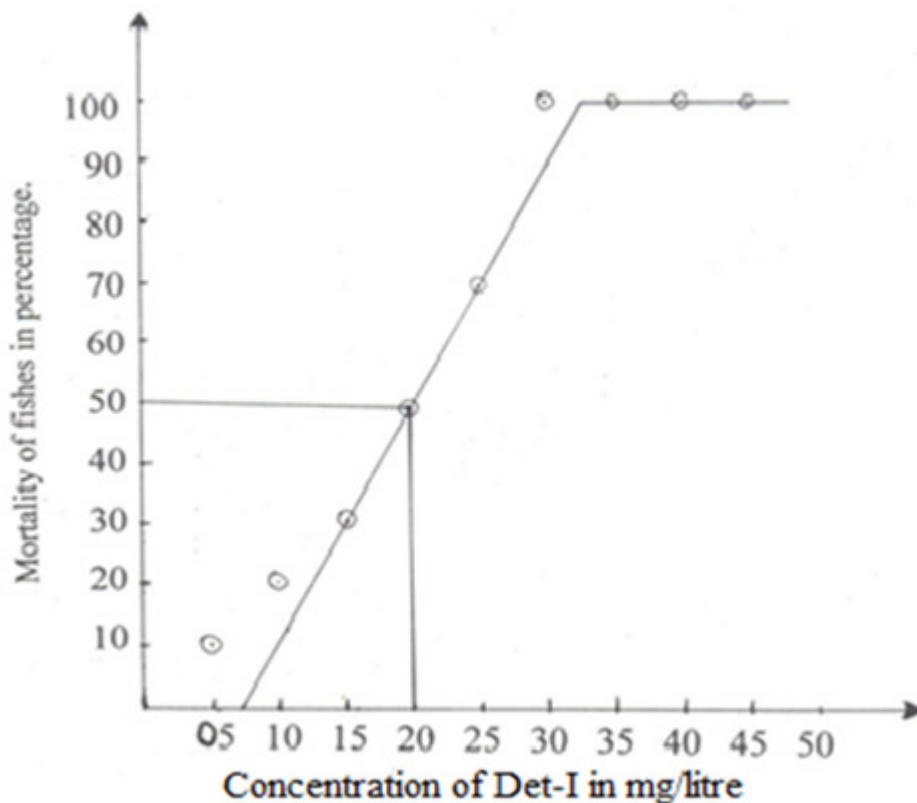


Figure-1
Graphical presentation of LC₅₀ value in *Mystus montanus* with Det-I

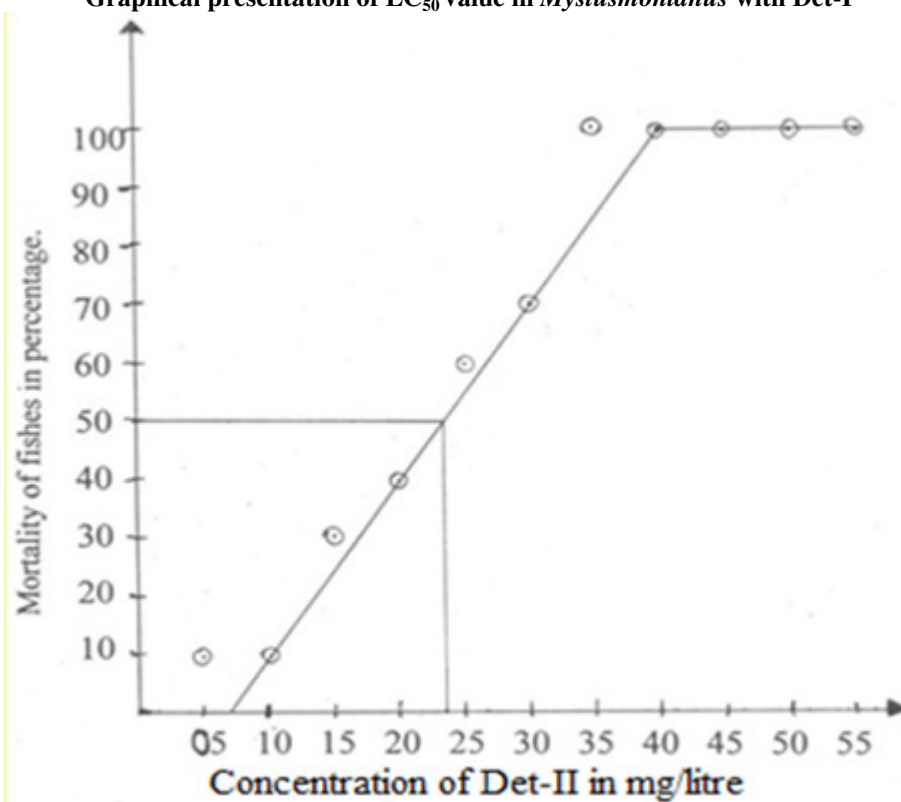


Figure-2
Graphical presentation of LC₅₀ value in *Mystus montanus* with Det-II

The 96 hours LC₅₀ value by the Litchfield and Wilcoxon (1949) graphical method: The 96 hours LC₅₀ value for Det-I is 20.0mg/litre. The 96 hours LC₅₀ value for Det-II powder is 23.5 mg/litre.

The 96 hours LC₅₀ for the Surf, Besto and Key detergents are 12.734, 77.624 and 32.292ppm respectively to *Rasbora elongata*⁹. The LC₅₀ values of Ariel detergent as 35ppm for 48 hours to freshwater teleost *Oreochromis mossambicus*¹⁰. Mortality rate of fish *Tilapia sp.* was 80% at 50ppm, while 100% mortality was in 51ppm of detergent water¹¹.

The toxicity of ionic and nonionic surfactants to six freshwater fishes was studied; 48 hours LC₅₀ value of SDS for *Trout* was 33.61mg/litre, in *Gambusia* was 40.15mg/litre, in *Goldfish* was 38.04mg/litre, whereas in *Cirrhinawas* 30.81 mg/litre¹². The 96 hours LC₅₀ value of Wheel detergent for *Lamellidens marginalis* (Lamarck) was 400 ppm¹³.

The 96 hours LC₅₀ of the household detergents Det-I is 28.5mg/litre and Det-II 41.75mg/litre in *Garramullya*. They also noticed changes in behavioural responses of *Garamullya* like opercular movement was 12-15 times more faster than controlled, frequent surfacing, loss of nervous control, try to jump out of media. In dead fishes opercular region becomes blackish, haemorrhaging at lower lip, along mid ventral line behind the mouth and between pectoral fin, at the base of anal and pelvic fins. In surf at higher concentrations swimming movements of fish immediately slow down with the addition of toxicants¹⁴.

Median lethal concentration (LC50) of LAS for different exposure periods (24 hr, 48 hr, 72 hr and 96 hr) by using a software, Trimmed Spearman-Kärber method. The LC50 values for different exposure periods 24, 48, 72 and 96 hrs were obtained as 0.48, 0.28, 0.18 and 0.03 ml/l respectively was determined¹⁵.

In the present investigation, *Mystus montanus* exhibited a variety of behavioural responses like opercular movement was 20-25 times more faster than controlled, loss of nervous control, try to jump out of media. In dead fishes opercular region becomes blackish, hemorrhaging occurs of gill filaments amongst, along the belly, at the base of pectoral, anal and pelvic fins. Body was slimy due to mucus secretion from epithelium of gills. The fishes were surfacing frequently. Affected fishes were swimming on lateral side of the body; nervous control and equilibrium were lost. The body colour of dead fishes turn to yellow. In higher concentrations of Det-I swimming movements of fish immediately slow down with the addition of toxicants. During exposure period, the test fish exhibited several behavioural changes before death such as restlessness, rapid swimming and respiratory distress. Opercular ventilation rate as well as visual examination of dead fish indicates lethal effects of the detergent on the fish.

Conclusion

With the various concentrations of detergents fishes shows various types of behavioural changes, like slow swimming movement, bleeding through the gills. Hemorrhage occurs at the base of body appendages (fins) and along the belly. There occurs loss of nervous control, fishes along lateral side of body. Body was slimy due to mucus secretion from epithelium of gills. Even at the low concentration fishes are died due bleeding through gills with both detergent powders. This becomes important in the light of the fact that the fishes forms staple diet and has commercial value. Further to sustain its natural population, pollution from washing activities needs proper control and management. The use of detergent in homes cannot be discontinued. However, better method of disposing the 'after wash' needs to be worked out. There is a need of development of ecofriendly detergents and soaps, so that aquatic fauna of various water bodies will be preserved. If the present rate at which they are introduced into aquatic environment is not checked, then continuous existence of aquatic fauna is in serious threat.

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