INTRODUCTION :

Nowadays industrialization is increasing rapidly in our country. The modern industries are making use of various heavy metals such as iron, steel, copper, nickel, platinum and lead. Among the different types of pollutions, chemical pollution appears to be the major type which threatens the living systems very extensively. Among the different habitats aquatic environment is the major target of pollution. Most of the heavy metals are natural constituents of the aquatic environment. Some of them are biologically essential, but some metals like cadmium, lead and mercury are highly hazardous to aquatic biota and normally occur in low concentration [1]. It is clearly known the common forms of lead poisoning result from the mining, processing and commercial dissemination of lead [2]. The primary source of lead exposure to animals are contaminated soils, lead paints that remain on older structures, water from plumbing systems that contain lead, and lead based products, especially batteries, used crankcase oil, and linoleum [3]. The lead containing gasoline fumes from automobile exhausts constitute the chief and wide spread source of lead contamination in urban environments. A Major source of lead to waterfowl and other wildlife is spent lead shot, bullets, cartridge, and lead sinkers used in sport fishing [4].

MATERIALS AND METHODS :

1. Material :

*Anabas testudineus* which is selected as test species in the typical representative of Anabantoid fishes in South India. It is fresh water, euryhaline and eurythermal teleost.

Lactate dehydrogenase post exposure recovery from Lead intoxicated freshwater fish

*Anabas testudineus.*

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**Abstract:** Lactate dehydrogenase are important amongst the several molecules available in the cells. Carbohydrates play an important role in the cellular process. Under extreme stress conditions, carbohydrate enzyme such as lactate dehydrogenase have been known to act as the energy supplier in metabolic pathways and biochemical reactions. In the present investigation fish treated with an equitoxic dose of 10 ppm of lead nitrate and lead acetate intoxicated fish After a period of 15 days of exposure a batch from lead nitrate exposed fish and a batch from lead acetate exposed fish were transferred to lead-free water. Fishes were scarified on 1, 4, 8, 12 and 15 days for the analysis of recovery pattern in tissues viz. liver, muscle, kidney, gill and brain. It is found that lead intoxicated fishes were recovered after 15 days depends upon physical condition of the fish.

**Key word:** Carbohydrate, lead, anabas.
These fishes are well known for their air breathing ability, and they can survive out of water in moist air for six days. Slender fish with large scales, spines on gill cover, Scales on the head rigidly attached to the skull bone, strongly ctenoid, Grey brown to silver colour, with a dark spot on the base of caudal fin. Omnivorous feeds on macrophyte vegetation, different invertebrates, small fish. No parental care. No sexual dimorphism, the fish will spawn in the evening between plants, and the egg hatch in 24-36 hours. This fish is extremely adaptable, and can be kept in any water, soft, hard, alkaline and acidic, even in brackish water. They are nicknamed as 'Climbing perch' since they ascent banks and even lower branches of trees. These fish are cultured in ponds in and around Kolleru belt and they have a very good commercial value, because of their nutritive value and taste. The fish Anabas scandens has been selected as the test animal because of its euryhaline and eurythermal nature, and unique position in food chain. They are quite sturdy and ideally suited for experimentation in laboratory for longer periods.

2. Methods:

Biochemical assays were made in different tissues from both experimental (exposed to toxicant) and Normal (toxicant free) fishes. Fish approximately of same size and weight were selected and grouped into 6 batches. 2 batch of fish served as controls, 2 batches of fish were exposed to lead nitrate and the remaining two batches were exposed to lead acetate for a period of 15 days. After a period of 15 days of exposure a batch from lead nitrate exposed fish and a batch from lead acetate exposed fish were transferred to lead-free water and scarified at the same intervals to observe the recovery responses. In all the experiments, a minimum of six individual observations were made. The values of different parameters were expressed as mean with their standard error. Significance of the values obtained were tested using student 't' test. The glucose content in the tissues were estimated by the method [5]. 10% (W/V) homogenates of liver, kidney, gills, muscle and brain were prepared in 0.25 M icecold sucrose solution and centrifuged at 2,500 rpm for 15 minutes. The clear supernatant was used for the assay of enzyme activity. The total reaction mixture contained in 2 ml volume: 100µ moles of phosphate buffer (pH 7.4), 2 µ moles of INT, 50 µ moles of sodium lactate (pH 7.4), 0.1µ moles of NAD and 0.5 ml enzyme extract. The reaction initiated by adding enzyme extract. After incubating the reaction mixture at 37°C for 30 minutes, the reaction was stopped by the addition of 5 ml glacial acetic acid. Zero time controls were maintained for each tissue separately by adding 5 ml of glacial acetic acid to the reaction mixture, prior to the addition of homogenate supernatant. Colour was extracted by adding 5 ml of toluene and kept overnight in the refrigerator. The extracted formazan was measured at 495 nm in a spectrophotometer. The enzyme was expressed as µ moles of formazan formed/mg protein/hour.

RESULTS AND DISCUSSION:

1. Results:

The activity of lactate dehydrogenase was increased in the early periods of exposure. However the activity was inhibited in the later periods of exposure in all the tissues.

On 1st day of exposure maximum activity was noticed in liver (+8.05% lead nitrate P < 0.05; +8.47% lead acetate P < 0.01), kidney (+7.33% for lead nitrate, +8.62% lead acetate, P < 0.05) followed by gill (6.45% lead nitrate P < 0.05 + 8.06% for lead acetate P < 0.001) muscle (+5.67% lead nitrate, P < 0.01, +7.01% lead acetate, P < 0.05). Insignificant elevation in activity was recorded in brain (+4.39% for lead nitrate and +5.49% for lead acetate).

On the 4th day of exposure all the tissues recorded an increase in LDH activity except kidney wherein significant inhibition was noticed (-10.16% for lead nitrate and -10.38% for lead acetate P < 0.05). Maximum enhancement was found in liver (+14.69% for lead nitrate and + 17.00% for lead acetate; P < 0.001) followed by gill (+11.75% for lead acetate).
nitrate, $P < 0.01$: +14.29% for lead acetate; $P < 0.001$), muscle (+11.38% for lead nitrate + 13.69% lead acetate; $P < 0.01$) and brain (+9.62% for lead nitrate $P < 0.01$; +11.54% for lead acetate $P < 0.05$).

On 8th day of exposure inhibition of LDH activity was recorded in all the tissues. Inhibition of enzyme activity was noticed till the end of exposure period (i.e. upto 15th day). Maximum inhibition was noticed in kidney (-17.65% lead nitrate $P < 0.05$, -18.30% lead acetate $P < 0.01$ followed by muscle (-10.06% lead nitrate, -11.49% lead acetate $P < 0.05$) and brain (-9.09% lead nitrate; $P < 0.01$; -8.36% lead acetate, $P < 0.05$).

On 12th day of exposure similar response was found in all the tissues. However the magnitude of depletion was more. The values were significant at $P < 0.001$; $P < 0.01$ and $P < 0.05$. The percent inhibition ranged between -14.04% to -24.69% for lead lead nitrate and -15.44% to -25.51% for lead acetate).

On 15th day of exposure the maximum inhibition was observed in kidney (-28.19% lead nitrate, -30.43% lead acetate, $P < 0.001$) followed by liver (-28.55% lead nitrate, -29.83% lead acetate; $P < 0.001$) gill (-23.44% lead nitrate; $P < 0.05$; -25.6% lead acetate $P < 0.001$ and a brain (-22.41% for lead nitrate and -23.79% lead acetate; $P < 0.001$).

After transferring fish to lead free water the inhibitory response was continued with less magnitude of response in comparison to exposure period. The inhibition was gradually reduced during the subsequent exposure periods. The rates of reduction the inhibition was tissue-specific. On 15th day of recovery period the inhibition was narrowed down and the difference between the control and experimental values were statistically insignificant in liver (-2.12% lead nitrate, -2.69% lead acetate) muscle (-2.37% lead nitrate, -3.11% lead acetate) kidney (-1.44% lead nitrate, -1.03% lead acetate). Brain tissue recovered on 8th day. The gill of fish exposed to lead nitrate recovered on on 8th day, but gills from lead acetate exposed fish regained the normal levels on 12th day (Fig 1).

Fig. 1: Lactate dehydrogenase activity in the
tissues of Anabas testudineus during exposure and recovery period after Lead nitrate and Lead acetate intoxication

2. Discussion:

Lactate dehydrogenase an enzyme which catalyses the interconversion of lactate and pyruvate exhibited a tissue-specific and time-dependent responses. The responses appeared more in the organic form of lead in comparison to inorganic. In the initial stages of toxicosis i.e., upto 4th day of exposure the LDH activity was found enhanced in the liver, muscle, gill and brain. However, in kidney the enhancement was recorded only on the first day of exposure. The observed increase in LDH activity in all the tissues is in agreement with the earlier studies in fishes [6-8]. The activity patterns of LDH in liver, muscle, gill and brain from 8th day onwards and in kidney from 4th day are simply reflected in the lactate levels of the tissues. The activity patterns of LDH during the early stages of toxicosis do not commensurate with the accumulation of lactate, and this seems to be an interesting deviation from observations recorded from the various animal models during metal toxicosis. The enhancement in LDH activity suggests the possibility of conversion of pyruvate to lactate. In evidence to this the accumulation of lactate and depletion of pyruvate content was recorded during the early stages of lead toxicosis in the present study.

Though the levels of pyruvic acid, lactic acid content depends on the activity patterns of LDH, a clear cut relationship in the responses could not be seen between these profiles, suggest the involvement of other factors for the observed responses in lactate, pyruvate content of the tissues.

REFERENCES:


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