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Fish behavioural changes in Exploited Ecosystems: A Laboratory Study

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Abstract

Water the natural biota of fish has been exploited by multifarious anthropogenic activities resulting in chronic pollution of the hydrosphere. One such mode of pollution is the enrichment of water bodies by nutrients. It can be of a natural origin (natural eutrophication) but is often dramatically increased by human activities (cultural or anthropogenic eutrophication). There are three main sources of anthropogenic nutrient input: erosion and leaching from fertilized agricultural areas, and sewage from cities and industrial waste water. Further indiscriminate use of shampoos and detergents also cause water pollution. These compounds affect the water quality and in turn the fish and their behaviour. Deviation from normal behaviour is the first sign of some internal physiological stress. This study investigates the effect of cultural eutrophication together with shampoos and detergents on the behavioural pattern of *Oreochromis niloticus*. Observations were done on the food response, opercular activity, and swim patterns. Behaviours of fish in eutrophic and detergent contaminated waters were compared with weight matched controls under laboratory conditions.

INTRODUCTION

Eutrophication can be a natural process that occurs over time due to natural runoff of soil nutrients and the decay of organic matter. Cultural eutrophication is the process that speeds up natural eutrophication because of human activity. Agricultural fertilizers are one of the main human causes of cultural eutrophication. Fertilizers, used in farming to make soil more fertile, contain nitrogen and phosphorus. The use, or overuse, of fertilizers can cause these nutrients to runoff of the farmer's field and enter waterways. The same fertilizers that were intended to enhance crop growth now enhance the growth of algae and aquatic plants.

Indiscriminate use of shampoos and detergents are increasing day by day by. They degrade very slowly and thus remain in the aquatic system for a longer time. They enter in the food chain of aquatic animals or are absorbed through the gills of fishes. Generally shampoos are anionic or cationic and non-ionic detergent based surfactants. These shampoos which are discharged in the water may change pH, total alkalinity, free CO_2, Dissolved Oxygen [DO_2] and also affect the rate of photosynthesis. It often also leads to eutrophication. Synthetic detergents are reported to be acutely toxic to fish in concentrations between 0.4 and 40 mg/lit (Najam Ahad et al., 2010).

Fishes live in a three dimensional environment that is largely inaccessible to us. Their behavioural manifestations thus act as direct indicators of water quality in which they sustain (Walsh et al., 2004). For the present study, *Oreochromis niloticus* were procured because they are considered to be good bio-indicator of aquatic toxicity. To understand and compare the fish behaviour in the two different above mentioned conditional of polluted waters, food response, operculum activity and swim pattern of the fish were studied.

METHODOLOGY

Experimental models

Oreochromis niloticus were used as experimental models.

Experimental Setup for studying the effect of detergent pollution

Two aquaria with equal amount of water were taken 1 serving as the control and the other serving as experimental (20 liters of water) to which 1gm of detergent was added (conc. 50mg/lit). The temperature and aeration was kept constant for the two aquariums. 3 fishes per experimental set was selected, of which two fishes were of 3-4 inches length and weighed 60-62gms while the 3rd was 6 inches in length and weighed approximately 85gms. 3 fishes were kept in each aquarium and food in equal quantity, at regular time intervals were given. The pH of the water was measured daily.

Experimental Setup for studying the effect of cultural eutrophication

The effect of eutrophication on fish behaviour was also studied maintaining the same experimental setup with minor changes. Two aquaria with equal amount of water were taken; 1 serving as the control and the other serving as experimental (20 lits of water) to which 3 macrophytes were added simultaneously in considerable amount: *Lemna* sp., *Wolfia* sp and *Pistia* sp. The aim was to enhance the nutrient contents of the water. 3 fishes per experimental set were selected, 3-4 inches length and weighed 60-62gms. 3 fishes were kept in each aquarium and same food in equal quantity, at regular time intervals were given. The pH of the water was measured daily. After acclimatization, their behaviour were studied under the following heads for the two different experimental setup –

i. Food response
ii. Operculum activity
iii. Swim patterns

After acclimatization the fishes were observed for 2 weeks continuously for 1hour at 9-10 am in the two different experimental setup.

RESULTS AND DISCUSSION

pH Estimation of the water sample of the two aquariums

0.5ml of water sample was collected from the two aquariums every day for testing the pH of the water. With the help of a watchmaker forceps that was heat sterilized, a strip of pH paper was dipped in the water sample that was collected. The process was repeated for the two water samples separately and with extreme care. The color change in the pH paper was observed and according to that the pH of the water sample was determined.

Table 1a: pH Estimation of detergent contaminated waters.

Sl. No.	Date	pH	
		Control	Experimental setup
Day 1	26.11.2011	7.0	8.5
Day 2	28.11.2011	7.0	8.5
Day 3	29.11.2011	7.0	8.5
Day 4	01.12.2011	7.0	8.5
Day 5	02.12.2011	7.0	8.5
Day 6	03.12.2011	7.0	8.5
Day 7	05.12.2011	7.0	8.5
Day 8	07.12.2011	7.0	8.5
Day 9	08.12.2011	7.0	8.5
Day 10	09.12.2011	7.0	8.5
Day 11	10.12.2011	7.0	8.5
Day 12	12.12.2011	7.0	8.5
Day 13	13.12.2011	7.0	8.5
Day 14	14.12.2011	7.0	8.5

Table 1b: pH Estimation of eutrophic waters.

Sl. No.	Date	pH	
		Control	Experimental setup
Day 1	5.03.12	7.0	7.5
Day 2	6.03.12	7.0	8.5
Day 3	7.03.12	7.0	8.5
Day 4	9.03.12	7.0	8.5
Day 5	10.03.12	7.0	9.0
Day 6	12.03.12	7.0	9.0
On the 5th day 2 fishes in the experimental set died. On the 6th day the remaining fish also died. The old set of aquatic macrophytes were changed. The aquarium was cleaned and fresh macrophytes were introduced. A fresh batch of three fishes were then introduced.			
Day 7	13.03.12	7.0	7.5
Day 8	14.03.12	7.0	8.5
Day 9	15.03.12	7.0	8.5
Day 10	16.03.12	7.0	9.0
Day 11	17.03.12	7.0	9.0
On the 11th day 3 fishes in the experimental set died and two in the normal setup died. The aquarium of the experimental setup was cleaned thoroughly and air dried. Three fishes in the experimental aquarium were reintroduced on 28.3.12. and two fishes were introduced in the normal aquarium. Reading was again taken from 29.3.12.			
Day 12	29.03.12	7.0	7.5
Day 13	30.02.12	7.0	8.5
Day 14	31.02.12	7.0	8.5
Day 15	2.04.12	7.0	9.0
Day 16	3.04.12	7.0	9.0
On the 16th day 3 fishes in the experimental set died			

Table 2a: Food response of fishes thriving in detergent contaminated waters.

Sl. No.	Date	FOOD RESPONSE	
		Control	Experimental setup
Day 1	26.11.2011	Normal (feeding form all sections of the aquarium-surface, column and bottom)	Normal
Day 2	28.11.2011	Normal	Normal
Day 3	29.11.2011	Normal	Loss of appetite (evidenced by lack of interest in the food. When food is introduced in the aquarium waters, show total lack of interest)
Day 4	01.12.2011	Normal	Lack of interest in food continues but at time seen to gush towards the food particle and gulp it very fast.
Day 5	02.12.2011	Normal	Takes time to detect food but eagerly gulps up the food.
Day 6	03.12.2011	Normal	As above.
Day 7	05.12.2011	Normal	As above.
Day 8	07.12.2011	Normal	As above.
Day 9	08.12.2011	Normal	As above.
Day 10	09.12.2011	Normal	As above.
Day 11	10.12.2011	Normal	As above.
Day 12	12.12.2011	Normal	As above.
Day 13	13.12.2011	Normal	As above.
Day 14	14.12.2011	Normal	As above.

Table 2b: Food response of fishes thriving in eutrophic waters.

Sl. No.	Date	FOOD RESPONSE	
		Control	Experimental setup
Day 1	5.03.12	Normal (feeding form all sections of the aquarium- surface, column and bottom)	Normal
Day 2	6.03.12	Normal	Normal
Day 3	7.03.12	Normal	Loss of appetite (evidenced by lack of interest in the food. When food is introduced in the aquarium waters, show total lack of interest)
Day 4	9.03.12	Normal	As above
Day 5	10.03.12	Normal	No food intake.
Day 6	12.03.12	Normal	No food intake
On the 5th day 2 fishes in the experimental set died. On the 6th day the remaining fish also died.			
Day 7	13.03.12	Normal	Normal
Day 8	14.03.12	Normal	Normal
Day 9	15.03.12	Normal	Loss of appetite (evidenced by lack of interest in the food. When food is introduced in the aquarium waters, show total lack of interest)
Day 10	16.03.12	Normal	As above
Day 11	17.03.12	Normal	No food intake.
On the 11th day 3 fishes in the experimental set died and two in the normal setup died.			
Day 12	29.03.12	Normal	Normal
Day 13	30.02.12	Normal	Normal
Day 14	31.02.12	Normal	Normal
Day 15	2.04.12	Normal	Loss of appetite (evidenced by lack of interest in the food. When food is introduced in the aquarium waters, show total lack of interest)
Day 16	3.04.12	Normal	No food intake.
On the 16th day 3 fishes in the experimental set died			

Table 3a: Opercular activity of fishes thriving in detergent contaminated waters.

Sl. No.	Date	OPERCULUM ACTIVITY [per minute] Normal range: 130-150	
		Control	Experimental setup
Day 1	26.11.2011	137 [even though there were three fishes the one which was the largest was considered for this parameter and all the data reflects that particular fish's data]	162 [even though there were three experimental fishes the one which was the largest {weight matched with the normal control} was considered for this parameter and all the data reflects that particular fish's data]
Day 2	28.11.2011	137	165
Day 3	29.11.2011	139	164
Day 4	01.12.2011	139	161
Day 5	02.12.2011	137	165
Day 6	03.12.2011	135	160

Day 7	05.12.2011	135	160
Day 8	07.12.2011	137	155
Day 9	08.12.2011	137	155
Day 10	09.12.2011	137	150
Day 11	10.12.2011	137	150
Day 12	12.12.2011	139	148
Day 13	13.12.2011	138	145
Day 14	14.12.2011	139	145

pH is a measure of hydrogen ion concentration in water. It has direct effects on fish growth and survival of food organisms. Hence, to achieve good fish production, pH of the water should be monitored regularly to ensure its optimum range of 6.5-7.5. It also exerts considerable influence on toxicity of ammonia and hydrogen sulphide as well as solubility of nutrients and thereby water fertility. In the experimental setup, with administration of detergent, the pH was maintained in a moderately high condition throughout (Table. 1a). With addition of 1gm detergent to 20 litres of water, the pH measured was found to be 8.5 (alkaline). The experimental setup was maintained in such condition for a fortnight with the pH being checked daily. When the experiment was conducted with aquatic macrophytes, the increase in pH showed similarity with the previous setup (Table.1b), but after every 5th day of addition of the macrophytes the pH increased to 9.00 which was followed by high fish mortality. The eutrophication induced phytoplankton bloom which intensified photosynthesis. As a result decrease in the pCO_2 followed which simultaneously enhanced the pH thereby causing high mortality.

Estimation of Behavioural Responses

Food response

The food response was remarkable as the fish in normal water took food at regular intervals and some leftover food was also observed, every time the food was administered. They would detect the food very quickly and would go toward it in a relaxed manner but especially in the morning hours it was observed that they were eagerly gulping the food particles. But the fishes in detergent treated water would take a longer time to detect the food but, once detected, would rush towards it gulping every bit, may it be morning, noon, or evening hours. Almeida et al., 2009, has seen the eagerness for food in rainbow trout as stress management strategies. They confirmed that reactive and proactive stress coping as seen in mammals is also present in rainbow trouts. Thus this observed greediness of *Oreochromis* for food in the present study can also be attributed to stress coping mechanisms. The behavioural response to artificial food in nutrient enriched water seemed to be very drastic. Especially after two day of introduction of the fishes to eutrophic waters, total loss in appetite is observed. In the 4th and the 5th day of administration of the aquatic macrophytes, food intake totally ceases. It seems that the nutrient enrichment in closed condition seems to have an adverse effect on the dietary behaviours.

Operculum activity

Respiratory activity of fish is often the first physiological response to be affected by the contaminants in the aquatic environment. Opercular movement is one of the early warning systems as an indicator of respiratory stress. In case of fish exposed to detergent treated water the opercular movement ranges from 145 -165 per minute, which is far more than the opercular movement in normal water

Table 3b: Opercular activity of fishes thriving in eutrophic waters.

Sl. No.	Date	OPERCULUM ACTIVITY [per minute] Normal range: 130-150	
		Control	**Experimental setup**
Day 1	5.03.12	138 [even though there were three fishes the one which was the largest was considered for this parameter and all the data reflects that particular fish's data]	137 [even though there were three experimental fishes the one which was the largest {weight matched with the normal control} was considered for this parameter and all the data reflects that particular fish's data]
Day 2	6.03.12	138	155
Day 3	7.03.12	138	170
Day 4	9.03.12	137	171
Day 5	10.03.12	139	160
Day 6	12.03.12	139	155
On the 5th day 2 fishes in the experimental set died. On the 6th day the remaining fish also died. On the 6th day the remaining fish (the fish on which the data is based) also died. Again 3 fishes reintroduced and the data is based on the fish with greater weight.			
Day 7	13.03.12	138	140
Day 8	14.03.12	137	165
Day 9	15.03.12	138	170
Day 10	16.03.12	138	150
Day 11	17.03.12	139	150
On the 11th day 3 fishes in the experimental set died and two in the normal setup died. Again 3 fishes reintroduced and the data is based on the fish with greater weight.			
Day 12	29.03.12	135	142
Day 13	30.02.12	135	165
Day 14	31.02.12	137	165
Day 15	2.04.12	135	148
Day 16	3.04.12	135	145
On the 16th day 3 fishes in the experimental set died			

Table 4a. Swim pattern of fishes thriving in detergent contaminated waters.

Sl. No.	Date	SWIM PATTERNS [per minute] [H=Horizontal; V=Vertical; B=Backwards]	
		Control	**Experimental setup**
Day 1	26.11.2011	H-6 ; V-1 [even though there were three control fishes the one which was the largest was	H-10 ; V-3 [even though there were three experimental fishes the one which was the largest {weight matched with the

		considered for this parameter and all the data reflects that particular fish's data]	normal control} was considered for this parameter and all the data reflects that particular fish's data]
Day 2	28.11.2011	H-5; V-1	H-10; V-5
Day 3	29.11.2011	H-6 ;V-2	H-12 ;V-4
Day 4	01.12.2011	H-5; V-2	H-10; V-3; B-2
Day 5	02.12.2011	H-5; V-1	H-10; V-3; B-1
Day 6	03.12.2011	H-5; V-1	H-10; V-3; B-1
Day 7	05.12.2011	H-6 ; V-1	H-10; V-3;
Day 8	07.12.2011	H-6 ;V-2	H-10; V-3
Day 9	08.12.2011	H-5; V-2	H-10; V-2
Day 10	09.12.2011	H-6 ; V-1	H-8 ;V-2
Day 11	10.12.2011	H-5; V-1	H-9 ; V-2
Day 12	12.12.2011	H-6 ; V-1	H-9 ; V-2
Day 13	13.12.2011	H-6 ;V-2	H-8 ;V-2
Day 14	14.12.2011	H-5; V-2	H-9 ; V-2

Table 4b: Swim pattern of fishes thriving in eutrophic waters.

Serial No.	**Date**	**SWIM PATTERNS [per minute] [H=Horizontal; V=Vertical; B=Backwards]**	
		Control	**Experimental setup**
Day 1	5.03.12	H-5 ; V-1 [even though there were three control fishes the one which was the largest was considered for this parameter and all the data reflects that particular fish's data]	H-5; V-1 [even though there were three experimental fishes the one which was the largest {weight matched with the normal control} was considered for this parameter and all the data reflects that particular fish's data]
Day 2	6.03.12	H-6; V-2	H-7; V-1
Day 3	7.03.12	H-5 ;V-1	H-12 ;V-4
Day 4	9.03.12	H-6; V-1	H-10; V-3; B-2
Day 5	10.03.12	H-5; V-2	H-3; V-1; B-1
Day 6	12.03.12	H-5; V-1	H-2; V-1; B-1
On the 5th day 2 fishes in the experimental set died. On the 6th day the remaining fish also died. Again 3 fishes reintroduced and the data is based on the fish with greater weight.			
Day 7	13.03.12	H-5 ; V-2	H-5; V-2;
Day 8	14.03.12	H-5 ;V-1	H-10; V-3
Day 9	15.03.12	H-6; V-1	H-10; V-2
Day 10	16.03.12	H-5; V-2	H-3 ;V-1
Day 11	17.03.12	H-6; V-2	H-3 ; V-2
On the 11th day 3 fishes in the experimental set died and two in the normal setup died. Again 3 fishes reintroduced and the data is based on the fish with greater weight.			
Day 12	29.03.12	H-5 ; V-2	H-5 ; V-1

Day 13	30.02.12	H-5 ;V-1	H-8 ;V-2
Day 14	31.02.12	H-6; V-1	H-9 ; V-2
Day 15	2.04.12	H-5; V-2	H-2 ;V-1
Day 16	3.04.12	H-5 ;V-1	H-2; V-1; B-1
On the 16th day 3 fishes in the experimental set died.			

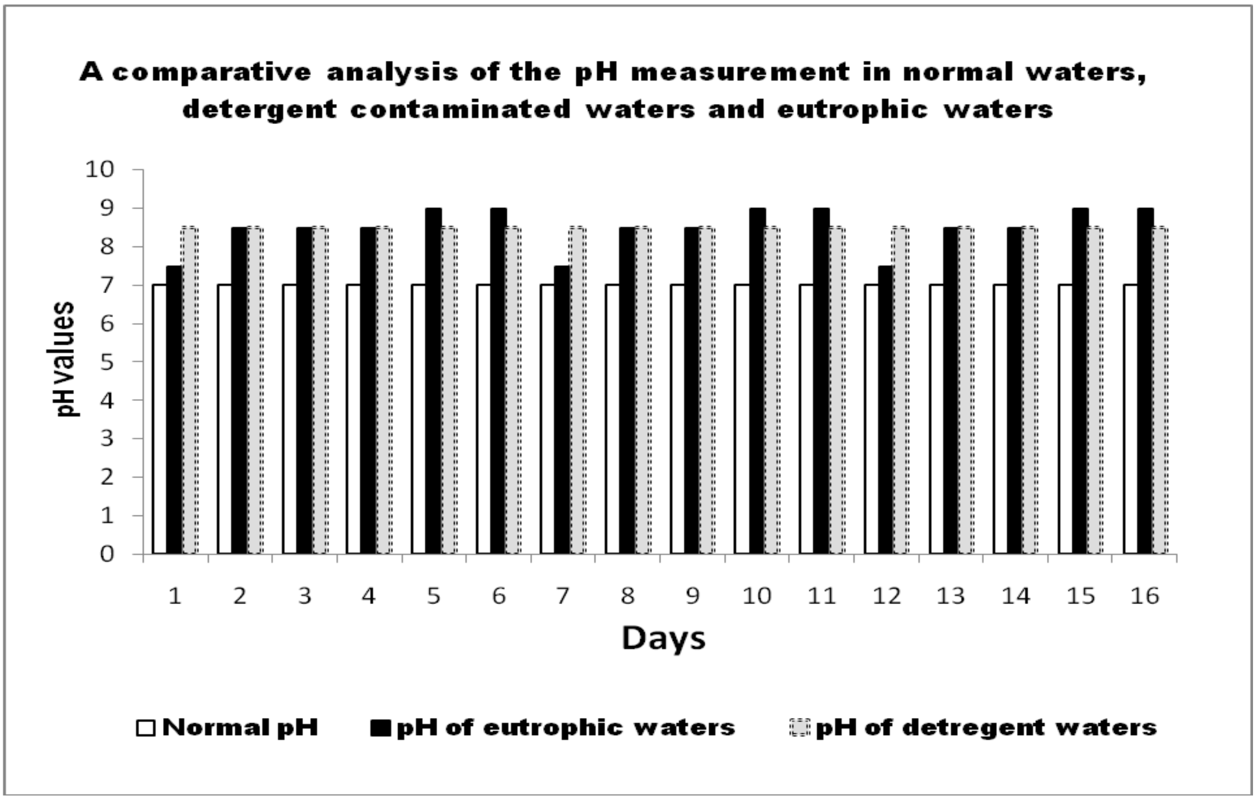

Fig.1: **A comperative analysis of the pH measurement in normal waters, detergent contaminated waters and eutrophic waters.**

that is 135-139, a clear indication of respiratory stress. Since fishes breath in water in which they live, changes in chemical properties there will surely be reflected in their respiratory activity. Schjolden et al., 2005, had studied stress in *Tilapia* with results in the same line. In nutrient enriched water the opercular activity increases drastically in the 3rd and 4th day most probably due to decrease in oxygen concentration (due to higher nutrient enrichment). Moreover the increased alkaline nature (9.0) of the water induced a high physiological stress as a result of which the fishes tend to become flaccid.

Swim patterns [per minute]

Fishes swim using undulatory movements of their body and their paired and unpaired fin. In this type of movement a backward bending is generated by the sequence of myotomes from head to tail (Altringhnam et al., 1999), body and caudal fin generating a forward thrust. Different species use their myotome muscle to generate thrust in different ways, in spite of many features being common. Swimming of these fishes were noted in horizontal and vertical directions, But backward swimming was noted in the fish in detergent treated water, that too very rarely, but never in fishes of the normal water. In fishes thriving in eutrophic waters also we observe backward swimming. Backward swimming has been studied in fishes like eel (Kristian and Peter 1999) where it was found that elongated swimmers can swim backwards in addition to longitudinal undulatory movements. Islam et al., 2006, studied the neural coordination underlying swimming and motor pattern where intrinsic function of spinal network is well understood. Hale et al., 2006 studied dog fish for the inter relationship of forward and backward swimming relationship. But no report for such movement in *Oreochromis* has been reported hitherto reported. Thus we can point out that there is some mis-coordination developing in the neural network in fishes thriving in both detergent contaminated waters and eutrophic waters.

CONCLUSION

Behaviours allow organisms to adjust to external and internal stimuli in order to best meet the challenges to survive in a changing environment. It is a selective response that is constantly adapting through direct interaction with physical, chemical, social, and physiological aspects of the environment. Observation in the above experimental setup point out the indiscrepancy in swimming movements, hyper excitation and respiratory dysfunction [as the fishes hovered around the oxygenater] in fishes exposed to detergent treated waters or nutrient enriched waters. An important indicator of any water body is the pH .Comparing the pH changes in the two experimental waters with respect to normal waters (Fig.1), it was evident that the pH fluctuation were more hyper in nutrient enriched waters. The data simulates with another finding by Lapointe et al., 2001 where he also conducting similar experiments in Florida Bay observed a pH value of 9.4 in the Bay water due to eutrophication. Cleary the pH affects the survival capacity of the fishes as documented by the observation of high mortality of fishes that were kept in the eutrophic waters.

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