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Effect of Aluminium Chloride on Testicular Marker Enzyme in Rat and the Effect of Vitamin-E

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ABSTRACT

To investigate the effect of Aluminium chloride on the activities of testicular enzymes in rat. Forty male Albino rats were divided into four groups (n=10). Treatment was given for a period of 45 days daily. Group-I Control given only distilled water, Group-II treated with aluminium chloride 50 mg/Kg body wt. Group-III treated with Vitamin E 20mg/Kg body weight. Group-IV treated with aluminium chloride+ Vitamin-E. The animals were sacrificed after 24 hours duration. Biochemical studies were observed in testis. Marker enzymes ALP, ACP and LDH, SDH activities were decreased significantly with control. Lipid peroxidation was increased significantly in aluminium chloride treated rats. The administration of $AlCl_3$+Vit E shows protective effect from the toxicity.

Keywords: Aluminium chloride, ALP, LDH, Marker enzymes, Vitamin-E

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INTRODUCTION

Aluminium (Al), abundant metal existing about 8% in the earth's crust[1]. Aluminium gets exposed through various routes like drinking water, food residues cooking untensil packaging food and beverage and as a constituent in medicine preparation[2]. Aluminium induces oxidative stress and act as a biomarker of cellular lipid peroxidation[3].The toxic effect of aluminium affects the tissues by free radical generation[4]. Aluminium chloride affects the process of spermatogenesis in mouse[5]. Vitamin E protect the cell constituent from damaging effects of free radicals. If the free radicals are not checked they may damage the tissue and cause cancer development[6]. Testis comprises various cell types including germ cells, Sertoli cells and Leydig Cells. The main function of testis is attributed to the process of spermatogenesis and steroidogenesis[7].Present studies were undertaken to investigate the toxic effect of aluminium chloride on rat testis on biochemical marker enzyme and the effect of Vitamin E

MATERIALS AND METHOD:

Aluminium Chloride was purchased from Aldrich chemical company USA. Vitamin E obtained from Triveni Interchem Pvt. Ltd Gujarat, India. Healthy adult male albino rats of Wistar strain (Rattus norvegicus (90 days old) weighing 190-210 gram obtained from the Central Animal House, Jawaharlal Institute of Postgraduate Medical Education and Research (JIPMER), Pondicherry, were used for the present investigation. The animals were maintained and handled as per the guidelines given by the Committee for the purpose of Control and Supervision of Experiments on Animals (CPCSEA), Government of India and Institutional Animal Ethical Committee (IAEC).No 845/GO/ac/04 CPCSEA. Animals were housed in propylene cages under controlled temperature and hygienic conditions with 12 hours of light and dark cycle through the experimental period. The animals were provided free access to drinking water ad libitum. Rats were grouped into 10 for control and test animals.

Rats were segregated into control and treated groups containing ten animals each with the following experimental design. The treatment period was 45 days daily. Group I control rats were given distilled water as vehicle orally, daily Group II Aluminium chloride treatment .Rats were given aluminium chloride dissolved in distilled water at a dose of 50 mg/Kg body weight , orally. Group III Vitamin E treatment. Rats were given Vitamin E alone orally at a dose of 20mg/Kg body weight daily. Group IV aluminium chloride with Vitamin E treatment. Rats were treated with aluminium chloride at a dose of 50mg/Kg body weight orally along with Vitamin E (20mg/Kg body weight). At the end of the 45th day the animals were anesthetized and the testis

tissue were taken and rinsed in ice cold 1.15% KCl solution, precut into small pieces and taken for homogenization with a Teflon pestle in the appropriate buffer to obtain 10% (W/V) tissue homogenate through the homogenization process, the tissue homogenate was maintained on crushed ice in an ice bucket. The tissue homogenate was then centrifuged in a refrigerated high-speed centrifuge at 4º C and at 10,000×g for 30 minutes. The clear supernatant obtained from each tissue homogenate was used an enzyme source for the investigation. Acidphosphatase[8], Alkalinephosphatase[9], Succinate dehydrogenase[10,11], Lactate dehydrogenase[12]. Lipid peroxidation was measured[13].

Statistical analysis:

Results are expressed as Mean ± SEM. The differences between mean value were evaluated by ANOVA followed by unpaired student't'- test, two tailed 'p' value. A difference at $P < 0.05$ was considered statistically significant.

RESULT AND DISCUSSION:

Table-1 shows the lipid peroxidation measured by Malondialdehyde (MDA) level. The aluminium treated group shows statistically significant increase $p<0.01$, compare with control. When Aluminium chloride plus Vitamin-E ($AlCl_3$+ Vit-E) treated group shows protective effect of lipid peroxidation and resume normal level. Administration of aluminium chloride over a period of 45 days treatment, Alkaline phosphatase (ALP) and Acid phosphatase (ACP) activity was decreased significantly with control. When $AlCl_3$+Vit-E was given together it get protected from the toxic effect. The activity of lactade dyhydrogenase (LDH) and Succcinate dehydrogenase (SDH) activity was decreased significantly with control group. When $AlCl_3$+Vit-E was given simultaneously it shows protective effect and the toxicity was reversed.

Lipid peroxidation affects lipids and produce oxidative damage to the cells[14]. Cellular organells like mitochondria, plasma membrane, and lysosome gets damage produced by lipid peroxidation and detrimental to the survival and function of the cells[15]. Aluminium exposure induces oxidative stress in rats[16].Oxidative stress was one of the factor that aluminium induce testicular dysfunction[17].

The results of present investigation indicate that aluminium chloride is toxic to rats and affect the testis[18]. The germ cells use TCA cycle rather than glycolysis .Spermatogonia depend more on glycolysis and whereas spermatocytes are intermediate and also depend on glycolysis. For energy demand the spermatids use TCA cycle as main source[19].

Table 1. Showing the effects of aluminium chloride activity of testicular enzymes and changes in the levels of Malondialdehyde expressed as n moles of MDA/hr/mg/protein

Parameter	Control	$AlCl_3$	Vit-E	$AlCl_3$+Vit-E
ALP	1.79±0.056	1.14±0.042**	1.85±0.061	1.68±0.072
ACP	1.96±0.098	1.37±0.08 ***	2.16±0.082	1.81±0.068
LDH	32.73±1.49	24.35±1.23 **	34.39±1.86	30.74± 2.34
SDH	47.35±2.46	32.75±2.13**	49.24±2.12	41.75±1.76
MDA	18.26±0.876	24.68±1.235 **	19.56±0.943	19.78±1.108

Note *P<0.05, ** p<0.01, ***p<0.001

Values are given as mean ± SEM for groups of 10 animals each. Values are statistically significant at *p< 0.05. $AlCl_3$ treated rats were compared with control rats.

The ALP activity is correlated to the spermatogenic mitosis cell division and also transport of nutrient glucose. Whereas ACP is located in the sub cellular organelle like lysosome of the Leydig cells. It performs the synthesis of protein by abduction of sex hormones. Alteration in the activity of ALP and ACP may be useful tool in determining the spermatogenic function. Present study there was a decrease in the activity of ALP and ACP when treated with aluminium chloride. Decreased enzyme activities shows testicular degeneration due to suppressed testosterone level showing affected nature[20].Decreased ALP and ACP activities was observed in chromium treated rat liver and kidney and testis[21].

Succcinate dehydrogenase (SDH) is important enzymes of Kreb's cycle, and the qualitative changes occur during pathological condition[22]. SDH is an oxidative enzyme affected by the action of metals. It is a marker enzyme for identifying the presence of TCA cycle in tissues[23]. SDH activity increases entire period of maturation of germ cells[24]. The lactade dyhydrogenase (LDH) is an important role in carbohydrate metabolism and converts the lactate to pyruvate. It is associated with the metabolic activity of the cells and inhibition in enzyme may be due to the damage caused to the plasma membrane. LDH present widely in spermatogenic cells and Sertoli cells, and play an important role in energy production and biotransformation. Inhibition of LDH may cause damage to spermtogenic cells[25]. In this study there is a decrease in LDH and SDH activity. Such a decrease was observed in cadmium treated rats[26] and 3, 4, Dichloro aniline treated rats[27].

Vitamin E is present in nuts and seeds and vegetable oils are the best sources of alpha tocopherols and they are available in green leafy vegetable and cereals[6]. Vitamin E protects the cell constituents from the damaging effect of oxidative stress and free radicals[6]. Vitamin E has been proved to be a powerful antioxidant activities, it prevent oxidation of lipid molecules[28].

CONCLUSION:

Present study reveals that administration of aluminium chloride affects the marker enzymes ACP, ALP, SDH, LDH in rat testis and its activities was decreased by lipid peroxidation and oxidative stress. When the enzyme activities inhibited, it will affect the process of spermatogenesis and may induce infertility. Whereas administration of Vitamin E prevents oxidative stress and the toxic effect was reversed. So, Vitamin-E can be given therapeutically to counteract the toxic effect of aluminium chloride.

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