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MODULATORY INFLUENCE OF *ALOE VERA* AGAINST RADIATION AND CADMIUM INDUCED QUANTITATIVE CHANGES IN THE TESTICULAR CELLS OF MICE

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ABSTRACT

The threat of accidental or hostile exposure to radiation and heavy metal is of great concern. Ionizing radiation inflicts its adverse effect through the generation of oxidative stress that unleash large scale destruction or damage of various biomolecules. Disorders of reproduction and hazards to reproductive health have become prominent issues in recent past after several reports of adverse effects of ionizing radiation on reproductive function. In view of the fact, the present study was planned to evaluate the protective efficacy of *Aloe vera* against radiation and cadmium induced quantitative changes in the testicular cells of mice. For the study, the Swiss albino mice were exposed to 3.5 or 7.0 Gy of gamma radiation with or without cadmium chloride treatment. The *Aloe vera* was administered seven days prior to radiation or cadmium chloride treatment in the experimental animals. Alterations in the population dynamics of germinal epithelium were studied quantitatively. Spermatogonia A, I and B, primary spermatocytes at various stages of their meiotic prophase and spermatids were counted from the transverse sections of the testis from each set of experiments. The cells were counted in the form of increase or decrease. The changes were more found more severe after combined treatment of radiation and cadmium chloride as compared to their individual treatment. An early and fast recovery in the *Aloe vera* pre treated groups showing the protective potential of *Aloe vera*.

KEYWORDS: Gamma Radiation, Cadmium Chloride, Swiss Albino Mice, *Aloe Vera*, Testes.

INTRODUCTION

The hazards to the public health arising from radiation produced by man, concern to a large degree, to those low doses and low dose rates relating to carcinogenesis or leucomogenesis, developmental abnormalities and the production of genetic mutations in the gonads, which are passed on to the offspring. In the present time, of course, the medical x-rays, both diagnostic and therapeutic, represent the largest man-made source of radiation exposure to the general population. Metal toxicity and radiation effects on organisms are manifested in the form of various pathological, histological and biochemical alterations. Ionizing radiation is one of the environmental factors that may contribute to reproductive dysfunction by a mechanism involving oxidative stress.[1]

Cadmium is an environmental toxicant and an endocrine disruptor in humans. Several organs (e.g., kidney, liver) are affected by cadmium and recent studies have illustrated that the testis is exceedingly sensitive to cadmium toxicity. More important, cadmium and other toxicants, such as heavy metals (e.g., lead, mercury) and estrogenic-based compounds (e.g., bisphenols) may account for the recent declining fertility in men among developed countries by reducing sperm count and testis function. The cadmium -induced toxicity to the testis is probably the result of interactions of a complex network of causes. This is likely to involve the disruption of the blood-testis barrier (BTB) via specific signal transduction pathways and signaling molecules, such as p38 mitogen-activated protein kinase (MAPK). The current studies on factors that confer the testis sensitivity to cadmium, such as cadmium transporters and metallothioneins and the impact of cadmium on the testis as an endocrine disruptor, oxidative stress inducer and how it may disrupt the Zn^{+2} and/or Ca^{+2} mediated cellular events. While much work is needed before a unified mechanistic pathway of cadmium -induced testicular toxicity is emerged, recent studies have helped to identify some of the likely mechanisms and/or events that take place during cadmium -induced testis injury. Furthermore, some of the recent studies have shed lights on potential therapeutic or preventive approaches that can be developed in future studies by blocking or minimizing the destructive effects of cadmium to testicular function in men.

In humans and other mammals, cadmium causes various damages to different organs and tissues of the body. The main observation of the effect of cadmium is destruction of the seminiferous tubules with severe necrotic areas. Damage is to all stages of developing germ cells by inducing their structural changes and the apoptotic cell death. Sertoli supporting cells

are considered the most vulnerable cells. Their damage results in cytoplasmic rearrangement and disruption of inter-Sertoli tight junctions resulting in increased permeability of the blood-testis barrier, structural changes in the Leydig cells and decreased testosterone secretion. After long time of cadmium exposure an increase of the amount of interstitial connective tissue occurs. In blood vessels cadmium exposure causes various morphological and physiological changes in vascular endothelial cells and smooth muscle cells. In humans and other mammals, the range of effect depends on the dose, route, ways, and duration of exposure. After necrosis of the sensitive cells cadmium produced lesions in surrounding tissue and activates free cells. Atrophy of the seminiferous tubules is followed by Leydig cell regeneration and interstitial revascularization. In birds, spermatogenic cells underwent irreversible degeneration or atrophy of seminiferous tubules in the absence of significant vascular lesions.[2]

Combined effects of chemicals and radiation have mostly been studied in unborn babies because of their higher sensitivity to these toxicants. The general aspects of the interaction between radiation and chemicals during prenatal development were also summarized.[3] The testicular damages mediated by oxidative stress in Swiss albino rats exposed to lead acetate and gamma rays co-toxicity and possible protective role of taurine has also been studied.[4] Testis is highly radiosensitive organ owing to its extremely active divisional status. As the effects of radiation and cadmium exposure are primarily genetic and fertility is the first to be adversely affected to identify such damage is of prime importance.

Despite the large number of reports, there is scarcity of systemic data on the combined effect of environmental agents and ionizing radiation. Hence, it may be a field of great importance, which needs further exploration of information. A supra-additive or additive type of synergism may be expected due to the combined use of radiation and cadmium chloride.

The usage of compounds to safeguard against the deleterious effects of radiation was endeavored following World War II with the awareness of the requirement to protect humans from the military use of atomic weapons. The pre‑treatment of amino acid cysteine protected rats from the detrimental effects of X-rays was first demonstrated by previous workers.[5] Radioprotective agents, although widely studied in the past four decades and including several thousand agents, have not reached the level of providing the field of medicine with an agent that conforms to all criteria of an optimal radioprotectant, including effectiveness,

availability, specificity and tolerance. In addition, their high toxicity at ideal protective doses prohibited their clinical use.[6,7] In addition, these compounds were inept in providing post irradiation protection. The attention of protection research became more therapy oriented following the appreciation that normal tissue protection during radiotherapy is as imperative as the destruction of cancer cells. The significant toxicity associated with thiol compounds demanded exploration for alternative agents. It was presumed that products/compounds isolated from natural sources could be of significant use as non-toxic radioprotectors. Consequently, attention was focused towards the plant and natural products. Plants have been described to play an extremely important role in the drug discovery and development process.[8] Therefore, investigation of plants and natural products is a beneficial example for radioprotection. The benefit of plants and natural products is that they are used in numerous traditional systems of medicines. They are generally considered non-toxic and extensively acknowledged by humans. Their use as radioprotectors needs systematic assessment and validation. Subsequent to the completion of these requirements their use as radioprotectors could be more successful than synthetic chemicals.

Aloe vera has been appreciated as an important herbal plant of the Indian system of medicine and used in Ayurvedic preparation for the treatment of various ailments. Due to pharmacological and therapeutic properties, present study has been undertaken to find out the possible protective potential of the *Aloe vera* against individual and combined exposure of radiation and cadmium chloride on testicular cells of Swiss albino mice.

MATERIALS AND METHODS

Experimental Animals

For the study, adult healthy male Swiss albino mice (6-8 weeks old) were procured from Lala Lajpat Rai University of Veterinary and Animal Sciences, Hisar. The animals were housed in polypropylene cages and maintained on balanced mice feed and tap water *ad libitum*. They were acclimatized to laboratory conditions before use. Occasionally tetracycline water was provided as a precaution against infection. The temperature of the room was maintained between 22-27ºC. The Govt. Dungar College, Bikaner is registered under CPCSEA, New Delhi (Reg.No. 1066/GO/Re/S/07/CPSEA) and has its own Institutional Animal Ethics Committee (IAEC). The experiments were designed in accordance with the relevant guidelines given by the IAEC of the College.

Cadmium Chloride Treatment

The aqueous solution of the cadmium chloride (SDS, Chemicals, India) was prepared by dissolving 20 mg of cadmium chloride in 1000 ml of the glass distilled water, thus giving a concentration of 20 ppm and then administered orally in drinking water.

Source 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 of 0.69 Gy/min during the first year and 1.22 Gy/min during the next year. 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.

Aloe vera: The fresh leaves of *Aloe vera* were cleaned, cut into small pieces, air dried, powered and extracted with double distilled water (DDW) by refluxing for 36 hrs. (12 hrs. x 3). The extract thus obtained was vacuum evaporated so as to make it in powder form. The extract was redissolved in DDW just before oral administration. An approximate 38 per cent yield of the extract was obtained. The extract was given at the dose of 1000 mg/kg body wt./animal/ day from seven days prior to cadmium chloride treatment or irradiation.

Design of experiment

Group – I: (Sham-irradiated animals)

The animals of this group were sham- irradiated and served as control (normal) group.

Group – II: (Cadmium chloride treated animals)

All the animals of this group were orally fed with cadmium chloride solution at the dose of 20 ppm *ad libitum* in drinking water continuously till the end of experiment.

Group – III: (Irradiated animals)

Sub-group III a: 3.5 Gy.

Sub-group III b: 7.0 Gy.

Group – IV: (Animals treated with radiation and Cadmium chloride)

Sub-group IV a: 3.5 Gy + Cadmium chloride.

Sub-group IV b: 7.0 Gy + Cadmium chloride.

Group – V: (Cadmium chloride and drug treated animals)

The animals of this group were orally fed with cadmium chloride (20 ppm) and also received *Aloe vera* orally at a dose of 1000 mg/kg body weight/animal/day from seven days prior to cadmium chloride treatment and continued up to the last autopsy interval.

Group – VI: (Radiation and drug treated animals)

Sub-group VI a: 3.5 Gy + *Aloe vera.*

Sub-group VI b: 7.0 Gy + *Aloe vera.*

Group – VII: (Radiation, Cadmium chloride and drug treated animals)

Sub-group VII a: 3.5 Gy + Cadmium chloride + *Aloe vera*

Sub-group VII b: 7.0 Gy + Cadmium chloride + *Aloe vera*

Autopsy: A minimum of five animals from each groups II to VII were sacrificed by cervical dislocation and autopsied at each post-treatment intervals of 1, 2, 4, 7, 14 and 28 days. Five sham-irradiated mice were also sacrificed in the similar manner. The weight of the animals was recorded and their testes and vas deferens were removed.

For identifying the various cell types in the specific stages of sprematogenic cycle, the 12 stage classification as proposed by the early scientists was used.[9] Various sprematogenic cell types were counted in the following tubular stages.

Cell	Type	Stages
Spermatogonia	A	V
Intermediate	I	III
	B	V
Spermatocytes	Resting (R)	VII
	Leptotene (L)	IX
	Zygotene (Z)	XI
	Pachytene (P)	V
Spermatids	Cap Phase generation	II and III

Selection of tubular cross section for counting the cell types was done by identifying the specific stages in the cycle of the seminiferous epithelium. Six types of typical cell association, i.e. cyclic stages were selected for counting the germ cells.

II Stage - Spermatogonia A and I, plenty of pachytene spermatocytes, spermatids in maturation phase as well as in cap phase generation. This stage was selected for counting cap phase generation spermatids both in control and experimental animals.

III Stage - Spermatogonia A and I, plenty of pachytene spermatocytes, spermatids in maturation phase as well as in cap phase generation. This stage was selected for counting sprematogonia I and cap phase generation spermatids both in control and experimental animals because most of the products of spermatogonial divisions transform into the intermediate type.

V stage - Type A and B, plenty of spermatocytes, spermatids in maturation phase as well as in cap phase. This stage was selected for counting spermatogonia type A, B and pachytene spermatocytes in control and experimental animals.

VII stage – At this stage rare type A spermatogonia are present. Resting type spermatocytes may be seen along the basement membrane, with their nuclei containing crust like chromatin accumulations. Spermatocytes of the older generation are at the pachytene stage (P). This stage was selected for counting resting spermatocytes (R).

IX stage – At this stage type A spermatogonia are scarce. The nuclei of the younger spermatocytes undergo transformations leading to the leptotene stage (L), while the older spermatocytes are still at the pachytene stage (P). The spermatids form a solid layer. This stage was selected for counting leptotene spermatocytes.

XI stage – Rare type A spermatogonia are present along the basement membrane. Zygotene and late acrosome phase of sprematids occurs. This stage was selected for counting the zygotene spermatocytes.

The following criteria were used to identify the cell types in the testes

Spermatogonia type A	These are the largest among spermatogonia. Cells have an ovoid, pale nucleus with a thin nuclear membrane, chromatin material is distributed as fine “dust like” granules.
Spermatogonia type I	These are similar to type A but with more chromatin. They are somewhat ovoid in shape with a thin layer of cytoplasm surrounding a large nucleus.
Spermatogonia type B	These are the smallest among spermatogonia, have a dark spherical nucleus with coarse “crust like” chromatin.
Resting Spermatocytes	These are morphologically similar to type B spermatogonia but their nuclei are small.
Primary Spermatocytes	Leptotene, zygotene and pachytene stages of primary spermatocytes are easily identified by the virtue of their typical chromosmal configurations.
Spermatids	Cap phase generation spermatids are characterised by the presence of proacrosomal granules in the idiosome and fusion of proacrosomal granules into a single large granule near the nucleus.
SeminiferousTubular	Tubular diameter of the seminiferous tubules was measured in the testes of each

diameter	group at each interval with the help of an ocular micrometer. Hundred circular tubules, randomly selected from the cross sections of the testis were measured at their two points. Average of the measurement was taken separately to get the actual diameter.
Diameter of leydig cell nuclei	Leydig cell nuclei diameter was measured with the help of an ocular micrometer. Hundred leydig cells were randomly selected from the cross sections of every interval. Two perpendicular diameters of each leydig cell nuclei were measured, averaged and expressed in terms of mean leydig cell nuclei diameter.

RESULTS

The following variations in the counts of different types of germ cells were noted

i. Spermatogonia type A counts decreased after treatment, it was found to be minimum at day 14 in non drug treated groups and at day 7 in the *Aloe vera* treated groups. The number of spermatogonia A recovered to a certain extent up to day 28 in all the experimental groups.

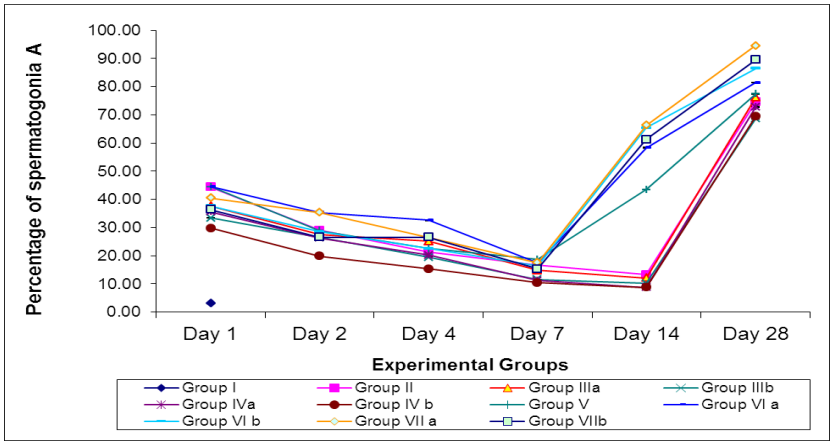
ii. Spermatogonia type I and B reduced to a great extent quantitatively up to day 2, remained negligible up to day 7 and recovered to a great extent upto day 28 in all the experimental groups.

iii. All the types of spermatocytes decreased in number after treatment. The number of resting and leptotene spermatids reduced at a faster rate than the zygotene and pachytene spermatocytes. All these types recovered quantitatively at day 28.

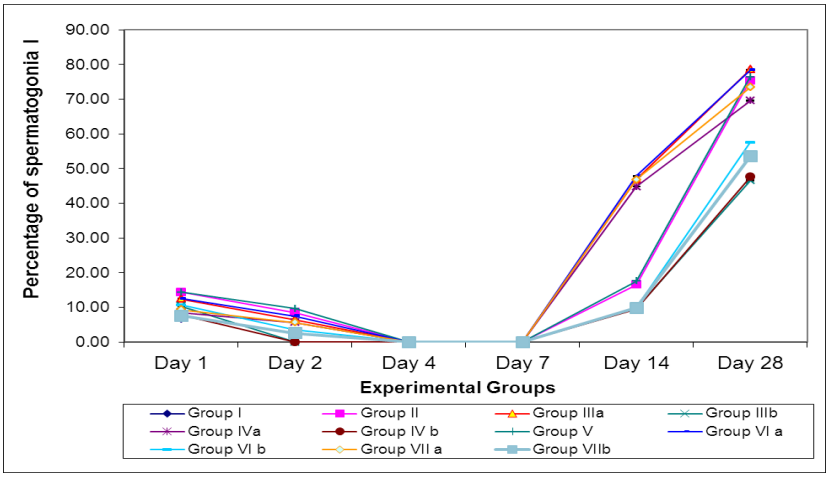
iv. The percentage of spermatids decreased gradually up to day 28 in all the experimental groups. In the combined exposure groups this decrease was comparatively more rapid. This was an indication of the synergistic effect. A less severe decrease was noted in the *Aloe vera* treated experimental animals.

v. Quantitative study of tubular diameter showed decreasing trend till day 14 in the non drug treated groups and day-7 in the *Aloe vera* treated experimental groups. However, tubular diameter recovered to a certain extent on day 28 in all the experimental groups.

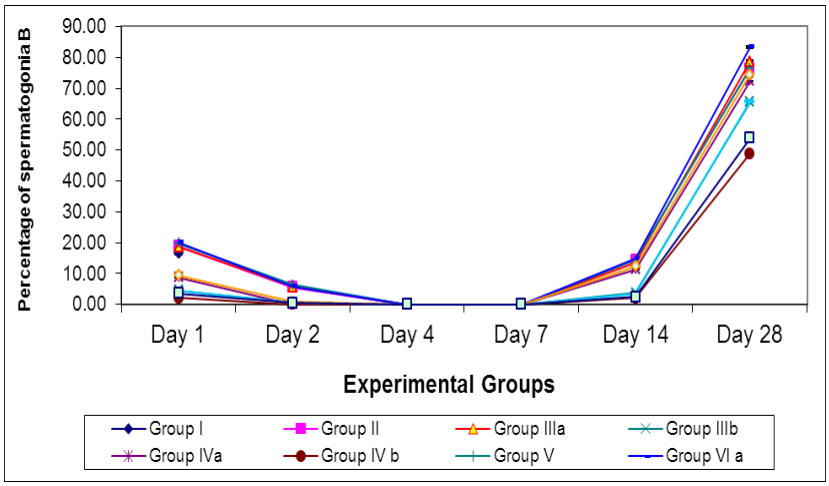
vi. Leydig cell nuclei diameter showed an increasing trend till day 14 in the non drug treated groups and day-7 in the *Aloe vera* treated experimental groups. Thereafter, it declined up to day 28 in all the experimental groups (Figs. 1 to 9).



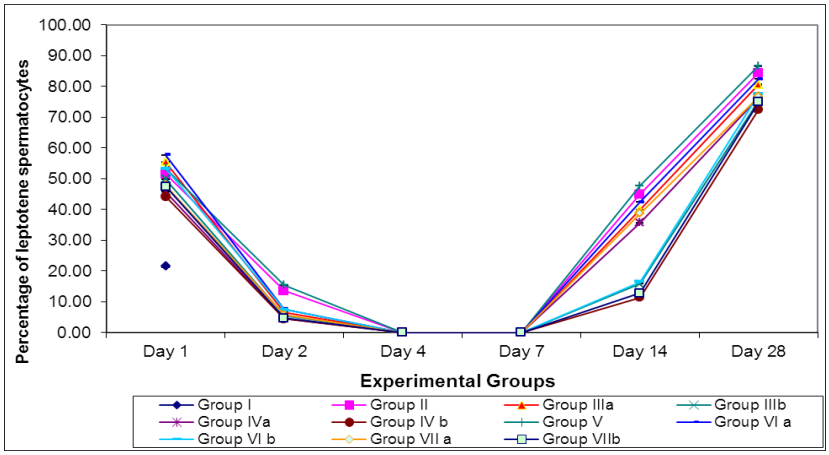
Graph. 1- Variations in the percentage of spermatogonia A per tubule section (Mean + S.E.).



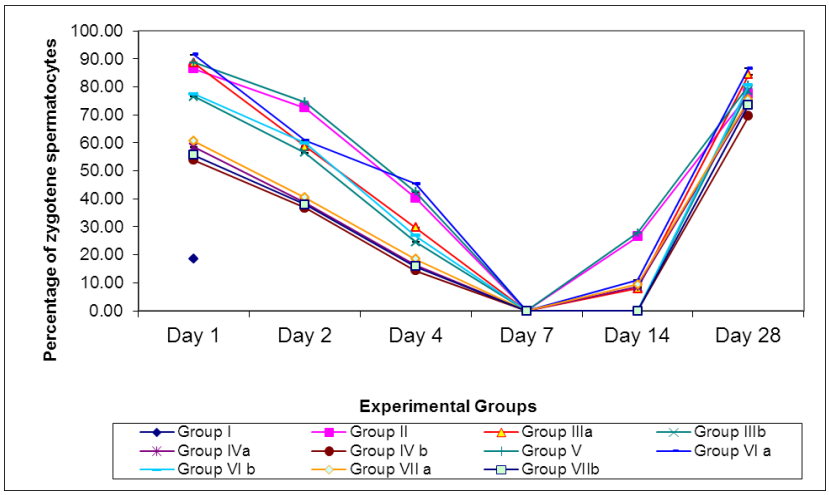
Graph. 2- Variations in the percentage of spermatogonia I per tubule section (Mean + S.E.).



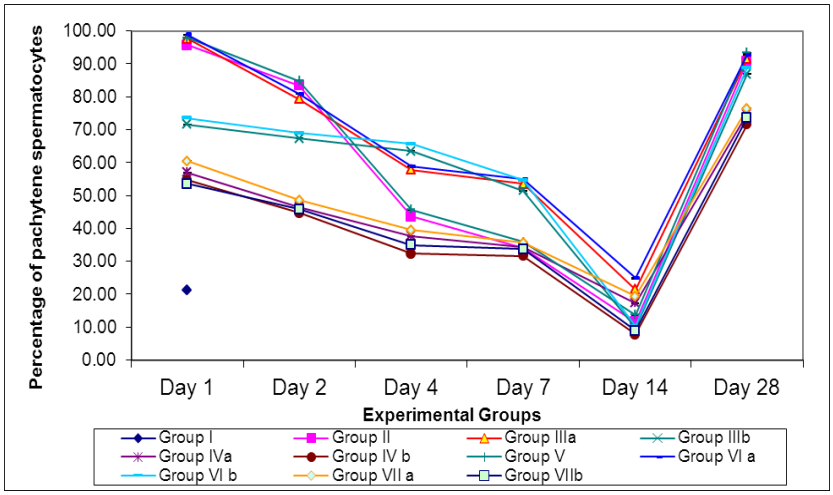
Graph. 3 -Variations in the percentage of spermatogonia B per tubule section (Mean + S.E.).



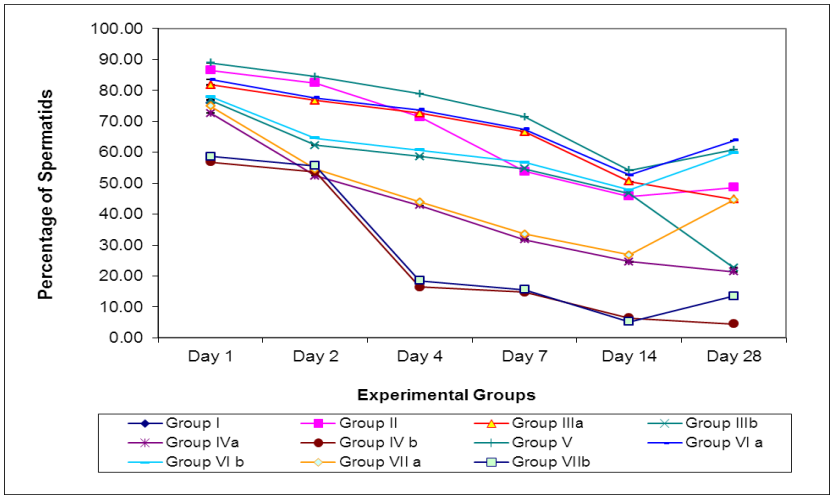
Graph. 4 – Variations in the percentage of leptotene spermatocytes per tubule section (Mean + S.E.).



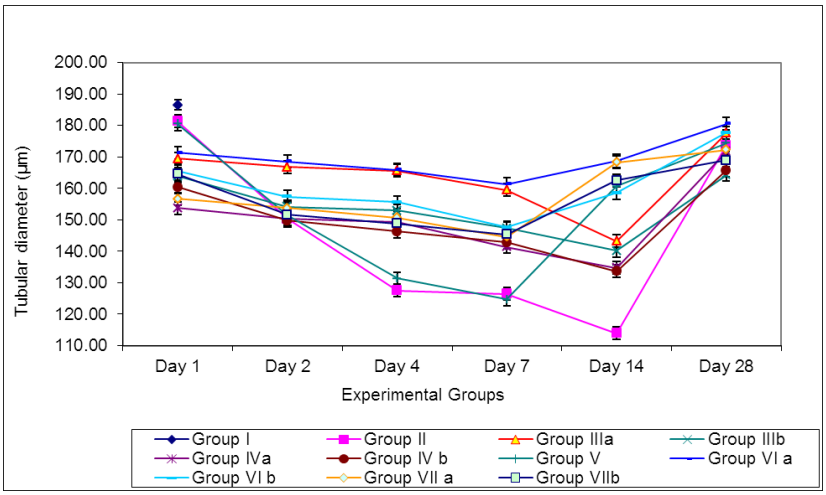
Graph. 5- Variations in the percentage of zygotene spermatocytes per tubule section (Mean + S.E.).



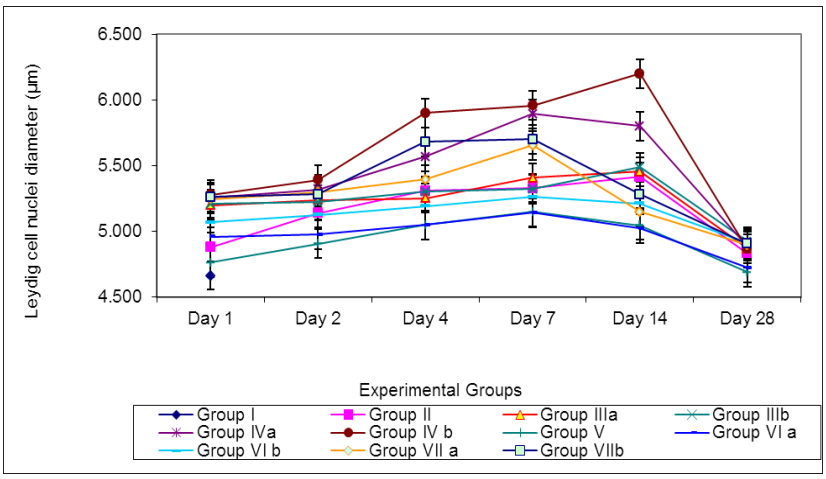
Graph. 6- Variations in the percentage of Pachytene spermatocytes per tubule section (Mean + S.E.).



Graph. 7 - Variations in the percentage of spermatids per tubule section (Mean + S.E.).



Graph. 8 - Variations in the tubular diameter (µm) of the Seminiferous tubule section (Mean + S.E.).



Graph. 9 - Variations in the Leydig cell nuclei diameter (µm) (Mean + S.E.).

DISCUSSION

Quantitative study of various spermatogenic cells of irradiated group (Group III) shows that all types of cells are affected by radiation in all the irradiated sub-groups (IIIa and IIIb), but the degree of depopulation and regeneration depends upon the dose delivered. In the 3.5 Gy exposed groups, there is a considerable reduction in spermatogonia type A, I and B up to 14 days after irradiation. On the other hand, in 7.0 Gy exposed groups, there is a drastic reduction of spermatogonia type A and complete disappearance of type I and B very early after exposure. Repopulation is also slow to follow depopulation process due to severe damage caused by radiation mainly in the precursor compartment; mild regeneration in precursor compartment is observed at two weeks post-irradiation.

Depopulation of various spermatogonial cell stages in the testes in the present experiments is not only due to diminishing or cessation of proliferative activity in the precursor compartment (which is very-sensitive to radiation) leading to maturation depletion but also due to distinct mitotically connected cell death, shortly after exposure, in the dividing maturing pool at the time of irradiation. Similar pattern has also been described on mouse germ cells after exposure to 1000 R or more.[10]

After exposure to 3.5 Gy gamma rays, there is a decline in the precentage of spermatogonia A on day-1, which continues up to day 14. Thereafter repopulation of spermatogonia A is noticed up to the last interval studied. A severe decline in their counts is observed after the dose of 7.0 Gy gamma radiation. At day-14 only 8.27 percent spermatogonia A are seen distributed sparsely in the germinal epithelium of the tubules. The signs of recovery are observed at day-28, but these are not significant. Even at day 28, the counts reach only up to 68.78 percent of the normal value. Irradiation causes an immediate depletion of spermatogonia A in the seminiferous tubules, which is due to cell killing. Similar type of depletion is also observed by many workers.[11-13] It leads to an initial rapid decrease in the population of spermatogonia A. Therefore, gradual decline in their number is found, which continues further due to the death of the damaged cells and moreover by spermatogonial differentiation to the more mature types.[14,15] As stated, the spermatogonia A have heterogenous sensitivity towards irradiation.[16] Most of the spermatogonia are killed by the moderate doses of gamma radiation like 3.5 Gy. It was further demonstrated that all the spermatogonial cells were in continuous cycle and could not find any evidence for a non-dividing reserve stem cell population.[17] Some interpretations can be given for the results

obtained in our experiments because the death of the cells after irradiation also depends upon the stage of the cell cycle at which they have been irradiated[18] and the most radio-resistant parts of the cell cycle occurs during G_1.[19,20] It is observed in our experiments that a high proportion of spermatogonia dies within twenty four hours after irradiation which may be due to the cell death in the interphase or following the mitotic division. At later intervals, the depletion in their population is due to the death of spermatogonia at mitotic divisions and further maturation depletion due to the lesser number of cells in the dividing pool. Spermatogonia in the mitotic state start appearing at early intervals (day-7) in *Aloe vera* treated groups. Regenerating spermatogonia are also observed simultaneously and at the later intervals also.

To explain the lack of increase in the stem cells up to day 14, it was postulated that the surviving stem cells do not resume their usual, pre-irradiation steady state proliferation differentiation pattern for at least 14 days after exposure. Without a change in the cell loss (differentiation) pattern, the stem cell pool will not regenerate, which could explain the prolonged duration of detectable radiation injury to the testes.[21] The regeneration of the spermatogonia A population is found dose dependent, higher the dose, lesser and slower is the regeneration as recorded in the present study. This repopulation was greater in the *Aloe vera* treated experimental animals.

In the present findings, after cadmium chloride treatment (Group II), the spermatogonia A counts decline up to day 14, then start increasing up to day 28 reach up to 72.13 percent of the normal counts. This is a similar type of depletion as in irradiated group, which may be due to the cell killing, which leads to an initial rapid decrease in the population of spermatogonia A. Therefore, gradual decline in their number is found, which continues further due to death of the damaged cells and more over by spermatogonial differentiation to the more mature types. In the cadmium chloride and *Aloe vera* administered group (Group V) the spermatogonia A counts decreased up to day-7 thereafter it increased up to day-28. This may be due to the protection provided by the drug.

Similar types of depletions in the population of spermatogonia A was also observed after combined treatment of cadmium chloride and radiation (Group IV) but there was found more depletion as compared to respective doses of radiation and cadmium chloride individually. These results have shown an “additive synergistic” type of nature of cadmium chloride with

the increased dose of radiation. An early recovery and less pronounced changes in the *Aloe vera* administered group VII were observed showing protective effect of *Aloe vera*.

The response of spermatogonia to various doses of radiation has been investigated by several investigators. After 48 hours both of these cell types are found to be absent from the tubules at stage III and V respectively in all the experimental groups in our present study. This indicates that irradiation and cadmium chloride inhibit or delay the mitotic division to yield the primary spermatocytes. Both of these spermatogonial types restore a fine percentage of their original number at day 28 in all the experimental groups. This profound recovery in their counts must be due to the re-establishment of cell renewal activity in the stem cell compartment and is the sign of progressing maturation differentiation process[22,23] The decline in the both types of spermatogonia in *Aloe vera* pre-treated animals was lesser due to the protection provided by the drug.

All the types of spermatocytes were also found to be decreased in number after exposure of radiation, cadmium chloride and with combined exposure of both. The number of resting and leptotene spermatocytes decreased at faster rate than zygotene and pachytene spermatocytes. All these types recovered quantitatively at day 28. Resting and leptotene spermatogonia are highly prone to the immediate cadmium and radiation death. This decrease was lesser in *Aloe vera* treated animals in present study.

Direct damage of resting spermatogonia of rat was reported in the dose range of 81-2996 R.[24] The direct damage to the resting spermatocytes after 283 and 350 R has also been reported.[25-26] Death of a small number of resting spermatocytes at 300 R has also been noticed.[27]

The division and further differentiation of spermatogonia form the resting primary spermatocytes. So the number of resting primary spermatocytes is directly affected by the damage to the spermatogonia B population. The resting spermatocytes were noted to be the most affected by irradiation after spermatogonia B. They were reduced to zero level at day 4 and 7 in all the experimental groups in the present study. Depletion in the number of leptotene, zygotene and pachytene spermatocytes were also noted. This decline was less pronounced in the *Aloe vera* treated groups. It is evident from tables 3, 4 and 5 the number of spermatogonia I, B and primary resting spermatocytes is significantly lower in the combined treatment sub groups (IV and IVb) as compared to respective irradiated sub-group (IIIa and

IIIb). This depicts that the cadmium chloride provides synergistic additive action to the spermatogonia I, B and primary resting spermatocytes towards immediate cell killing or radiation injury. This reduction in the spermatogonia I, B and resting primary spermatocytes was less severe in the *Aloe vera* treated groups showing protective effect of *Aloe vera*.

In the present investigation percentage of spermatids per tubule section declined on day-1 which continued up to day-14 in both the dose groups. In the combined treatment group this decline was more severe due to synergistic effect of radiation and cadmium chloride. The reduction in the spermatid percentage was lesser in the *Aloe vera* treated groups showing protection provided by the drug. Similar observations were made by other workers.[28,29]

Mechanism of protection provided by *Aloe vera*

The possible mechanisms of action of *Aloe vera* may be as under

(1) Radiation has been shown to induce DNA strand breaks and mutation and induced peroxidative changes to lipid and proteins. *Aloe vera* extracts has been shown to have significant antioxidant activity, which reduces the oxidative changes induced by radiation.

(2) *Aloe vera* extract was also found to inhibit mutagenesis by direct binding to certain mutagens as well as by inhibing carcinogen activation.

(3) It stimulates haemopoiesis thus reducing the myelosuppression induced by radiation.

(4) Moreover, it produces a protective layer in stomach thus reduces the mucosal damage of gastrointestinal linings during irradiation.

(5) It has been shown that the exogenous application of *Aloe vera* increases glutathione levels in the tissues on one hand and maintains-SH groups and increases protein synthesis on the other hand.

(6) The protection offered by *Aloe vera* has been explained by scavenging or oxidizing free-radicals Thus it can be concluded that *Aloe vera* may inhibit the Lipid peroxidation by.

(i) Reducing the formation of free radicals,
(ii) Scavenging of free radicals (antioxidant mechanisms).[30]
(iii) Exuadating the repair mechanism of damaged cell membrane.
(iv) Delay of cellular division and inducing hypoxia in the tissues.[31]

(v) *Aloe vera* provides powerful anti-oxidant action, due to amongst other properties, its vitamin content, especially vitamin A, E and C.[32] In addition to its innate anti-oxidant properties and constituents, Aloe *vera* has the ability to stimulate the body's own anti-oxidant activities. This results in reduced oxidative stress, which has been shown to play "plays an important role in age related disease".[33]

CONCLUSION

Based on the above promising results, it can be concluded that *Aloe vera* has the potential to mitigate the testicular injuries against lethal dose of gamma radiation, which in turn reflected in the form increased survival inhibited pathological alterations, which in turn reflected in the form increased survival inhibited, significant decline in LP levels and an enhancement in GSH content in *Aloe vera* extract pre-treated irradiated group as compared to irradiated control group.

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