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EVALUATION OF OXYGEN TOLERENCE IN FRESH WATER FISHES

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ABSTRACT

Dissolved oxygen (DO) is one of important environmental variable. It has changed so drastically, in such a short period of time that causes loss of fisheries, loss of biodiversity and alteration of food webs. Dissolved oxygen is required to meet the respiration needs of fish fauna. Once the supply of oxygen is cut off in water and the oxygen consumption rate exceeds resupply, the fish fauna suffocate and results into moribundity.

In the present study four different fresh water fishes such as Gambusia affinis (Baird and Girrard), Clarias magur (Linnaeus), Xiphophorus clemenciae (Alvarez) and Labeo rohita (Hamilton) were selected for finding lethal oxygen concentration .We found oxygen consumption rate of all fishes. This study shows that smaller size fish has moderately high rate of dissolved oxygen consumption than the larger ones.

***KEYWORDS**: Dissolved oxygen (DO), Oxygen consumption rate, Moribundity*

INTRODUCTION

The economics of most modern aquaculture operations require that animals be cultured at high densities. Various factors affecting this intensive culture include proper concentration of dissolved oxygen level, the availability of food, and the optimum range of various physical and chemical factors. The vital physical factors are temperature, light etc. whereas important chemical factors include the sustainable range of pH, carbon dioxide, ammonia, salinity, etc.

Dissolved oxygen concentration (DO) is considered the most important water quality variable in fish culture, in the broadest sense. However, dissolved oxygen concentration is no more important than other environmental variables because any factor that is outside the range tolerated by fish can cause stress or death.

The availability of oxygen is the most striking feature, which remains as top priority requirement for aquatic life. The dissolved oxygen content in aquatic environment also varies much than in the air because availability of oxygen is limited. To begin with, small differences in the metabolism of aquatic life can dramatically change dissolved oxygen concentration. If dissolved oxygen concentration is consistently low, the growth of aquatic animals will be affected and will be more susceptible to infectious diseases. If still concentration falls to further low level, the fish may die. This level of dissolved oxygen is considered as lethal oxygen concentration and the dissolved oxygen concentration above this which is enough for just survival but not for growth and health is called sublethal concentration.

What makes dissolved oxygen concentration so important in intensive fish culture is the speed with which it can change, over a matter of hours, or sometimes even minutes, from optimum to lethal level. No other environmental variable in fish culture is so dynamic.

The dynamic nature of dissolved oxygen concentration results from the interaction of three factors. First, oxygen is not very soluble in water so water has only a limited capacity to hold oxygen. Second, the rate of oxygen used by aquatic biota (fish, plankton and other organism) living in the pond and its bottom mud can be high. Third, oxygen diffuses very slowly from the atmosphere into undisturbed water. The combination of these three factors, i.e. limited solubility, rapid use and slow replenishment can cause drastic change in dissolved oxygen concentration (P. Mickel: Ecological methods for field and laboratory investigations).

As the pressure on aquaculture industry increases, the fresh water fishes play an important role. The production of fish has become increasingly intensive during the last few decades with reduced flow of water as one of the key feature (Rosten et al. 2004). This is made possible with the extensive use of hyper oxygenation in the inlet water, increasing the oxygen saturation typically up to 160% (Wedemeyer, 1996; Lygren et al., 2000).

Metabolism and growth of fishes are dependent on the availability of oxygen (Doudoroff and Shumway, 1970; Fry 1971; Davis, 1975; Brett, 1979; Kutty, 1981). Thus all factors affecting changes in dissolved oxygen concentration, including the lowering of oxygen and resultant hypoxia and diet flux of oxygen can affect production of fish in pond. There is considerable information on how ambient oxygen limits growth in fishes. For example, Hermann (1975) found that growth of the salmonid, *Onchorhyncus kisutch* is proportional to dissolved oxygen between 4 and 8 mg/l at 20ºC.

The problem of hypoxia occurred in fish is the reduction of respiratory frequency, which causes accumulation of acidosis (Bernier and Randell, 1998).

Several researchers have reported that dissolved oxygen levels can also affect the toxicity of ammonia to aquatic organisms. Thurston et al. (1981) showed an increase in ammonia toxicity to rainbow trout (*Onchorhyncus mykiss*) at reduced levels of dissolved oxygen.

There are numbers of ways to measure dissolved oxygen concentrations. These are selected on the basis of 1) the number of tanks/ experimental bottles to be measured, 2) the level of accuracy required 3) the cost of the measurement technique. The titration based "drop-count" method fairly rapidly assesses, whether or not there is sufficient dissolved oxygen in water. The drop-count method is in expensive and appropriate.

The purpose of this study intend to check the oxygen tolerance level for some fresh water fishes named as *Gambusia affinis* (Baird & Girard), *Clarias magur* (Linnaeus), *Xiphophorus clemenciae* (Alvarez) *and Labeo rohita* (Ham.).

MATERIALS AND METHOD

I. Selection of Fish

Four different fresh water fishes were selected for the experiment. These are *Gambusia affinis* (Baird & Girard), *Clarias magur* (Linnaeus), *Xiphophorus clemenciae* (Alvarez) *and Labeo rohita* (Ham.).

1. *Gambusia affinis* (Baird & Girard) – It is a mosquito fish. It is an actinoptergian fish, native to North and Central America, distributed in Mississippi river basin from central Indiana and Illinois in USA to south of Gulf of Mexico. Most abundant in lower reaches of streams. Its maximum size is 40 cm. Total length (male/ Unsexed); 7 cm. (female) and reported age 3 year. In India it is exotic fish but very well established all over.
2. *Clarias magur* (Linnaeus) **–** It is an actinoptergian cat fish, distributed in India, Pakistan, Nepal, Sri Lanka, Bangladesh, Thailand, Myanmar, Philippines, and Indonesia. USA, Singapore. Its approximate size is about 60 cm. experiment was done on fish fingerlings of size 10 cm approximately.
3. *Xiphophorus clemenciae* (Alvarez) – It is an aquarium fish, it belong to class Actinopterygii, its size is about 4 cm. it is a native to Central America, Mexico.
4. *Labeo rohita* (Ham.) – This also belongs to class Actinopterygii. Its common name is rohu. It is a carp fish, attaining a maximum size of about 200 cm. weight about 45.0 kg; it is distributed throughout the Asia.

All the above selected fishes have specific importance. They have different oxygen tolerance. The fish were collected from different sources. 50 fish of each sample were collected and transferred to glass aquaria or in buckets. The fish were maintained in aerated, dechloroinated municipal tap water. They were fed on artificial diet once a day, around noon, with 3% to 4% of total fish biomass given in a form of dry pellets which were floating type.

II. Acclimatization

This part is important for fish for and it's stocking in new environmental condition. In the laboratory, the fish were acclimatized in the experimental tanks for a week prior to the conducting of an experiment. The acclimatization period varies for different species of fish. During acclimatization oxygen level was maintained above 5 ppm and water temperature was maintained at 24ºC to 30ºC.

III. Starvation

Prior to starting of an experiment the fishes were starved. The fishes were transferred from aquarium to the plastic containers containing dechlorinated water. The fishes were starved for 24 hours before starting an experiment. This was done to avoid excess contamination of water due to fecal discharge of fishes. 3-4 fishes were stocked in each plastic container and the water was changed after 24 hours.

IV. Setting of an Experiment

The acclimatized and starved fish were transferred to BOD bottle of capacity 150ml, 250ml, 500ml and 1 liter as per the weight of fishes.

The MCD tap water was used for experiment. It was dechlorinated by leaving it open for 24 hr. before its use in the experiment or storage of fish, acclimatization etc. The pH of the water was checked by using the digital pH-meter. The temperature of water was also determined with the help of laboratory thermometer. The dissolved oxygen of the water sample was determined by using Winkler’s method. The fish was introduced into the BOD bottles already filled with dechlorinated water up to the brim. A small muslin cloth net was used for transferring the fish from the acclimatization tank to the BOD bottle. A large funnel was put on the mouth of BOD bottle, so that fish was handled least with hands. Thereafter the BOD bottle was stopper using airtight stopper taking care that no air bubble gets trapped below the stopper. The time of starting the experiment was recorded. The number of fishes in each BOD bottle depended upon the weight and length of the fish. Each fish was mandatory provided with 1 liter of water/ 5 gms of body weight. During experiment the fish was observed for the following activities

- Locomotion
- Opercular movement
- Movement of fins
- Quiescence
- Death (moribundity) time.

The fish was observed till it reached the moribundity which was symptomized by the absence of any movement of operculum and fins and more frequently turning of fish upside down. At this point the pH, temperature and dissolved oxygen was again calculated in the experimental water. The fishes were than taken out for recording their length and weight.

OBSERVATIONS AND RESULTS

Gambusia affinis

Experiment (Table 1): In all 9 fishes were tested. The weight of the test fishes ranged between 0.03gm to 0.171gms. Each fish was provided with around 0.45 liters water/ gm body weight. pH of water at the start of experiment was 7.5 and temperature ranged between 30ºC±1ºC .The Dissolved oxygen was 5ppm. All the fishes reached moribundity or died between 220 minutes with average death time of 211.6 minutes.

The perusal of Table 1 shows that the initial dissolved oxygen was 5ppm. The dissolved oxygen at the end of the experiment ranged between 1.7ppm and 2.1ppm. The average lethal oxygen concentration was 1.86ppm.

Total dissolved oxygen consumed was 3.16ppm i.e. 3.92mg/gm fish, @1.112mg oxygen/hr./gm wt. of fish.

Clarias magur

Clarias magur available for the experiment were medium sized and weight ranged between 6.538 gm and 10.896 gm hence large bottles were used (250ml or 350ml or 500ml)

Experiment (Table 1): The first set of experiment was on lines similar to that of other 3 test fish.

The average weight of the fish was 9.23gms and ranged between 6.538 and 10.846gm. Each fish was provided around 250 ml or more of water. The initial pH of the water was 7, temperature range was 27ºC to 28ºC and dissolved oxygen 7.0ppm.

A perusal of table 3 shows that the *Clarias magur* died faster than *Gambusia affinis* with mean death time much lower than i.e. only 95 minutes (range 65- 120 minutes). The most striking fact was that this fish had a very a high Lethal dissolved oxygen value with an average of 4.00ppm; the least lethal concentration for a fish was 3.8ppm. The total oxygen consumed was 3ppm i.e. 2.25 mg i.e. 0.081 mg/gm at the rate of 0.51 mg/gm fish/hr.

Xiphophorus clemenciae

Experiment (Table 1): The experiment with *Xiphophorus clemenciae* (Yellow sword tail) was conducted with 9 fishes divided into 3 lots of 3 fishes each. The range of weight was 0.17 gm to 0.420gms, with an average of 0.327gms. The length of fishes ranged between 2.6 and 3.5cms. The temperature remained at 25-27ºC and pH was 7. The fishes reached the moribundity in 172-215minutes, with an average of 181.66 minutes (2 hours 2 minutes)

The dissolved oxygen at the beginning of the experiment was 6ppm but at the time of moribundity it ranged between 3.6-4.2ppm with an average of 3.86ppm.

A perusal of table 3 shows that the rate of oxygen consumption in case of *Xiphophorus clemenciae* was 0.0993mg of oxygen/gm body weight/hour (Table 2).

Labeo rohita

The experiment (Table 1) with *Labeo rohita* fingerlings was performed with sample of 9 fishes divided into 3 lots of 3 fishes each. Each set of 3 fishes was kept in a glass bottle of 250 ml capacity. The initial temperature of the water was 29ºC and the pH was 7. The average weight of *Labeo rohita* fingerlings was 1.56gms with a range 0.539 to 2.539. The maximum and minimum length of test fish was 6.5cm and 3.7 cm respectively. The initial dissolved oxygen was 9ppm and total dissolved oxygen consumed was 4.05ppm.

The moribundity was achieved in 70-90 minutes with an average of 80 minutes (1 hour and 20 min.) the dissolved oxygen being zero in all samples. The rate of oxygen consumption in case of *Labeo rohita* was 5.06mg/gm of body weight/hour (Table 3).

The experiments were carried out 30 times and the average readings have been presented in the paper.

Table 1

Length, Weight, Mortality Time for the Four Test Fishes

S.No	Name of fish	Length of fish in cm		Weight of fish in gm		Mortality time limits in minutes	
		Range	Average	Range	Average	Range	Average
1	*Gambusia affinis*	1.5-2.7	2.23	0.03-0.171	0.117	190-215	211.6
2	*Clarias magur*	10-12.8	11.1	6.538-10.846	9.233	65-120	95
3	*Xiphophorus clmenciae*	2.5-3.5	2.93	0.170-0.420	0.327	170-215	196.6
4	*Labeo rohita*	3.7-6.5	5.31	0.530-2.539	1.56	70-90	80

Table 2

pH, Temperture and Rate of O_2 Consumption in Four Test Fishes

S.No	Name of Fish	Initial pH	Final pH	Initial Time	Final Time	Initial DO	Final DO
1	*Gambusia affinis*	7.5	6	30	29	5	1.86
2	*Clarias magur*	7	6	28	27	7	4
3	*Xiphophorus clmenciae*	7	3.5	27	25	6	3.8
4	*Labeo rohita*	7	6	26	25	9	0

Table 3

Rate of O_2 Consumption of the Four Test Fish

S.No.	Name of fish	Total dissolved oxygen consumed in ppm	Average weight of O_2 consumed in mg	Average rate of O_2 consumed mg/gm fish	Average of O_2 consumed mg/gm fish/ hr
1	*Gambusia affinis*	3.16	0.153	3.92	1.112
2	*Clarias magur*	2.25	0.249	0.081	0.051
3	*Xiphophorus clmenciae*	1.73	0.106	0.0163	0.0993
4	*Labeo rohita*	4.05	0.749	6.75	5.06

DISCUSSION

Large number of fishes have been subjected to experimentation for finding out the effect of lethal and sub-lethal concentration of dissolved oxygen. (Salmo, Trout and Char-Albaster and Lloyd, 1980; Trout and small mouth bass – Burdick et al., 1954; channel cat fish – Andrew's et al., 1978; Seven fresh water fish species of India – Singh, G. and Virdi, G.S., 1983). In the present study the four fishes selected for finding out of the lethal oxygen concentration belonged to four different categories.

Gambusia affinis is a small fresh water pond and lake dweller feeding on various aquatic larvae and is highly valued for its ability to kill mosquito larvae. It is a warm water fish and known to be a hardy one. The current experimental data clearly indicate that this small fish has a moderately high rate of dissolved oxygen consumption (1.112 and 0.193 mg/gm fish/hr).

The *Clarias magur* is another pond dwelling fish of warm waters rich in organic matter and can survive long periods of low dissolved oxygen concentration. This ability of the fish is primarily because of the fact that it can breath atmospheric air with the help of accessory air breathing organs. The fish died on an average 95 minutes when kept in air tight water containing bottles and the dissolved oxygen consumed was much less as compared to other fishes like *Gambusia* and *Labeo* (Table 1). The same fish when given only 40% atmospheric air above the water was able to live for 25 hrs (1500 minutes) and consumed more oxygen to deplete it to 0.8 ppm. The two experiments with *Clarias* show that the rate of consumption was very low (0.05 mg/gm fish/hr) when fish was unable to access the atmospheric air because it is an air breathing fish. The *Xiphophorus clemenciae* is a valuable aquarium fish, commonly known as yellow sword tail primarily it is also a fish of clean warm waters but is an active mid water dweller. It is a small fish with relatively high rate of oxygen requirement (0.0993 mg/gm fish/hr) in the experiments.

Labeo rohita is one of the most valuable fresh water carp. It is although running water breeder it is stocked in a lower mid water dweller and very fast rate of growth and very agile habit. Its oxygen requirement is appropriately very high consuming the total oxygen on lowest time before its death. The rate of O_2 consumption is as high as 5.06 mg/gm/hr. This is highest amongst all the four test fishes including *Clarias.*

It can be summarized that the small fishes like *Gambusia* and *Xiphophorus* had a larger survival time, whereas *Labeo* and *Clarias* died faster.

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