Influence of Sago Effluent on the Levels of the Enzyme Lactate Dehydrogenase in the Liver of the Fresh Water Fish *Clarias batrachus*

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Abstract

The levels of the enzyme lactate dehydrogenase has been observed in the liver of fresh water fish Clarias batrachus exposed to different concentrations of treated sago effluent. . The concentration chosen were 25%, 50% and 75% of treated sago effluent. The levels of LDH were increased with increase in concentrations of the effluent. The control group recorded 20u/l whereas the experimental groups such as 25%, 50% and 75% treated sago effluent showed 54.2u/l, 82.9u/l and 104.4u/l respectively.

Key words*: Lactate dehydrogenase, Sago effluent, Clarias batrachus.*

1. Introduction

The aquatic environment is the ultimate sink for all the environment pollutants any chemical pollutant either natural or synthetic is most likely to reach the aquatic environment sooner or later. The toxicity may be either acute or chronic to all forms of biota in aquatic system and also varies to different aquatic organisms. The toxic effects may include both lethal and sublethal concentrations, which may change the growth rate, development, reproduction, histopathology, biochemistry, physiology and behavior (Rand & Petrocelli, 1985).

Alterations in the physiological and biochemical parameters of toxicant treated fish have recently emerged as an important tool for the water quality assessment and to know the pathological status of fish in the field of environmental toxicology (Racicot *et al*., 1975; Wieser & Hinterleitner, 1980). The alteration in various physiological and biochemical parameters of an aquatic animal due to exposure of different toxicant has been shown to be directly or indirectly related to the behaviour, immune system, neurotranmission, energy matabolism and reproduction (Ekwoezor *et al*., 2001; Adeyemo, 2005). Accumulation of the environmental pollutants and toxicants has been shown to cause alteration in the activity of many enzymes concerning to cellular energy metabolism (Niwelmski, 1990; Claireaux & Dutil, 1992; Sebert *et al*., 1993; Almeida *et al*., 1995).

Enzymes are biological catalysts produced by living cells they catalyze metabolic reactions. They are soluble and colloidal substances characterized by great activity, specificity and susceptibility to the influence of pH, temperature and other environmental changes. Enzymes are the most important tools of the livings cells. Cells cannot be without enzymes. They function as catalysts in a wide variety of biological reactions. They alter the speed of reactions without themselves undergoing any permanent change. Pollution monitoring method using enzyme inducement of enzyme depression in fish or other organism has been proposed for studying polluted environments. Enzymes getting into the blood after cell necrosis of certain organism may be used to indicate the tissue damage.

Fish are sensitive indicators of pollutants present in water. These pollutants cause various physiological and physical alterations in fishes. In the present work alternations in the activities of enzyme Lactate dehydrogenase (LDH) has been evaluated in liver of fresh water fish *Clarias batrachus*. The enzyme LDH is generally associated with cellular catabolic activity (Abston &

Yarbrough, 1976). LDH has been used for demonstrating damage in fish for a long time (Nemcsok & Boross, 1982).

LDH, a cytoplasmic biomarker in the glycolytic pathway, which catalyses a reversible reaction of reduction of pyruvate in to lactate plays an important role in the regulation of glycolysis. It is an extensively studied carbohydrate metabolizing enzyme which is crucial for normal cellular functions. The inhibition in the activity of this enzyme induced by pollutants has been shown due to changes in the conformation of active site (Valarmathi & Azariah, 2002). The activity of this prime enzyme is present in virtually all tissues of the organisms. They were involved in the energy release by the biological oxidation of food stuffs inside the mitochondria and also in the production of reduced potential (NADPH) required in the biosynthetic and detoxification mechanisms as stated by Gupta (1987).

The cytoplasmic enzyme LDH is widely used as marker of organ or tissue lesions in toxicology and clinical chemistry. This enzyme commonly reflects the metabolic capacities of a tissue are key factors in the metabolism with high sensitivity to pollutants (Osman *et al*., 2010).

Many investigators have studied the LDH activities in fishes. The enzyme activity was recorded in *Catla catla, Labeo rohita and Cirrhinus mrigala* – Chlorpyrifos (Kondal & Saxena, 2003), *Cyprinus carpio* – Distillary effluent (Ramakritinan *et al*., 2005), *Heteroneutes fossilis* – Vegetable oil factory effluent (Tilak *et al*., 2005) and *Channa striates* – Ferlizer industry (Archana *et al*., 2007).

2. Materials and methods

The Sago industry effluents were collected from a private Sago industry, situated at Ponnachi near Ammapet of Erode District, Tamil Nadu, India. The effluent from the industry was collected and transported to the laboratory and used for further experiments. Fingerlings of healthy *Clarias batrachus* were brought to the laboratory and acclimatized for 15 days. The fish were well fed during the acclimatized period. Feeding was stopped one day before commencement of the experiment.

For the assay of LDH, the liver of the fishes were cut and homogenized with cold distilled water and centrifuged at 7000 RPM for about 7 minutes. The supernatant was taken for assay. LDH activity was estimated by GCCA / kinetic method of Anonymous (1970, 1972) and Webinar *et al*., (1975).

3. Result and Discussion

The LDH activity in the liver tissues of the fish showed increase with increase in the concentration. The control group recorded 20u/l whereas the experimental groups such as 25%, 50% and 75% treated sago effluent showed 54.2u/l, 82.9u/l and 104.4u/l respectively.

In any diseased condition, pathological changes are preceded by biochemical changes. Thus, health of an individual depends on the harmonious equilibrium of all the biochemical reactions accruing in the body, which the diseased condition reflects the abnormalities in the normal biochemical reactions.

In the present investigation as a biochemical aspect of analysis LDH enzyme assay was analyzed. Enzymes are biocatalysts made up of protein. All the metabolic activities of organisms (catabolic and anabolic reactions) are assisted by them. Hence their normal activity is essential for the growth and survival of an individual. Any factor which affects the protein content of an individual will directly or indirectly manifested in malfunctioning of any specific enzyme of the body.

Basically there are four different processes that suggest the responses of enzymes to specific or non-specific chemical stress. They are direct enzyme induction, enzyme induction by specific

classes of chemicals, elevation of serum enzymes via tissue damage and alterations in enzyme activity as a result of changes in metabolic pathways or fluxes.

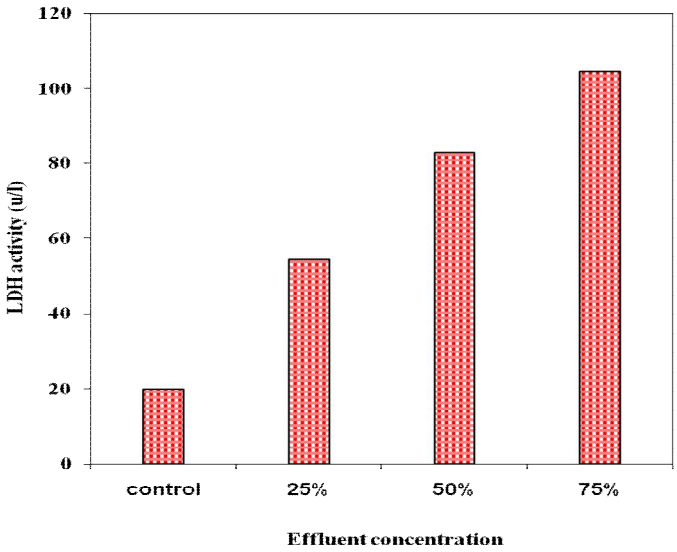

Figure 1
Lactate dehydrogenase levels in the liver tissues of *Clarias batrachus* on exposure to control and different concentrations of treated sago effluent

Enzyme activity is generally regulated such that specific substrates or entire pathways may be homeostatically adjusted toxicity compensate for endogenous or exogenous changes was suggested by Mayer *et al.*, (1992).

Measurement of enzymes that are indicative of cellular dysfunctions or damage is of diagnostic value in the clinical assessment of the diseased state. Both physiological and pathological states influence the blood enzyme levels or altered activities. Elevated levels of serum/plasma enzymes may be resultant of leakage from the necrotic or damaged cells/tissues/organs due to ischemia, neoplasia, carcinoma, decrease in their normal clearance and impairment in the normal process of excretion, as seen in biliary obstruction.

Application of optimized and standardized enzyme assay conditions is a perquisite for the assessment of clinically important diagnostic enzymes. This aspect of analysis is a major component of clinical enzymology.

Alterations in the enzyme activity have been used as gauge to monitor environmental pollution. Enzyme assays has recently emerged as an important diagnostic tool in the fields of environmental toxicology (Baskaran, 1991). Pollutants in the aquatic organisms affect growth and enzymes (Webb & Brett, 1973). Quantification of enzymes can serve as a valuable biomarker of pollutant exposure (Mayer *et al.*, 1992).

However, many authors have reported the enzyme activities of various organs of fish exposed to toxicants (Kabeer Ahamed, 1979; Vermal *et al*., 1978). Enzymes play an important role in metabolism. They are exceedingly efficient and very specific in terms of nature of reaction catalyzed and the substrate utilized. The synthesis and final concentration of enzyme is under genetic control and is greatly influenced by very small molecules of substrate. These cellular catalysts control the formation of biochemical intermediates essential to all physiological functions. Hence, change in enzyme levels is one of the fundamental steps to assess the effects of toxicants (Rana *et al*., 2003).

The enzyme LDH is predominantly present in heart and skeletal muscles, liver, kidney and in erythrocytes. Any damage to these tissues result in the release of enzyme into peripheral circulation consequently a rise in serum LDH levels. Thus this enzyme is an important diagnostic marker for the evaluation of myocardial infarction and muscle dysfunction.

Activity of LDH in normal serum is relatively low. An elevated enzyme level is observed in myocardial infarction. Serum LDH levels are also enhanced in muscular dystrophy and liver diseases such as hepatitis, liver cell necrosis, cirrhosis, liver tumors and obstructive jaundice. Abnormal levels of this enzyme are also associated with haematological and renal diseases like tubular cell necrosis renal infarction and renal tumors.

This enzyme expressed higher levels when lymphocytes are divided or more commonly, when cells are distressed and damaged particularly red blood cells. Elevated LDH is an indirect indication of disease progression sharp increases can indicate transformation.

LDH is present in most animal tissues. It serves as a pivotal enzyme between the glycolytic pathway and tricarboxylic acid cycle. LDH, a cytoplasmic biomarker in glycolytic pathway plays an important role in regulating glycolysis and is therefore, crucial for normal cellular functions. The activity of this prime enzyme is present in virtually all tissue of the organisms (Gupta *et al*., 1987). It maintains glucose substrate for vital tissues and prevents acidosis due to an excess of lactate.

LDH has been used for demonstrating damage in fish for a long time (Nemcsok *et al*., 1982). In this present investigation the levels of LDH were increased with increase in sub lethal concentrations of treated sago effluent. The increase in LDH activity may indicate the glycolytic rate might be stepped up during toxicity stress as pointed out by Sreeramalu Chetty *et al.*, (1980). A significant increase in LDH activity has been reported by many authors (Mary Chandravathy & Reddy, 1995; Suresh *et al.*, 1993).

The increased lactate levels of sub lethal concentrations exposure indicate a decrease in aerobic and increase in anaerobic respiration. The increased lactate level is due to increased muscular activity (Ray & Cremer, 1979). Martin e*t al*., (1983) State that the LDH activity increases during conditions favoring anaerobic respiration to meet the energy demands when aerobic oxidation in lowered. The earlier reports on fish support the present study (Sastry & Sharma, 1979; Anastasi & Bannistor, 1980; Vijaya Laksmi & Tilak, 1996).

Ramakritinan *et al*., (2005) have observed the elevated levels of LDH activity of muscle, liver and brain tissue of *Cyprinus carpio* chronically exposed to distillery effluent results from damage to tissues. Similar observation has been made in the fish *Channa striatus* exposed to sublethal concentrations of fertilizer industry effluent (Archana *et al*., 2007).

Satyaparameswar *et al*., (2006) have studied the elevated levels of LDH in the fresh water mussel, *Lamellidens marginalis* under sulphate toxicity. Tilak *et al*., (2005) have investigated the increased levels of LDH in the three fishes (*Catla catla, Labeo rohita and Cirrhinus mrigala*) with increase in the concentrations of the toxicants. Kondal and saxena, (2003) have studied the similar observations in the liver of the fresh water teleost, *Heteropneutes fossilis* exposed to vegetable oil factory effluent.

Osman *et al.,* (2010) have observed the elevated levels in the activity of LDH in the liver and muscle tissues of the African catfish *Clarias gariepinus* collected from Rosetta and Damitta branches comparing to the other sites.

4. Conclusion

Liver is one of the richest sources of LDH and the leakage of enzyme from even small mass of damaged liver tissue can increase the observed level to significant extent. The increase in the activity of enzymes after exposure to some pollutant was explained as a result of destruction of liver cells and increased cell permeability leading to a leakage of the enzymes from the damaged liver cells in to the serum.

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