IJBPAS, August, 2017, 6(8): 1589-1604

ISSN: 2277–4998

International Journal of Biology, Pharmacy and Allied Sciences (IJBPAS)

'A Bridge Between Laboratory and Reader'

www.ijbpas.com

PROTECTIVE INFLUENCE OF *MORINGA OLEIFERA* AGAINST RADIATION AND MERCURY INDUCED HEPATOTOXICITY IN MICE

PUROHIT RK*, BANOT J, PARIHAR N, VYAS S, BANA MK, MEENA BM, AGARWAL M AND CHAKRAWARTI A

Radiation Biology laboratory, Department of Zoology, Govt. Dungar College, Bikaner, India

***Corresponding Author: E Mail: dr_rajendra_purohit@yahoo.co.in**

Received 6th Nov. 2016; Revised 5th Jan. 2017; Accepted 1st April 2017; Available online 1st August 2017

ABSTRACT

Radiation induced damage and lethality to the normal tissues can be partially reduced by the use of radio-protectors that lower down the damaging effects of radiation. In recent years, extensive research work has been carried out on chemical protection against radiation and heavy metals induced toxicity. Several synthetic chemicals have been tested for their radio-protective action in mammals but their practical applications is found to be limited in various fields owing to their high toxicity at their optimum dose levels.

Therefore, a worldwide hunt is on to find an ideal radio-protective agent for its uses against planned and unplanned radiation exposure. For this purpose Swiss albino mice were divided in various groups. Group I was sham irradiated and served as normal. Group II was given mercuric chloride solution at the dose of 0.5ppm. Group third was exposed to 2.5 Gy of gamma radiations;

Group IV was treated with gamma radiation and mercuric chloride. Group V, VI and VII were given *Moringa oleifera* seven days prior to radiation or mercuric chloride or combined treatment. The animals from all experimental groups were sacrificed by cervical dislocation at each post-treatment interval of 1,2,4,7, 14 and 28 days. After sacrificing the animals, pieces of the liver were taken out and kept at -20^{0}c for various biochemical parameters.

The value of total proteins, glycogen, acid phosphatase & alkaline phosphatase activities and RNA increased whereas the values of cholesterol and DNA declined. Almost normal values were noticed on day-28. After combined treatment of radiation and mercuric chloride the changes were more severe showing synergistic effects. An early and fast recovery in the drug treated groups showing protection provided by the drug.

Keywords: Radiation, Mercury, Liver, Mice, *Moringa*

INTRODUCTION

Discovery of radioactive rays, over a hundred years ago, has proved a landmark in the history of medical research. Extensive studies on radio-exposure have yielded plenty of information on its biological effects on living systems at the molecular, cellular and neuro-endocrine levels. Addition of nuclear armament in war arsenal marks a clear departure from conservative warfare and confronts us with the horrors of atomic warfare. The vast potential of atomic energy, while opening new vistas of a promising horizon, also exposure the global population to hazards of a nuclear accident like the Chernobyl accident that occurred at Ukraine in 1986. Therefore, understanding the biological effects of radiations of human body becomes an essential prerequisite in order to improve measures of adequate protection and management of injury when it inevitably occurs. Most of the organisms are not adapted to deal with high concentration of trace metal, the problem of metal toxicity arises **[1]**. As a result of geological phenomena such as ore formation, weathering of rocks, leaching excessive levels of trace metals may occur naturally. By metabolic active metals are not usually readily detoxified. Thus their release into the environment must be fully monitored and controlled. Following metals have been

identified as potentially hazard to human kind.

Mercury (Hg) is ubiquitously distributed in the environment and is non-essential and toxic to the human body. Mercury is considered to be one of the major environmental pollutants, is widely used in industry, agriculture, and medicine, and circulates in ecosystems, but is never destroyed. Chemically, mercury exists in various forms as elemental (or metallic, Hg^0) mercury, inorganic mercury compounds, and organic mercury compounds. Elemental mercury is liquid at room temperature, and it can be released easily into the atmosphere as mercury vapor because of its high vapor pressure.

In recent times, there has been an increased interest on plant research in exploring their advantageous aspects owing to their natural origin, cost effectiveness and less side effects. One such plant, *Moringa oleifera* Lam. (Sy. *Moringa pterygosperma* Gaertn, Family: Moringaceae) is referred as "Miracle tree" in tropics and sub-tropics. *Moringa oleifera* is coming to the forefront as a result of scientific evidence that *Moringa* is an important source of naturally occurring phytochemicals and this provides a basis for future viable developments **[2]**. Hence, in the present study an attempt has been made to investigate the possible prophylactic role of *Moringa* against radiation and mercury induced biochemical changes in the liver of Swiss albino mice.

MATERIALS AND METHODS

For the present investigation, the adult healthy male Swiss albino mice (6-8 weeks old) were procured from Lala Lajpat Rai University of Veterinary and Animal Sciences, Hissar (India). The animals were irradiated at the dose rate ranging from 0.95 Gy/min to 1.97 Gy/min. The dose was calculated at the midpoint by multiplying dose rate and tissue air ratio. The tissues of Swiss albino mice were assumed to be equivalent to human soft tissues. The mercury salt in the form of mercuric chloride of analytical grade was administered orally in drinking water at the dose of 0.5ppm. The dried powder of *Moringa oleifera* was procured from the Umalaxmi organics private limited, Jodhpur (India) and aqueous extract of the same was obtained in the department. The plant extract of *Moringa* was fed orally at the dose of 150 mg/kg body weight. The *Moringa* extract was given daily from seven days prior to individual or combined treatment of mercuric chloride and radiation and continued up to the last autopsy interval.

The animals were divided into following groups:

The animals of group first were Sham-irradiated and served as normal group. The group second animals were fed with mercuric chloride (0.5 ppm) orally *ad libitum* up to the end of the experiment. The an**i**mals of group third were exposed to 2.5 Gy of gamma rays from Co^{60} source. Group fourth animals were orally fed with mercuric chloride at the dose of 0.5 ppm and also exposed to 2.5 Gy of gamma radiation. The groups fifth animals were orally fed with mercuric chloride at the dose of 0.5 ppm and were also administered *Moringa.* The animals of group sixth were exposed to 2.5 Gy of gamma radiation from Co^{60} source. The animals of group seventh were orally fed mercuric chloride at the dose of 0.5 ppm and also irradiated with 2.5 Gy of gamma radiation. The *Moringa* extract was given orally for seven days prior to irradiation and mercuric chloride feeding till the last autopsy day of the experiment.

Five animals from all the groups were autopsied after 1, 2, 4, 7, 14 and 28 days of the treatment. The animals were sacrificed by cervical dislocation. Prior to autopsy the animals were weighed. Five sham-irradiated mice were also autopsied. Immediately after autopsy, the liver was removed, blotted and weighed on electric monopan balance. The liver was kept at -20ºC for various biochemical estimations. The biochemical parameters taken into consideration were: total proteins **[3]**, glycogen **[4]**, cholesterol **[5]**, acid and alkaline phosphatase activities **[6]**, DNA **[7]** and RNA [**8**].

RESULTS

The value of total proteins, glycogen, acid & alkaline phosphatase activities and RNA increased up to day-14 in the non drug treated groups and declined thereafter till day-28 without reaching to the normal. In the *Moringa* treated groups the value increased up to day 7 and declined thereafter till day 28 but could not reach to the normal level. The value of total proteins and glycogen showed a decreasing trend in the mercuric chloride treated groups second. The cholesterol content and DNA showed a decreasing trend till day 14 in the non drug treated groups and day 7 in the *Moringa* administered groups and increased thereafter till day 28 without attaining the normal level. In the combined treatment group the increase and decrease in the value was more severe showing synergistic effects. An early and fast recovery in the *Moringa* pre-treated groups may be due to the protection provided by the drug.

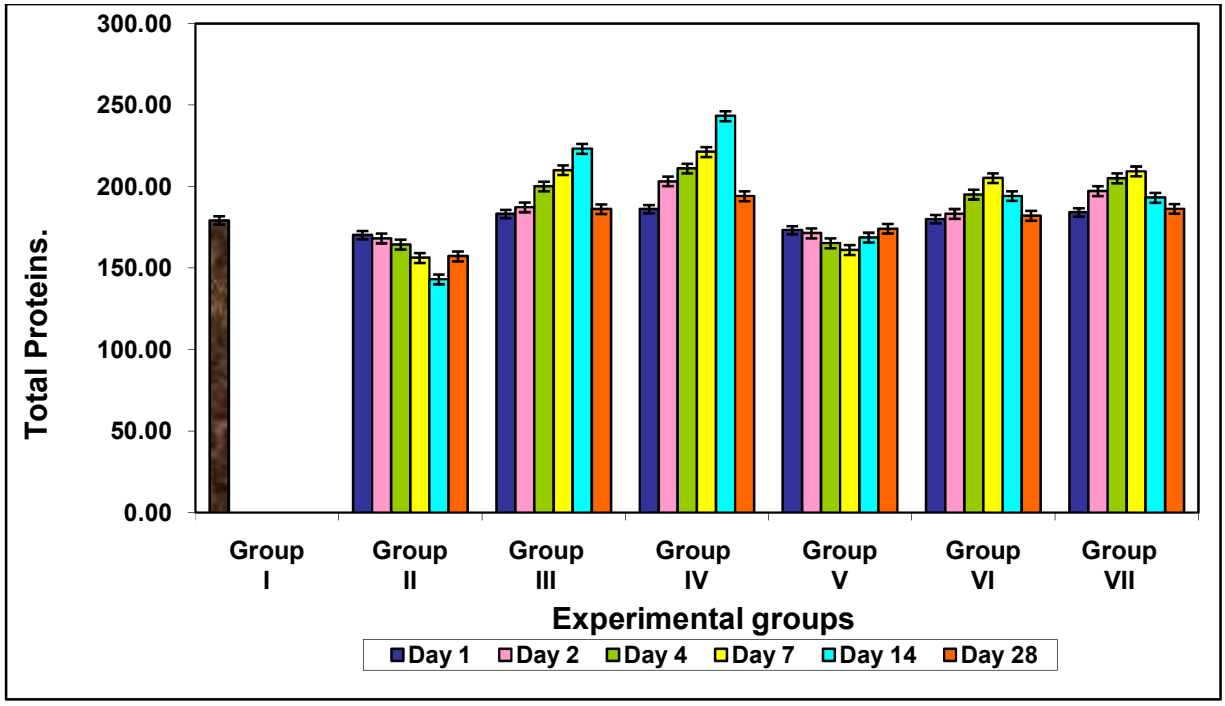

Fig. 1: Total proteins

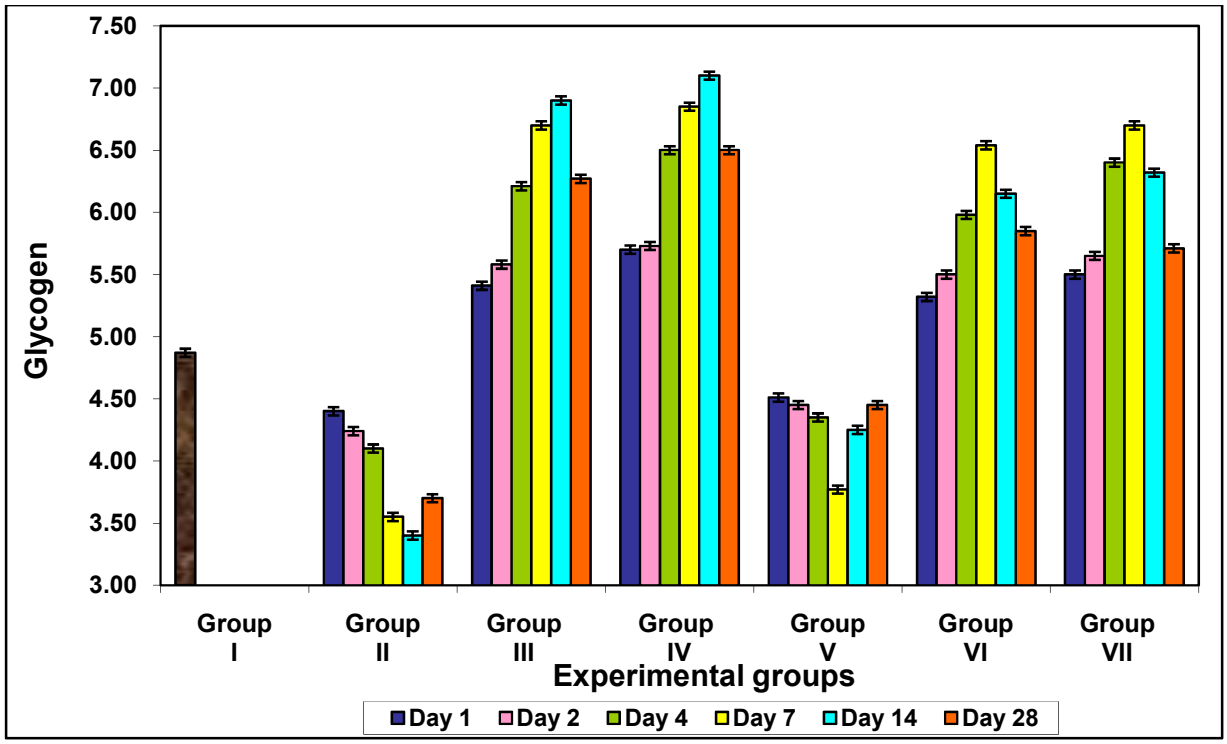

Fig. 2: Glycogen

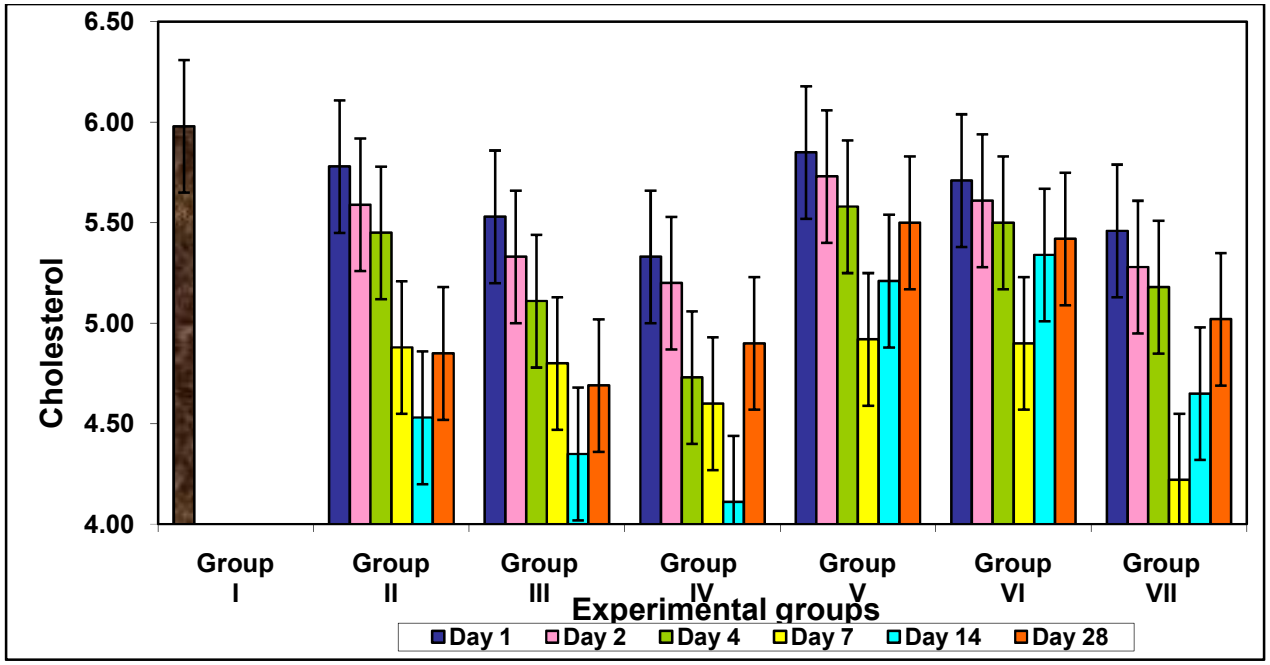

Fig. 3: Cholesterol

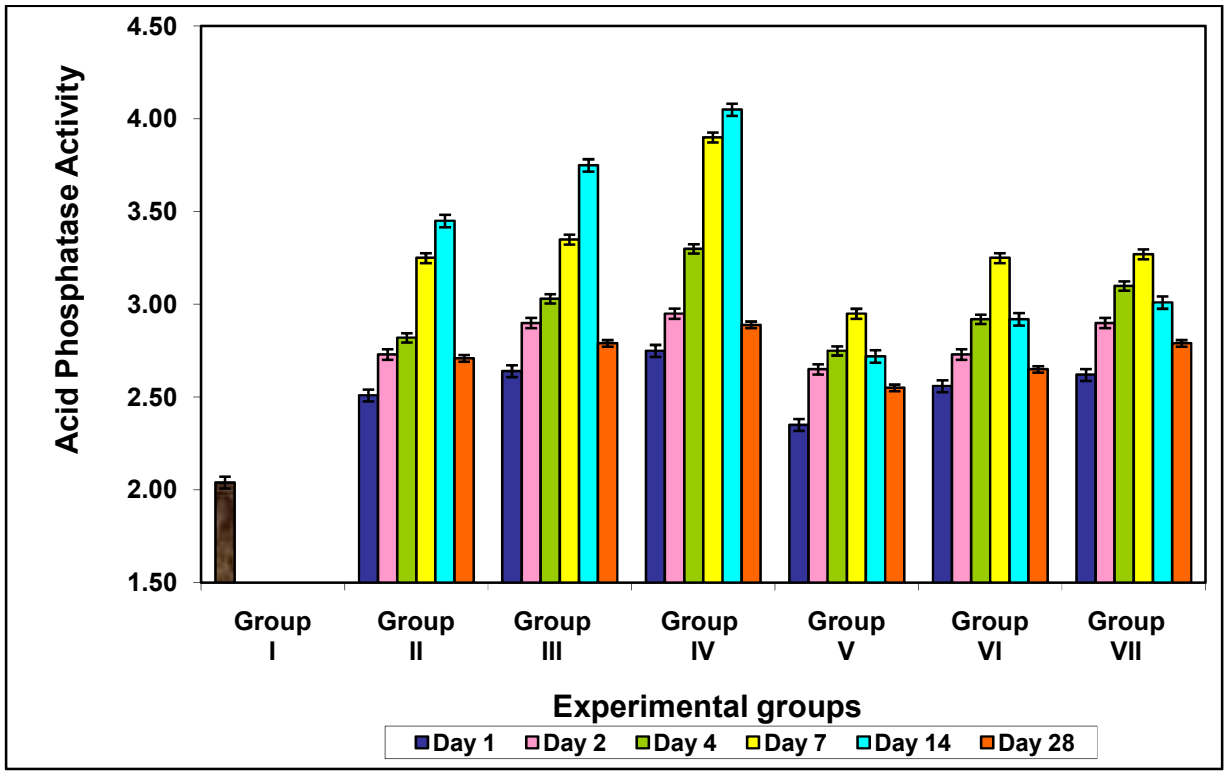

Fig. 4: Acid Phophatase Activity

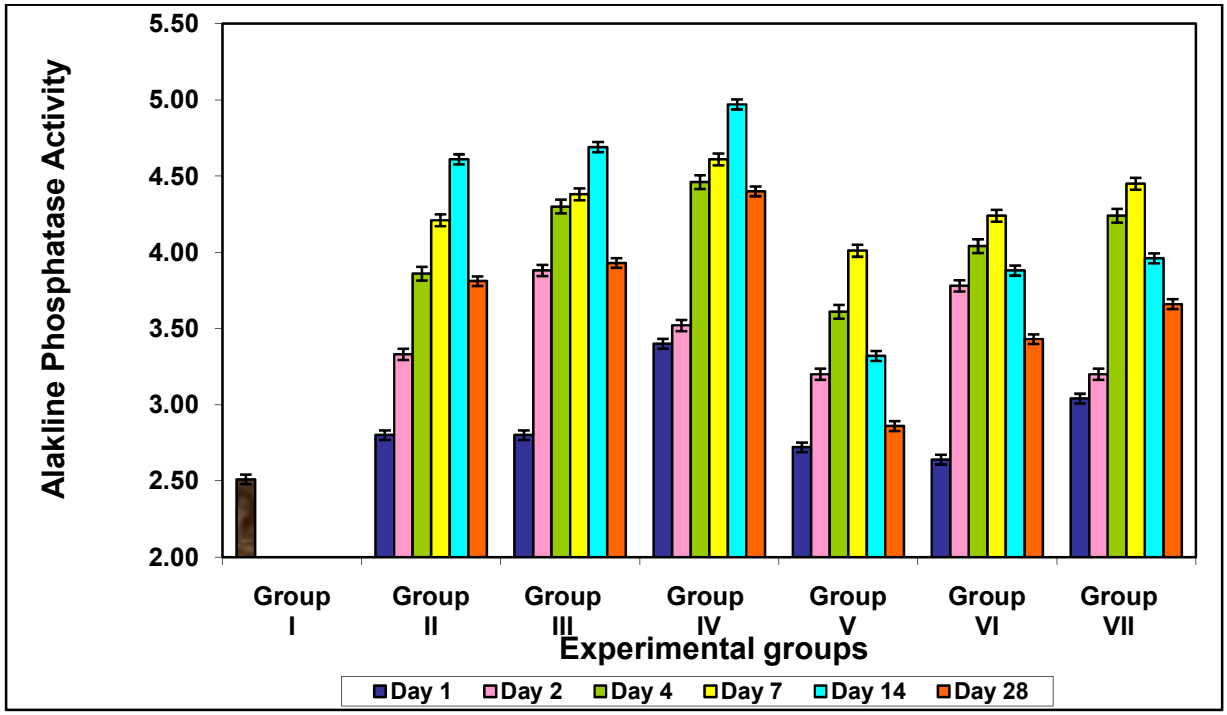

Fig 5: Alkaline Phophatase Activity

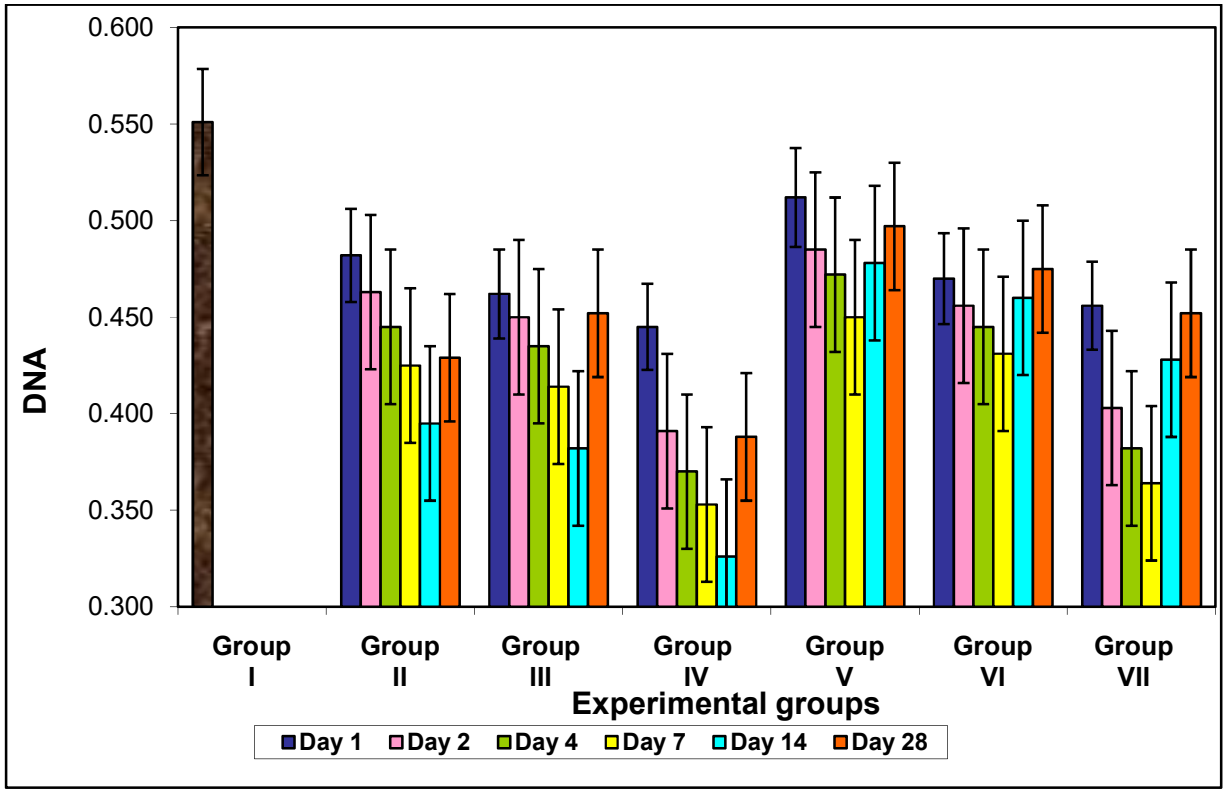

Fig 6: DNA

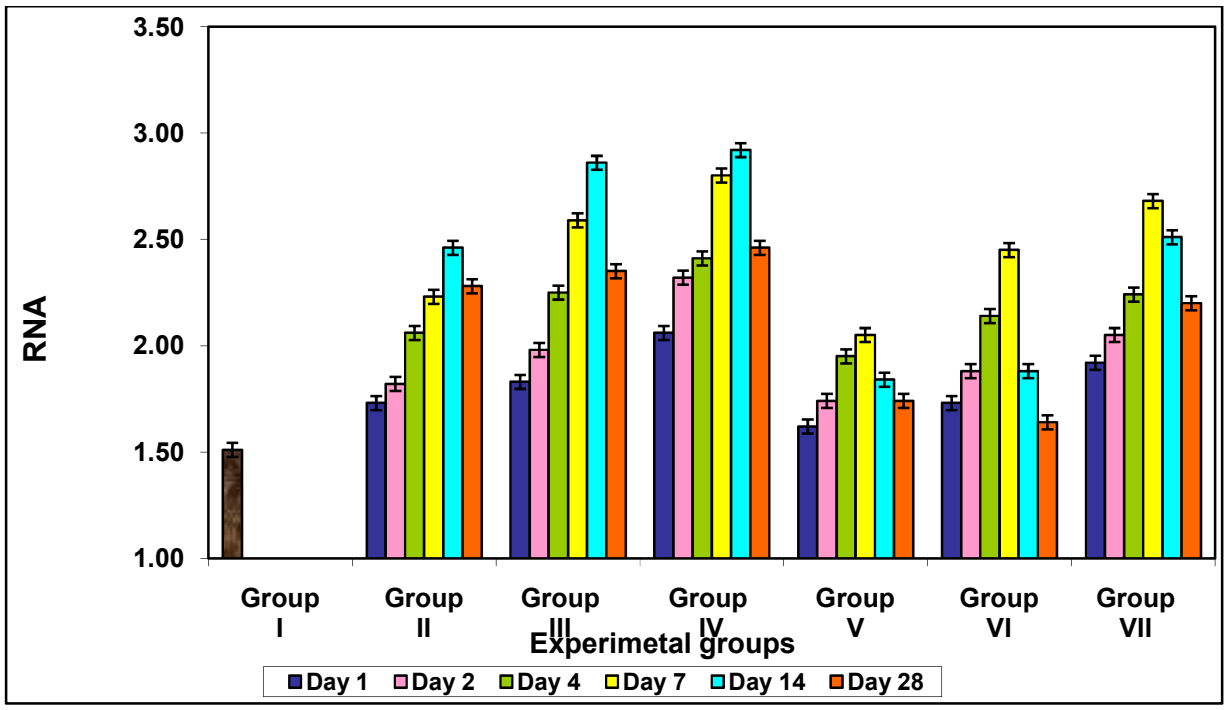

Fig. 7 RNA

DISCUSSION

The toxicity of mercury sources can be expected to depend on its nature, i.e., salts vs. organomercury compounds vs. elemental mercury. One mechanism of mercury toxicity involves its irreversible inhibition of selenoenzymes, such as thioredoxin reductase (IC50=9 nM). Although it has many functions, thioredoxin reductase restores vitamins C and E, as well as a number of other important antioxidant molecules, back into their reduced forms, enabling them to counteract oxidative damage. Since the rate of oxygen consumption is particularly high in brain tissues, production of reactive oxygen species (ROS) is accentuated in these vital cells, making them particularly vulnerable to oxidative damage and especially dependent upon the antioxidant protection provided by selenoenzymes. High mercury exposures deplete the amount of cellular selenium available for the biosynthesis of thioredoxin reductase and other selenoenzymes that prevent and reverse oxidative damage, which, if the depletion is severe and long lasting, results in brain cell dysfunctions that can ultimately cause death. Mercury in its various forms is particularly harmful to fetuses as an environmental toxin in pregnancy, as well as to infants. Women who have been exposed to mercury in

substantial excess of dietary selenium intakes during pregnancy are at risk of giving birth to children with serious birth defects. Mercury exposures in excess of dietary selenium intakes in young children can have severe neurological consequences, preventing nerve sheaths from forming properly. Mercury inhibits the formation of myelin **[9]**.

Ionizing radiation is known to induce various physiological, and biochemical changes in humans and animals. Several molecular mechanisms of ionizing-radiation have been proposed, including cumulative damage by ROS, dislocation in replicative cells, genome instability, mutation, or altered expression of specific enzymes and cell death **[10]**. The oxidative stress due to free radical-formation was greatly augmented during ionizing-radiation exposure **[11]**. It was likely that animal particular antioxidants generally decreased the level of oxidation in such systems by transferring hydrogen atoms to the free radical structure **[12]**.

Total Proteins

Total serum proteins are, diagnostically, of relative importance in assessing the state of health of an organism, their increase appearing especially in inflammatory process and tissue dysfunction after irradiation **[13]**. The present study revealed that irradiation resulted in continuous augmentation in total proteins in liver tissue up to day 7^{th} that probably as a result of an increased transport of amino acid through plasma membrane as a consequence of permeability changes in irradiated cell membrane **[14]**. In addition, increased synthesis of m- RNA and ribonucleoprotein could also be added to the radiation induced increased level in proteins **[15]**.

It has been reported that total serum proteins are, diagnostically, of relative importance in assessing the state of health of an organism, their increase appearing especially in inflammatory process and tissue dysfunction after irradiation. In the present findings also, biochemical alterations are correlated with histopathological changes **[16]**.

In the present study, increase lipid peroxidation, due to toxic effects of mercuric chloride were accompanied by significant reductions in glutathione levels, glutathione peroxidase (GSH-Px), reduced glutathione (GST), catalase activities of the liver tissue, implicating of oxidative tissue damage. Furthermore, these tissue injuries caused functional impairment as evidenced with hepatic function tests, elevated serum of serum protein, albumin levels and transamnases (AST, ALT and ALP) activities demonstrated the severity of mercury

induced tissue damage. The pro-oxidant properties of mercury are well established. An imbalance in the antioxidant protective mechanisms leading to oxidative stress in the cells is being identified as a common factor in mercuric chloride exposure **[17]**.

Moringa treated animals prior to irradiation showed a significantly lower concentration of protein in liver than control. Increased level of it was observed up to day-14 after 5.0 Gy irradiation, respectively. Thereafter, the protein level tended to recover on later autopsy intervals. It is suggested that protection of protein is due to the hydrogen atom donation by the protector **[18]**.

Glycogen

Higher level of hepatic glycogen after irradiation could be due to the stimulation of the pituitary-adrenal system **[19, 20].** Since it has been shown that irradiation did not increase the hepatic glycogen in the hypophysectomized rats **[21]**.

Fatty degeneration, necrosis, increase in connective tissues are the changes produced by heavy metals, which have been described by a number of workers **[22]**. Result of our experiments on Swiss albino mice treated with mercuric chloride exhibit a fall in glycogen values. The loss of glycogen from hepatocytes was statistically significant when compared with the values of normal group. The loss of glycogen in liver takes place before the cell necrosis and it can also drop in physiological circumstances. The present observations are in agreement with those of **[23]** who also reported decrease in glycogen content due to mercury toxicity, this change attributed to the increased glycogenolysis after mercury treatment.

In the present study, when *Moringa* extract was given before mercury treatment, the change in glycogen content remained similar to that of control group (without *Moringa*), but the values were found to less prominent than the controls. Similar results were also observed with the *Aloe vera* **[24]**.

Cholesterol

After irradiation the reduction of cholesterol concentration in liver during early intervals might be due to the stress response caused by radiation which stimulates the synthesis of steroid hormones via hypothalamic-pituitary system **[25]**.

In the present investigation cholesterol showed a significantly declining pattern till day-14 in the mercuric chloride treated group II and day-7 in the drug treated groups V but afterwards there was a significant elevation in cholesterol. It was suggested that the decrease in cholesterol level may be related to its enhanced utilization in corticosteroidogenesis and/or a decreased *de*

novo synthesis. Involvement of thyroid hormones has also been suggested in cholesterol metabolism and an enhanced breakdown in hyperthyroidism is known to result in hypochoelesterolemia **[26]**.

In the present investigation, *Moringa* treated groups showed decreasing trend in value of cholesterol up to day-7 then increased on day-14 which continued up to day-28. *Moringa* is a major antioxidant which affect cholesterol metabolism through its antioxidant effect **[27]**.

The animals treated with both radiation and mercuric chloride also exhibited a decrease in the level of cholesterol in liver. While *Moringa* minimize the level of variation of cholesterol in liver showing its protective effect.

Acid and Alkaline phosphatase activity

Radiation exposure resulted in elevation of liver phosphatase activity which may be attributable to the tissue impairment and per-oxidation of membrane lipids leading to activation of suppressed acid hydrolyases **[28, 29]**. An increase in acid phosphatases activity after radiation exposure in the present experiment could be ascribed either to a direct effect of radiation which results in enhanced Golgi activity **[30]** and per-oxidation of lysosomal membranes by mercuric chloride causing lysis of cellular membranes of hepatocytes, which in turn leads to an increase in the permeability of cell membranes and facilitates the passage of cytoplasmic enzymes outside the cells leading to the increase in both enzyme activity in liver **[31-33]**.

The *Moringa* caused an early recovery to normalcy in both the enzyme level, which was evident as statistically lowered values in these groups in comparison to control. Active principles of *Moringa* in terms of augmentation of oxidative free radical scavenging enzymes, concomitant with reduction in radiation induced lipid peroxidation. It is quite possible that the *Moringa* retards the formation of the toxic lipid peroxidase responsible for radiation damage.

DNA

The depletion in the DNA content of a tissue *in vivo* is due to reduction in or absence of the essential factors controlling the DNA synthesis **[34]**. These factors are the substrates (Four deoxyribonucleoside triphosphates); enzymes (Polymerase), template activity of deoxyribonucleo proteins activators (Mg^{++} and other divalent ions). The enzymic incorporation of deoxyribonucleotides into DNA by the mammalian testis has been studied. The principal type of enzyme involved seems to

be a replicative DNA nucleotidyl transferase, which catalyzes the incorporation in DNA of deoxribonucleotides. There was a general agreement that interference with DNA was one of the important biological effects of irradiation **[35]**. Similar results were also observed with *Moringa* **[36]**.

RNA

The results of the effects of ionizing radiation *in vivo* synthesis of nucleic acids in a mammalian radiosensitive tissue depends to a great extent on two important factors **[37]**:

(i) more or less rapid cytolysis of large proportion of cells, and

(ii) changes in population of cells after irradiation.

RNA metabolism may be influenced by a number of factors. Reasons for the increase in RNA due to irradiation could be due to an increase in the RNA concentration of the surviving cells after radiation insult. Causes of this increase in the cellular RNA may be:-

1. Ability of DNA to transcribe RNA is not affected quantitatively but the length of the chain of RNA molecules reduces **[38]**.
2. Increase in the nuclear RNA polymerase activity may contribute to the post-irradiation increase in the cellular RNA **[39]**.
3. Increased gonadotropin secretion after irradiation may accelerate the RNA synthesis after higher doses of irradiation **[40]**.

Protective mechanisms of *Moringa oleifera*

In the present study, inhibition of LPO in biomembranes has been caused by antioxidants present in *Moringa oleifera*. It was also observed that, radiation caused depletion in GSH levels in entire test period. Under normal conditions, the inherent defense system like glutathione protects against oxidative damage. GSH is a versatile protector and executes its radio protective function through free radical scavenging, restoration of the damaged molecules by hydrogen donation or by reduction of peroxides and maintenance of thiols in the reduced state. The decrement of GSH could be due to an enhanced utilization of the antioxidant system during detoxification of the free radicals generated by radiation. This depletion of glutathione further enhanced the lipid peroxidation **[41]**.

REFERENCES

[1] Pillai, K.C. (1985): Heavy metals in aquatic environment. In C. K. Varsnney (Ed.) (1985), water pollution and management, wiley Estern Limited New Delhi.

[2] Paliwal, R., Sharma, V. and Pracheta (2011): A Review on Horse Radish Tree (*Moringa oleifera*): A Multipurpose Tree with High Economic and Commercial Importance. Asian Journal of Biotechnology, 3: 317-328.

[3] Lowry, O.H., Rosenbrough, M. J. Ferr, A.L. and Randell, R.J. (1951): J. Biol. Chem., 194: 265.

[4] Montgomery, R. (1957): Determination of glycogen. Arch. Biochem. Biophys., 67: 378.

[5] Oser, B.L. (1965): Hawk's physiological chemistry, McGraw Hill, New York, 246.

[6] Fiske, C.H. and Subbarow, Y. (1925): The colorimetric estimation of phosphates. J. Biol. Chem., 66: 375.

[7] Ceriotti, G. (1952): The microchemical determination of deoxyribonucleic acid. J. Biol. Chem., 198: 297.

[8] Ceriotti, G. (1955): Determination of nucleic acid in animal tissue. J. Biol. Chem., 214: 59.

[9] Nicholas, V.C.R. and Laura J. R. (2010). "Dietary selenium's protective effects against methylmercury toxicity." Toxicology. 278 (1): 112–123.

[10] Hart, B.A., Potts, R.J. and Watkin, R.D. (2001): Cadmium adaptation in the lung—A double-edged sword? Toxicology; 160: 65–70.

[11] Cuypers, A., Plusquin, M., Remans, T., Jozefczak, M., Keunen, E., Gielen, H., Opdenakker, K., Nair, A.R., Munters, E. (2010): Artois TJ. Cadmium stress: An oxidative challenge. Biometals; 23: 927-40.

[12] Tawfik, S.S., Abbady, M.I., Azab, KhSh., Zahran, A.M. (2006): Anticlastogenic and haemodynamic efficacy of flavonoid mixture challenging the oxidative stress induced by gamma rays in male mice. Egypt J. Rad. Sci. Applic.; 19: 195-210.

[13] Nwokocha, C.R., Nwokocha, M., Mounmbegna, P., Orhue, J., Onyezuligbo, O., Olu-Osifo, E.H., Okojie, E., Asuquo, E., Ejimofor, T., Ikenna, N. and Judith M. (2012): Proteins and liver function changes in rats following cumulative total body irradiations. West Indian Med. J.; 61(8):773-7.

[14] Mukerjee, H. and Goldfeder, A. (1974): Protein biosynthesis in the liver of x-irradiated mice. Int. J. Radiat. Biol.; 25: 445.

[15] Hidvegi, E.J., Bolonic, E., Holland, J., Lonal, P., Antoni, F. and Verteresz, V. (1968): The effect of whole body x-irradiation of guinea pigs on liver ribosomes. Biochem J.; 109(4): 495-505.

[16] Nwokocha, C. R., Nwokocha, M., Mounmbegna, P., Orhue, J., Onyezuligbo, O., Olu-Osifo, E.H., Okojie, E., Asuquo, E., Ejimofor, T., Ikenna, N. and Judith M. (2012): Proteins and liver function changes in rats following cumulative total body irradiations. West Indian Med J. 61(8):773-7.

[17] Necib, Y., Bahi, A. and Zerizer, S. (2013): Amelioration of mercuric chloride toxicity on rat liver with argan oil and sodium selenite supplements. Int. J. Pharm. Bio. Sci.; 4(2): (B) 839-849.

[18] Grant, G.A. (1969): Mechanism of action of aminothiol radioprotectors. In "Radiation Damage and Sulphydryl Compounds". IAEA, Vienna; p.95.

[19] Kay, R.E. and Entenman, C. (1956): The conversion of glycine –1-C14 to protein C14 and 14Co2 in liver slices from x-irradiated rats. Arch Biochem Biophys.; 62:419.

[20] Hansen, L. (1967): The increase in liver glycogen in non-fasted rats after irradiation. A comparison with pair fed animals. Int J Radiat Biol.; 12:367-72.

[21] North, N. and Nims, I.F. (1949): Time dose study of biochemical response of rats to x-irradiation. Fed Proc, 1949; 8:119.

[22] Shastri, D., Kumar, M. and Kumar, A. (1999): Modulation of lead toxicity by *Spirulina fusiformis.* Phytother Res.; 13(3):258-60.

[23] Dhane, R.R. and Patil, G.P. (1977): Blood glucose and hepatic glycogen interrelationship in *Channa punctatus* (Bloch), exposed to heavy metal pollutants. Proc. 84th session. Ind Sci Cong.; pp 77.

[24] Purohit, R.K., Bhati, S.L., Kanwar, O., Agarwal, M., Chakrawarti, A. and Meena, D. (2014): Therapeutic potential of *Aloe vera* against radiation and cadmium induced changes in the brain, kidney and liver of Swiss albino mice. WJPPS. 3(7): 608-616.

[25] Gould, R.G., Lotz, L.V. and Lilly, E.M. (1956): Effect of x-irradiation on hepatic cholesterol synthesis. Fed Proc.; 15: 264.

[26] Wassermann, M., Wassermann, D. and Aronovaski, I. (1970): The effects of organochlorine insecticides on serum cholesterol level in people occupationally exposed. Bull Environ Contam. Toxicology; 5:368-72.

[27] Mansour, H.H., Abd El Azeem, M.G. and Ismael, N.E. (2014): Protective effect of *Moringa oleifera* on gamma radiation induced hepatotoxicity and nephrotoxicity in rats. Am. J. of Phtomed. and Clin. Therapeu.; 2(4): 495-508.

[28] Sharma, A. and Goyal, P.K. (2005): Radioprotective capacity of *Emblica officinalis* against radiation induced biochemical alterations in liver of Swiss albino mice. J cell Tissue Res.; 51:341-4.

[29] Rene, A.A., Dorden, J.H. and Parker, J.L. (1971): Radiation induced ultra structural and biochemical changes in lysosomes. Lab Invest.; 25: 230-3.

[30] Bhartiya, H.C. and Khan, A.S. (1983): Modification of acid phosphatase activity in the liver of Swiss albino mice by 2-mercaptopropionylglycine against gamma irradiation. Radiobiologia Radiotherapia; 24(2):235-8.

[31] Wilson, R., Doell, B.H., Groger, W., Hope, J. and Gellatey, J.B. (1970): The physiology of liver enlargement. In "Metallic aspects of food safty." Pub-black well scientific oxford.

[32] Ramadan, L.A., Roushdy, H.M., Abu Senna, G.M., Amin, N.E. and El-Deshw, O.A. (2002): "Radio protective effect of silymarin against radiation induced hepatotoxicity". Pharmacol Res.; 45(6): 447-54.

[33] Shastry, K.V. and Shukla, V. (1988): Acute and chronic toxicity of some heavy metals to Heteropneustes fossils. Threat Hab.; pp 321-34.

[34] Altman, K.I., Gerber, G.B. and Okada, S. (1970a): In "Radiation Biochemistry", Vol. I., Acad. Press, NY., p. 187.

[35] Bollum, F.J., Anderegg, J.W., MeElya, A.B. and Potter, V.R. (1960) : Nucleic acid metabolism in regenerating rat. VII. Effects of x-irradiation on enzymes of DNA synthesis. Cancer Research, 20 : 139.

[36] Sheikh, A., Yeasmin, F., Agarwal, S., Rahman, M., Islam, K., Hossain, E., Hossain, S., Karim, M.R., Mikkon, R., Saud, Z.A. and Hossain, K. (2014): Protective effects of *Moringa oleifera* Lam. leaves against arsenic-induced toxicity in mice. Asian. Pac. J. Trop. Biomed., 4 (Suppl 1): S353-S358.

[37] Bacq, M. and Alexander, P. (1961): Fundamentals of radiobiology. Pergamon, New York.

[38] Leadon, S.A. and Ward, J.F. (1981) : The effect of γ-irradiated DNA on the activity of DNA polymerase. Radiat. Res., 86: 445.

[39] Vandergoten, R. and Goutier, R. (1966): Effect of total body x-irradiation and of chemical protector AET administration on the activity of nuclear RNA polymerase in regenerating rat liver. Int. J. Rad. Biol., 11: 449.

[40] Ellis, L.C. (1970): "Radiation effects" In "The testis" (A.D. Johnson, W.R. Gomes and N.L.Vandemark eds.), **3**: 231, Acad. press, NY.

[41] Bump, E. A. and Brown, J. M. (1990): Role of glutathione in the radiation response of mammalian cells in vitro and in vivo, Pharmac Ther, 47: 117.