



Assessment of Antioxidant Status of *Oreochromis mossambicus* in Fresh and Brackish Water of Thiruvarur District, Tamil Nadu from India

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

Antioxidant biomarkers have been used to demonstrate the effects of various environmental stressors on certain aquatic organisms. The objective of the study is to analyze the antioxidant status of freshwater and brackish fish such as *Oreochromis mossambicus* in 2022. Antioxidant enzymes superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx), glutathione reductase, glutathione S transferase (GST) and Glutathione content (GSH) was with gills, and Livers and muscles of fresh water fish of *Oreochromis mossambicus* analyzed. The results of the study showed that superoxide dismutase, catalase, glutathione peroxidase, glutathione reductase,

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glutathione S transferase and glutathione contained in different tissues (gills, gills and various tissues) Antioxidant enzymes such as glutathione content have been demonstrated and the liver and muscles have been reduced in *Oreochromis mossambicus*. The decreased activities of these enzymes may be due to contaminant exposure. This modulation in enzyme activity could be due to the mechanism by which Tilapia *Oreochromis mossambicus* circumvents the salinity stress caused by contaminants. The research will provide more detailed information on the response of antioxidant defenses to tilapia species in dark water environments and the mechanisms of regulation of this response. This future research will undoubtedly benefit some aspects of aquaculture and aquaculture production as well.

Keywords: *Oreochromis mossambicus*; antioxidants; freshwater; brackish water; pollutants.

1. INTRODUCTION

Tilapia inhabit a variety of freshwater habitats. Traditionally, they were very important in small-scale commercial and subsistence fishing around the world, especially in Africa and Asia. It is the third most common farmed fish after the family of carp and salmon [1]. World production has been strongly influenced by the rapid expansion of Mozambique Tilapia (*Oreochromis mossambicus*), which is grown in China, the Philippines, India and Egypt [2]. Tilapia fish are nutritious, with less fat (0.5 - 3.0%) and more protein (16 - 25%) forming a healthy part of a balanced diet. %) and [3] which replace seafood recipes well.

Oxidative stress is an imbalance between the production of reactive oxygen species (ROS) and the body's antioxidant defense capabilities. Various antioxidant compounds have been used to combat free radicals generated by the oxidation process. Biological oxidation is a primitive process, and given the inevitable consequences of O₂ toxicity, evolution has provided the appropriate defense strategies. As more complex aerobic life forms developed, diversified and adapted the development of antioxidant defense systems to new situations. The first line of defense is the use of antioxidants such as vitamin C, vitamin E, uric acid, glutathione and carotenoids. In addition, the various antioxidant enzymes prevent the cascade of oxidation reactions, block and inactivate reactive oxygen intermediates in order to close the lipid peroxidation cycle [4].

Antioxidants are important in countering oxygen toxicity when the supply of other antioxidants is insufficient or depleted [5]. Antioxidants and enzymes form the so-called primary antioxidants [6]. Like all aerobic organisms, fish are susceptible to attack by reactive oxygen species and have developed an antioxidant defense

system, which is known to be highly potent, which was mainly proven by research from the 1980s. Special adaptive enzymes such as superoxide dismutase, catalase, glutathione peroxidase, and glutathione reductase, Glutathione S transferase and glutathione have been detected in most fish species examined to date [7,8]. In this study, the antioxidant status of freshwater and brackish fish under the name *Oreochromis mossambicus* was examined.

2. MATERIALS AND METHODS

2.1 Collection Fishes

Fishes were weighing (370.50 ± 34.70 g) and (25.32 ± 4.12 cm) long *Oreochromis mossambicus*, a brackish water and fresh water environment 5 to 8 months old in the Indian district of Thiruvavur, Tamil Nadu. Fish were collected from brackish and freshwater environments using an experimental network with multiple plates and mother filaments. This technique is known as 'non-selective' fishing technique and uses nets with different mesh sizes of 16w150mm telescopic net, so that the sample represents the fish population. The network was set to U800 0600 h overnight for about 12 hours and removed the next day. The fish were then removed from different panels and divided into separate trays depending on the type and size. Individual *Oreochromis mossambicus* fish were identified and collected from the research area and transported to the laboratory for biochemical analysis.

2.2 Sample Collection

Up to 10 fish were collected with the weight of, their tissues were synthesized for analysis. The collected fish must be placed in a clean, labeled polyethylene bag, which is handled by the staff with latex gloves. Fish must also be dissected with a thin 3 hour catch.

2.3 Preparation of Homogenate

Muscle and liver Samples of fish analysis were taken from the left side of each fish, the meat was dissected and washed with ice-cold saline. The 1 g fabric was weighed and homogenized using a Teflon homogenizer. Tissue homogenates were produced in a 0.1 m Tris Hcl buffer (pH 7.4)) and used to estimate various biochemical parameters.

2.4 Antioxidants

The superoxide dismutase activity is [9], analyzed according to the method of Kakkar et al. The activity of the catalase was determined by the methods of beer and sieve [10]. The glutathione reduction was developed according to the method of Moron et al. [11] determined. The activity of glutathione peroxidase was determined using the estimation of Rotruck et al. [12]. GST is developed by Habig et al. [13] analyzed. The activity of glutathione reductase was determined using Staal et al. [14] measured.

2.5 Statistical Analysis

The value is expressed as the average $\pm$ SD of 10 fish. The data were calculated by the student's t-test (independent sample, P-value 2 tail) using MS-excel. 2013. Statistically significant level 0. 05.

3. RESULTS

The activities of SOD, GPX, GR, CAT, GST and the content of GSH in the gill of fish from fresh and brackish water are presented in Table 1. The SOD activity of freshwater fish was

27.64 $\pm$ 1.43U while brackish water fish was 31.47 $\pm$ 1.09 U. The CAT activity of freshwater fish was 5.32 $\pm$ 0.34 μ mol while brackish water fish was 7.05 $\pm$ 0.25 μ mol. The GPX activity of freshwater fish was 0.27 $\pm$ 0.04 μ mol while brackish water fish was 0.35 $\pm$ 0.02 μ mol. The GST activity of freshwater fish was 218.43 $\pm$ 2.36 μ mol while brackish water fish was 231.07 $\pm$ 2.94 μ mol. The GR activity of freshwater fish was 0.03 $\pm$ 0.01 μ mol while brackish water fish was 0.07 $\pm$ 0.01 μ mol. The GSH content of freshwater fish was 0.11 $\pm$ 0.01nmole while brackish water fish was 0.08 $\pm$ 0.01nmole. The fishes showed a significant (P < 0.01) increase in activity of all the five enzymes and GSH content in gill of brackish water fish as compared with freshwater fish.

The activities of SOD, GPX, GR, CAT, GST and the content of GSH in the liver of fish from fresh and brackish water are presented in Table 2. The SOD activity of freshwater fish was 51.62 $\pm$ 0.36U while brackish water fish was 57.84 $\pm$ 0.41U. The CAT activity of freshwater fish was 8.54 $\pm$ 0.17 μ mol while brackish water fish was 9.61 $\pm$ 0.23 μ mol. The GPX activity of freshwater fish was 0.43 $\pm$ 0.05 μ mol while brackish water fish was 0.52 $\pm$ 0.04 μ mol. The GST activity of freshwater fish was 251.39 $\pm$ 0.92 μ mol while brackish water fish was 264.25 $\pm$ 1.17 μ mol. The GR activity of freshwater fish was 0.05 $\pm$ 0.01 μ mol while brackish water fish was 0.09 $\pm$ 0.01 μ mol. The GSH content of freshwater fish was 0.13 $\pm$ 0.02nmole while brackish water fish was 0.09 $\pm$ 0.01nmole. The fishes showed a significant (P < 0.01) increase in activity of all the five enzymes and GSH content in the liver of brackish water fish as compared with freshwater fish.

Table 1. Antioxidant status of *Oreochromis mossambicus* gills tissue in fresh and brackish water (N =10)

Antioxidant	Unit	<i>Oreochromis mossambicus</i> gills tissue	
		Freshwater	Brackish water
SOD	U / mg protein in tissues	27.64 $\pm$ 1.43	31.47 $\pm$ 1.09*
Cat.	μ mol H ₂ O ₂ / mg prot./min in tissue	5.32 $\pm$ 0.34	7.05 $\pm$ 0.25*
GPx	μ mol / mg prot./min	0.27 $\pm$ 0.04	0.35 $\pm$ 0.02*
GST	μ mole / mg prot./min	218.43 $\pm$ 2.36	231.07 $\pm$ 2.94*
GR	μ mole / mg prot./min	0.03 $\pm$ 0.01	0.07 $\pm$ 0.01*
GSH	nmol / mg protein in tissues	0.12 $\pm$ 0.02	0.08 $\pm$ 0.01*

The value is expressed as the average $\pm$ SD of 10 fish. The data were calculated by the student's t-test (independent sample, P-value 2 tail) using MS-excel. 2013. Statistically significant level 0. 05.

Table 2. Antioxidant status of *Oreochromis mossambicus* liver tissue in in fresh and brackish water (N =10)

Antioxidant	Unit	<i>Oreochromis mossambicus</i> liver tissue	
		Freshwater	Brackish water
SOD	U / mg protein in tissues	51.62±0.36	57.84±0.41*
Cat.	µmol H ₂ O ₂ / mg prot./min in tissue	8.54±0.17	9.61±0.23*
GPx	µmol / mg prot./min	0.43±0.05	0.52±0.04*
GST	µmole / mg prot./min	251.39±0.92	264.25±1.17*
GR	µmole / mg prot./min	0.05±0.01	0.09±0.01*
GSH	nmol / mg protein in tissues	0.13±0.02	0.09±0.01*

The value is expressed as the average ± SD of 10 fish. The data were calculated by the student's t-test (independent sample, P-value 2 tail) using MS-excel. 2013. Statistically significant level 0. 05.

Table 3. Antioxidant status of *Oreochromis mossambicus* muscles tissue in in fresh and brackish water (N =10)

Antioxidant	Unit	<i>Oreochromis mossambicus</i> muscles tissue	
		Freshwater	Brackish water
SOD		13.27±0.94	16.54±0.85*
Cat.	U / mg protein in tissues	3.15±0.63	5.34±0.71*
GPx	µmol H ₂ O ₂ / mg prot./min in tissue	0.14±0.01	0.17±0.01*
GST	µmol / mg prot./min	185.51±3.25	194.49±2.19*
GR	µmole / mg prot./min	0.02±0.01	0.05±0.01*
GSH	µmole / mg prot./min	0.07±0.02	0.03±0.01*

The value is expressed as the average ± SD of 10 fish. The data were calculated by the student's t-test (independent sample, P-value 2 tail) using MS-excel. 2013. Statistically significant level 0. 05.

The activities of SOD, GPX, GR, CAT, GST and the content of GSH in the muscle of fish from fresh and brackish water are presented in Table 3. The SOD activity of freshwater fish was 13.27±0.94U while brackish water fish was 16.54±0.85U. The CAT activity of freshwater fish was 3.15±0.63µmol while brackish water fish was 5.34±0.71µmol. The GPX activity of freshwater fish was 0.14±0.01µmol while brackish water fish was 0.17±0.01µmol. The GST activity of freshwater fish was 185.51±3.25µmol while brackish water fish was 194.49±2.19µmol. The GR activity of freshwater fish was 0.02±0.01µmol while brackish water fish was 0.05±0.01µmol. The GSH content of freshwater fish was 0.07±0.02nmole while brackish water fish was 0.03±0.01nmole. The fishes showed a significant ($P < 0.01$) increase in activity of all the five enzymes and GSH content in the muscle of brackish water fish as compared with freshwater fish.

4. DISCUSSION

The natural aquatic system was [15] the final recipient of the pollutant. The aquatic ecosystem contaminated by a variety of pollutants has been a problem in recent decades [16]. Change in the

geological matrix or accumulation and persistence of pollutants and toxic substances due to pollutants emitted by anthropogenic sources such as industrial wastewater and mining waste, [17] has become a major threat to biological life. Aquatic animals were the keystone species of many ecosystems [18]. Fish are among the most common organisms in aquatic ecosystems and reflect the biological effects of pollution. Contamination of aquatic systems has attracted the attention of researchers around the world. Many environmental pollutants or their metabolites can cause oxidative stress in aquatic organisms, including fish [19]. It is known that the release of pollutants into the aquatic environment has a negative impact on the environment and organisms, This gives great interest in examining the reaction to oxidative stress in aquatic organisms [20].

Antioxidant biomarkers have been used to demonstrate the effects of various environmental stressors in certain aquatic organisms [21]. Chemical pollution usually occurs in aquatic environments and can significantly affect aquatic species such as fish. [22]. Chemical residues in aquatic ecosystems and their effects on fish have been investigated using several biomarkers [23].

Fish exposure to various pollutants in the aquatic ecosystem is known to result in an excess of reactive oxygen species (ROS), including, This can adversely affect cell macromolecules [24]. Fish have a complex antioxidant system, including superoxide dismutase, catalase, glutathione S transferase and more, glutathione reductase and glutathione peroxidase, Oxidative damage to reactive oxygen species (ROS) has occurred. These antioxidants protect cellular components from oxidative damage caused by ROS. The imbalance of ROS production and neutralization by these antioxidant mechanisms in vivo is known as oxidative stress. This has become an important topic for the study of terrestrial and aquatic toxicity [25]. In this study, antioxidant enzymes such as GST, CAT, SOD, GPX and GR are increased in the gills, liver and muscles of brackish water bodies in relation to freshwater fish.

Antioxidative defense enzymes have been proposed as biomarkers for oxidative stress, which are transmitted by contaminants in various organisms in sea, fresh water and brackish water, and their induction reflects specific reactions to pollutants [26]. Fish cells excrete antioxidant enzymes to reduce the effect of ROS on cell function [27]. Antioxidative defense enzymes are also associated with changes in environmental factors such as temperature, salinity, food availability, dissolved oxygen levels, etc., as well as intrinsic biological factors such as gonadal development and the reproductive cycle [28]. However, due to the conditions of the brackish water, free radicals accumulate due to the salinity. The results showed that it overcomes the effects of free radicals on brackish water and that fish are beginning to separate enzymes such as SOD, GPX, GR, CAT and GST. The induction of antioxidant enzymes in fish that are exposed to various pollutants can be seen as a biological indicator of oxidative stress [29]. Rueda-jasso et al. [30] reported that the levels of activity of the antioxidant enzymes CAT and SOD in the liver of the fish *Solea senegalensis* were higher.

As the most common non-protein thiol in the body and a pronounced cellular antioxidant, GSH plays an important role in preventing oxidative stress [31]. Based on this study, it can be assumed that the ROS changes in fish caused by brackish water in the medium via the first route of the antioxidant enzyme CAT were in the gills, The liver and muscle tissue, followed by the second track, was GSH. As a result, the use of these two routes significantly reduces the GSH

content in brackish water compared to freshwater fish. This is similar to research by Dudley and Krasen [32], which shows a decrease in GSH in line with a decrease in the activity of GPx, GR and GST enzymes in tissues.

5. CONCLUSION

Based on the results obtained the enzymes as superoxide dismutase, catalase, glutathione peroxidase, glutathione reductase, glutathione S transferase and various tissues (gill, liver and muscle) and is clear that Gil's glutathione content, which has been reduced in the liver and muscles of tilapia due to exposure to pollutants. This regulation of enzyme activity can be attributed to the mechanism by which Tilapia *Oreochromis mossambicus* avoids salt stress caused by impurities. The study of oxidative stress in fish has paved the way for a number of research areas aimed at expanding knowledge about fish physiology and toxicology. In addition, such studies provide more detailed information about the antioxidant defense reaction in tilapia species in a dark water environment and the mechanism for regulating this reaction

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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