Volume-4, Issue-4, April-2015 • ISSN No 2277 - 8160

Research Paper | Biotechnology

Antioxidant Effect of Vitamin A & E on Heavy Metal (Cd) Induced Renal, Testicular as Well as Hepatic Injury in Male Albino Mice

Rajesh Ku. Meher	Research Student, P.G Dept. of Biosciences and Biotechnology, Fakir Mohan University, Balasore- 756020
Namita Nayak	Research Student, P.G Dept. of Biosciences and Biotechnology, Fakir Mohan University, Balasore- 756020
Bhaskar Behera	Associate professor P.G Dept. of Biosciences and Biotechnology, Fakir Mohan University, Balasore- 756020

ABSTRACT

Heavy metal toxicity is world threatening problem for human as well as other organism. Heavy metal is widely distributed in environment and some of them occur in food, water, air. Exposure of heavy metal induced oxidative stress and renal and testicular as well as hepatic injury were evaluated in male mice. Present study is desired to find out whether vitamin A & E is able to protect the heavy metal (Cd) stress injury to kidney, liver, testicular abnormality. For these purpose 15 numbers of male albino mice were taken and acclimatization on laboratory condition for about 15 days. 15 mice were divided in to 3 different group each group contain 5 mice first group served as control and was given only distilled water, the second group received distilled water supplemented with Cd 10mg per Kg body weight, third group received distilled water supplemented with 10 mg Cd/kg body weight with Vitamin Cd/kg per day for 90 days. Tissue were collected from both control and treated animals and studied. Mice exposure to Cd show histopathological changes in the liver, kidney, testis but not in Cd supplemented with vitamin A & E

KEYWORDS : Heavy metal, Toxicity, Albino mice, Histopathology, Vitamin A& E

Introduction:

The exploration and exploitation of natural resources using modern technology and the exponential growth of population have inadvertently resulted in the release of varied types and amounts of industrial wastes into the environment. These industrial wastes are complex mixtures of several classes of pollutant such as hydrocarbons and heavy metals. These have contributed immensely to the heavy metal load in the environment. Heavy metals also bio accumulate in one or several

Compartments across food webs as shown by several scientific observations. Among several elements of periodic table, there are 35 metals are associated with commonly and occupational exposure. Out of these 23 are described as heavy metals. Presently, there is steady increase in their concentration in all habitats owing to mining, electroplating, plants and dye, battery making industries etc. are the release is rapid in rapidly growing technology and heavy metal application in these industries. Heavy metal become toxic when they are not metabolized by the body and accumulated in tissue .Out of the several heavy metal in the industries waste streams, cadmium is reported to be associated with the effluents of battery, electroplating and metal finishing, mining and meteorology and paints and dye industries. Most of the heavy metal exhibit toxicity through the formation of coordination complex and cluster in the animal cells. Cadmium is considered as one of the most toxic heavy metals. It is a non essential element to living organism.

Water is mainly effect by diluted cadmium due to contamination of river and other water sources. I t is important to note that cadmium is highly toxic element for all mammals. Cadmium levels have constantly been increasing, and consequently, the research on cadmium has become quite topical and urgent.

In mammals cadmium can cause a number of structural and morphological changes in various organ as well as genomic level. The present study aimed to investigate that toxicological effect of heavy metal (Cd) on mammalian system (albino mice)

Materials and method:

Albino mice were brought from them/s. Scientific Trader, Balasore, Orissa. All animal were acclimatized for 15 days before starting the experimental procedure. The mice were assigned randomly to either a control or test group and were housed in individual cages (19 x 19 x 12 cm) with solid plastic side and stainless-steel grid tops and floors in a room with temperature approximately 30 ±1 , 60-75 humidity and a 12h light/dark cycle.

III.3 Study design

The genotoxicity assays were performed at the department laboratory. Animals were kept in the cage and were fed regularly with balanced diet. Tissue analysis was performed by bone-marrow chromosome preparation and protein assay method. The protocol was approved by the animal Ethics committee of the Faculty of Fakir Mohan University, Balasore. 15 mice were acclimatized for 15 days before starting the experimental procedure. The mice were assigned randomly to either a control or test groups. Mice were feed with balanced diet which consists of bun cake, carrot and cabbage. Sanitation was maintained properly inside the cages .The cage was cleaned properly twice a week.15 mice were divided to3 groups each groups contain 5 numbers of mice 1st group which contain 5 mice were taken as control was given only distilled water. The second group received distilled water supplemented with 10mg Cd/Kg body weight per day orally, 3rd group received 10mg Cd/Kg with Vitamin A& E orally per days for 90 days .Tissue were collected from both control and treated animals by sacrificing the mice.

TABLE 1: study design

No. Of mice	Control Mice 1st group	Treated with 10mg $CdCl_2$ 2nd group	Treated with CdCl2 with vitamin A& E. 3rd group
1	No treat		
2	No treat		
3	No treat		
4	No treat		
5	No treat		
6		Treated with Cd	
7		Treated with Cd	
8		Treated with Cd	
9		Treated With Cd	
10		Treated With Cd	
11			Cd+vit A& E
12			Cd+vit A&E
13			Cd+vit A& E
14			Cd+vit A& E
15			Cd+vit A& E

Histological preparation:

For sectioning tissue (liver, kidney &testis) were collected from mice after sacrifice. Then the tissue were cut into small pieces and washed

with physiological solution and immerged in fixative (Bouin's fluid) for 24 hours. Then the tissue was washed in running tap water for 24 hours till removal of fixative. Then tissue were dehydrated through upgrade of alcohol, Viz, 30%, 50%, 75%, 90% and 100% then the tissue were transfer to malted paraffin wax and section cutting was done by using microtome in thickness of 6µm, grease free slide were taken, one surface of slide was marked with diamond pencil. A droplet of Mayer's albumin was smeared on the marked surface, a thin film of water was put on the slide and paraffin ribbons with section were placed on it. Then the slide was heated on a hot plate and the sections were properly stretched. The water was drained off and the slides were dried on hot plate. Then the slide was transferred to absolute alcohol 100%, 90%, 70%, 50%, 30% each for 15-20 minutes. Then transferred to distilled water. Section of tissue were usually stained with two dyes, means double staining haematoxylin and eosin. At first the slides were stained with haematoxylin then put on running tap water for removal of excess stain. Then the slides were transferred to acetic water, then from acid water to distilled water and finally transferred to alcohol of different concentration (30%, 50%, 70%, 90%). After this the slides were stained with another stain i.e. eosin from this stain the slides were transferred to absolute alcohol 100%. Then a small amount of DPX as putted on the slide, depending on the size of cover slip.

Results and Discussion

. 1 Results

The oral administration at different concentration of cadmium to Mice shows many symptoms of toxicity such as cellular abnormalities, deposition of heavy metals on tissues, tumour formation, paralysis and death.

Histopathological changes:

Histopathological changes were observed in the liver, kidney & testis for different treated groups. Lesions were essentially similar for all the treatment & the exposure time, although the intensity of cell damage increases with the increasing concentration & the time of exposure.

Exposure of CdCl2 to liver:-

Normal liver cell of climbing perch no pathological lesion observed in control & Vitamin with $CdCl_2$ in Albino mice. There are disarrangement of hepatic cell, necrosis & thickness of hepatic cell was observed in 10 mg/kg bw which is highest dose given to the climbing perch, showed the necrosis & faded hepatic cell in the liver.

Exposure of CdCl2 to kidney:-

In 10mg kg bw of CdCl2 with Vitamin A& E to kidney cells does not show any changes, but in 10 mg/kg bw only $CdCl_2$ cause the cell shrinkage takes place, black patches & degeneration of cell was observed.

Exposure of CdCl2 to testis:-

Exposure of 10mg/ kg bw show hemorrhagic, interstitial edema degeneration of somniferous tubules of testis, but no changes occur in control and treated with vitamins like A& E on Albino mice.

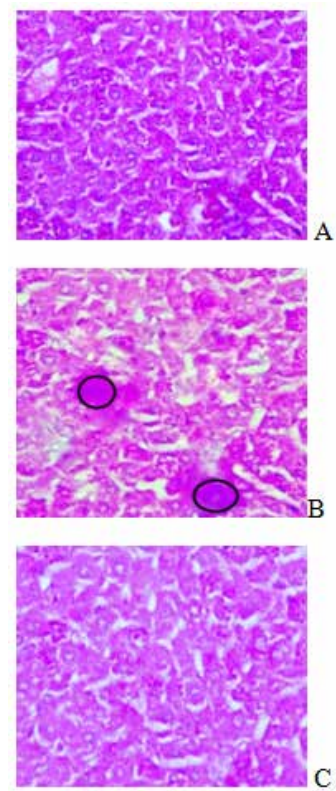

Fig: A control of liver tissue stained with Hematoxylin & eosin , B treated with 10 mg $Cdcl_2$,marked region shows cellular abnormality C treated with $CdCl_2$ with vitamins A& E.

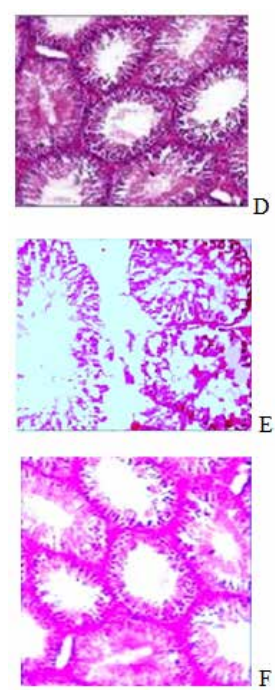

Figure 4: D control of testis & E treated with 10 mg $CdCl_2$ / kg b.w .fig shows hemorrhagic, interstitial edema degeneration of somniferous tubules, but control is almost normal and fig F treated with both Cd and Vitamins shows no toxic effect in testis. Stained with H& E

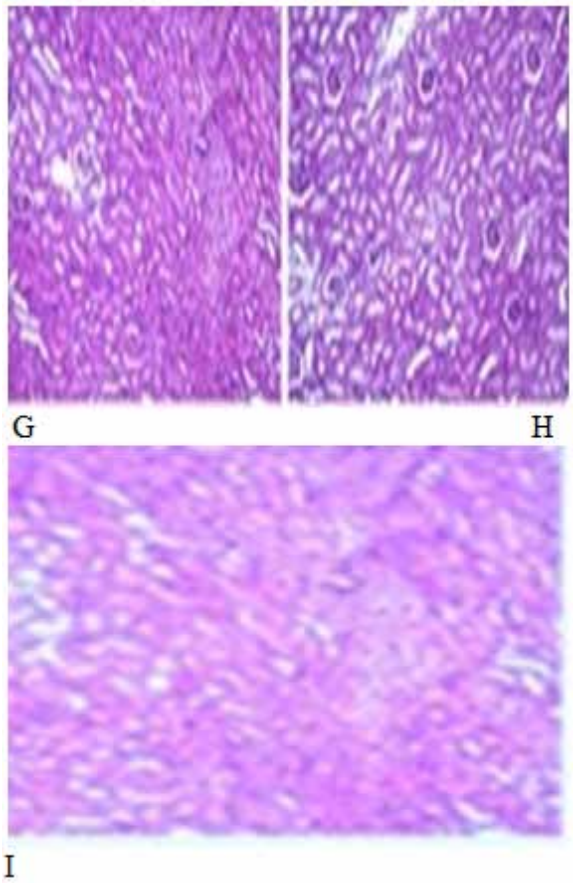

Fig: G control, H treated with 10 mg $Cdcl_2$/Kg b.w, I treated with 10 mg $CdCl_2$ with vitamins. Stained with H& E.

Discussion:

Exposure of heavy metal (Cd) is common, because exploration and exploitation of natural resources using modern technology and the exponential growth of population have inadvertently resulted in the release of varied type and amounts of industrial wastes and heavy metals into the environment. The heavy metals are release into environment by process like weathering of rocks, volcanic eruption mining and exposure through water, air and food. Hence their exposure to population is inescapable consequences. Exposure of Cd cause histopathological changes like cell shrinkage, cell necrosis due to free radical production.

In mice from the above analysis it was observed that long term exposure to Cd shows cellular abnormalities, tumour formation and death of the animals can protect by administration of vitamin A & E. Due to similar physiology of human and mice so it may be work on human

Conclusion:

Prolong exposure of the heavy metals like $CdCl_2$, caused tissue as well as cellular abnormality which leads to tumour formation, which can be control by administration of vitamin like A & E due to its antioxidant properties. Histopathological changes increase with increase concentration of the cadmium chloride which can be controlled by antioxidant compound. Thus the present study suggests that anti oxidant activity of Vitamin A and E can protect from Cd toxicity in Albino mice.

Acknowledgement:

We complete this work by the kind co-operation, valuable suggestion and proper guidance of Dr. Bhaskar Behera faculty of P.G Dept. of Biosciences and Biotechnology, Fakir Mohan University and also valuable suggestion of Dr. Bisnu Prasad Dash, Dr. Bhabatosh Mittra and Dr. Biswajit Ratha, faculties of Fakir Mohan University P.G Dept. of Biosciences and Biotechnology. So we convey our deep sense of gratitude and healthy thanks to all of them.

REFERENCES

1. Abdel-Moneim A.M, Said K.H. Acute effect of cadmium treatment on kidney of rats: biochemical and ultrastructural studies. Pak.J.Bio.Sci. 2007; 10: 3497-3506 | 2. Al- Madani et.al, renal toxicity of mercuric chloride at different time intervals in rats.Biochem.Insights. 2009. | 3. Chukwu, 1991. Bio accumulation of heavy metal on food web. | 4. IARC: overall evaluation of carcinogenicity: an updating of IARC Monographs volumes 1 to 42. 1987. |