CHANGES IN METABOLITES IN THE PLASMA OF BLACK JAW TILAPIA (SAROTHERODON MELANOTHERON) EXPOSED TO DIMETHOATE IN THE LABORATORY

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Abstract

This study evaluated the metabolic activities in Black Jaw Tilapia (Sarotherodon melanotheron) exposed to dimethoate at varied concentrations of 0.00 (control), 0.05, 0.10, 0.15, and 0.20 mg/L in order to determine the amount of metabolic alterations in fish exposed to chemical in aquatic environment. Six water quality indicators were measured during the trial: temperature, pH, salinity, dissolved oxygen, nitrite, and ammonia. A total of 180 of S.melanotheron, with 60 in each of the three categories of fish sizes used in the study: Group 1 consisted of juveniles with a mean length of 12.45cm±1.98SD and a mean weight of 66.23g±3.04SD; Group 2 consisted of sub-adults with a mean length of 15.22cm±3.03SD and a mean weight of 100.34g±11.00SD); and Group 3 consisted of adults with a mean length of 19.04cm±6.09SD and a mean weight of 142.05g±12.54SD. After the experiment, blood samples from the fish were taken, and metabolite profiles were examined in them using normal laboratory techniques. The results of the study demonstrated that urea levels were significantly greater ($P<0.05$) in the exposed fish compared to control values, although creatinine, total bilirubin, and total protein values were significantly lower ($P<0.05$) in the exposed fish. In contrast, the young fish that had been exposed to the toxin showed higher signs of these changes. In the future, comparative studies of metabolic stress in aquatic biota from contaminated coastal habitats and efficient bio-monitoring of the aquatic biota may benefit from the baseline data that this work offers.

Keywords: *Metabolites, Dimethoate, Contaminants, Tilapia, Toxicology.*

INTRODUCTION

When organisms are exposed to extreme concentrations and conditions that cause their mortality, changes in the quality of the water have a negative impact on them [1]. Herbicides used in agriculture and industry might be to blame for this. Heavy chemicals are used by humans to protect crops from insects and rodents, starting at the pre-planting stage and continuing through weed control and crop cultivation and storage, all in an effort to boost agricultural productivity. Fish and other aquatic life as well as people are all harmful to these substances [2, 3]. Any modification in the behaviour of fish provides insight into changes in behaviour that may be connected to physiological indicators in aquatic animals [4]. Because behavioural bioassays are quicker, more sensitive, and more relevant to the environment, they are frequently utilized in toxicity assessments [5]. Pollutant concentrations in water bodies have the potential to significantly increase aquatic habitat mortality. On the other hand, low concentrations cause the contaminants to bioaccumulate and biomagnify, which ultimately reaches humans through the food web [6]. To guarantee that we prioritize eating healthful fish, the problem of water contamination needs to be taken very seriously and addressed with the utmost care [7,8].

Although there are many advantages to using chemicals in farming, there are also significant drawbacks, such as environmental degradation and pollution, which are directly related to the usage of chemicals [9]. The safety of the environment is seriously threatened by agrochemical contamination, and exposure to these chemicals can have harmful health effects like cancer and damage to the neurological system [10]. Due to the use of agricultural chemicals, which pose a serious threat to the ecosystem and all living things and easily contaminate water bodies, causing extensive harm to non-target species, including fish, the presence of these chemicals in ecosystems has become a major cause for concern that causes significant social and scientific anxiety worldwide [11]. Agrochemical pollution of the environment has grown to be a major concern for both human health and wildlife conservation worldwide [12]. Water can get contaminated, whether intentionally or accidentally, by substances that are applied directly into aquatic systems, spray drifts, atmospheric fallout such as dust and rain, sewage, industrial effluent, and rarely, spills.

Unlike other pollutants, pesticides are purposely added to the environment with the goal of using their poisonous qualities to lessen pests and insects that spread illness. Less than 0.1% of insecticides used on intended targets actually reach the pests, while 99.9% end up in different environmental media [13]. Because to torrents, dilution, partitioning in water or air, attachment to sediment particles, accumulation in the tissues of aquatic organisms, or burial in sediment, pesticides in streams eventually disappear [14]. The main method of pesticide transmission from land to aquatic bodies is surface runoff from sporadic rainfall [15], which exposes non-target species like fish to pesticides on a pulse basis. Pulse exposure in lentic (still) systems may be caused by pesticide dissipation, while lotic (flowing) systems may be related to their hydrology (continuous replenishment by water movement) [16]. Pulse exposure to low pesticide concentrations can occur through torrents, dilution, partitioning in water or air, attachment to sediment particles, accumulation in the tissues of aquatic animals, or burial in the sediment [17].

Sub-organism responses in organisms that can show exposure to or the impact of environmental contaminants are known as biomarkers [18]. Among the biomarkers frequently assessed in fish are oxidative damage, haematological changes, biochemical and histological changes, genotoxicity, and mutagenicity. Fish health may be tracked using biomarkers, which can also serve as early warning systems for environmental dangers [19]. The biochemical reactions of *S. melanotheron* to pesticides remain poorly understood, despite over 20 years of research on aquatic creatures exposed to pesticides or other toxicants. The effects of these animals on the environment and economy are extensive. There is not much information in the literature about dimethoate's impact on *S.melanotheron* metabolites. As a result, this study evaluated the metabolic reactions of *S. meleanotheron* subjected to various lab-based dimethoate concentrations.

MATERIALS AND METHODS

Experimental Location and Fish

The study was conducted at the African Regional Aquaculture Center in Buguma, Rivers State, Nigeria, which is a branch office of the Nigerian Institute for Oceanography and Marine Research. During low tide, a total of 180 *S. melanotheron*, used for the experiment were sourced from the recruitment ponds in the centre. They were later sorted and grouped into three based on their sizes. Group 1(Juveniles) were of the size (mean length 12.45cm±1.98SD and mean weight 66.23g±3.04SD), Group 2 (Sub-Adults) were of the size (mean length 15.22cm±3.03SD and mean weight 100.34g±11.00SD) and Group 3 (Adults) were of the size (mean length 19.04cm±6.09SD and mean weight 142.05g±12.54SD). The fish were brought to the lab in six open, 50-liter plastic containers, where they acclimated for seven days.

Preparation of Test Solutions and Exposure of Fish

In the present study, dimethoate was used. Dimethoate is a white crystalline solid, with a camphor-like odor, white to grayish crystals for technical product. This material is a contact and systemic organophosphate insecticide effective against a broad range of insects and mites when applied on a wide range of crops, was used to make the stock solutions. The pesticide was purchased from a commercial outlet in Port Harcourt, Nigeria. *S.melanotherom* were exposed to the chemical at the concentrations of 0.00 (control), 0.05, 0.10, 0.15, and 0.20 mg/L in triplicates. Five fish were randomly distributed into each test tank. The experiment lasted for a period of 15 days. The water in the tanks was renewed daily. The fish were fed twice daily at 3% body weight with a commercial feed

Determination of blood Plasma Metabolites

A 2ml sample of fresh blood was taken at the conclusion of each experimental period by puncturing the caudal artery with a tiny needle and pouring the sample into heparinized sample vials. Serum was separated by centrifugation in a TG20-WS Tabletop High Speed Laboratory Centrifuge for 5-8 minutes at 10,000 rpm. Following the guidelines provided by APHA [20], the samples were examined for the metabolites creatinine, total bilirubin, total urea, and total protein. There were three copies of each test run. The methods APHA[20] were also used to determine water quality parameters.

Statistical Analysis

The mean and standard deviation of the mean were used to express all the data. The data analysis was done using SPSS Version 22, a statistical program. Using two-way ANOVA, the means were split, and the two means were deemed significant at 5% ($P < 0.05$).

RESULTS

The parameters of water quality (Table 1) were all within the same range, with the exception of DO, where lower values were recorded at larger chemical concentrations. Table 2 shows how the chemical dimethoate affected the metabolites in the plasma of juvenile *S.melanotheron*. With rising dimethoate concentrations, it was seen that the values of creatinine, total protein, and total bilirubin levels dropped. When compared to the control values, while the values of urea considerably increased. Additionally, Table 3 shows how the chemical dimethoate affected the metabolites in the plasma of sub-adult sizes of *S.melanotheron.* With rising dimethoate concentrations, it was seen that creatinine, total protein, and total bilirubin levels dropped. When compared to the control values, however, the values of urea considerably increased.

Table 1: Physico-chemical Parameters of Water in Experimental Tanks (Mean± SD)

Parameters	Concentrations of Dimethoate (mg/L)				
	0.00	**0.05**	**0.10**	**0.15**	**020**
Temperature (^{0}C)	28.87±1.77^a	28.91±1.82^a	28.71±1.66^a	28.44±1.46^a	28.89±1.87^a
pH	6.65±1.11^a	6.67±1.07^a	6.66±1.88^a	6.67±1.02^a	6.04±1.77^a
Ammonia (mg/l)	0.17±0.01^a	0.38±0.01^{ab}	0.44±0.17^b	0.49±0.08^b	0.58±0.45^c
DO (mg/l)	6.65±0.07^c	6.40±0.57^c	5.50±0.69^b	4.05±0.33^b	3.79±0.55^a
Nitrite (mg/l)	0.02±0.01^a	0.06±0.01^b	0.07±0.01^b	0.08±0.01^b	0.13±0.02^c
Salinity (ppt)	11.45±1.04^a	11.45±3.89^a	11.47±1.03^a	11.46±3.67^a	11.47$\pm$2.04^a

Means within the row with different superscripts are significantly different (P<0.05)

Table2: Metabolite Activities in Juveniles of *S. melanotheron* Exposed to Dimethoate in the Laboratory

Concentration	Metabolites (mg/dl)			
	Creatinine	**Urea**	**Total Bilurubin**	**Total Protein**
0.00	80.01±1.02 ^{c}	2.11±0.53 ^{a}	10.43±1.11 ^{c}	24.99±1.76 ^{c}
0.05	70.44±5.11 ^{b}	3.01±0.66 ^{a}	10.00±1.02 ^{c}	18.02±1.04 ^{b}
0.10	65.03±5.03 ^{b}	4.66±1.23 ^{b}	8.12±1.88 ^{b}	15.03±1.01 ^{a}
0.15	58.00±6.02 ^{a}	5.98±1.01 ^{b}	7.02±0.33 ^{b}	14.03±1.02 ^{a}
0.20	50.22±8.42 ^{a}	6.68±1.33 ^{b}	4.04±0.65 ^{a}	12.01±1.51 ^{a}

Means within the same row with different super scripts are significantly different (P<0.05)

Table3: Metabolite Activities in Sub-Adults of *S. melanotheron* Exposed to Dimethoate in the Laboratory

Concentration	Metabolites (mg/dl)			
	Creatinine	**Urea**	**Total Bilurubin**	**Total Protein**
0.00	90.44±4.99 ^{c}	5.01±0.12 ^{a}	22.02±1.77 ^{c}	35.01±3.77 ^{c}
0.05	82.11±8.01 ^{b}	4.77±0.88 ^{a}	21.72±1.87 ^{c}	29.02±8.01 ^{b}
0.10	72.22±6.98 ^{b}	4.32±1.02 ^{b}	14.01±1.44 ^{b}	24.55±6.87 ^{b}
0.15	68.29±7.19 ^{a}	6.08±1.77 ^{b}	13.02±0.88 ^{b}	21.73±3.43 ^{b}
0.20	56.77±3.78 ^{a}	7.88±1.99 ^{b}	11.77±0.99 ^{a}	17.03±1.45 ^{a}

Means within the same row with different super scripts are significantly different (P<0.05)

Table4: Metabolite Activities in Adults of *S. melanotheron* Exposed to Dimethoate in the Laboratory

Concentration	Metabolites (mg/dl)			
	Creatinine	**Urea**	**Total Bilurubin**	**Total Protein**
0.00	94.01±2.01 ^{c}	5.02±0.07 ^{a}	22.11±1.03 ^{b}	35.03±3.63 ^{c}
0.05	80.55±9.12 ^{b}	5.89±0.17 ^{a}	20.03±1.77 ^{b}	29.51±1.03 ^{b}
0.10	72.99±8.19 ^{b}	6.87±1.04 ^{b}	16.12±1.09^a	26.11±6.54 ^{b}
0.15	60.01±4.88 ^{b}	6.98±1.33 ^{a}	12.78±0.44 ^{a}	23.33±3.03 ^{b}
0.20	55.02±4.03 ^{a}	7.92±1.09 ^{c}	10.88±0.91 ^{a}	20.01±1.47 ^{a}

Means within the same row with different super scripts are significantly different (P<0.05)

DISCUSSION

In general, changes in metabolites may arise from the influence of water quality characteristics, particularly in hazardous water. In this investigation, the metabolites' changes and fluctuations following the chemical's administration are unrelated to the water utilized in the experiment. This is due to the fact that there was no discernible difference (P>0.05) in temperature, salinity, pH, ammonia, and nitrite between the solution's varied concentrations and the control. On the other hand, the dissolved oxygen readings showed decreased values. The trend in the water quality indicators is comparable to the findings of earlier writers who exposed fish to different concentrations of chemicals in a salinity chamber [21]. The primary function of blood or plasma is to carry waste products to different excretory organs so they can be expelled from the body, as well as absorbed metabolites and nutrients (both organic and inorganic) across the body[22].One crucial clinical correlate is the presence of metabolites in the blood, whether at high or low concentrations. As the chemical solution's concentration rose, the readings of total bilirubin decreased. Damage to the liver cells is indicated by a drop in plasma total bilirubin [23]. Lower bilirubin levels in this investigation suggest that S.melanotheron's liver cells may have suffered harm. The inability of the liver to convert bilirubin into bile and urobilin, a condition that gives human urine its yellow color, may also be the cause of a drop in total bilirubin concentrations in the plasma [24].

Using a range of both in-vivo and in-vitro techniques, urea and creatinine have been utilized as significant markers for the assessment of the effects of stress on the kidney [25]. The experimental fish's creatinine decreased during the course of the exposure period, while its urea levels rose as the chemical solution's concentration did. According to Calbreath [26], there was a decrease in glomerular filtration rate and an increase in urea content, indicating that the kidney was unable to eliminate these waste products. Total protein content is a crucial non-specific immunological measure and is utilized as a fundamental indicator of fish health [27]. In the current investigation, the experimental fishes' serum protein levels decreased. Reduced amino transferase activity, altered liver structure, and poor fluid balance regulation can all contribute to a decrease in total protein levels. This outcome supports Anyanwu et al.'s [28] findings on *Sarotherodon melanotheron* exposure to different salinities. The reduced or perturbed production of microsomal proteins was most likely the cause of the protein content drop. The breakdown of proteins raises the possibility of increased proteolytic activity and the potential use of the result for metabolic purposes [29].An increase in protein content may be linked to an increase in protein synthesis because of an increase in the enzyme activity involved in protein synthesis, whereas an occasional reduction in protein content may be caused by a decrease in protein synthesis and an increase in proteolysis [30]. Nonetheless, researchers have noted a drop in fish protein content after stress.

CONCLUSION

The results of this investigation have demonstrated that various dimethoate concentrations have a substantial impact on the metabolites of *S.melanotheron*, with this effect being particularly noticeable in young fish. This is consistent with other research showing that stress can modify the internal functioning of fish organs, particularly the liver. In summary, lower total protein levels observed in this study indicate either decreased protein synthesis or protein loss through excretion, both of which are indicative of problems with the kidneys. A drop in creatinine levels in fish exposed to the toxin suggests the fish are under stress. Increased urea is an indication that the kidneys are unable to get rid of excess waste. It is possible that the liver was unharmed by the toxin if fish exposed to it showed variations in total bilirubin. The results of this study suggest that the amounts of total protein, total bilirubin, creatinine, and urea in the probe organism's plasma could serve as useful indicators of dimethoate's sublethal effects on aquatic organisms.

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