



Fluoride induced alterations in the arginase activity of freshwater catfish, *Clarias batrachus* (Linn)

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Abstract

Fluoride induced alterations in arginase activity in liver, muscle, kidney, gill and brain of freshwater cat fish, *Clarias batrachus* (Linn.) were carried out. The effect of fluoride was observed in different concentrations like 1, 10, 20 and 30 ppm in 1, 30 and 60 days of exposure. The arginase activity was found increased in all the tissues, throughout the exposure span. Maximum activity was observed on 60th day of exposure in 30 ppm of fluoride concentration. In most of the cases the elevation of arginase activity significantly differed from the controls. Gill showed maximum activity followed by liver, brain, kidney, and muscle. The increase in arginase activity indicates production of urea or ornithine.

Keywords:- *Clarias batrachus*, Fluoride, Toxicity, Arginase

Introduction

Fluoride levels in unpolluted freshwater generally range from 0.01-0.3 mg/l. Fluoride ions can be removed from aquatic phase by the precipitation of calcium carbonate, calcium phosphate, calcium fluoride and magnesium fluoride (Camargo, 2004). Fluoride ion must be considered as a serious pollutant since, its concentration in many aquatic systems is significantly increasing as a consequence of man's activity (Camargo and Lawpoint, 1995). Fluoride containing pesticides and plants manufacturing bricks, ceramics and fluoride chemicals are however leading to increased local fluoride levels up to 100 times the natural background level (Camargo, 2003). Important anthropogenic sources of fluoride to the aquatic environment include waste waters and effluents from fertilizer producing plants and aluminum refineries (Gigore and Campbell 1985).

Fluoride toxicity increases with increasing fluoride concentration, exposure time, and water temperature (Angelovic *et al.*, 1965, Wright, 1977). The effect of fluoride on living systems have been considered at various levels of cellular tissue organisation. The wide range of responses in enzymes have been observed to an increased concentration of fluoride (Iwase, 1972).

Jenkins *et al.*, 1972). Fluoride inhibits many enzymes through different mechanisms. Fluoride induced enhancement of ammonia levels were observed by Devi (1988). Not much information is available on fluoride induced metabolic alterations on arginase activity in fishes, especially in *Clarias batrachus*. The main objective of the study is to know the effect of different concentrations of fluoride on arginase activity of freshwater cat fish, *Clarias batrachus* in different exposure spans.

Materials and Method

Clarias batrachus fish was obtained from the Kaikaluru fish farms, Krishna district, Andhra Pradesh, carried to the laboratory in aerated polythene water containers from their natural habitats. Animals of weighing about 75-100 gm and measuring about 20-25 cm were used for experiment. They were left in the water tanks for acclimatization as suggested by Klontz and Smith (1969) for 20 days. During acclimatization fish were fed with fish feed and egg albumen. Fish of same sex and weight were selected and transferred to plastic containers. The experimental tubs were set up in parallel with control. Feed was given to the both experimental and control fish and water was renewed. The feeding was stopped 24 hours prior to experiment to avoid metabolic

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differences if any due to differential feeding. Sodium fluoride was added in 1, 10, 20, 30 ppm and the fish were exposed to different exposure spans of 1, 30 and 60 days. After the exposure period, the fish were dissected and various organs like liver, kidney, gill, muscle and brain were used for estimation by modified method of Archibald (1944), Vanslyke and Archibald (1946). The statistical tests were employed as per Bailey (1959).

Results and Discussion

Arginase activity was increased in all the tissues throughout the exposure span (Fig. 1 to 5). Time dependent percent enhancement was recorded. Maximum arginase activity was observed on 60th day of exposure in 30 ppm of fluoride concentration. Gill showed maximum activity (+91.83%; $P<0.001$), followed by liver (+64.78%; $P<0.001$); brain (+44.35%; $P<0.001$, kidney (37.38%; $P<0.001$) and muscle (+25.94%; $P<0.001$). All tissues showed statistically significant values over controls. However on first day of exposure at 1 ppm concentration, all tissues showed statistically insignificant values.

Discussion

During urea cycle arginase converts L-arginine into L-ornithine. This L-ornithine is then utilized in different pathways in liver (a) by ornithine carbonyl transferase in the urea cycle, (b) by ornithine transaminase to form glutamate semialdehyde ultimately converted in to either glutamate or L-proline and (c) for formation of polyamines by ornithine carbamylase (Bolkenius and Sieler, 1981; Matsui and Pegg, 1982).

In the present investigation, arginase found increased in all the tissues throughout the exposure span. The elevation is progressive and reached maximum in 60th day of exposure. Liver and gill showed maximum activity followed by brain, kidney and muscle. However, the percent enhancement was more marked in all the experimental tissues at all exposure spans.

Little is known about arginase activity in different fishes. The increase in arginase activity indicates production of urea or ornithine. Conversion of ammonia to urea is significant in the detoxification of ammonia (Krebs, 1952). The enhancement of arginase activity

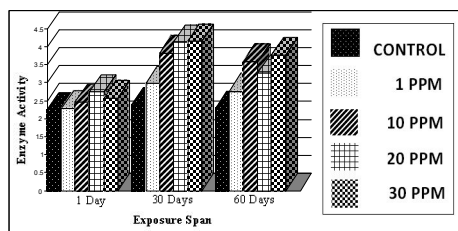


Fig. 1: Arginase activity (μ moles of urea/ mg protein/ hr) in liver of *Clarias batrachus* exposed to various concentration of fluoride

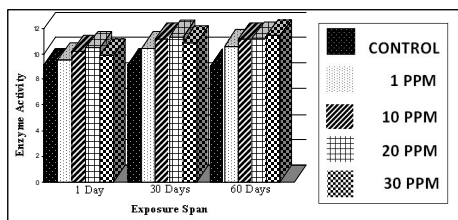


Fig. 2: Arginase activity (μ moles of urea/ mg protein/ hr) in muscle of *Clarias batrachus* exposed to various concentration of fluoride

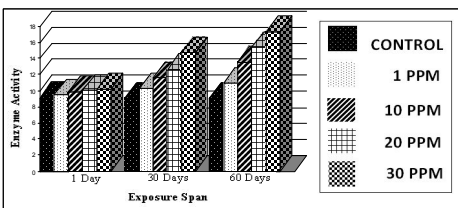


Fig. 3: Arginase activity (μ moles of urea/ mg protein/ hr) in kidney of *Clarias batrachus* exposed to various concentration of fluoride

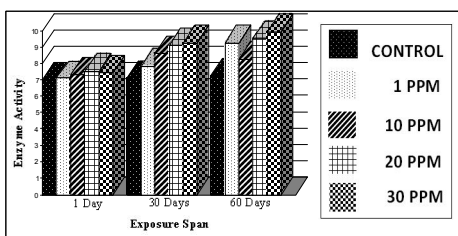


Fig. 4: Arginase activity (μ moles of urea/ mg protein/ hr) in gill of *Clarias batrachus* exposed to various concentration of fluoride

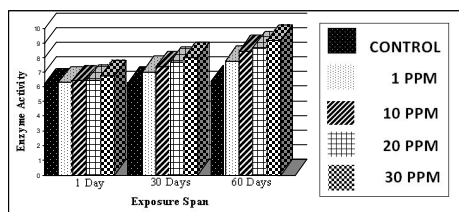


Fig. 5: Arginase activity(μ moles of urea/ mg protein/ hr) in brain of *Clarias batrachus* exposed to various concentration of fluoride.

indicates the increased utilization of ammonia towards urea synthesis, to avoid ammonia toxicity. The inconsistency in the activity levels of arginase were reported in the fish model (Geethanjali, 1988) and this inconsistency attributed the defect in the ornithine cycle at some level by inhibiting the enzyme systems.

References

- Angelovic, J. W., Sigler, W.F and New Hold J.W., 1965. Temperature and fluosis in Rainbow trout. *J. water. Pollut. Control. Fed.*, 33: 371 - 381.
- Archibald, R.M., 1944. Determination of citrulline and Allontoin and demonstration of citrulline in blood plasma. *J. Biol. Chem.*, 156:121.
- Bailey, N.T.J., 1959. *The statistical methods in biology*. The ECBS and English unix press Ltd., Great Britain 1959).
- Bolkenius, F.N. and Sieler, N., 1981. Acetyl derivatives as intermediate in polyamine catabolism. *Int. J. Biochem.*, 13:287-292.
- Camargo, 2004. J.A Bull. Environ. Contam. *Toxicol*, 72: 579-585.
- Camargo, 2003. J.A. *Chemosphere*, 50(3): 251-264.
- Camargo, 1995. J.A, T.W. Lawpoint. *Arch. of Environ. Contam. Toxicol*, 29: 159-163.
- Devi, J.A., 1988. Fluoride induced alterations in the protein metabolism of fresh water fish *Clarias batrachus*. Ph.D. thesis Osmania university, Hyderabad.
- Geethanjali, 1988. Alterations in nitrogen metabolism during BHC intoxication in fresh water fish *Channa punctatus*. Ph.D. thesis Osmania university, Hyderabad.
- Gigure, A. and Cambell, P.G.C. Fluoride Toxicity towards fresh water organisms and hardness effects. *Rev. Sci. Env.*, 17(3): 373-393.
- Iwase, 1972. Studies on the Glycogen and phosphorylase variations on myocardium, skeletal muscle and liver in experimental fluorosis influence of fluorine on, phosphorylase part II Shikoku *Acta Medica*, 12: 624-669.
- Jenkins, G.N., Venkateswarlu, P.P. and Zipkin, I., 1972. *Effects physiologiques de faibles doses de fluorures*. In fluor et sante . WHO monographies, 59: 167-135.
- Klontz, G.W. and Smith, L.S., 1975. *Methods of using fish as biological research I*. Methods of animal experimentation. 3rd ed. Gar, W.I. pp: 323-385. Makay *et al.* Academic press, Newyork (1969).
- Krebs, H.A., 1952. *The enzymes*. Academic press, New York. pp: 2: 866
- Matsui, I. and Pegg, A.E., 1982. Induction of spermidine N'acetyl transferase by dialkyl nitrosamines. *Cancer. Res.*, 42: 2990-2995.
- Nachlas, M.M., Margulus, S.P. and Seligann, A.M., 1960. Sites of electron transfer to tetrazolium salts in the succino oxidase system. *J. Biol. Chem.*, 235:2735.
- Vanslyke, D.D. and Archibald 1946 gasometric and photometric measurement of arginase activity. *J. Biol. Chem.*, 165: 293 - 309.
- Wright, D.A, 1977 Toxicity of fluoride on brown trout fry (*Salmo trutta*) Environ pollute 12:57-62.

