

## Radioprotective Effect of *Panax ginseng* on the Phosphatases and Lipid Peroxidation Level in Testes of Swiss Albino Mice

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The *Panax ginseng* has been used as traditional medicine for past several years among oriental people. The present investigation has been made to assess the radioprotective efficacy of ginseng root extract in the testicular enzymes of Swiss albino mice. The Swiss albino mice were divided into different groups. (i) Ginseng treated group: The animals were administered 10 mg/kg body weight ginseng root extract i.p. (ii) Radiation treated group: The animals were exposed to 8 Gy gamma radiation at the dose rate of 1.69 Gy/min at the distance of 80 cm. (iii) Combination group: Animals were administered ginseng extract continuously for 4 d and on 4th day they were irradiated to 8 Gy gamma radiation after 30 min of extract administration. The animals from above groups were autopsied on day 1, 3, 7, 14 and 30. Biochemical estimations of acid and alkaline phosphatases and Lipid peroxidation (LPO) in testes were done. In ginseng treated group acid and alkaline phosphatases activity and LPO level did not show any significant alteration. In irradiated animals there was a significant increase in acid phosphatase activity and LPO level. However, significant decline in alkaline phosphatase activity was observed. The treatment of ginseng before irradiation causes significant decrease in acid phosphatase and LPO level and significant increase in alkaline phosphatase activity. One of the cause of radiation damage is lipid peroxidation. Due to lipid peroxidation, lysosomal membrane permeability alters and thus results in release of hydrolytic enzymes. So, an increase in acid phosphatase was noticed after radiation treatment. The alkaline phosphatase activity is associated with membrane permeability and different stages of spermatogenesis. Due to membrane damage and depletion of germ cells of testes after irradiation the enzyme activity was decreased. Ginseng markedly inhibits lipid peroxidation. It acts in indirect fashion to protect radical processes by inhibition of initiation of free radical processes and thus reduces the radiation damages in testes of Swiss albino mice.

**Key words** testis; phosphatase; lipid peroxidation; ginseng

It is a well known fact that radiation are used for the treatment of tumor. However, killing of tumor is not difficult, it is important to save the normal cells. Hence if radioprotective agents are combined with radiotherapy there could be a possibility to differentially protect the normal cells and kill the tumor cell. There is now a long list of active chemicals, which can modify the radiation response when given before exposure. These substances have diverse chemical structures and include agents such as sulfhydryl compounds, amino acid groups, fatty acids, hormones, a number of pharmacological agents and metabolic inhibitors. Several synthetic compounds such as 2-mercaptopyruvyl glycine,<sup>1–3</sup> WR-2721,<sup>4</sup> lipoic acid,<sup>5</sup> deoxyspergualin<sup>6</sup> and adenine monophosphate<sup>7</sup> have been tested for their protective action against radiation. But, they have their limited use due to their inherent toxicity.

Various plant extracts such as Spirulina,<sup>8</sup> Garlic,<sup>9</sup> Mentha,<sup>10</sup> Aloe<sup>11</sup> and Ocimum<sup>12</sup> are being investigated to evaluate their radioprotective effects, because these plant products have minimum or no side effects. *Panax ginseng* (family: Araliaceae) is a native plant of Korea. Its roots are used for the extraction purpose. It acts on the CNS, cardiovascular systems and endocrine secretion, promotes the functioning of immune system and metabolism; possesses biomodulation action, anti stress and anti ageing activities.<sup>13</sup> Ginseng is also acclaimed as a haemopoietic stimulant.<sup>14</sup>

In the present study an attempt has been made to investigate the radioprotective effect of *Panax ginseng* root extract by evaluating the values of phosphatases and lipid peroxidation in the testes of Swiss albino mice against gamma radiation.

### MATERIAL AND METHODS

**Animals** Adult male Swiss albino mice (6–8 weeks old) weighing  $22 \pm 2$  g from an inbred colony were used for the present study. The animals were maintained on the standard mice feed (procured from Hindustan lever Ltd., India) and water *ad libitum*.

**Irradiation Source** Animals were irradiated by <sup>60</sup>Co Source (ATC-C9) at the cancer treatment centre, Radiotherapy Department, S.M.S. Medical College and Hospital, Jaipur. The animals were exposed to 8 Gy whole body gamma radiation at a dose rate of 1.69 Gy/min at a distance of 80 cm from the source.

**Ginseng Extract** The *Panax ginseng* root extract was obtained from Amsar Pvt. Ltd., Indore (India) in powder form.

**Experimental Design** Mice selected from inbred colony were divided into three groups.

Group I: The animals were administered 10 mg/kg body weight *Panax ginseng* root extract (in D.D.W.) for 4 consecutive days intraperitoneally.

Group II: Animals were exposed to 8 Gy gamma radiation.

Group III: Animals were administered ginseng extract for 4 d and on 4th day they were irradiated to 8 Gy gamma radiation after 30 min of extract administration.

The animals from the above groups were autopsied at 1, 3, 7, 15 and 30th day after the treatment and testes were removed for biochemical estimation of alkaline and acid phosphatase activity<sup>15</sup> and Lipid peroxidation level.<sup>16</sup>

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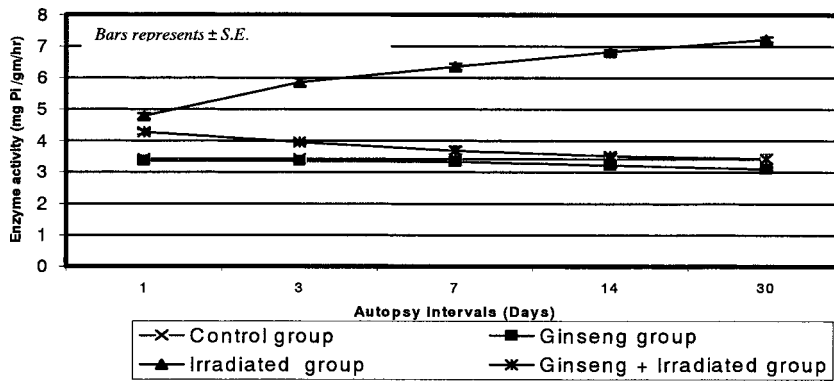


Fig. 1a. Variation in Testicular Acid Phosphatase Activity in Different Experimental Groups

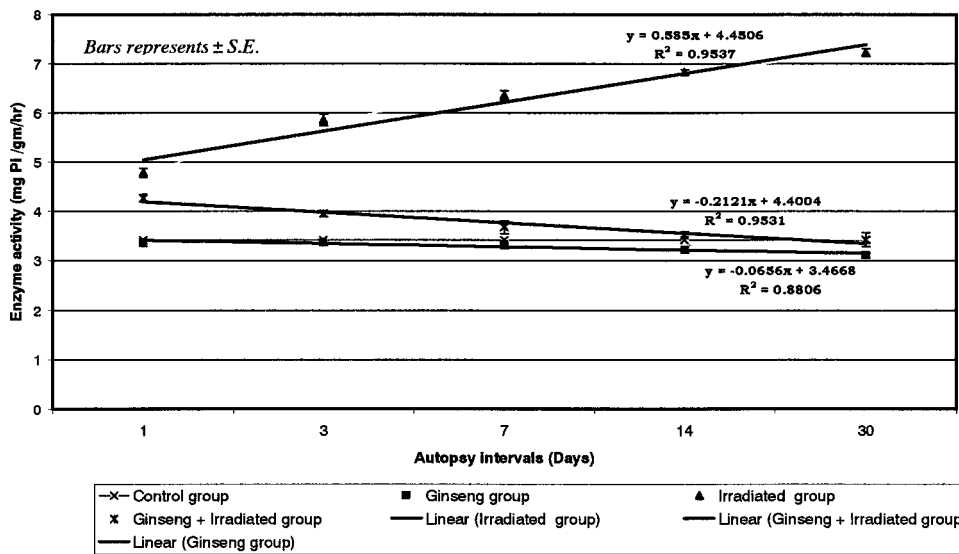


Fig. 1b. Graph Showing Trendline of Testicular Acid Phosphatase Activity in Different Experimental Groups

**RESULTS**

**Radiation Sickness and Mortality** The animals exposed to 8 Gy of radiation showed anorexia, diarrhoea, weight loss, lethargy and epilation. 70% of the animals died within 30 d after exposure to 8 Gy radiation. However, radiation sickness was not observed in the irradiated animals, which were pretreated with ginseng intraperitoneally for 4 d. Further there was only 20% mortality within 30 d in pretreated ginseng group.

**Biochemical Observation** The data are presented as per Figs. 1, 2 and 3.

**Ginseng Treated Group** In ginseng treated group the testicular acid and alkaline phosphatase activity and lipid peroxidation level did not show any significant alterations at all autopsy intervals. The values were found near normal.

**Irradiated Group** In irradiated animals the testes acid phosphatase activity and LPO level exhibited significant elevation ( $p < 0.001$ ) at all autopsy intervals. However the alkaline phosphatase activity showed a significant decline ( $p < 0.001$ ) at all autopsy intervals as compared to normal animals.

**Ginseng Extract+Irradiated Group** The pretreatment of ginseng extract before irradiation significantly altered the

biochemical changes in testes as compared to irradiated group. A significant decline was observed in acid phosphatase activity and LPO level ( $p < 0.001$ ). Whereas alkaline phosphatase activity exhibited significant elevation.

**DISCUSSION**

Present study revealed a significant increase in acid phosphatase activity in the testes of Swiss albino mice after irradiation. Acid phosphatase is localized in cellular lysosomes and irradiation causes lipid peroxidation of lysosomal membrane. Increased acid phosphatase activity may be attributed due to breakdown of lysosomal membrane and liberation of the enzyme. Present studies are in agreement of the findings of Jacob and Maini<sup>17)</sup> and Samarth *et al.*<sup>10)</sup> Increased activity may also be due to synthesis of new lysosome as a consequence of radiation exposure.<sup>18)</sup>

In the present study decrease in alkaline phosphatase activity was observed in the testis after irradiation. The alkaline phosphatase activity is associated with the differentiation and growth processes of cells. Martin and Jacoby<sup>19)</sup> suggested its importance in the process of spermatogenesis. Radiation depletes germ cell population.<sup>11,20)</sup> So, decrease in alkaline phosphatase activity is correlated with the state of germ cells

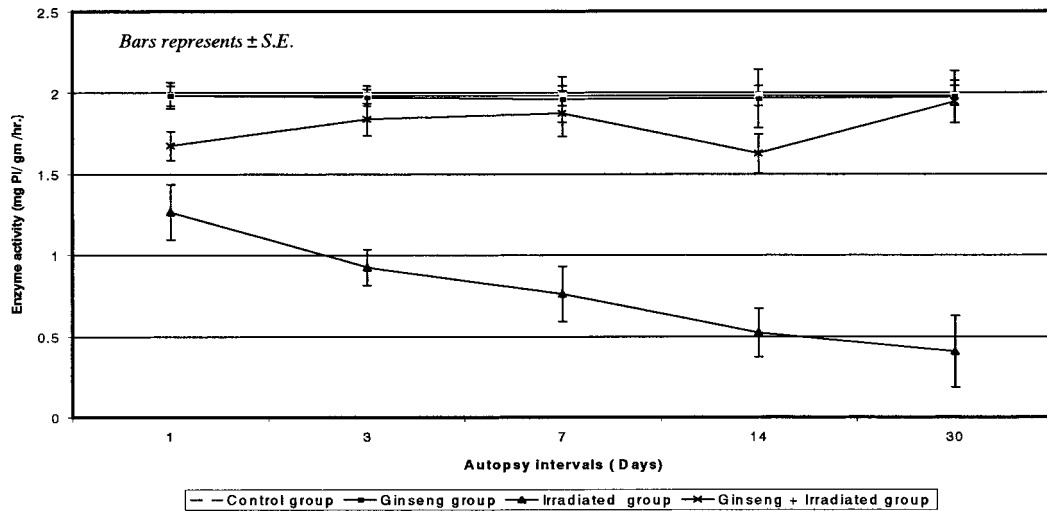


Fig. 2a. Variation in Testicular Alkaline Phosphatase Activity in Different Experimental Groups

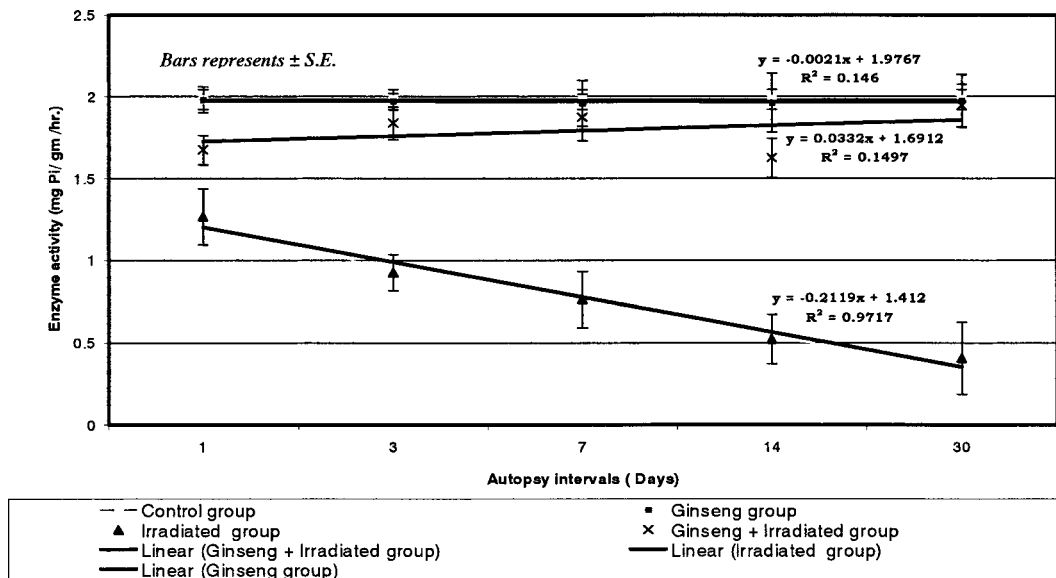


Fig. 2b. Graph Showing Trendline of Testicular Alkaline Phosphatase Activity in Different Experimental Groups

population. According to Cowler and Grodzinski<sup>21)</sup> the acrosomic system of sperm head is composed of alkaline phosphatase. In our previous reports we have observed that radiation causes loss of sperms from the testes.<sup>11,20)</sup> So, loss of sperms by irradiation in the present study may be one of the causes of low alkaline phosphatase activity.

Alkaline phosphatase also plays an important role in maintaining the cell membrane permeability.<sup>22)</sup> The radiation damages the cell membrane thus decline the alkaline phosphatase activity. The decrease in alkaline phosphatase activity may be due to the changes in the amino acid residues and catalytic activity of alkaline phosphatase.<sup>23)</sup>

A significant elevation in testes LPO level in terms of TBARS (thiobarbituric acid reactive substances) or MDA (malondialdehyde) level was observed following radiation exposure. Lipid peroxidation is one of the measure to determine the cellular toxicity. Uma Devi and Ganasoundari<sup>12)</sup> also observed that radiation exposure significantly increases the lipid peroxidation. Radiation induces the formation of re-

active oxygen species such as superoxide radicals, which causes lipid peroxidation.<sup>24,25)</sup> Lipid peroxidation has been suggested as one of the main causes of radiation induced membrane damage.<sup>26)</sup> These findings supported our earlier findings on histopathological studies that suggest that radiation cause germ cell depletion.<sup>11,20)</sup>

In the present investigation, it has been observed that ginseng extract provides the protection by reflecting a significant decrease in testicular acid phosphatase activity LPO level and significant increase in alkaline phosphatase activity as compared to irradiated animals. It has been reported that ginseng extract contains active components Baicalcein ginsenosides Rf and Baicalin ginsenosides Rbl and Rg1. These active components are well known to suppress radiation induced lipid peroxidation and reduce the radiation induced cellular damage. Zhao<sup>27)</sup> has also observed that in ginseng treated group anti lipid peroxidative activity was increased and thus lipid peroxidation was inhibited.

Kim *et al.*<sup>28)</sup> also reported that water fraction and alkaloid

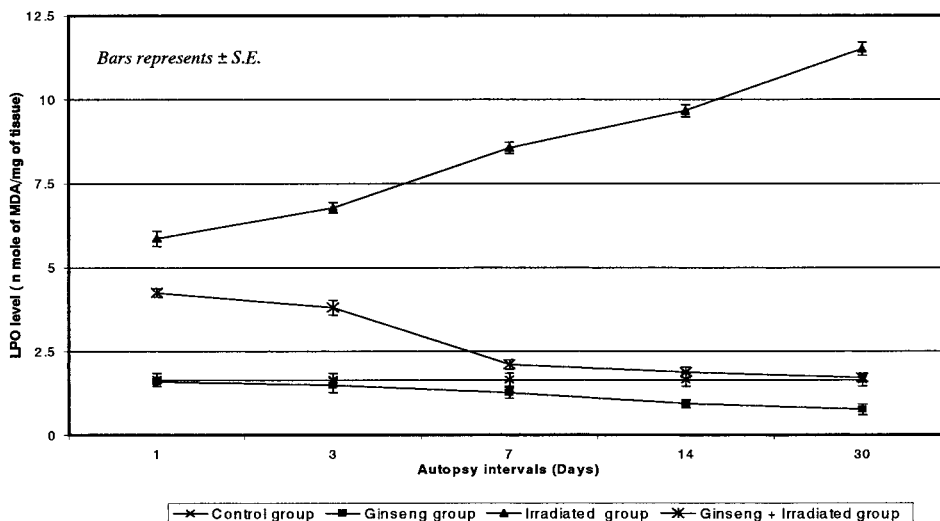


Fig. 3a. Variation in Testicular Lipid Peroxidation (LPO) Content in Different Experimental Groups

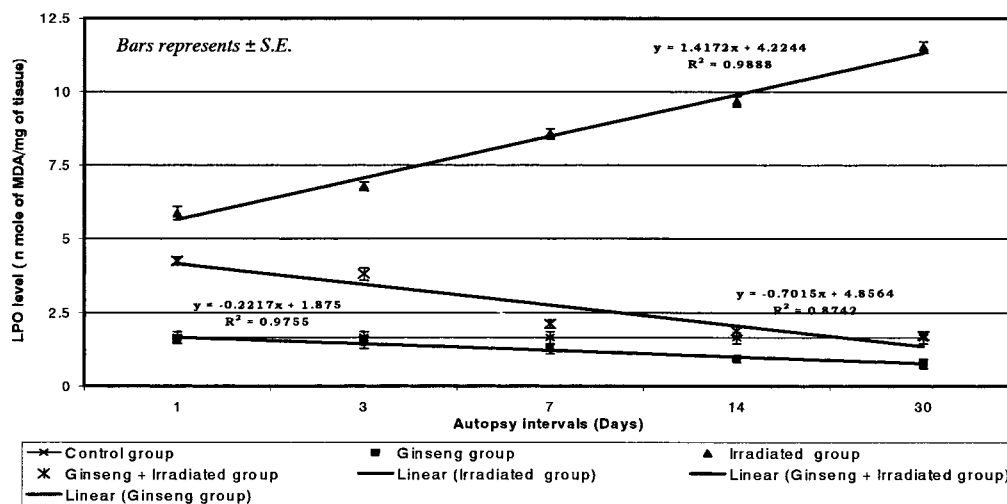


Fig. 3b. Graph Showing Trendline of Testicular LPO Content in Different Experimental Groups

fraction of ginseng may reduce cell damage, especially the damage to DNA molecules, caused by gamma rays and thus playing a role in the repair of regeneration process of damaged cells. It is possible that ginseng reduces DNA damage by antiradical action. Thus the present study suggests that radiation damage may be reduced by inhibiting lipid peroxidation by ginseng extract, which in turn is reflected by a decline of testicular acid phosphatase and LPO level and elevated alkaline phosphatase activities as compared with the irradiated animals. These findings confirm our earlier findings which shows that administration of ginseng before radiation treatment significantly protected the germ cell population against the gamma radiation in mice.<sup>20)</sup>

**CONCLUSION**

From the present study it can be concluded that the ginseng extract could give effective protection against radiation induced testicular damages.

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