Fluoride Induced Toxicity on Protein Profile of rat, Rattus rattus (Wistar)

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Abstract:-

Fluoride when present in higher concentrations in natural resources becomes a potential environmental and health hazard. The present study was designed to investigate the biochemical changes in liver of albino rats after exposing them to sodium fluoride. The data indicate significant reduction in total proteins, mitochondrial proteins , microsomal proteins, cytosolic proteins, soluble proteins, insoluble proteins, acidic proteins and basic proteins in the liver of experimental animals and amino acids were highly elevated in the exposed rats, it may due to impairment of protein synthesis as well as reduced incorporation of amino acids into proteins.

Key word:- Albino rat, biochemical changes, sodium fluoride, liver.

Introduction

Fluorine, considered to be one of the environmental toxins (Jaśkowski, 2000) does not occur free in nature. Fluoride intoxication is a serious public health problem in many parts of the world as a result of high fluoride content in ground water and airborne fluoride released from burning of fluoride load coal. Sodium fluoride is an inorganic chemical compound with the formula NaF, it is a source of the fluoride ion in diverse applications. It is an ionic compound dissolving to give separated Na^+ and F^- ions. Fluoride anions are widely distributed in the environment in different forms and their compounds are extensively used (ATSDR, 2003). Underground water sources are more likely to have higher levels of fluoride, whereas the concentration in seawater averages 1.3 ppm. Fresh water supplies generally contain between 0.01-0.3 ppm, while the ocean contains between 1.2 and 1.5 ppm. According to World Health Organization (1984), the permissible limit of fluoride in drinking water is 1.5 ppm. Over 50% of the groundwater sources in India have been contaminated by fluoride. Fluorosis is the phenomenon caused by excessive ingestion of fluoride for a prolonged period of time. Intrinsic geological resources and more recently the arrival of increased industrialization and attendant ecological contaminated waste have contributed seriously to the increasing incidence of fluoride-related human health problem. High stratum of fluoride in drinking water has become a future health hazard all over the world, approximately with 66.62 million victims in India alone. Fluoride enters the human and animal body through drinking water and to a slight extent through food (Susheela, 2007).

The present investigation was undertaken to elucidate the effects of sodium fluoride on rat liver for understanding the mechanism of fluoride action on Metabolism of liver through biochemical changes in level of different biochemical parameters.

Material and Methods

Experimental Animal

Albino rat , *Rattus rattus* weighting 150-200 g, were used. Animals were purchased from wadhwani pharmacy Collage Yavatmal and acclimatized for two weeks in Animal House in the Department of Zoology Govt. Vidharbha Institute of Science and Humanities Amravati. The Institutional Animal Ethical Committee already approved this study for the use of Rat. The rat were housed in well-ventilated animal house and caged also well, at room temperature and exposed to 10-12 h of daylight.

Rats were divided into four groups having five animals each.1st group was used for control and 2nd , 3rd and 4th groups were ingested with 0.02 gm, 0.04gm, and 0.06 gm of fluoride water respectively for 72 days. Animals from each dose group were deprived of food overnight and sacrificed at the end of 72 days. They were stunned by a blow on the head and operated. The liver was removed with adhering material by dipping in chilled normal saline and homogenized.

Chemical; All the reagents were purchased from Chaiga Traders, Yavatmal and were of analytical grade.

Biochemical Analysis

The estimation of total protein, mitochondrial protein, microsomal protein, cytosol protein, soluble and insoluble protein, acidic and basic protein were done from liver tissue by using Lowry method, 1951. And free amino acid by using Danilson and Harold method, 1958.

Statistical analysis

The results were expressed as the mean ± SEM. The data were statistically analyzed using one-way analysis of variance (ANOVA) . The level of significance was taken as $p < 0.05$.

Results

Table 1 depicts the levels of total protein and Acidic & basic protein in the liver of control and experimental groups of rats. There was a significant ($P<0.01$) decrease in Total protein and Acidic & basic protein in the liver of rat.

Present data showed the effect of sodium fluoride treatment on mitochondrial protein in rat liver. Results showed that NaF treatment caused significant ($P<0.05$) decrease in mitochondrial protein & Insoluble Protein in rat liver compared to the control group of rats.

Biochemical parameters are shown in Table After 72 days of treatment , Microsomal protein, Cytosol protein, Soluble protein showed significant ($P<0.001$) decrease compared to control group.

As shown in Table 1 total amino acid and free amino acid were significantly increased with higher doses of fluoride content as compared to control.

Table 1 Effect of fluoride on protein profile of liver of rat.

Parameters	Control	0.02gm/kg body weight	0.04gm/kg body weight	0.06gm/kg body weight
Total protein	0.46 ± 0.68	0.16 ±0.40**	0.20 ±0.45**	0.15 ±0.39***
Mitochondrial protein	0.13 ±0.36	0.11 ±0.33*	0.10 ±0.32*	0.10 ±0.32*
Microsomal protein	0.47 ±0.69	0.41 ±0.64**	0.35 ±0.60***	0.21 ± 0.46***
Cytosolic protein	0.16 ±0.41	0.11 ±0.33***	0.07 ±0.28***	0.04 ±0.21***
Soluble protein	0.17 ±0.41	0.07 ±0.26***	0.02 ±0.14***	0.02 ±0.17***
Insoluble protein	0.09 ±0.30	0.06 ±0.26*	0.06 ±0.25*	0.04 ±0.22*
Acidic& Basic protein	0.14 ±0.37	0.12 ±0.34*	0.11 ±0.33**	0.10 ±0.32**
Total amino acid	0.13 ±0.36	0.37 ±0.60***	0.36 ±0.61***	0.37 ±0.61***
Free amino acid	0.39 ±0.63	0.45 ±0.67***	0.43 ±0.66**	0.45 ±0.67***

Values are expressed as Mean ± SE *=p<0.05; **=p<0.01;***=p<0.001;where nothing is shown =Non Significant

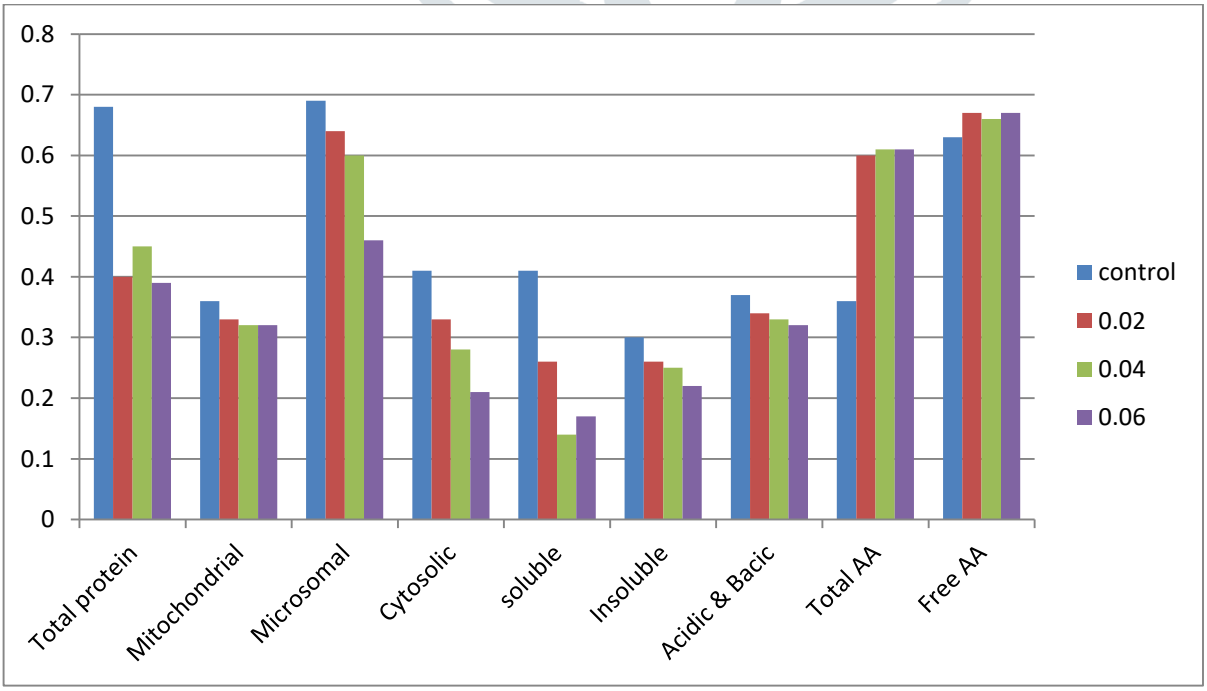

Fig. 1 Effect of fluoride on protein profile of liver of rat.

Discussion

Liver is the principal organ responsible for metabolism and involved in the metabolism of toxic compounds produced during systemic processes and exogenous toxins entering into the organisms from the environment (Gale *et al.,* 1978). Fluoride is useful in preventing dental caries and in the treatment of osteoporosis. But excessive intake of fluoride can be toxic. Although fluoride is considered as an essential trace element, but its exposure at high doses results in fluoride accumulation in body tissues. Besides, the vital organs like liver, kidney and brain are also susceptible to its toxic effects, where the pathological changes occur even before the development of overt clinical signs of fluoride toxicity.

Irregular protein metabolism is considered a sign of hepatic dysfunction. In the present study, the administration of fluoride resulted in a significant decrease in the attention of total protein as compared to the control group. Earlier studies reported a similar reduction in protein content in fluoride treated animals and related it to inhibition of decarboxylation of branched chain amino acids and simultaneously promoting protein breakdown (Shashi *et al.,* 1992). Fluoride affects cellular protein synthesis mainly due to the impairment of peptide chain initiation (Godehaux and Atwood ,1976).

In rats, fluoride-induced fluctuations in liver protein biosynthesis have been attributed to decrease in RNA transcription and inhibition of the methionin-activating enzyme of the liver, catalyzing certain stages of protein synthesis (Zahvoronkov and Strochkova,1981). In mice, decrease in protein levels in the liver were due to alteration in metabolism and change in osmotic balance (Chinoy *et al*.,1994). The results obtained in the present study also revealed that the concentrations of acidic proteins, basic proteins, and total proteins in the liver were reduced after NaF treatment. A similar decrease in the protein content of the liver of fluoride intoxicated experimental animals has also been reported by Kathpalia and Susheela, (1978). Ravel *et al* claim that NaF acts as a specific inhibitor of protein synthesis by interfering with a reaction associated with new peptide chains on ribosomes.

The increase in concentration of amino acids reflects a decreased shunting of amino acids into the tricarboxylic acid cycle for energy production as fluoride also inhibits enzyme enolase (Wiseman,1970). Fluoride affects the mechanism of glutamine synthesis, a stage in the deamination process and in Na+- and K+- activated ATPase which is essential for active uptake of amino acids (Whittam and Wheeler, 1970). The abnormal increase in hepatic amino acids may also be due to reduced incorporation of amino acids into proteins (Holland, 1979). In addition, fluoride affects the amino acid sequence of newly synthesized proteins in rat liver (Rymarscherbina, 1974).

Conclusion

From the results, it is clearly indicated that 72 days of sodium fluoride exposure to rats caused a significant decrease in total protein, mitochondrial protein, microsomal protein, cytosol protein, soluble and insoluble protein, acidic and basic protein but amino acids were highly elevated in the exposed rats, it may due to impairment of protein synthesis as well as reduced incorporation of amino acids into proteins.

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