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RESPIRATORY STRESS OF SALINITY ON *Oreochromis niloticus*

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AUTHORS' CONTRIBUTIONS

This work was carried out in collaboration among all authors. Author RSM managed the analyses of the study and drafted the manuscript. Author AUA designed the study and wrote protocol. Author GH carried out the experiments. Authors SS, RR and BR managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Oreochromis niloticus (Nile tilapia) commonly found in the freshwater bodies of Kerala was used in this study as the experimental fish so as to determine the effect of salinity on the respiration rate of the fish. The fishes were exposed to different salinities (5ppt, 10ppt, 15ppt, 20ppt) for a period of one hour. The sample with 0ppt salinity was taken as the control. Using Winkler's method, the dissolved oxygen level in the water taken at different intervals (5th minutes, 15th minutes, 30th minutes and 1 hour) from the sample solutions were estimated. In control (0ppt) average oxygen consumption during different time interval was found to be stable at 0.12 mg/ml/g body weight. The dissolved oxygen consumption by fish increased with increasing salinities from 0ppt to 10ppt, then decreased in 15ppt and 20ppt, besides this consumption of Oxygen decreased from 5th minutes to 60th minutes of exposure. The opercular beats of the fish was noted and it was found that in the control (with 0 ppt salinity), the rate of opercular beats was quite steady without a huge rise or fall and the average value noted was 122/minutes. In all other salinities (5, 10, 15 and 20 ppt), the opercular beats was decreased from 1st minute to 60th minute. The rate of opercular beats was lower in the control when compared with 5ppt,10ppt, 15ppt and 20ppt.Even though *Oreochromis niloticus* (Nile tilapia) is very sturdy fish, and tides over stressful environment conditions, salinity changes in this experimental setup caused changes in the respiration rate of the fish. So this study discloses how other less sturdy aquatic fauna could easily succumb to salinity change in their environment.

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1. INTRODUCTION

Terrestrial animals and plants are concerned with the quantity of oxygen present in the air, similarly all aquatic organisms also depend on oxygen dissolved in water. As we know, solubility of any gas in water depends on the temperature of water and its partial pressure; and oxygen being poorly soluble in water, the solubility of oxygen decreases with increasing salinity of water [1].

The survival of fish depends on its ability to extract the oxygen dissolved in water and transfer it to their blood stream. Extracting oxygen from water is more difficult and requires a greater expenditure of energy than does extracting oxygen from air. Water is thousand times denser than air and at 20°C it has 50 times more viscosity than air and contains only 3% as much oxygen as an equal volume of air. Fishes, therefore, have necessarily evolved very efficient systems for extracting oxygen from water; some fishes are able to extract as much as 80% of oxygen contained in the water passing over the gills, where humans can extract only about 25% of the oxygen from the air taken into the lungs. This is done by highly vascularized respiratory organs like gills, lungs, specialized chambers or skin itself [1].

Stress is any condition that causes physical or mental discomfort that results in the release of stress-related hormones or results in specific physiological responses. Stress is present in the lives of all living things. Fishes responds to the stressors like temperature changes, presence of pollutants and other chemical toxins, salinity etc [2], by either producing effects that disrupt homeostatic equilibrium or by inducing adaptive behavioural and physiological responses [3].

Salinity represents a critical environmental factor for all aquatic organisms, including fishes [4]. The simplest definition of salinity is that it is a measure of dissolved salts in a concentration of water. There are several salts in sea water, but the most abundant is sodium chloride (NaCl). The average salinity of sea water is 35 parts per thousand.

Tilapia has been referred to as the 'aquatic chicken'. Among the tilapia species, the Nile tilapia, *Oreochromis* sp. is the preferred species for culture as this fish dominates production in freshwater and brackish water ponds and cages. However, it has low tolerance to high salinity levels [5]. The present study is an evaluation of the effect of salinity on the fish *Oreochromis niloticus*. Oxygen consumption has been used as an indirect indicator of metabolism of fish and the energetic cost of ionic and osmotic regulations seems to play a significant role in growth rates. Therefore, it is important to know the oxygen consumption of tilapia in different salinities. Due to its wide range availability and its sturdy nature, *Oreochromis niloticus* (Nile tilapia) was used in this study compared to other fishes.

2. MATERIALS AND METHODS

Salt water collected from Cherai beach was mixed with normal water to achieve the required salinity, determined using portable refractometer and standardized by Mohr's titration method [6]. Experimental fishes were collected from Keezhillam fish farm. Moderate size fishes were selected for the experiment having weight ranging from 7 g to 14 g. The collected fish was acclimatized in laboratory conditions. Opercular frequency was recorded by visual counting intermittently for one minute each at different time interval such as 1st minute, 5th minutes, 15th minutes, 30th minutes and 60th minutes (1 hour) in four treatment groups (5ppt, 10ppt, 15ppt, 20ppt) and control (0ppt) and average values were calculated. In the present study, the respiration rate of the fish was measured for a 120h period. At the end of 120h exposure, each fish was transferred from the test chamber (10 L) to the respiratory chamber (10 L) and then the experiment was run for a period of 1h. After the experiment, the fishes were replaced in their respective test chambers. The same procedure was repeated 6 times. Controls were also run simultaneously to obtain information on the oxygen consumption of the fish in normal state. The dissolved oxygen was estimated with the Winkler's method at 1st minute, 5th minutes, 15th minutes, 30th minutes and 60th minute in 4 treatment groups and control. The oxygen consumed by the fish is expressed as mg/ml/g body weight of the fish. ANOVA was employed to test the significance of difference between the means of control and exposed. Statistical significance was set a priori at alpha >0.05.

3. RESULTS AND DISCUSSION

The opercular beats of the fish was found be quite steady without great fluctuations in the control group (with 0ppt salinity) (Fig. 1). Whereas, in 5ppt, 10ppt, 15ppt and 20ppt salinity, the opercular beats shows a declining trend from first minute to 60th minute. Average gill movement is decreased as the concentration of salinity increases with that in the control group averaged at 122/minutes (Fig. 2).

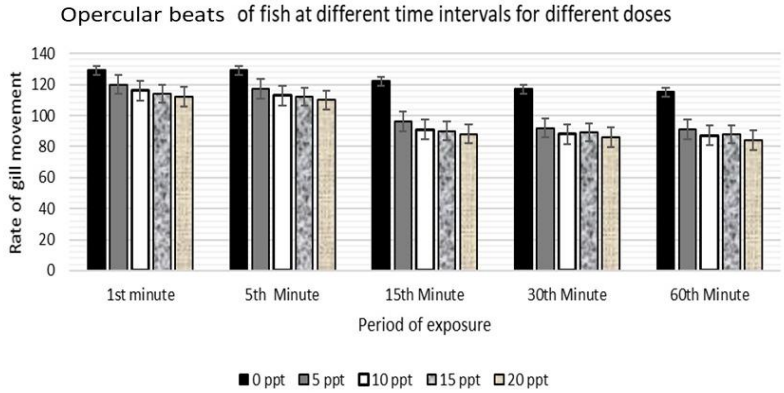

Fig. 1. Showing the opercular beats of fish in 0 ppt, 5 ppt, 10 ppt, 15 ppt and 20 ppt salinity at various time intervals

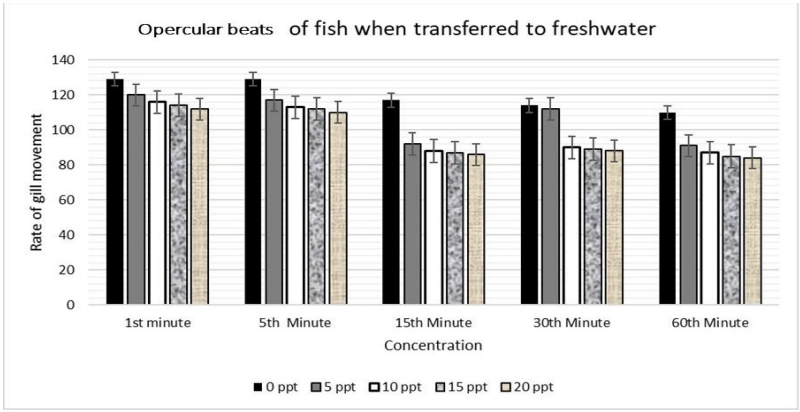

Fig. 2. Showing average opercular beats of fish in different dosages

The opercular beats of the control and treated groups on exposure to freshwater was noted .It has been noted as the concentration of salinity increases the rate of gill movement decreases and reaches a minimum value at 20ppt (Fig. 3).

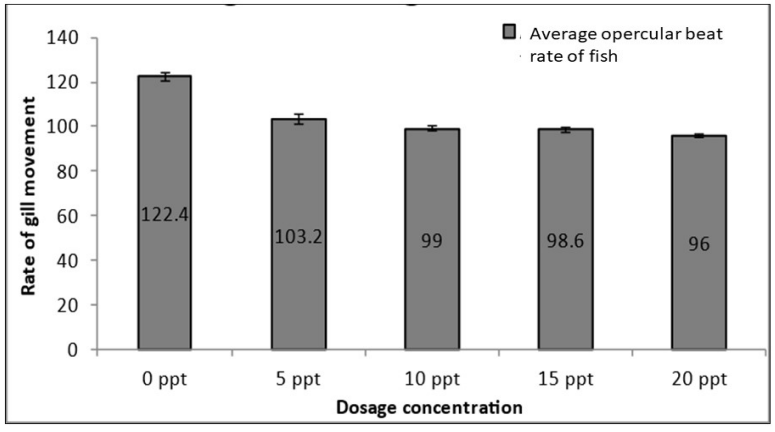

Fig. 3. Showing the opercular beats of fish in different salinities on exposure to freshwater

From the data presented above, it is found that the opercular beats of fish decreased from 5ppt to 20ppt, and the lowest rate of opercular beats was observed in the 20ppt. In all the above salinities, the opercular beats of the fish decreased from first minute to 60th minute.

The opercular movement recorded during the experiment and after the experiment (when transferred to freshwater). The control (0ppt), 15ppt and 20ppt showed a slightly declining trend in the rate of opercular movement. But in the case of 5ppt and 10ppt a decrease was noted in the rate of breathing during the experiment, the rates fluctuated when exposed to freshwater (Table 1).

The opercular beats showed variance at various salinities. In all the salinities, the opercular beats was higher in the initial minute and it decreased in the 5th, 15th, 30th and 60th minute. Though these fluctuation in the opercular beats could not be counted proportional to the changes in metabolic activity, it is definitely an indicator of stress. Increase in ventilation rate is probably due to increased oxygen requirement necessitated by increased salt excretion [7] or increased activity of the fish due to irritation in the ambient water. Though the ventilation rate increased with alternation in the ambient environment, the rate of oxygen uptake showed a decrease. Such decrease in oxygen uptake may be the result of increased water/blood barrier due to the deposition of mucus over the gills. The mucus probably reduces the exchange of gases as also salts and water. These changes could be interpreted as a compensatory mechanism facilitating the acclimation process [8].

The dissolved oxygen consumption by fish at 5th minute, 15th minute, 30th minute and 60th minute was calculated in all the samples (0ppt, 5ppt, 10ppt, 15ppt and 20ppt) (Table 2). In control (0ppt), the dissolved oxygen consumption by fish was 0.12 mg/ml/g body weight at 5th minute, 15th minute, 30th minute and 60th minute. The data obtained (Table 2) reveal that indifferent of the dosage, the time exposure had a negative relation with the amount of oxygen consumed.

From the data presented above, it is found that the dissolved oxygen consumption increased from 5ppt to 10ppt and showed a decrease to 15ppt and 20ppt (Fig. 4) The oxygen consumption was highest in 10ppt and it was lowest in 20ppt and was stable at 0.12 mg/ml/g body weight in the control group.

The oxygen consumption by fish *Oreochromis nilotica* in the study has decreased from 5th minute to 60th minute in all the salt concentrations (5ppt, 10ppt, 15ppt and 20ppt). In fingerlings of tilapia, oxygen consumption at 0, 10, 20 and 30 ppt salinity was found to be 2.14, 0.71,1.43 and 1.42 ml/liter/h respectively. Oxygen consumption rates thereafter decreased with increasing salinity, although the lowest consumption occurred at 20 ppt. Similar results were obtained in earlier studies on different fish [9-13]. Further it was noted that, fish at higher salinity were visibly less active than those at fresh water.

Ion regulatory systems on the gills of euryhaline teleosts are known to undergo physiological and morphological restructuring in response to altered salinity, including adjustments of the activity and protein abundance of Na+/K+-ATPase [14-15]. This reorganization must incur an energetic cost [16] and studies of the time-course of these changes show an initial adjustment phase (3–12 h) with increased Na+/K+-ATPase activity followed by a regulatory phase (48–96 h) with increased in Na+/K+-ATPase mRNA and protein abundance [17]. According to [18] the decrease in oxygen uptake at progressively higher salinities could be attributed to the decreased thyroid activity and dehydration of blood as a result of osmotic imbalance. The studies of Md. Abdul Awal et al. [19] and Ron et al. [13] showed that the change in oxygen consumption rate is linked to a decrease in the metabolic rate, suggesting that lower consumption at high salinities might be a result of reduced activity, which in itself was salinity modulated.

Table 1. Showing the opercular beats of fish during and after the experiment at various concentrations and time intervals

opercular beats during different salinity and different time intervals	Time interval	0 ppt	5 ppt	10 ppt	15 ppt	20 ppt
	1st Minute	129	120	116	114	112
	5th Minute	129	117	113	112	110
	15th Minute	122	96	91	90	88
	30th Minute	117	92	88	89	86
	60th Minute	115	91	87	88	84

P-value= 0.0246 (ANOVA), hence significant

Table 2. Showing the oxygen consumed by fish during the experiment at various time intervals

Time interval	Oxygen consumption (mg/ml/g body weight)				
	0ppt (control)	5ppt	10ppt	15ppt	20ppt
5th Minute	0.12	0.39	0.4	0.121	0.11
15th Minute	0.12	0.23	0.25	0.11	0.1
30th Minute	0.12	0.166	0.17	0.094	0.09
60th Minute	0.12	0.132	0.14	0.088	0.062

P-value= 0.0246 (ANOVA), hence significant

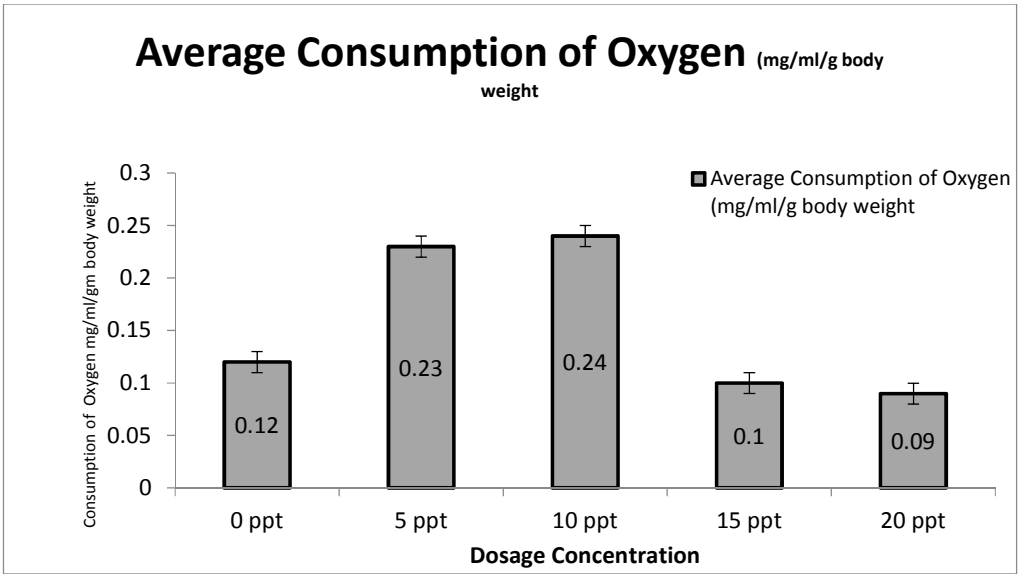

Fig. 4. Average amount of oxygen consumed by the fish at different concentrations

4. CONCLUSION

It was observed in the present study that oxygen consumption by fish in all the salinities decreases from 5 minute to 30 minute and later increases after 1 hour. The opercular ventilation rate of the fish also decreases from initial minute to 30th minute and later it slightly increases at 60th minute. It was also found that the oxygen consumption decreases with increasing salinity from 5 ppt to 20 ppt, although the lowest consumption occurs at the 15 ppt. This experimental study gives a good idea about some of the effects that salinity could have on fishes and other higher or even lower organisms of the aquatic environment.

The results of the study it is reveals that more research could be conducted in this direction. The specific effects of salinity alone as a stressor as well as the effect of salinity on the toxicity of different types of chemicals on the survival of different species of fish can be envisaged. So this study can be considered as the base line study in this topic and further research can be conducted in the same area with sophisticated techniques and purified methods. The current study can be elaborated with much better laboratory facilities, time and more utilization of scientific knowledge.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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