



# International Journal of Fisheries and Aquatic Studies

E-ISSN: 2347-5129

P-ISSN: 2394-0506

(ICV-Poland) Impact Value: 5.62

(GIF) Impact Factor: 0.549

IJFAS 2018; 6(4): 81-86

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www.fisheriesjournal.com

Received: 23-05-2018

Accepted: 25-06-2018

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## Changes in the biochemical profile of *Anabas testudineus* Bloch 1792 on exposure to aquatic toxicants of Buckingham canal, Chennai, Tamil Nadu, India

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### Abstract

Toxicants affect biologically active molecules, viz., carbohydrates, proteins and lipids. Carbohydrate, protein and lipid metabolism of fishes are disturbed under the condition of toxic stress and are known to cause increase or decrease in the biochemical constituents of the tissues. In the present study, the biochemical effects caused to *Anabas testudineus* on exposure to aquatic toxicants present in the Buckingham canal, Chennai, Tamil Nadu, India were investigated. The per cent change recorded in the tissues of experimental *Anabas testudineus* was found to be in the order of liver > kidney > muscle > intestine > gill; muscle > gill > kidney > liver > intestine; intestine > liver > kidney > muscle > gill in carbohydrate, protein and lipid respectively. The depletion in the metabolites indicated the fact that the whole metabolic pool of the fish gets disturbed/ altered under the toxic stress. Further, the change in the biochemical profile indicates their rapid utilization to provide excess energy to cellular biochemical process in order to cope with the stressed condition.

**Keywords:** *Anabas testudineus*, biochemical parameters, carbohydrate, protein, lipid

### 1. Introduction

The interaction between the contaminants and biomolecules is the first step in the generation of toxic effects. Understanding the alterations induced by the exposure to pollutants may contribute to the prediction of toxic effects that may occur at higher level of biochemical organization<sup>[1]</sup>. Toxicants affect biologically active molecules, viz., carbohydrates, proteins and lipids<sup>[2]</sup>. Carbohydrate, protein and lipid metabolism of fishes are disturbed under the condition of toxic stress<sup>[3]</sup>. Bacterial pathogens, pesticides and heavy metal pollutants are known to cause increase or decrease in the biochemical constituents of the tissues. Therefore, in the present study, the biochemical effects caused to *Anabas testudineus* on exposure to aquatic toxicants present in the Buckingham canal, Chennai, Tamil Nadu, India were investigated.

### 2. Materials and methods

#### 2.1 Study area

Chennai situated on the eastern coast of India, on 13° 04' north latitude and 80° 15' east longitude has three water ways that flows through the city, viz., Cooum river, Adayar river and Buckingham canal. The Buckingham canal is a man-made water canal linking the above mentioned two rivers. The canal extends from Nellore in Andhra Pradesh to Marakkanam near Puducherry. The length of this canal in Andhra Pradesh is 257km, and 163km is in Tamil Nadu. Within the city of Chennai, the canal is badly polluted from sewage and industrial effluents, and the silting up of the canal has left the water stagnant, creating an attractive habitat for mosquitoes.

#### 2.2 Test organism

Taxonomic position

Kingdom : Animalia

Phylum : Chordata

Sub Phylum : Vertebrata  
 Class : Pisces  
 Order : Anabantiformes  
 Family : Anabantidae  
 Genus : *Anabas*  
 Species : *testudineus*

*Anabas testudineus* commonly called climbing perch is an extremely hardy, small, brown or dark greenish-brown fish, native to Southeast Asia. It inhabits the majority of drainage systems across its native range and has been recorded in many different habitat-types including swamps, marshes, lakes, canals, pools, small pits, rice paddies, puddles, tributaries and main river channels. It is highly adapted to life in a seasonal tropical environment. It can tolerate very turbid and brackish water conditions and under extreme circumstances it can aestivate for several weeks by burying itself into moist ground [4].

**2.3 Biochemical studies**

Biochemical studies, viz., total carbohydrates, total proteins and total lipids were analyzed in control and experimental fish groups. Tissues of *Anabas testudineus*, viz., gill, muscle, liver, intestine and kidney from the control and experimental groups

were selected for the present study.

**3. Results**

The values of total carbohydrate, protein and lipid in the control and experimental tissues of *Anabas testudineus*, viz., gill, muscle, liver, intestine and kidney are presented in Table 1 and Figure 2. The order of carbohydrate per cent change recorded in the tissues of experimental *Anabas testudineus* was found to be liver > kidney > muscle > intestine > gill. In the tissues of control fish, the total protein content was in the order of muscle > gill > liver > intestine > kidney. The variation in distribution suggests the gradual difference in metabolic caliber of various tissues under the influence of aquatic pollutants and the total protein contents was found to decrease in all the tissues of the experimental fish. Maximum decrease was observed in muscle and gill and minimum in kidney and intestine. The per cent decrease of total protein content in pollutant affected *Anabas testudineus* was found to be in the order of muscle > gill > kidney > liver > intestine. The decreased total lipid content of tissues in pollutant affected *Anabas testudineus* was found to be in the order of intestine > liver > kidney > muscle > gill whereas the per cent change in the lipid content over the control fish was found to be in the order of gill > liver > kidney > intestine > muscle.

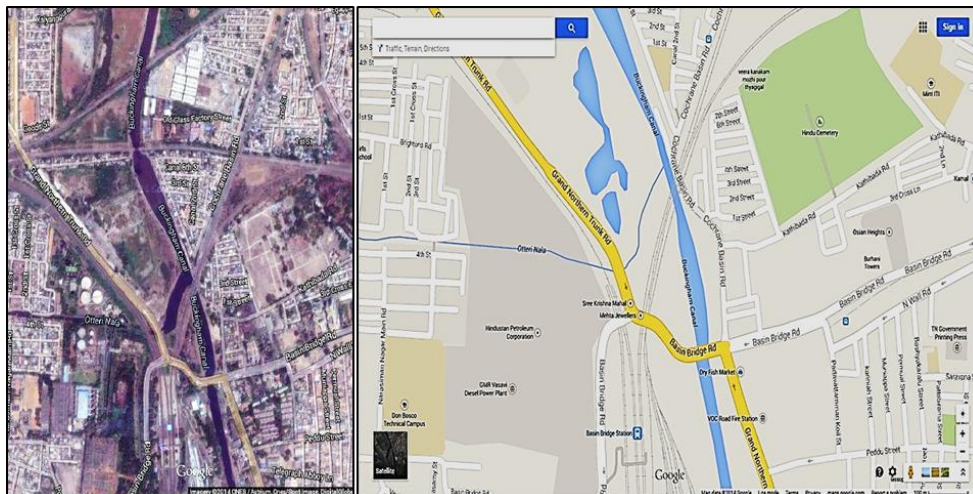
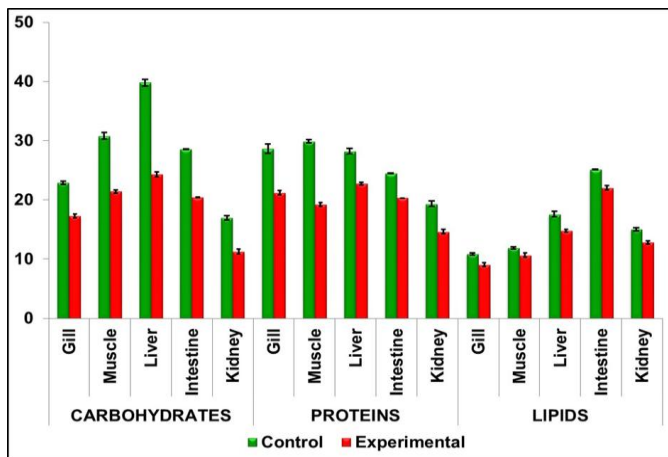


Fig 1: Study area - Buckingham canal

Table 1: Effect of aquatic toxicants on the biochemical parameters of different tissues of *Anabas testudineus*

Total Carbohydrate Content		
Tissue	Control fish	Experimental fish
Gill	22.90 ±0.30	17.29 ±0.29 (24.50%)*
Muscle	30.83 ±0.52	21.43 ±0.27 (30.48%)*
Liver	39.81 ±0.54	24.35 ±0.42 (38.28%)*
Intestine	28.56 ±0.02	20.45 ±0.04 (28.39%)*
Kidney	16.98 ±0.34	11.27 ±0.39 (33.63%)*
Total Protein Content		
Gill	28.67 ±0.82	21.20 ±0.36 (26.10%)
Muscle	29.90 ±0.31	19.25 ±0.31 (35.62%)*
Liver	28.23 ±0.46	22.76 ±0.26 (19.40%)*
Intestine	24.47 ±0.05	20.32 ±0.01 (16.95%)
Kidney	19.37 ±0.44	14.65 ±0.34 (24.37%)*
Total Lipid Content		
Gill	10.88 ±0.21	9.09 ±0.34 (16.45%)*
Muscle	11.91 ±0.18	10.64 ±0.37 (10.66%)
Liver	17.59 ±0.45	14.81 ±0.22 (15.80%)*
Intestine	25.15 ±0.06	22.05 ±0.35 (13.71%)
Kidney	15.05 ±0.28	12.84 ±0.27 (14.68%)*

Values expressed as mg/g wet weight of tissue; each value is mean ±standard error of five individual; Values in parenthesis denote per cent change; \*denote P<0.05 level of statistical significance.



**Fig 2:** Effect of aquatic toxicants on the biochemical parameters of *Anabas testudineus*. Values expressed as mg/g wet weight of tissue.

#### 4. Discussion

Carbohydrates are the chief sources of energy for any organism<sup>[5]</sup> and are found in large amounts in the liver and muscle tissues and in small amounts in kidney and heart. The decrease in carbohydrate content in the muscle and brain may be due to glucose utilization to meet excess energy demand imposed by severe anaerobic stress of mercury intoxication. A decrease in the glucose content of the liver, muscle and kidney tissue of *Clarias batrachus* was observed on exposure to sodium arsenite<sup>[6]</sup>. Neethirajan and Madhavan<sup>[7]</sup> also reported changes in the carbohydrate of liver, muscle, and gill tissue of *Mystus vittatus* when exposed to sumidon. Agarwal<sup>[8]</sup> noticed that mercuric chloride declined blood glucose of *Channa punctatus*. Srivastav *et al.*<sup>[9]</sup> investigated the effect of aldrin and carbamate on *Heteropneustes fossilis* and found a significant decline in blood glucose levels along with hyperchloremia and hypochloremia. Hypoglycemic condition may be due to increased muscular activity in the fish, which require more oxygen to meet the energy demand and consequently more amount of glucose rapidly utilized. Probably, the fish tries to meet this condition by increasing respiratory rate by drawing more amount of oxygen from contaminated water which may be the reason why low amount of blood glucose level are observed in pollution affected fish.

Vutukuru<sup>[10]</sup> reported an appreciable decline in different biochemical constituents (glycogen, total lipid and total protein levels) in liver, muscle and gills of *Labeo rohita* in chromium stressed condition when compared to that of control fish. The decrease in glycogen content of the tissues of *Labeo rohita* can be due to its enhanced utilization as an immediate source to meet energy demands under metallic stress. It could also be due to the prevalence of hypoxic or anoxic conditions, which normally enhances glycogen utilization<sup>[11]</sup>. The decrease in the glycogen content during the present study indicates its lipid utilization by the respective tissue as a consequence of pollutant stress. The results of the present study were comparable with the result of a study on *Channa punctatus*<sup>[12]</sup>; *Sarotherodon mossambicus*<sup>[13]</sup> exposed to chromium, and *Puntius conchoniensis* to nickel<sup>[14]</sup>. Further, the observations made by Murty and Devi<sup>[15]</sup> in *Channa punctatus*, exposed to bleached kraft pulp mill effluent; and Bengeri *et al.*<sup>[16]</sup> in *Labeo rohita*, exposed to parathion supports the present findings in *Anabas testudineus*. Metals intoxication generally leads to glycogen depletion due to generalized disturbance of carbohydrate metabolism.

Tissue hypoxia and changes in the buffering system also leads to glycogen depletion in tissues<sup>[3]</sup>. The mechanism involved in the glycogen depletion may be due to glycogenolysis and inhibition of glycogenesis through stress induced release of adrenal catecholamine<sup>[17]</sup>. Glucose level significantly decreased in pollution affected *Anabas testudineus* from Buckingham canal in the present study. Similarly, a significant fall in blood glucose at 8ppm in *Clarias batrachus*<sup>[18]</sup> was attributed to the utilization of glucose by the tissues of extremely active fish to the enhanced production of insulin by the beta cells of the islet of Langerhans or of somatostatin by D cells of the pancreatic islet of Langerhans which has an inhibitory action<sup>[19]</sup>. In the present study, aquatic toxicant caused a significant fall in the level of total carbohydrates and the results were observed to be in consistent with previous reports.

Proteins are involved in major physiological events and therefore the assessment of the protein content can be considered as a diagnostic tool to determine the physiological phases of organism. Depletion of protein content has been observed in the muscle, intestine and brain of the fish *Catla catla* as a result of mercuric chloride toxicity<sup>[20]</sup>. Freshwater fish, *Cyprinus carpio* exposed to sub lethal concentration of cypermethrin showed marked changes in protein fractions of liver, brain and muscle tissue<sup>[21]</sup>. Similar results were observed in *Anabas testudineus* and *Anabas scandens*, when exposed to copper, lead nitrate and mercuric chloride<sup>[22]</sup>. De Smet and Blust<sup>[23]</sup> reported that proteolysis was intended to increase the role of proteins in the energy production during cadmium stress. A decrease in the total protein of the liver of *Channa punctatus* on tannery effluents exposure was observed<sup>[24]</sup>. A good number of references are available regarding decrease in the level of blood protein on the administration of sublethal dose of agrofane in *Oreochromis mossambicus*<sup>[25]</sup>; and heavy metals in *Perna viridis*<sup>[26]</sup>.

Proteins are the most fundamental and abundant biochemical constituent present in fishes. Proteins are the most important energy source to spare during chronic period of stress. Animal exposed to toxicants even at the sub-lethal levels experience great stress at the metabolic level during the period of detoxification of the toxicant<sup>[27]</sup>. Namrata *et al.*<sup>[28]</sup> observed depletion of proteins under exposure to kelthane, an organochlorine insecticide and in starvation of *Channa punctatus*. Depletion in the protein level in different tissues/organs of experimental animals was found under the stress of various metals in traces. Appreciable decrease in the protein level of tissue of *Channa punctatus* was noticed after the fish were exposed to heavy metals<sup>[29,30]</sup>. Protein depletion in liver and gonads of *Anabas testudineus* under the stress of nickel chloride was observed by Jha and Jha<sup>[31]</sup>. The present observation on total protein and free amino acid levels are in agreement with that of Jha<sup>[32]</sup>. The reduction in total protein content in the present study was further comparable with the findings of Gupta and Sastry<sup>[33]</sup> on *Heteropneustes fossilis*, exposed to mercuric chloride by Ramos and Herrera<sup>[34]</sup> on fish exposed to pesticides. The decreased protein level recorded during the present study was an indicative of increased proteolysis<sup>[35]</sup> resulting in to a shift in nitrogen metabolism<sup>[36]</sup>. Inhibition of ribosomal activity<sup>[37]</sup> resulting in protein degradation<sup>[38]</sup> is also one of the possible reasons for protein depletion.

Lipids are important constituent of cellular structures. Lipids are also essential for maintenance of normal cell permeability and structural integrity of cell membranes. Synthetic



pyrethroid and cypermethrin was found to induce a decrease in lipid content of liver and muscle tissue of *Labeo rohita* [39]. Govindan *et al.* [40] reported a significant decline in the level of total lipids in the muscle, liver and brain of *Gambusia affinis* exposed to a pesticide, phosphamidon. A decrease in total lipid content during the present study in gill, muscle, liver, intestine and kidney tissue denoted the effect of metals present in the Buckingham canal effluent on lipid as tissue specific of the species. The findings of Ram and Sathyanesan [41] on *Channa punctatus* intoxicated with mercuric chloride and Jha [32] on *Channa punctatus* under lead exposure support the present study. Loss of lipid may be a consequence of inhibition of lipid synthesis and mobilization of stored lipids [24, 42]. The decrease in tissue lipids and proteins might be partly due to their cell repair and tissue organization with the formation of lipoproteins, which are important cellular constituents of cell membranes and cell organelles present in cytoplasm [43]. Decrease in the lipid concentration observed in the pollution affected *Anabas testudineus* from Buckingham canal can also be attributed to its utilization in cell repair and cell organelles. The depletion in tissue proteins of *Channa punctatus* and *Oreochromis mossambicus* from Arakkonam lake may be due to impaired or nil protein synthesis and their pollution are due to their utilization in the formation of macroproteins, which are eliminated in the form of mucus by the fish for the direct and/or indirect utilization of proteins and lipids for energy needs were also reported [44, 45].

Normally carbohydrates, proteins and lipids which constitute the major components of the body play an important role in growth and energy metabolism. A reduction of carbohydrates, proteins and lipids were observed in *Labeo rohita* under the influence of heavy metal, lead [46]. The effluent from dye [47], tannery [48] and distillery [49] reported a severe reduction in the carbohydrate, protein and lipid contents among the fish population. Sujatha [50] observed that the total sugars and protein content in muscle, liver, brain and kidney revealed a mixed trend in *Catla catla* in the different experimental groups and different days of the exposure. It is clear from the results of the present study that there is an appreciable decline in different biochemical constituents of fish under pollution stress. Fishes are sensitive to contaminations of water, and pollutants that may significantly damage certain physiological and biochemical processes, when they enter the organs of these animals [51]. Several cases have been reported in which toxic effects of pollutants may be altered due to changes in pH, temperature, hardness and dissolved oxygen content of water [52]. The harmful effects especially sub-lethal effect of the pollutants retard growth and adversely affects the metabolic activity of surviving individual [53]. The biochemical profiles of glycogen, total protein and total lipid contents underwent a significant depletion of varying degree in the tissues of the pollution affected fish. During the present study, it was observed that metabolic levels were organs specific, indicating differential sensitivity of the tissues against the toxicant toxicities. Further, it was difficult to explain the changes under metallic stress in an organism since such alterations differ not only from metal and from species to species, but also from one experimental period to another [54]. Nair *et al.* [55] while studying biochemistry and haematology of *Anabas testudineus* found serious alterations in these factors when the fishes were exposed to titanium dioxide effluent. According to them, glycogen, being the chief source of energy for the fish is the first metabolite to be affected by any stress. It was further pointed out that the increased liver

glycogen suggests a stimulated glycogen synthesis contemporaneous with the increased liver glycogenolysis and subsequent blood glucose elevation. The dose-dependent hyperglycemic state indicated a typical stress response. Such a response might be due to general stress or it could also be a result of hypoxic state caused by a strictly mechanical and/or heavy metal action on the functioning of the gill. In general, more energy is required to cope up for the stressful condition. This energy may be obtained from the organic constituents as carbohydrates, proteins and/or lipids [56, 57]. Therefore during toxicant exposure, the fish tries to detoxify the toxicant by spending more energy and thereby showing a reduction in glycogen, protein and lipid contents.

A severe dysfunction was noticed in the liver and muscle tissue of *Anabas testudineus* exposed to aquatic pollutants. Significant depletion in glycogen, protein and lipid contents were also observed. The depletion in the metabolites indicated the fact that the whole metabolic pool of the fish gets disturbed/alterd under the toxic stress. Further, the change in the biochemical profile indicates their rapid utilization to provide excess energy to cellular biochemical process in order to cope with the stressed condition. The results of the present study further confirm that aquatic pollution is one of the major reasons for decline in *Anabas testudineus* population and most probably in the population of the other freshwater fish residing in Buckingham canal.

## 5. Conclusion

The present study showed that aquatic pollutant induced alteration at the biochemical level, more pronounced changes acquiring or also time dependent this change may probably affect the enzyme mediated bio-defense mechanism of these fishes. Further research should focus on the effect of pollutant toxicity on *Anabas testudineus* at cellular and sub cellular levels.

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