

## Studies on Oxygen Consumption of the Fish *Catla catla* (Hamilton) as Influenced by Toxic Synergism

M. ILAVAZHAHAN<sup>1</sup> and R. TAMILSELVI<sup>2</sup>

<sup>1</sup>Department of Zoology, Sir Thyagaraya College, Chennai - 600 021, India.

<sup>2</sup>Department of Chemistry, Bharathi Women's College, Chennai - 600 108, India.

(Received: November 15, 2011; Accepted: December 12, 2011)

### ABSTRACT

An investigation was carried out to assess the impact of pathogens (*Pseudomonas putida*), pesticide (Methyl parathion) and metal (Ferrous sulphate) on the oxygen consumption of the fish *catla catla* (Ham.). The effect of oxygen consumption were studied by exposing the fish *catla catla* to the toxicant. The changes in the oxygen increment was noticed with increasing exposure time.

**Key words:** *Pseudomonas putida*, Methyl parathion, Ferrous sulphate, *Catla catla*.

### INTRODUCTION

Pollution is an undesirable change in the physical, chemical or biological characteristics of our land, air or water that may or will harmfully affect human life or that of desirable species. Freshwater ecosystem consists of a large number of fauna and flora in them. These aquatic organisms are very sensitive to even a slight alteration in the environment. The causes for pollution are mostly man-made and therefore the prevention and control measures must be taken as much as possible.

Bacterial diseases are a constant threat to fish farming, and because of the rapid course and severity of manifestation, they represent a significant part in fish pathology and also have a great economic importance (Jeremic *et al.*, 2005). Harmful effect of bacterial diseases on fishes are increased morbidity and Mortality rate, decreased feed conversion efficiency, decreased growth rates, weakening of reproductive capability.

Pesticides are mainly used against pests of crop and disease causing vectors, but their improper use in agricultural practice has posed a serious threat to human life and his environment. Highly effective pesticides are used extensively and

indiscriminately, which on entering the aquatic environment bring multiple changes in organisms by altering the growth rate, nutritional value, behavioral pattern.

In nature, aquatic animals are constantly exposed to toxicants including metallic ions. Concentrations of metals in water are determined by geochemical processes and large scale release into the aquatic environment by human activities. Intensive activity in industrial and agricultural sectors has inevitably increased the levels of heavy metals in natural waters (Jordao *et al.*, 2002). Heavy metals play a major role among pollutants of environmental concern (Singer *et al.*, 2005).

Although, trace metals are essential for normal physiological process, abnormally high concentrations can be toxic to aquatic organisms (Javed, 2003). Heavy metals being non-biodegradable primarily necessitate knowledge on their uptake, distribution and persistence in tissues of organisms (Lewis *et al.*, 2004).

Toxicity of a substance is known by its capacity to cause adverse effects on the living organisms. Toxic impact may bring about physiological, biochemical or pathological

alterations in the organisms; the signs of toxicity may reveal symptoms of illness varying from simple local effects - structural and behavioral (Shivakumar *et al.*, 2005) to complex disorders resulting in mortality. The intoxication includes a sequence of events that start with the exposure of a substance to an organism. Subsequently, the toxic substances are absorbed into the viscera by various routes causing an internal exposure (Tilak *et al.*, 2005). Within the body of an organism, the substances are converted into metabolites which may either be more toxic or less toxic. The sequence of events and interaction of toxic substances with target molecules of organism depend on various factors such as the nature of the toxicant, duration of exposure, physiological state of the organism, biotic and abiotic factors of environment (Subramaniam, 2004).

Any variation in the environment acts as a stress on the organisms. When a pesticide or any pollutant reaches the aquatic ecosystem, the fish are exposed to severe stress and as a natural instinct, the fish tend to adapt themselves by reacting suitably to overcome the stress. When two toxicants act together, the stress shall be more and severe and cause irreparable damage. In an aquatic environment the pollutants reach easily as runoff from pesticide residues and heavy metals like Iron, copper, mercury, nickel, lead and zinc pollute the aquatic system through industrial and municipal wastes. In such a scenario, the metal compounds react with the chemical pesticides to form complexes that may have entirely different properties than the original one. Such complexes formed may be more toxic than the individual toxicant or may even be less toxic. When the fish are exposed to such complexes the reaction to stress by the fish may be different.

Hence an attempt has been made in the present study to know the effects of a pathogen, pesticide and a heavy metal compound acting independently and combined. For this study, a pathogen, *pseudomonas putida*, a pesticide, Methyl parathion and a heavy metal compound, Ferrous sulphate ( $\text{FeSO}_4$ ) were chosen as the toxicants and one of the commercially important Indian Major Carp, *Catla catla* chosen as the target animal to test the toxic effects on.

## MATERIAL AND METHODS

Fingerlings of *Catla catla* of relatively same size ranging from (8 to 10 cm) and weight about (5-8 gm) were collected from culture ponds of Bharath Fish Farm, Poondi, Thiruvallur district, Tamil Nadu. The fish, after conditioning, were oxygen packed in tins and brought to the lab. They were slowly released into cement tanks, half filled with bore water and seasoned over night. These were maintained in stocking tanks, where the fish were quarantined and acclimatized for two days.

The fish feed was prepared with rice bran, groundnut oil cake, tapioca powder and mineral mixture (Ramaiah, 1982).

The fish were fed daily with pelleted feed at 5% body weight in two split doses, in the morning and evening. Feeding was started one day after the fish were stocked and stopped 24 hr prior to experiment.

All the fish were maintained in the glass aquaria of the size 1'L x 2'B x 1'H throughout the period of experimentation. The tanks were disinfected with potassium permanganate thoroughly, before and after use. These tanks were filled with known volume of water per fish (30 litres for 10 fish) and covered with nylon mesh to prevent the mosquitoes breeding in the tanks and also to restrain the fish jumping out.

Healthy fish without any observable pathological symptoms were chosen for the experiments. Fish were divided into groups of ten each and exposed to the different toxicants viz. Bacteria, Methylparathion and ferrous sulphate independently and synergistically. In the synergistic toxicity experiments, the fish were exposed to two toxicants together.

Fingerlings of *Catla catla* were treated with Pathogen, pesticide and heavy metal individually and synergistically to determine 96hr  $\text{LC}_{50}$  values for the test toxicants. Groups of fish were maintained in separate tanks and considered as experimental groups which were categorized as follows:

- Independent toxicity
  - Bacterial Pathogen
  - Methyl Parathion
  - Ferrous Sulphate
- Combined synergism
  - Bacteria + Methyl parathion
  - Bacteria + Ferrous sulphate
  - Ferrous sulphate + Methyl parathion

Suitable controls were maintained in normal bore well water for all the experimental groups, without dissolving the chemicals.

Groups of fish were exposed to the toxicants at SLC and sacrificed on 4, and 7, days post exposure to study of their physiology.

Rate of Oxygen consumption were analyzed on zero day control and on 4<sup>th</sup>, and 7<sup>th</sup> day after exposing the fish with the toxicants in the experimental groups of independent toxic exposure and combined synergistic toxic exposure.

Rate of oxygen consumption was studied in the control and experimental groups by following the modified Winkler's method. The dissolved oxygen content of the water in the respiratory chamber was estimated, before the fish was released. After one hour, water from the respiratory chamber was carefully siphoned out avoiding air bubbles, and the dissolved oxygen content was estimated. The fish were weighed accurately to the nearest milligram after the fish were carefully wiped on a blotting paper. From this data, the unit oxygen consumption was calculated for each fish and expressed as ml/gm/hr. The dissolved oxygen in water was calculated using the formula.

$$\frac{K \times 200 \times 0.698 \times \text{Titre Value}}{\text{Volume of Sample}}$$

$$\text{Rate of Oxygen consumption: } \frac{\text{Oxygen Consumed}}{\text{Wt. of fish}} = \text{ml/gm/hr.}$$

For all the parameters analysed, ten estimations were made and subjected to standard statistical methods to arrive at Mean, Standard Deviation and Test of Significance. The results were

tabulated and the data represented in suitable tables and histograms.

### RESULTS AND DISCUSSION

Oxygen consumption by the fish was estimated both in the control as well as treated groups exposed to bacterial pathogen (*Pseudomonas putida*), pesticide (Methyl parathion) and a heavy metal (Ferrous sulphate) individually and combined as bacteria + Fe SO<sub>4</sub>, bacteria + methyl parathion and FeSO<sub>4</sub> + methyl parathion synergistically. The experimental analyses were done on 4<sup>th</sup> day and 7<sup>th</sup> day post exposure to toxicants. The experimental groups which were exposed to bacteria, pesticide and heavy metal independently recorded a higher rate of oxygen consumption on 4<sup>th</sup> day and 7<sup>th</sup> day compared to control groups with 0.19 and 0.16 ml/gm/hr respectively.

In the experiments of combined synergism, where the test fish were exposed to two toxicants together viz., bacteria + FeSO<sub>4</sub>, bacteria + methyl parathion and FeSO<sub>4</sub> + methyl parathion also, similar observations were recorded on 4<sup>th</sup> day and 7<sup>th</sup> day of experimentation with a two fold increase.

Several workers take up oxygen consumption as an important study to know the effects of toxicant. Prashanth *et al.*, (2003) observed that the rate of oxygen consumption increased when exposed to pesticide, cypermethrin. Tripathi *et al.* (2003) studied the effect of bacterial pathogen on freshwater fish *Labeo rohita* and observed increased opercular movements under the influence of bacterial pathogen.

Metals like cadmium compounds also brought down the oxygen consumption in the freshwater fish *Ctenopharyngodon idella* (Espina *et al.*, 2000). The decline in the consumption of dissolved oxygen may be due to gill damage that reduces the efficiency of oxygen uptake. Alam and Mangalam (1995) observed that the lead has also been reported to accumulate in the body of fishes from the surrounding water and detritus and there by produce its adverse physiological effects through biological mechanism.

Sampoorani *et al.* (2001) studied in fresh water fish *Labeo rohita*, an increased rate of oxygen consumption due to textile effluent treatment, while treatment with dairy effluent brought down the rate of oxygen consumption. The rate of oxygen consumption in *Catla catla* treated with DTC, CuSO<sub>4</sub> and ZnSO<sub>4</sub> showed an increasing trend in all the experimental groups as the duration of exposure increased (Sujatha, 2006). The test organism exposed to sub lethal concentrations of pathogen, pesticide and a heavy metal independently showed that rate of oxygen consumption was increasing in all the experimental groups as the duration of the exposure increased. The differences observe in the group were highly significant, when compared to the control fish.

Oxygen consumption can yield important information as an index of the metabolic rate. The alteration or failure of respiratory metabolism is one of the early symptoms of pesticide poisoning in any organism. Decrease in the rate of respiration with increasing concentration to toxicants was recorded in fishes.

In an aquatic environment, one of the most important manifestations of the chemical toxicant is over stimulation or depression of respiratory activity (Murheed Thomson, 1977). Tilak *et al.* (2002) stated that the rate of oxygen consumption decreased with an increase in the duration of exposure resulting in absorbance of more toxicant through gill. At higher concentrations of metals, the

**Table 1: Oxygen consumption in catla : Independent toxicity**

(Catla)		Consolidate table : Oxygen consumption (ml / l / hr)				
		zero day control	4 <sup>th</sup> day control	4 <sup>th</sup> day treated	7 <sup>th</sup> day control	7 <sup>th</sup> day treated
(Exposed to bacteria)	Mean	0.19	0.19	0.28	0.16	0.32
	S.d.	0.012	0.005	0.032	0.031	0.015
	P <		0.001		0.001	
(Exposed to feso <sub>4</sub> )	Mean	0.19	0.19	0.47	0.16	0.39
	S.d.	0.012	0.005	0.049	0.031	0.035
	P <		0.001		0.001	
(Exposed to parathion)	Mean	0.19	0.19	0.33	0.16	0.34
	S.d.	0.012	0.005	0.014	0.031	0.024
	P <		0.001		0.001	

**Table 2: Oxygen consumption in catla : Combined synergistic toxicity**

(Catla)		Consolidate table : Oxygen consumption (ml / l / hr)				
		zero day control	4 <sup>th</sup> day control	4 <sup>th</sup> day treated	7 <sup>th</sup> day control	7 <sup>th</sup> day treated
(Exposed to Bacteria + FeSO <sub>4</sub> )	mean	0.19	0.19	0.34	0.16	0.39
	S.d.	0.012	0.005	0.024	0.031	0.013
	P <		0.001		0.001	
(Exposed to Bacteria+ Parathion)	mean	0.19	0.19	0.35	0.16	0.41
	S.d.	0.012	0.005	0.032	0.031	0.045
	P <		0.001		0.001	
(Exposed to FeSO <sub>4</sub> + parathion)	mean	0.19	0.19	0.20	0.16	0.31

dissolved oxygen contents of the test medium decreased significantly. This shows that high concentrations of metallic ions induced stress in the fish that resulted in significantly more oxygen consumption and thus, dissolved oxygen concentrations of the test medium declined. Environmental conditions such as oxygen concentration, temperature, hardness, salinity and presence of other metals may modify metal toxicity to the fish.

Toxicants in the environment mainly enter into fish by means of their respiratory system. A mechanism of toxicant uptake through gill probably occurs through pores simple diffusion and is then observed through gill membranes (sampoorni *et al.*,2001). From the results obtained, it is clearly evident that the toxicants affect the oxygen consumption of *Catla catla* under all exposed concentrations. The observed increased in oxygen consumption by the whole animal may be due to respiratory distress as a consequence of the impairment of oxidative metabolism.

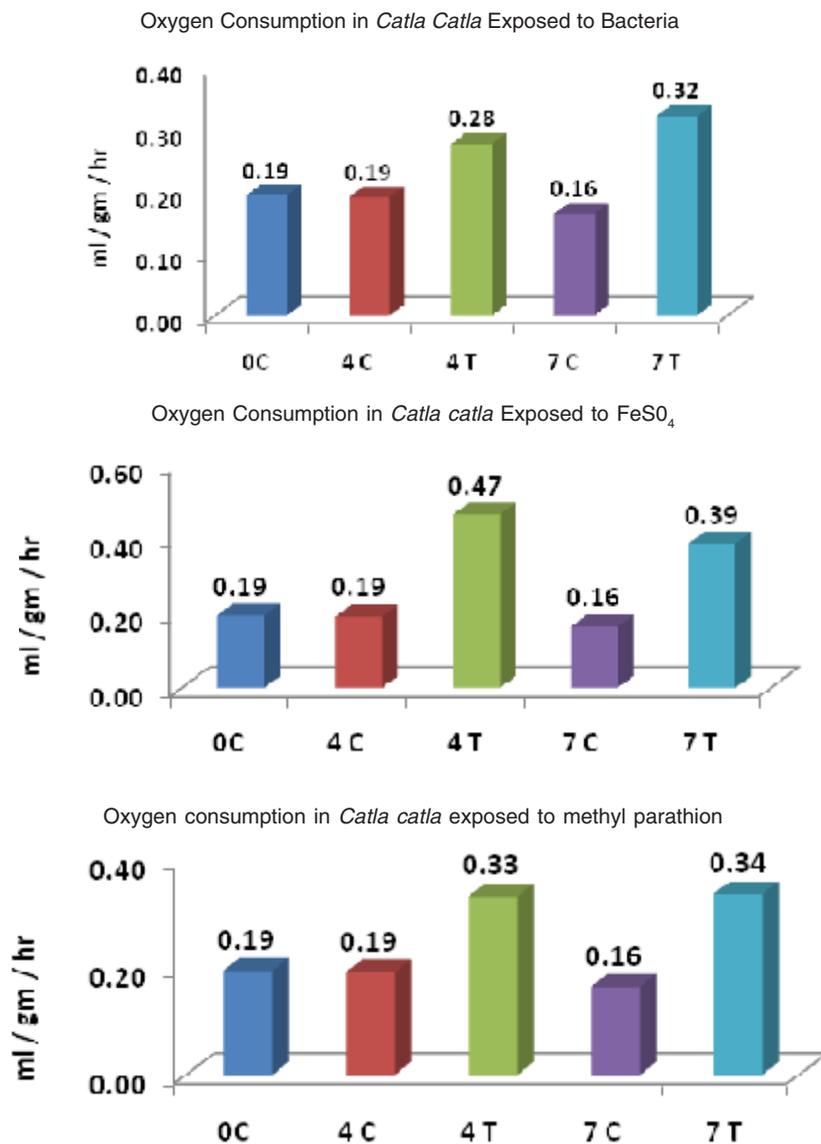


Fig. 1: Independent toxicity

The elevation in the oxygen consumption by 50% in the experimental group of fish treated with pathogen independently revealed the nature of toxic effect and its impact on the test organism. These results corroborate with the observations of behavior like increased opercular beats, increase in surface behavior, increase in secretion of mucus, irregular swimming activity, rapid and darting

movement. Moreover, the higher rate of oxygen consumption may be due to increased metabolic rates in the experimental fish as an adaptation to overcome stress induced by the toxicants.

Sampoorani *et al.*, (2001) studied in fresh water fish *Labeo rohita*, an increased rate of oxygen consumption due to textile effluent treatment.

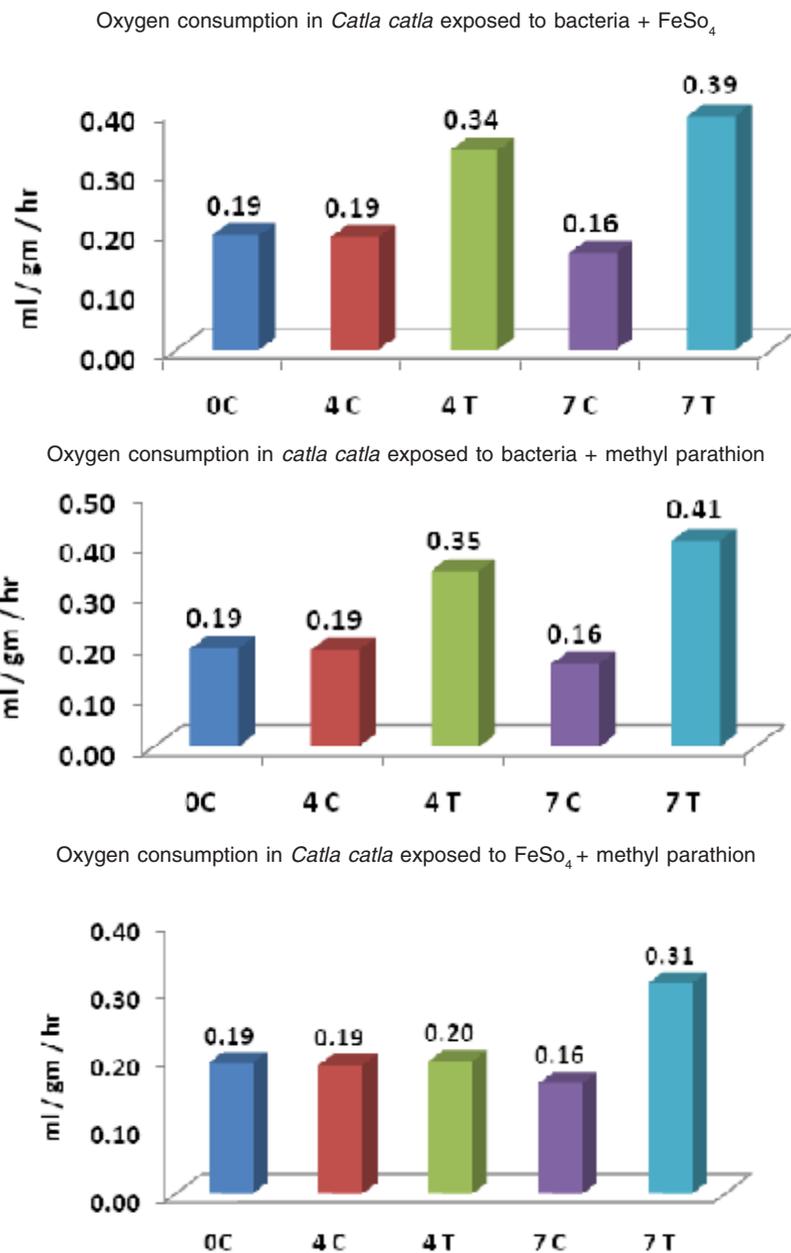


Fig. 2: Combined Synergistic Toxicity

Sujatha (2006) reported that the disturbance in oxidative metabolism leads alteration in whole animal oxygen consumption in different species of fish exposed to pesticides. The alternative reason for the elevation of oxygen consumption would be

due to the internal action of toxicants. The results of the present study confirm the earlier reports on oxygen consumption by fish in toxicants mixed water.

## REFERENCES

1. Alam, M.K. and O. C. Maughan. Acute toxicity of heavy metals to common carp, *Cyprinus carpio*. *Environ. Hazards. Subst. Control.* **30**(8): 1807-1816 (1995).
2. Espina, S., A. Salibian and F.Diaz. Water, air and soil Pollution. **119**(1-4): 1-10 (2000).
3. Javed, M. Relationships among water, sediments and planktons for the uptake and accumulation of heavy metals in the river Ravi. *I.J.P. Sci.* **2**: 326-331 (2003).
4. Jeremic, S., Radosavl Jevic and D. Jakic-Dimic. Current bacterial diseases of fresh water fishes. *Biotechnology in Animal-Husbandry*. **21** (3/4): 141-151(2005).
5. Jordao, C P and M G. Pereira. Assessment of water systems for contaminants from domestic and industrial sewages. *Environ. Monit. Assess.* **79**: 75-100 (2002).
6. Lewis, M. A., Scott, G. I., Bearden, D.W and E. D. Strozier. Fish tissue quality in near-coastal areas of the Gulf of Mexico receiving point source discharges. *The Science of the Total Environment*. **284**: 249-261 (2004).
7. Murheed Thomson , R.L. Pesticides and Freshwater Fauna. *Academic Press, New York*. 2451(1971).
8. Prashanth, M.S. David , M. and R.C. Kuri. Effect of cypermethrin on toxicity and Oxygen consumption in the freshwater fish, *Cirrhinus mrigala*. *J. Ecotoxic. And Environ. Monit.* **13**(4): 201-271(2003).
9. Ramaiah, N. Bacterial diseases of Indian Major Carps and effects of chemotherapeutic drugs on the host and pathogen. M.F.Sc., Thesis. Coll. Fish., Mangalore (1982).
10. Sampoorani, V., Dhanapackiam, P. and S. Eswari. Toxic effects of textile and dairy effluents on growth and respiration in fingerlings of *Labeo rohita*. *NCEBB-LIFE*.(2001).
11. Shivakumar, R. Mushigeri, S. B. and M. Davie. Effect of Endosulfan to freshwater fish *Ctenopharyngodon idellus*. *J. Ecotoxl. Environ. Monit.* **15**: 113-116 (2005).
12. Singer, C and S. Zimmermann. Induction of heat shock proteins (hsp70) in the zebra mussel (*Dreissena polymorpha*) following exposure to platinum group metals (platinum, palladium and rhodium): Comparison with lead and cadmium exposures. *Aquatic toxicology*. **75**:65-75 (2005).
13. Subramanian, M.A., Toxicology: Principles and Methods. *MJT Publishers*. p202 (2004).
14. Sujatha, L. B. Studies on the Physiology, Haematology and Histology in the Indian Major Carp, *Catla catla* (Ham.) As influenced by individual and synergistic toxic effects of a pesticide and two metallic compounds. Ph. D., Thesis. University of Madras, Madras (2006).
15. Tilak, K.S. and K. Yacob. Toxicity and effect of fenvalerate on fish *Ctenopharyngodon idella* exposed to technical fenvalerate and EC 20%. *Pollu. Res.* **20**(3): 387-393 (2002).
16. Tilak, K.S., Veeraiah, K. and Koteswara Rao. Histopathological changes observed in the Gill, Liver, Brain and Kidney of the Indian Major Carp *Cirrhinus mrigala* exposed to Chlorphrifos. *Pollu. Res.*, **24**(1): 101-111(2005).
17. Tripathi, N.K. Latimer, K. S, and P. M. Rakich. Columnaris disease in freshwater fish *Labeo rohita*. *Compendium on continuing Education for the practicing Veterinarian*. **25**(7): 528-536 (2003).