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Effect of Synthetic Detergents on Some of the Behavioral Patterns of Fish Fingerlings (*Cirrhina mrigala*) and Its Relation to Ecotoxicology

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With the wide spread use of synthetic detergents for household purpose, it is inevitable that these compounds would find their way into rivers and ponds. The effect of detergents on tissue of different fishes have been studied by Abel (1974) and Bromage et al. (1976). Gills, a primary site of osmoregulation, as well as respiration, may be highly vulnerable to lesions because they are in immediate contact with aquatic toxicants. The most prominent manifestation of the acute toxicological effect of detergents is on the gill tissue of the fish. Destruction of the gill epithelium is regarded as a consequence of the reduction of surface tension by the presence of surfactants (Bock 1965). Exposure of fish to elevated level of surfactants cause multiple hepatomas to develop on the gill tissue resulting in diminished oxygen uptake and impairment of electrolyte balance (Schmid and Mann 1961). Schooling patterns of fish have been reviewed by Partridge (1982). Detergent causes impairment of chemoreceptor organs (Bardach et al. 1965) and damage to epidermis and pharyngeal wall (Brown et al. 1968). Eleven species of fish exposed to a mixture of hard alkyl benzene sulphonate based detergent under varying environmental conditions and during different stages of development have shown wide differences in response (Thatcher 1966). Behavioural changes have been used successfully as rapid and sensitive indicators of toxic stress in fish (Sprague et al. 1965; Bengtsson 1974 and Besh et al. 1977).

Operculum movement and schooling pattern was measured as an index for behavioural toxicity in control and detergent exposed fish fingerlings (Cirrhina mrigala). On increasing the concentration of dissolved oxygen there was a significant change in the rate of opercular movement as compared to controls, but no marked change was observed in schooling pattern of fish fingerlings in control and experimental groups. The consumption

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of dissolved oxygen by fish fingerlings was studied with increasing time. Changes in lactic acid content and activity of lactic acid dehydrogenase in the gills of fish fingerlings was measured in control and detergent exposed cases. The objective of the present study was to evaluate the behavioural changes by means of opercular movement under various conditions of dissolved oxygen in presence of detergents. Such studies may be helpful in predicting stress due to pollutant in ecotoxicological cases.

MATERIALS AND METHODS

Ninety days old fish fingerlings (Cirrhina mrigala) of 2 inch size were obtained from the State Department of Fisheries and kept for two days in laboratory conditions for acclimatization. Chemicals used in the study were from BDH AnalaR or E. Merck extra pure. Aged tap water (tap water left in open for at least three days) having following characteristics: pH 7.2-7.5, Do 6.7-7.2 ppm, alkalinity 95-100 ppm, hardness 118-122 ppm at 25°C $\pm$ 1 measured by the method of American public health association (1975) and by using Century Portable Kit.

One litre solution of linear alkyl benzene sulphonate (obtained from the manufacturer in crude form) was adjusted to pH 7.0 with the help of sodium carbonate in aged tap water. A solution of linear alkyl benzene sulphonate of concentration 0.015 ppm (25% of LC_{50}) was prepared by the method of Lal et al. (1982). Two aquaria jars of 10 litre capacity was taken and filled with 6 litres of aged tap water. In one jar 0.015 ppm (25% of LC_{50}) of linear alkyl benzene sulphonate pH adjusted to 7.0 was introduced. The dissolved oxygen in both the jars was measured. Five fish fingerlings were introduced in each jars. The dissolved oxygen was measured at 0, 4, 8, 12, 16, 20 and 28 hrs. Twenty fish fingerlings were taken and divided into two groups ten in each as control and experimental. In experimental 0.015 ppm of linear alkyl benzene sulphonate was introduced. Four beakers of two litre capacity containing one litre aged tap water were taken. Dissolved oxygen was maintained at 1, 3, 5 and 7 ppm and one fingerling from the ten fingerlings were introduced one at a time and the opercular movement was measured after 15 minutes and three observations were taken at the time interval of five minutes. Mean of opercular movement by ten fingerlings were taken and then all the ten fish fingerlings were kept in jar and schooling pattern was noted.

For the determination of lactic acid content and

lactic acid dehydrogenase enzyme activity in gill tissue, five fish fingerlings were exposed to detergent solution upto 30 days for experimental purposes, then individual fish fingerlings were taken, weighed and homogenised in a Potter-Elvejhem type homogenizer. During the homogenization process the homogenizer tubes were packed in crushed ice to maintain the temperature 0°C. The homogenate strength was adjusted to 2.5% (w/v) with water. Protein was estimated by the method of Lowry et al. (1951).

Table 1. Opercular movement in fish fingerlings (Cirrhina mrigala) at different level of dissolved oxygen

Dissolved oxygen (ppm)	Opercular movement (per minute) Control[a]	Experimental[a]	Student 't' test (P value)
1.0	209.66±14.46	213.51±33.79	N.S.
3.0	106.83±22.48	139.20±28.29	<0.02
5.0	57.72±18.70	99.73±19.14	<0.001
7.0	53.11±19.70	60.76±17.42	N.S.

a- Values are expressed as the arithmetic mean ± S.D. of 20 animals (10 control + 10 experimental).
N.S.-Not significant.

Table 2. Alterations in lactic acid content and lactic acid dehydrogenase activity of gills in controls and detergent exposed fish fingerlings (Cirrhina mrigala).

Parameters	Control[a]	Experimental[a]	Student 't' test (P value)
Protein*	24.99±5.78	28.51±8.04	N.S.
Lactic acid*	0.09±0.01	0.11±0.01	<0.05
Lactic acid dehydrogenase**	0.45±0.13	0.59±0.16	<0.1

* Values are expressed in mg/g of gill tissue
** Values are expressed in μ moles of NADH oxidised per minute per mg protein
a- All values are presented as the arithmetic mean ± S.D. of 10 animals (5 control + 5 experimental).
N.S.- Not significant.

Lactic acid content was determined by the method of Barker (1963). Lactic acid dehydrogenase activity (E. C. 1. 1. 1. 27) was measured by the method of Oser (1965). The Student 't' test described by Fisher

(1950) was employed to calculate the statistical significance between control and experimental values.

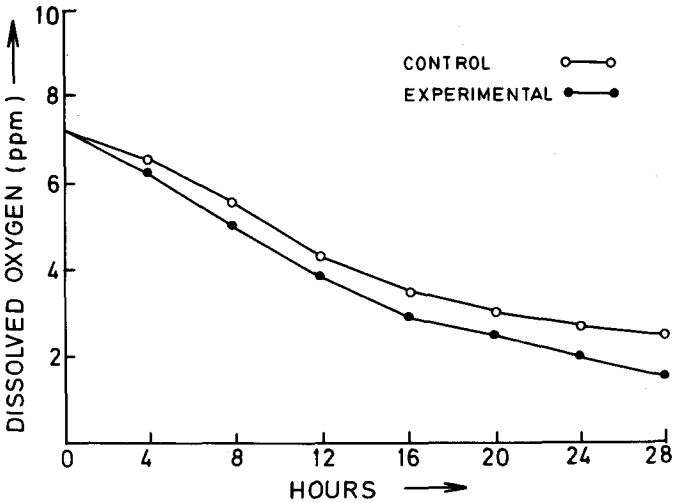

Figure 1 : The consumption of dissolved oxygen by fish fingerlings in control and experimental cases.

RESULTS AND DISCUSSION

The consumption of dissolved oxygen by fish fingerlings in control and experimental groups are given in Fig. 1. The figure shows that on increasing time from 0 to 28 hrs the D.O. decreases from 7.2 to 1.6 ppm in experimental cases. The significant increase (30.3%) and (72.78%) in the rate of opercular movement in fish fingerlings at 3.0 ppm ($P < 0.02$) and 5.0 ppm ($P < 0.001$) in detergent exposed cases as compared to controls are given in Table 1. Similarly there is (18.18%) ($P < 0.05$) increase in the lactic acid content with (23.73%) ($P < 0.1$) increase of lactic acid dehydrogenase when fish fingerlings was exposed for 30 days (Table 2). There was no marked change in the schooling pattern of fish fingerlings in control and detergent exposed groups.

Fishes have a variable behavioural pattern which enable them to select environments favourable for survival and

reproduction. These behavioural features provide useful measures of sublethal toxicity because they represent the integrated results of any biochemical and physiological processes (Anderson 1971).

The muscular expansion and contraction of the buccal and opercular cavities maintains a flow of water over gill surfaces in fishes. The respiratory movements are under physiological control of the respiratory centre and or modified by both internal (oxygen concentration, carbon dioxide concentration and pH) and external (temperature, oxygen depletion) factors (Shelton 1971). Increase in the rate of opercular movement in fish fingerlings in detergent exposed cases with decrease in dissolved oxygen, in above result clearly indicate the physiological stress to aquatic organisms. This stress leads to the accumulation of more lactic acid in gills along with increase in the activity of lactic acid dehydrogenase. McLeay and Brown (1975) have shown that muscular exertion in fish is accompanied by a marked increase in blood lactate. An increase in circulating catecholamines and plasmatic corticosteroids due to stress in fish have been studied by Mazeaud et al. (1977). This increase in catecholamines may increase blood lactic acid level of fish directly (Larsson 1973). Thus detergent stress apparently leads to higher metabolic rate which require an additional expenditure of energy. This increased energy demand perhaps interferes with the enzymatic systems of the metabolic pathway as well. This is evident from the rapid movement of the fish fingerlings and gulping of air by them. This situation may be analogous to mammalian skeletal muscle in heavy exercise, with increased breathing rate and lactic acid production from glycogenolysis. Although, respiratory distress imposed by detergent may have a detrimental effect on fish, there are a very few studies in this direction in which a sublethal effect has been proved. Such studies have importance as test system for behavioural ecotoxicology.

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