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Challenges in Biochemical Engineering and Biotechnology for Sustainable Environment

Influence of Seasons and Locations in the Hepatic Enzymological Changes in the Fish *Lates Calcarifer* from River Uppanar, Sipcot Complex, Cuddalore.

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Abstract: In the recent years, heavy metal pollution has grown up as a serious concern all over the globe, as these heavy metals create adverse effects on all forms of living organisms in the biosphere. The aim of the present study was to elucidate the influence of seasons and locations (Station I, II and III) over the enzyme contents in the liver of *Lates calcarifer*. Three different locations were chosen in favour of fish sampling along river Uppanar, near Sipcot Complex, Cuddalore. A significant reduction in acid and alkaline phosphatases and concomitant increase in alanine and aspartate transaminase activities had been observed in the liver tissue in station II. Acid and alkaline phosphatases exhibited substantial decrease and concomitant increase of alanine and aspartate transaminase activities were observed at summer seasons at all different locations. From the results, it could be inferred that the station II has notable heavy metal pollution followed by station III and I. During the seasonal study, the summer season had huge alterations in the enzymological activities. This might be due to the stoppage of water flow and increase in the heavy metals. Further results will be discussed.

Keywords: Heavy metal pollution, *Lates calcarifer*, Transaminase, Phosphatases.

Introduction

In the recent years, heavy metal pollution has grown up as a serious concern all over the globe, as these heavy metals create adverse effects on all forms of living organisms in the biosphere. These heavy metals are not readily degradable in the environment and thus accumulate in the animal and human tissues to a very high toxic levels leading to undesirable effects¹. The discharge of untreated or partially treated industrial waste water containing heavy metals into the water bodies, especially rivers, prevail in aquatic bodies and get bioaccumulated along the food chain. Aquatic organisms have the ability to accumulate heavy metals from various sources including sediments, soil erosion and runoff, air depositions of dust and aerosol and discharge of waste water^{2,3}. Fishes are often at the top of the aquatic food chain and may accumulate large amounts of metals prevalent in the water⁴.

Enzymes are crucial factors, which permit several bio-chemical reactions that represent life to proceed in the cells of the body and therefore any changes in enzyme concentration in tissue cells reflect the state of health. The liver is a major target organ for toxic compounds⁵. Amino transaminase (AST and ALT) are the reliable marker enzymes of liver and they are the first enzymes of liver in diagnostic enzymology whenever liver damage has occurred⁶. The measurement of phosphatase (ACP and ALP) activity is an useful indicator of liver function⁷.

Although several studies deal with the enzymological composition of many commercially important fishes, no marked work has been carried out on *Lates calcarifer* particularly from River Uppanar. The *Lates calcarifer* fish was chosen for the present study for the following reasons; It is an economically important food fish in the tropical and subtropical regions in the Asia –Pacific. They are medium to large-sized bottom-living fishes occurring in coastal seas, estuaries and lagoons in depths between 10 and 50m. They are abundant and the availability is high throughout the year. They have high economic and ecological importance. Therefore, the present study was undertaken to elucidate the impact of both seasonal and locational variations in the enzyme contents in the liver of *Lates calcarifer*.

Materials and Methods

The fish *Lates calcarifer* (Sea bass) was collected from the three different stations based on their heavy metal pollution and seasonal variations. The station I was noted as less polluted and is located near the Alappakkam village, the station II is located backside of the TANFAC and SIPCOT and near the Kudikadu village which was notified as highly polluted location. The third one noted as station III was medium polluted location 2 km away from the Bay of Bengal. All the three locations are marked in and around Cuddalore district, Tamil Nadu, India (Fig. 1). The sampling locations were notified as less polluted, medium polluted and highly polluted based upon the quantum of industrial discharge, the physic-chemical parameters of water and the heavy metal accumulation in the sediments and water (Fig. 2 and 3).

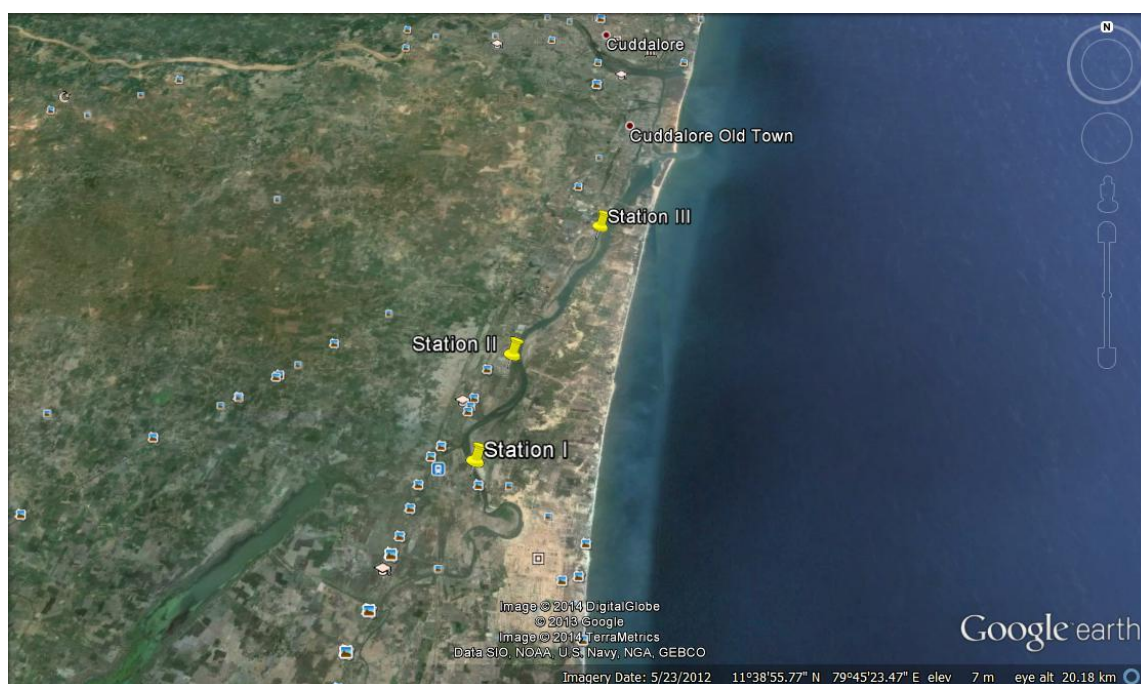


Fig. 1 Aerial view of the three sampling stations in river Uppanar, Cuddalore District.

The fish liver was collected for the bio-enzymological studies in all the three stations during different seasons. The AST and ALT were determined using the protocol of King⁸. Acid and alkaline phosphatase activities were assayed as per Tenniswood *et al*⁹

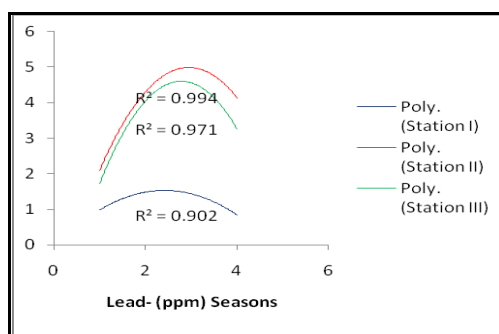


Fig. 2 Level of lead (ppm) in water

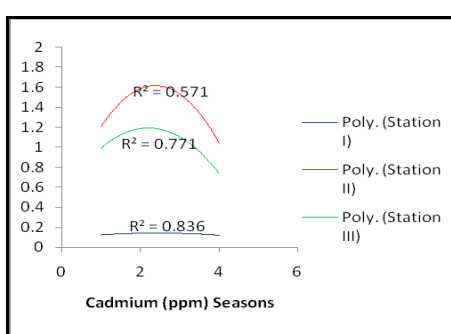


Fig. 3 Level of cadmium (ppm) in water

Results and Discussion

Transaminase activity

Fish liver is not only one among the vital detoxifying organs, but also the important compartment of heavy metal accumulation^{10,11}. The present study showed that the AST and ALT activity in the liver of *Lates calcarifer* increased when the heavy metal pollution increased. Among the three stations, station II had substantial level of transaminase (AST and ALT) activity followed by station III and station I because of the heavy metal pollution. Summer season had showed an increased amount of AST and simultaneously monsoon season had revealed an increased amount of ALT. Alterations in the activity of alanine and aspartate transminase enzymes will be reflected on the energy yielding TCA cycle and nitrogen metabolism. They also influence the gluconeogenic process and any change in the transaminase activity could be correlated with the protein and carbohydrate metabolism and thereby help in analyzing the metabolic shifts in the teleost fish *Anabas testudineus* treated with thiouracil¹².

Table 1. The activity of Asparate aminotransaminase (AST) and Alanine aminotransaminase (ALT) in the liver tissue of *Lates calcarifer* in different seasons and locations (Mean±SD)

Season	AST			ALT		
	Station I	Station II	Station III	Station I	Station II	Station III
Post monsoon	68.01±0.15*	77.57±0.35*	70.57±0.35*	65.04±0.25*	83.25±0.20*	76.84±0.16*
%COLP ^a		14.06	3.76		27.99	18.14
%COHP ^b			-9.02			-7.70
Summer	60.05±0.18*	79.94±0.36*	73.58±0.34*	65.94±0.21*	85.92±0.28*	78.21±0.18*
%COLP ^a		33.12	22.53		30.30	18.60
%COHP ^b			-7.96			-8.97
Pre Monsoon	70.51±0.42*	80.94±0.37*	76.32±0.32*	65.72±0.26*	85.21±0.34*	79.68±0.20*
%COLP ^a		14.79	8.24		29.65	21.24
%COHP ^b			-5.71			-6.49
Monsoon	67.54±0.43*	78.88±0.38*	75.37±0.25*	65.88±0.27*	84.85±0.32*	80.84±0.22*
%COLP ^a		16.79	11.59		28.80	22.71
%COHP ^b			-4.45			-4.73

^aPercentage change over less polluted; ^bPercentage change over highly polluted; *The values are significant at 5% level ANOVA

Phosphatase activity

In toxicological studies, ACP and ALP are important biochemical enzymes to be used to detect the alteration of physiological metabolism of such animals induced by metal exposure¹³. As ACP is a lysosomal enzyme, many environmental contaminates including heavy metals could be sequestered in the lysosomes of eukaryotic cells and some metals could alter the structure, permeability and integrity of lysosomal membranes and result in enzyme diffusion into cytosol¹⁴. The present study showed that the level of ACP and ALP in the liver of *Lates calcarifer* decreased when heavy metal concentration increased. Among the three stations, station II had huge amount of phosphatase (ACP and ALP) reduction followed by station III and station I. Monsoon season had revealed decreased amount of ACP and simultaneously summer season had exhibited a decreased amount of ALP. Shakoori *et al.*,¹⁵ suggested that a decrease (or) inhibition of ACP and ALP activities might be taken as an index of necrosis in hepatocytes.

Moreover, Atli *et al.* (2006) reported that the responses of enzyme activities in different tissues to heavy metals also depend on metal bioaccumulation ability of these tissues.

Table 2. The activity of Acid phosphatase (ACP) and Alkaline phosphatase (ALP) in the liver tissue of *Lates calcarifer* in different locations (Mean±SD)

Season	ACP			ALP		
	Station I	Station II	Station III	Station I	Station II	Station III
Post monsoon	26.50±0.05*	22.82±0.08*	23.84±0.45*	16.11±0.15*	13.91±0.15*	15.36±0.14*
%COLP ^a		-13.89	-10.04		-13.66	-4.66
%COHP ^b			4.47			10.42
Summer	29.70±0.15*	25.72±0.14*	27.06±0.34*	17.42±0.14*	12.40±0.16*	14.29±0.13*
%COLP ^a		-13.40	-8.89		-28.82	-17.98
%COHP ^b			5.21			15.24
Pre monsoon	29.20±0.16*	25.51±0.06*	26.64±0.25*	17.62±0.14*	12.64±0.17*	13.38±0.15*
%COLP ^a		-12.63	-8.77		-28.26	-24.06
%COHP ^b			4.43			5.85
Monsoon	28.61±0.019*	23.91±0.09*	24.91±0.29*	16.61±0.16*	13.32±0.12*	13.59±0.13*
%COLP ^a		-16.43	-12.93		-19.81	-18.18
%COHP ^b			4.18			2.03

^aPercentage change over less polluted; ^bPercentage change over highly polluted; *The values are significant at 5% level ANOVA

Conclusion

This study indicated that several key enzymatic systems in fish could be used as sensitive bioindicators of metal contamination in aquatic systems. The changes induced by the metals examined, demonstrate that modifications of different fish enzymes, at rather low concentrations might be practical endpoints for use in the biomonitoring of fish health in water bodies that are routinely polluted in many countries across the world.

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