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A Study on Radiation and Cadmium Induced Biochemical Changes In The Jejunum of Mice

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Abstract

Exposure to ionizing radiation increases the production of the reactive oxygen species (ROS) leading the irradiated cells into a state of oxidative stress. Furthermore, cadmium exposure along with ionizing radiation can potentially become toxic to the tissues due to the heightened oxidative stress. In the present study adult male Swiss albino mice were procured and divided into four groups. Group (II to IV) received sub lethal dose (1.25, 2.5 or 5.0 Gy) and /or cadmium chloride (20ppm) in drinking water *ad libitum*. Sham- irradiated animals of Group I served as normal. Animals of all the groups were autopsied at each post treatment interval of 1, 2, 4, 7, 10, 14 and 28 days. The value of total proteins, glycogen, cholesterol, DNA, RNA, Acid & Alkaline Phosphatase activities were observed in the form or increase or decrease. After the combined exposure of gamma radiation and cadmium, the damage observed were more than individual exposure of radiation or cadmium chloride indicating "synergistic "effect. Process of recovery started on day-14 after the combined treatment but the process was slow. The biochemical alterations were found dose dependent.

Keywords: Gamma Radiation, Cadmium chloride, Swiss albino mice, Jejunum.

Introduction

Radioactive materials that decay spontaneously produce ionizing radiation, which has sufficient energy to strip away electron, from atoms (creating two charged ions) or to break some chemical bonds. Any living tissue in the human body can be damaged by ionizing radiation in a unique manner. The body attempt to repair the damage, but sometimes the damage is of a nature that cannot be repaired or it is too severe or widespread to be repaired. Also mistakes made in the natural repair process can lead to cancerous cells. The exposure of mammals to ionizing radiation such as gamma radiation can cause the development of a complex dose dependant series of potentially fatal physiological and morphological changes. Oxidative stress contributes to normal tissue damage during tumor therapy with radiation[1]. The most common forms of ionizing radiation are alpha and beta particles or gamma and x-rays. In general, the amount and duration of radiation exposure affects the severity or type of health effect.

Intestine is the most sensitive tissue in the gastrointestinal tract mainly because its cell population in the villi is dynamic and under normal condition it is in a steady state. The cells are produced in the crypts, migrate up in the villi, and are finally sloughed off from the tips off villi, the immature cells and cells in active division are especially sensitive to irradiation. It is their presence is plenty in the intestine, which makes it relatively more radiosensitive. The intestinal epithelium is a cell renewal system, consisting of cells with different sensitivity. Crypts have been reported to be more vulnerable than the vilus epithelium. The extensive cell loss in the intestinal crypts after irradiation is responsible for morphological, biochemical and functional changes[2]. In view of the above, the present study was planned to evaluate the radiation and cadmium induced changes in the jejunum of Swiss albino mice.

Review of Literature

Heavy metals were considered to pose major health problems only to workers involved in mining and processing of ores and in industries using metals. Metals are inhaled primarily as dusts and fumes. Metal poisoning can also result from exposure to vapors (e.g., mercury) vapor in

the manufacture of fluorescent lamps. When metals are ingested in contaminated food or drink or through hand to mouth activity (implicated especially often in children), their gastro-intestinal absorption varies greatly with the specific chemical form of the metal and the nutritional status of the host. Once a metal is absorbed, blood is the main medium for its transport, with the precise kinetic dependent on diffusibility, binding forms, and rate of biotransformation, availability of intracellular legends, and other factors. Most metals are excreted through salivation, perspiration, exhalation, lactation, skin exfoliation, and loss of hairs and nails. Some metals, such as copper and selenium are essential to normal metabolic function as trace elements[3]. The metallic toxicants and radiations when administered simultaneously may prove the more disastrous to living beings. To understand the effect of cadmium and radiations, intestine study can provide vulnerable information. The changes were more severe in the combined treatment groups showing synergistic effects.

Absorption of cadmium from the gastrointestinal tract appears to be a saturable process with the fraction absorbed decreasing at high doses[4]. It is also important to distinguish true absorption from simple retention of cadmium in the microvilli of the small intestine[5]. Little is known about the mechanism involved in intestinal uptake of cadmium in mammals. The metal is primarily absorbed across the brush border of duodenum. Due to electrostatic attraction, cadmium is bound to the luminal surface of the mucosa cell. Uptake across the brush border membrane is not an active process since the metal is taken into the mucosa cell by a process related to membrane fluidity[6]. Cadmium is rapidly complexes by metallothionein once it enters the mucosal cells.

The effects of gamma radiation on small intestine of Uromastix exposed to 500 and 1000 r of gamma ray from a Co ^{60} source has also been studied. The radio- lesions were reported in the form of disarrangement of cells in the intestinal mucosa and appearance of dead cells at the base of villus folds. The lamina propria showed hydropic degeneration, muscular coat and sub mucosa appeared oedematous. In both groups, the number of goblet cells decreased after irradiation[7].

The mucosal permeability in irradiated rectum and non-irradiated sigmoid colon from patient subjected to radiation therapy has been reported. The passage of all markers was increased in irradiated rectum compared with non-irradiated sigmoid colon, whereas, in specimens from non-irradiated patients there were no differences between rectum and sigmoid colon[8].

The biochemical alterations in mouse testis after the combined treatment of radiation and cadmium chloride have been investigated. They exposed the animals with 1.25, 2.5 and 5.0 Gy of gamma radiations with or without cadmium chloride treatment. The values of acid phosphatase activity, alkaline phosphatase activity increased and sia lic acid concentration found decreased. The alterations in the values were found to be dose dependent and the more severe changes were observed in the combined treatment group showing additive or synergistic effects[9].

Materials and Methods

Procurement of Animals and Their Maintenance

In the present study, adult healthy male Swiss albino mice (6-8 weeks old) were purchased from Lala Lajpat Rai University of Veterinary and Animal Sciences, Hissar (India). The animals were housed in cages under good ventilation and illumination condition. They were fed with standard mice feed and water was given ad libitum. The temperature of the room was maintained between 22-27ºC. The Govt. Dungar College, Bikaner is registered under CPSCEA, Chennai (Registration no. 1066/ac/07/CPCSEA) and has its own Institutional Animal Ethics Committee (IAEC). All the experiments conducted in the present investigation were performed strictly under the supervision of IAEC of the college.

Cadmium

Cadmium salt in the form of cadmium chloride was procured from Ranbaxy Laboratories Ltd. Cadmium chloride was given in the drinking water at the dose of 20 ppm.

Source and Procedure of Irradiation

Cobalt-60 gamma radiotherapy source (Theratron) of AECL make, obtained from Canada was used to expose the animals. This facility was provided by the Radiotherapy Department of Prince Bijay Singh Memorial Hospital, Bikaner (Rajasthan). The animals were irradiated at the dose rate ranging between 0.97 Gy/min to and 1.97Gy/min.

Experimental Design

In the present study, the animals were grouped as under

Group I : (Sham-irradiated animal-normal)
Group II: (Cadmium chloride treated animals)
Group III: (Only irradiated animals)
Sub-group IIIa: 1.25 Gy
Sub-group IIIb: 2.5 Gy
Sub-group IIIC: 5.0 Gy
Group IV: (Animals treated with radiation and cadmium chloride)
Sub-group IVa: 1.25 Gy + Cadmium chloride
Sub-group IVb: 2.5 Gy + Cadmium chloride
Sub-group IVc: 5.0 Gy + Cadmium chloride

Parameters

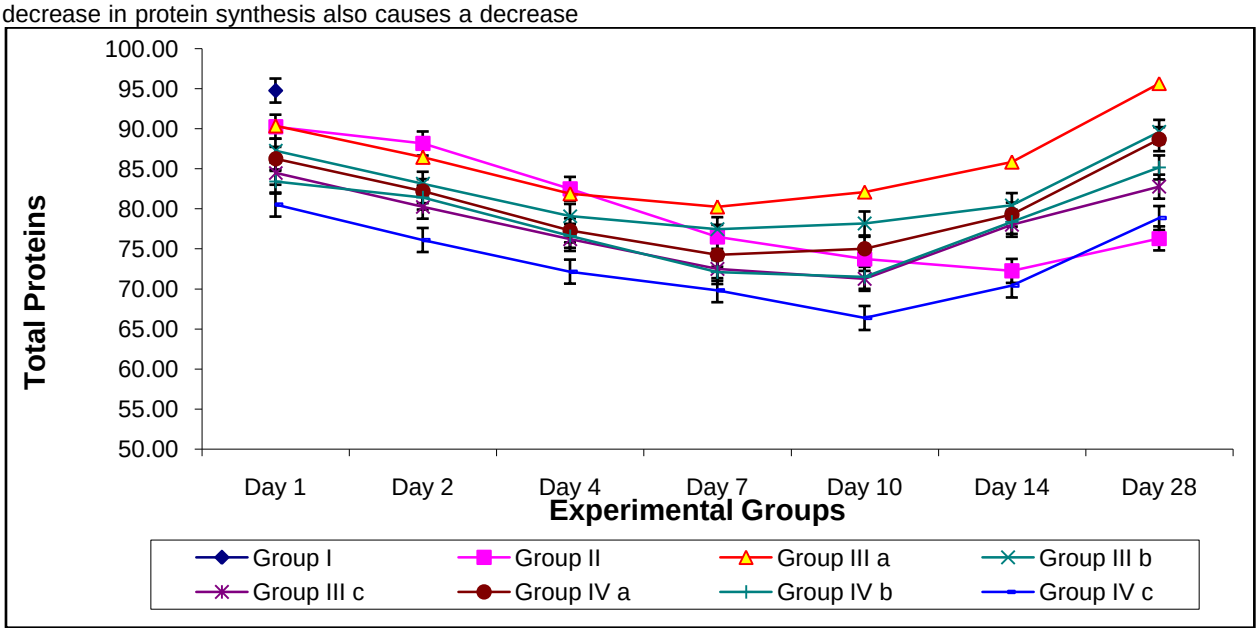
In the jejunum, total proteins[10], glycogen[11], cholesterol[12], DNA[13] and RNA[14] Acid & Alkaline Phosphatase activities[15], were studied.

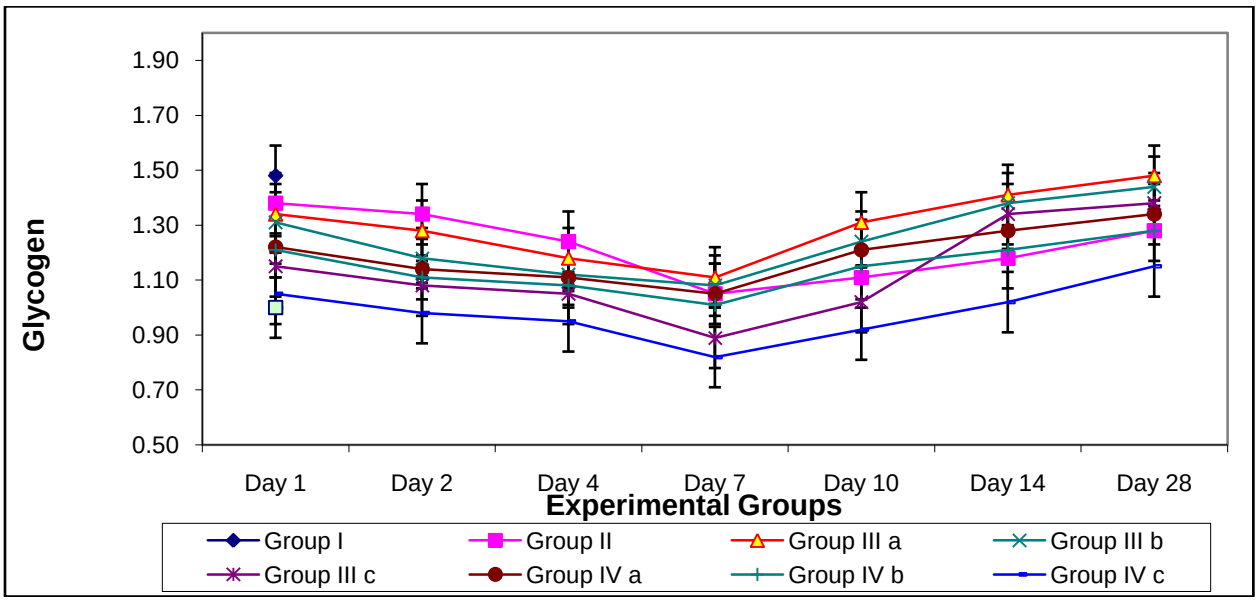
Result and Discussion

The values of total proteins, glycogen, cholesterol, DNA and RNA decreased up to day 10 in non-drug-treated groups and up to day 7 in the polybion-treated groups, and then increased in all the groups. The values of acid phosphatase and alkaline phosphatase activity increased up to day 14 in non-drug-treated groups and day 7 in the drug-treated groups, then declined in all groups.

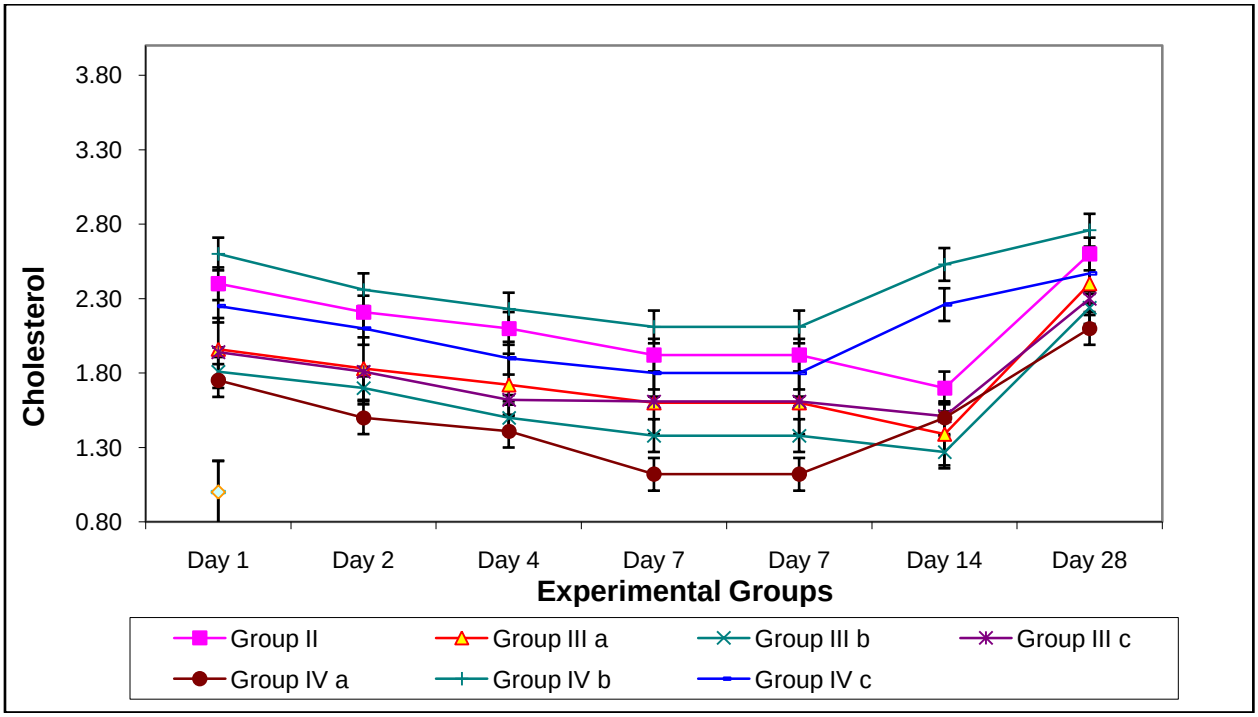
Severe changes were observed after combined exposure, showing a synergistic effect. The

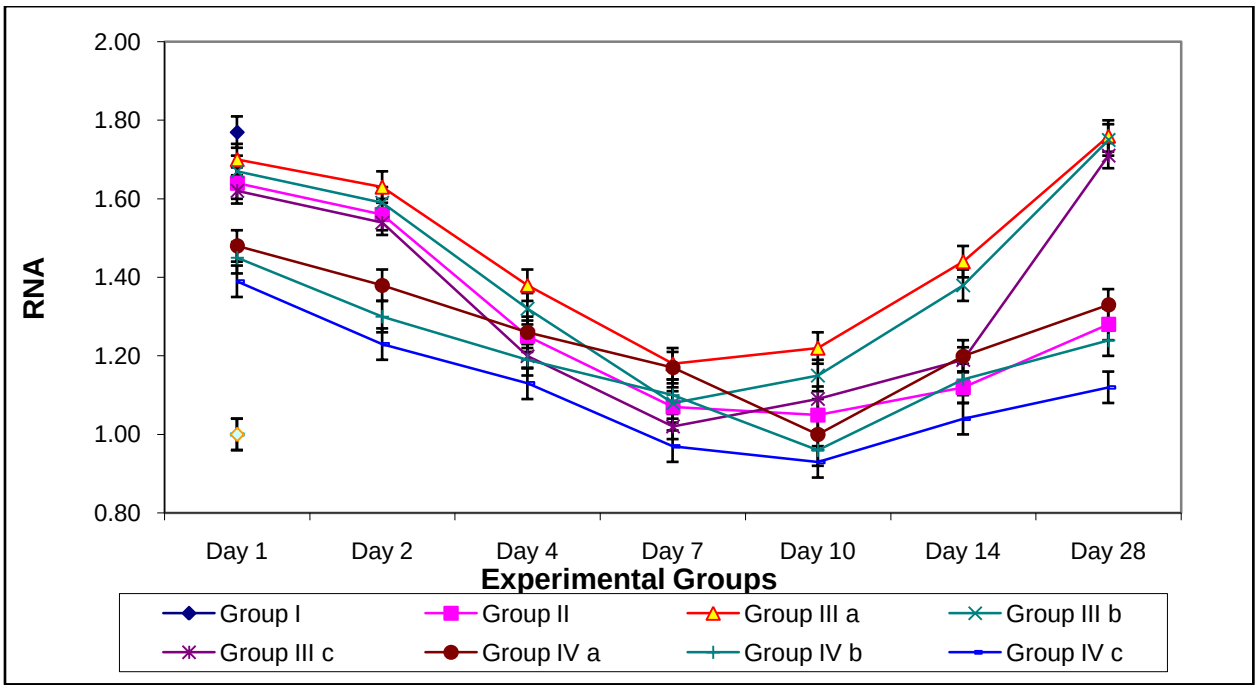
changes were found dose dependent. The decline in protein values reflects the combined effect of a decreased number of mature cells entering the villi and the presence of the damaged cells on the villi. The low values of total proteins corresponded to a decrease in the cellularity of intestinal epithelium. A decrease in protein synthesis also causes a decrease in the mitotic index of tissues[16]. The depletion in glycogen has been noticed for a number of chemicals present in the environment[17]. The decrease in glycogen level in the present study as a result of cadmium chloride treatment has also been supported by previous workers[18,19].



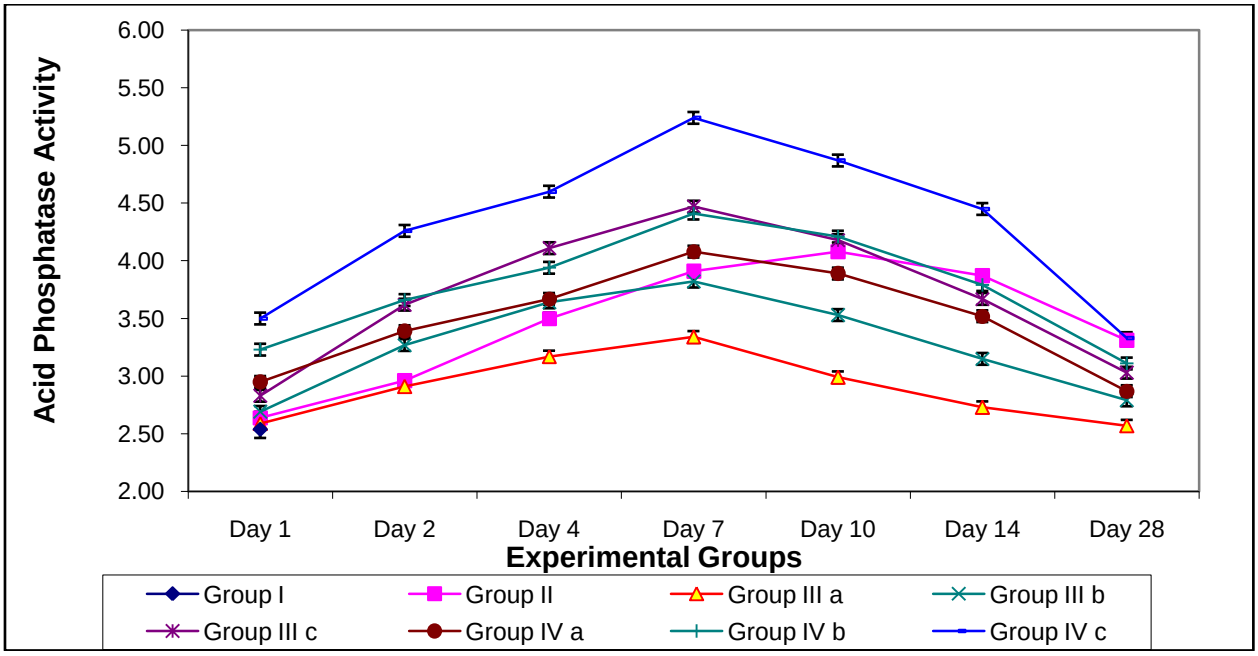


It has also been stated that although the total cholesterol decreases after whole body X-irradiation in rats, there is a great increase in the hepatic cholesterol biosynthesis[20, 21]. This suggested that the cholesterol is either excreted or destroyed by other means, causing a decrease in tissue cholesterol level. It has also be shown that post-irradiation acute cell death could lead to a loss of DNA in excess of that normally eliminated from the tissues. The prolonged interphase or delayed onset of DNA synthesis after irradiation could also lead to decreased DNA content[22]. Although irradiation disturbs RNA metabolism, it is less sensitive in comparison with DNA metabolism[23]. It was reported that when cells died and were lost due to irradiation, the intestinal RNA decreased[24].





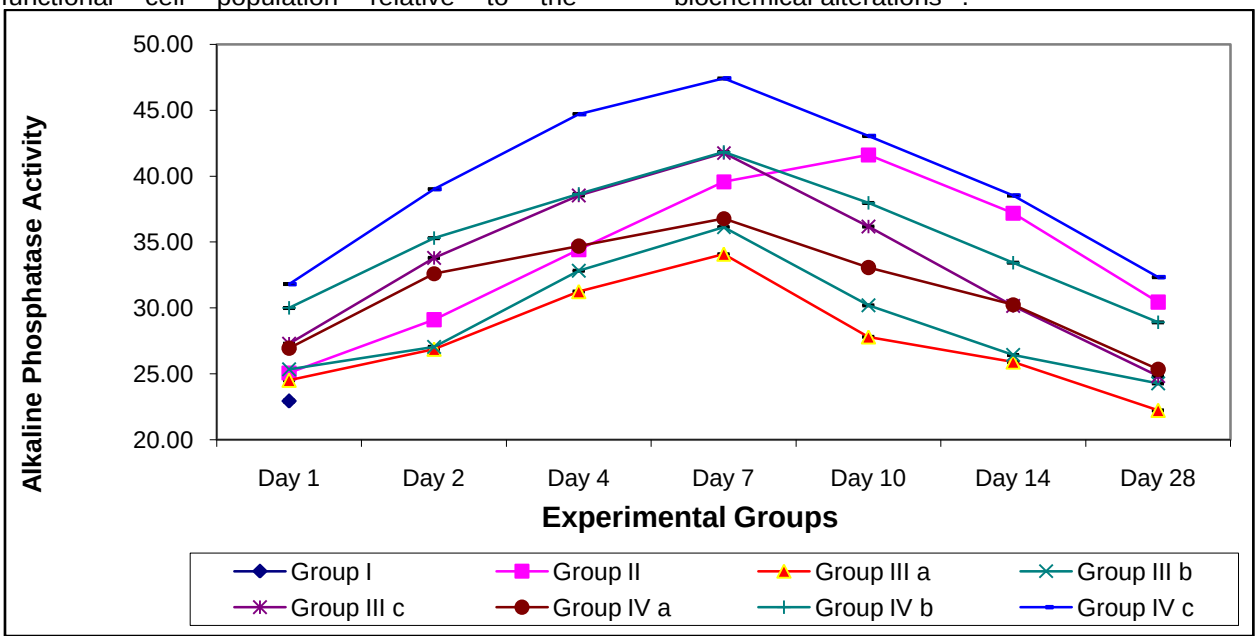
In present study, cadmium chloride treated group showed increased value of acid phosphatase activity at early intervals and this increase continued up to day-10 in lower dose groups and day-14 in the higher dose groups. The last autopsy interval (i.e.day-28) showed decline in value of acid phosphatase activity. Acid phosphatase is a lysosomal enzyme and is non-specific phosphomonoestrase. It helps in the autolysis of cells after death. It hydrolyses various phosphate esters and liberates phosphate. Heavy metals induce cellular damage in the tissue that in turn releases lysosomal enzymes thereby increasing the acid phosphatase activity[25]. The cellular damage might cause rupture of lysosomes and hence phosphatase activity increases due to heavy metal toxicity. It has been reported that increased acid phosphatase activity seems to be characteristics of tissue damage by radiation[26]. Lysosomal hydrolases are thought to contribute to the degradation of damaged cells, hence facilitate their replacement with normal tissue[27].



Alkaline Phosphatase Activity

Alkaline phosphatase is an enzyme present in many tissues. In intestine alkaline phosphatase activity is located mainly on the brush border and Golgi zone of the villi epithelial cells[28] Our observation in group II showed an increase in the values of alkaline phosphatase activity continuously up to day-10 in lower dose groups and day-14 in higher dose groups, thereafter, it decreased on day-28. The increased value of the alkaline phosphatase activity shows good agreement with the observation of previous workers[29,30].The possible reasons for increase may be due to increase in synthesis of enzyme, increase in the functional cell population relative to the proliferative epithelial cells; as the latter decrease rapidly due to the radiation induced cell death, disruption of lattice like structure of phospholipids and hydrogen bonds that separate enzymes from their intracellular substrates and altered physiological conditions, such as liver function, mediated by the alkaline phosphatase activity[31,32].

Both radiation and cadmium also induce damage to cell organelles by altering membrane permeability and enzymatic activity because both attack on –SH groups of proteins and reduce glutathione (GSH) level which is a natural ligand providing protection to the tissues. This lowered level of GSH may be responsible for the biochemical alterations[33].



Conclusion

The changes in the biochemical parameters were found dose dependent. More pronounced biochemical changes were registered after the combined exposure of cadmium chloride and gamma radiation. Thus it can be deduced that radiation and cadmium exert synergistic effect on the jejunum of Swiss albino mice when administered together.

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References

1. *Singariya, S., Songara, V., Agarwal, M., Chakrawarti, A. and Purohit, R.K., Radioprotective potential of Aloe vera against radiation and cadmium mediated alterations in differential leucocyte counts in Swiss albino mice. Int. J. Adv. Res., 3 (7): 1293-1593 (2015).*
2. *Pyarelal, Role of protection provided by Emblica officinalis Linn. against radiation and lead induced changes in the jejunum of Swiss albino mice. A Ph.D. thesis submitted to M.G.S.University, Bikaner (2015).*
3. *Goyer, R.A. and Cherian, M.G. (1979): Ascorbic acid and ethyl endediamine tetra acetic acid treatment of lead toxicity inrats. Life Sci., 24: 433-438.*
4. *Nordberg, G.F., Kjellstrom, T. and Nordberg, M. (1985): Kinetics and metabolism. In : Friberg, L., Elinder, C.G., Kjellstrom, T.and Nordberg, G.F. eds. Cadmium and Health : A toxicological and metabolism. CRC Press, Boca Raton, FL. 103. (Cited in ATSDR).*
5. *Foulkes, E.C. and McMullen, D.M. (1986) : Endogenous metallothionein as determinant of intestinal cadmium absorption : A re-evaluation. Toxicology, 38: 285.*
6. *Handy, R. D. (1996): Dietary exposure to toxic metals in fish. In E. W. Taylor, editor. Toxicology of Aquatic pollution: Physiological, Cellular and Molecular Approaches. Society for Experimental Biology, Seminar Series 57. Cambridge University Press, Cambridge. pp 29*
7. *Gupta, M. L. and Umadevi, P (1980) : The effect of gamma irradiation on the small intestine of Uromastix. Proc. Natl. Academy of sciences, India. Golden Jubilee session, Allahabad.*
8. *Nejdfors, P., Ekelund, M. and Weström, B.R. (2000): Intestinal permeability in humans is increased after radiation therapy. Disease of the Colon & Rectum. 43(11): 1582-1587.*
9. *Verma, C.L., Dev, K., Gupta, M. L. and Purohit, R.K. (2014): Biochemical alterations in mouse testis after the combined Treatment of radiation and cadmium chloride. Int. J. Adv. Res. 2 (6): 340-344.*
10. *Lowry, R.O. and Baker, O.G. (1972) : Protection against local irradiation injury to skin by locally and systemically applied drugs. Radiol. 105 : 425.*
11. *Montogomery, R. (1957) : Determination of glycogen. Arch. Biochem. Biophy., 67 : 387.*
12. *Oser, B.L. (1965) : Hawk's physiological chemistry, McGraw Hill, New York, 246.*
13. *Ceriotti, G. (1952): The microchemical determination of deoxyribonucleic acid. J. Biol. Chem., 198: 297.*
14. *Ceriotti, G. (1955): Determination of nucleic acid in animal tissue. J. Biol. Chem., 214: 59.*
15. *Fiske, C.H. and Subbarow, Y. (1925) : The colorimetric estimation of phosphates. J. Biol. Chem., 66: 375.*
16. *Baijal, K. (1978): Biochemical and histopathological effects of external irradiation of small intestine of mammals. Ph. D. thesis, University of Rajasthan, Jaipur, Rajasthan (India).*
17. *Ravdin, I. S., Vars, H. M., Goldschmidt, S. and Lingensmith, L. F. (1968): J. Pharmacol. Exptl. Ther., 64: 111.*
18. *Hodson, P. V. (1976): Temperature effects on lactate glycogen metabolism in Zinc-intoxicated rainbow trout. Salmo gairdneri. J. Fish Res. Bd. Can., 33: 1393.*
19. *Rana, S. V. S., Prakash, R., Kumar, A. and Sharma, S. B. (1985): Toxicol. Letters, 29: 1.*
20. *Gould, R. G., Loth, L. V. and Lilly, E. M. (1956): The effects of X-irradiation on hepatic cholesterol synthesis. Fed. Proc., 15: 264.*
21. *Gould, R. G. and Cook, R. P. (1958): Cholesterol metabolism in liver, In "Cholesterol"ed. Cook R. P., Pub. Acad. Press, Inc. Pub. N. Y. pp 145.*
22. *Nygaard, O.G. and Potter, R.M. (1960): Effect of X-irradiation on DNA metabolism in various tissue of rat. II RECNERY after sub lethal doses of irradiation. Radiat. Res., 12: 120.*
23. *Toal, J. N., Reid, J. C., Williams, R. B. and While, J. (1958): Effects of total body X-irradiation from near threshold to tissue lethal doses on small bowl epithelium of the rat. II/ Changes in nucleic acid and protein synthesis in relation to cell division. J. Natl. Cancer Inst., 21: 63.*
24. *Gerber, G. B., Gerber, G. and Altman, K. I. (1961): The catabolism of tissue nucleic acid. III. The Catabolism of ribonucleic acid after total body X-irradiation. Int. J. Radiat. Biol., 4 : 67.*
25. *Wilson, R., Doell, B.H., Groger, W., Hope, J. and Gellatey, J.B. (1970) : The physiology of liver enlargement in "Metallic aspects of food safety" Pub. Black-well scientific, Oxford*
26. *Wriggles, J.M.W. and Pover, W.F.R. (1967): Hydrolytic enzyme activity in rat's small intestine after whole body irradiation. Int. J.Radiat. Biol. 12: 243.*

27. *De Dave, L. and Watiaux, R. (1966): Function of Lysosome. Ann. Rev. Physiol., 28: 435.*
28. *Webster, H.L and Harrison, D.D. (1969): Enzymatic activities during the transformation of the crypt to columnar intestinal cells. Exptl. Cell Res., 56 : 245.*
29. *Kodama, M., Ogata, T. and Yamamori, K. (1982): Acute toxicity of zinc to rainbow trout Salmo gairdneri. Bull. Jap. Soc. Sci. Fish, 48: 593.*
30. *Sastry, K.V. and Shukla, K.V. (1988): Acute and chronic toxicity of some heavy metals to Heteropneustes fossilis Bloch. Threat. Hab. pp. 321-334.*
31. *Bacq, Z.M. and Alexander, P. (1961): Fundamentals of radiobiology (Pergamon press Ltd, London).*
32. *Tsubouchi, S. and Matsuzawa, T. (1973b): Recovery of function and cell population in the intestinal epithelium of X-irradiated mice. Radiat. Res., 56: 345.*
33. *Gajawat, S., Pareek, T. K. and Goyal, P. K. (2001): Effects of lead and radiation on some biochemical markers and its modification by vitamin E. Journal of Medical Physics, 26: 135.*