RESULTS

4. RESULTS

4.1 Effect of Radiation on mRNA Expression of glyoxalase I

Male *Swiss albino* mice (5-6 weeks old) were irradiated with 3Gy at dose rate 0.06 Gy/sec and the expression level of glyoxalase I was examined in the liver at different time intervals (0, 3, 6, 12. 24h). The results are shown in Figure 1. It is clearly seen that radiation is able to alter the mRNA expression of glyoxalase I at different time intervals (Figure 1 &2). Since, the expression was relatively higher at 6h, for the further study with different doses was carried out at 6h post irradiation. Also the size of the transcript was found to be approx. 1.8 kb. It may be mentioned that three different dose rate (0.24, 0.06, 0.015 Gy/sec) of gamma radiation were used in the present work, depends on the design of the experiment(s).

4.1.1 Effect of different doses of Radiation on mRNA Expression of glyoxalase I

To find the effect of different doses of radiation (0-7Gy) on the expression, the animals were irradiated at dose rate 0.06 Gy/sec. The mRNA level in the liver was increased with dose upto 5Gy and declined at 7Gy (Figure 2). It is clearly visible from the densitometry measurement (Figure 3 & 4).

4.1.2 Effect of different dose-rate on mRNA Expression of glyoxalase I

Mice were irradiated with 3 and 7Gy at three different dose rates (i.e. 0.24, 0.06, 0.015 Gy/sec) and the extent of mRNA expression was observed 6h post irradiation. The mRNA expression of glyoxalase I was found to decline progressively with decrease in dose rate (Figure 5).The lowest dose rate (0.015 Gy/sec) was most effective in inhibiting the expression. With this dose rate the expression of glyoxalase I was reduced to 14% and 46% at 3 and 7 Gy respectively compared to unirradiated control (Figure 5 & 6).

4.1.3 Split dose effect on the mRNA expression of glyoxalase I

The split dose effect on glyoxalase I expression was examined by using total 6 Gy which was splitted into two equal fractions (3+3 Gy). Animals were irradiated with first 3Gy and then to second fraction of 3 Gy at the time interval of 3, 6, and 12h. Then 6h after last exposure, the mRNA expression was determined in the liver. At all three time intervals

between two exposure (3+3 Gy), the level of expression (mRNA) was found to be decreased compared to single exposure of 6 Gy total dose. It may be mentioned that there was no regular pattern in change of expression that could be seen. However at 3h interval there was relatively more inhibition in the expression of mRNA (Figure 7 & 8).

4.1.4 Adaptive response and mRNA expression of glyoxalase I

Animals were irradiated with the conditioning dose of 0.5 Gy and subsequently higher dose of 3 Gy (challenging dose). Dose rate used was 0.015 Gy/sec. The time interval between the conditioning and challenging dose were 3, 6 and 12h. The mRNA level was determined 6h post irradiation. The results are shown in Figure 9 & 10s. The level of mRNA expression was lowered in the group of animals which were irradiated with conditioning dose of 0.5 Gy followed by challenging dose (3Gy) compared to irradiated control group of animals which received single dose of 3 Gy. Interestingly, the expression was maximum in the group of animals which received only conditioning dose of radiation (0.5 Gy). The sequence of extend of expression was as follows: Conditioning Dose > Challenging Dose > Adaptive Response.

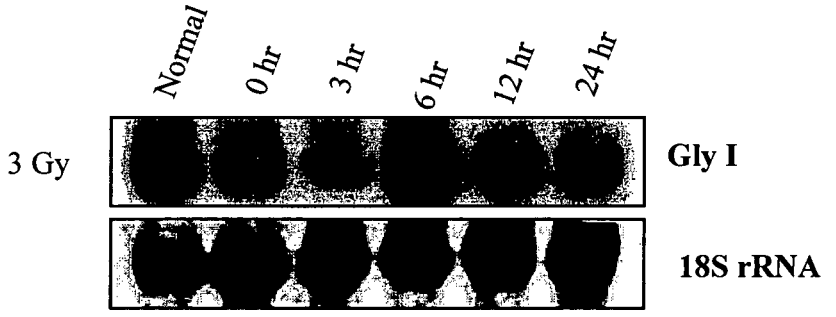

Figure 1: Northern Blot pattern of glyoxalase I (Gly I) expression in the liver of mice at different time interval after irradiation at 3 Gy. Dose rate used was 0.06 Gy/sec.

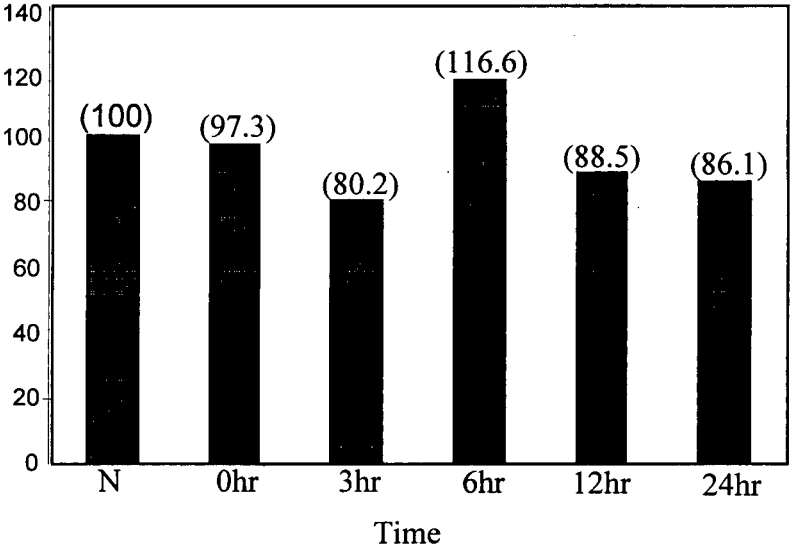

Figure 2: Percentage change in density of glyoxalase I (Gly I) bands expressed at different time intervals in liver of the mice irradiated at 3 Gy. Image Gauge V2.54 (Fujifilms) software was used for densitometry analysis. Values in the parentheses represents % change in density of glyoxalase I (Gly I) bands in Figure 1. N: unirradiated control

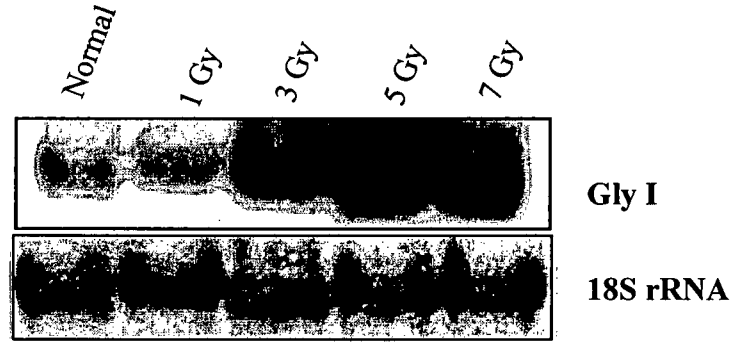

Figure 3: Northern Blot pattern of glyoxalase I (Gly I) expression in the liver of mice irradiated with different doses. Dose rate used was 0.06 Gy/sec.

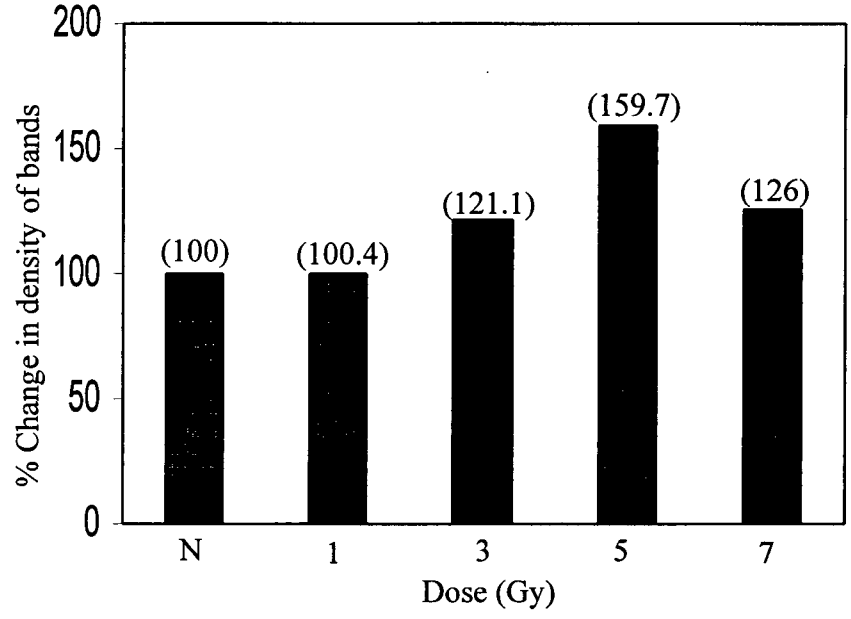

Figure 4: Percentage change in density of glyoxalase I (Gly I) bands expressed in the liver of mice irradiated with different doses. Image Gauge V2.54 (Fujifilms) software was used for densitometry analysis. Values in the parentheses represents the % change in the density of glyoxalase I (Gly I) bands in Figure 3. N: unirradiated control.

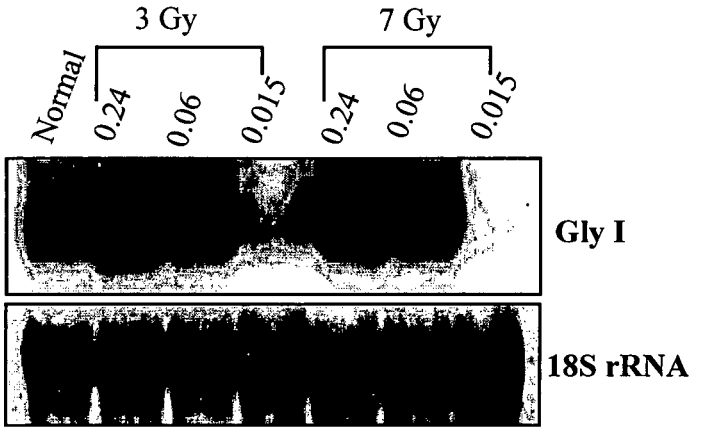

Figure 5: Northern Blot pattern of glyoxalase I (Gly I) expression in the liver of mice irradiated with different dose-rates (0.24, 0.06, 0.015 Gy/sec) at 3 Gy and 7 Gy respectively.

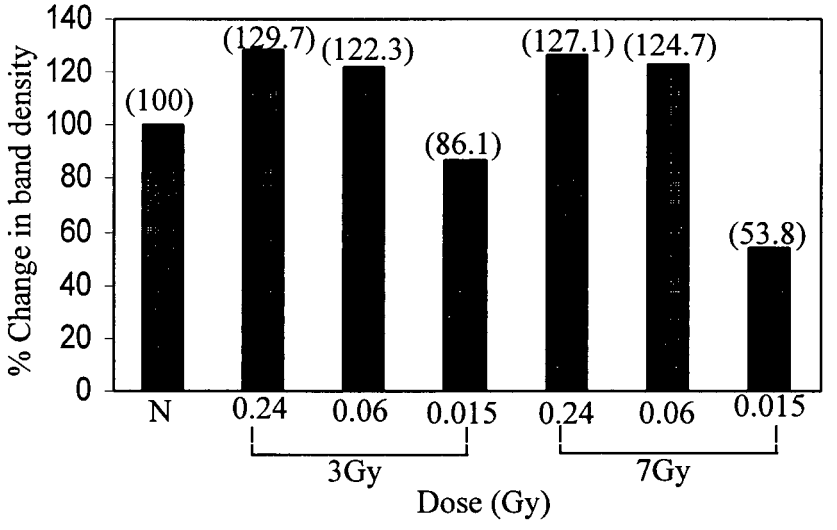

Figure 6: Percentage change in the band density of glyoxalase I (Gly I) expressed in the liver of mice irradiated with different dose-rates (0.24, 0.06, 0.015 Gy/sec) at 3 Gy and 7 Gy respectively. Image Gauge V2.54 (Fujifilm) software was used for densitometry analysis. Values in the parentheses represents the % change in the density of glyoxalase I (Gly I) bands in Figure 5.

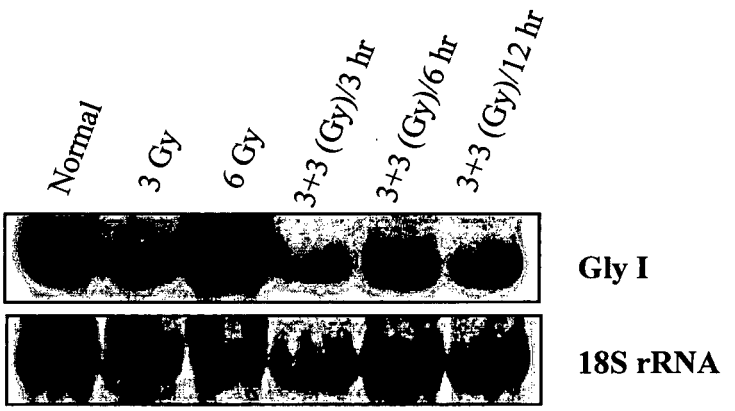

Figure 7: Northern Blot pattern of glyoxalase I (Gly I) expression in the liver of mice irradiated with two equal fractions of (3+3 Gy) doses separated with time intervals of 3, 6 and 12hr. Dose-rate used was 0.015 Gy/sec.

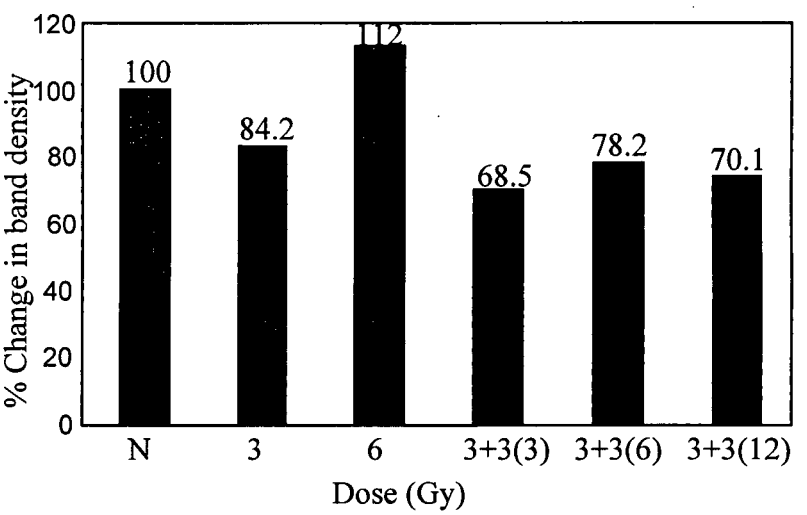

Figure 8: Percentage change in band density of glyoxalase I (Gly I) expressed in the liver of mice irradiated with two equal fractions (3+3 Gy) separated by time intervals of 3, 6 and 12hr. Image Gauge V2.54 (Fujifilm) software was used densitometry analysis. Values above the bars represents % change in the density of bands in Figure 7. N: unirradiated control

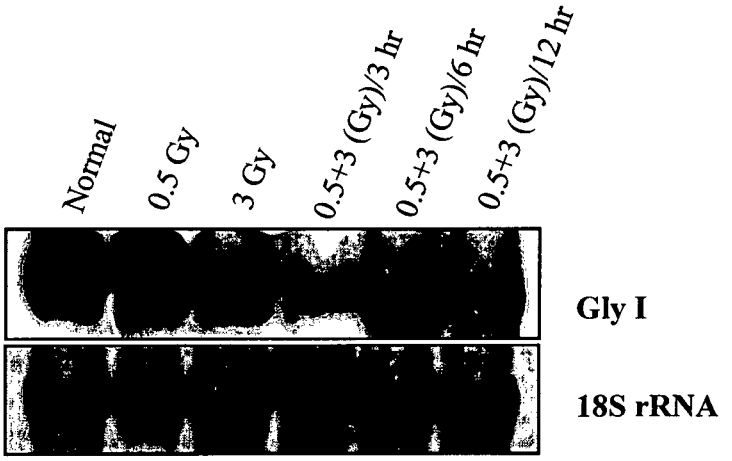

Figure 9: Northern Blot pattern of glyoxalase I (Gly I) expression in the liver of mice irradiated with conditioning dose (0.5 Gy) followed by higher dose (challenging dose = 3 Gy) with time intervals 3, 6and 12h between two exposure. Dose rate used was 0.015 Gy/sec.

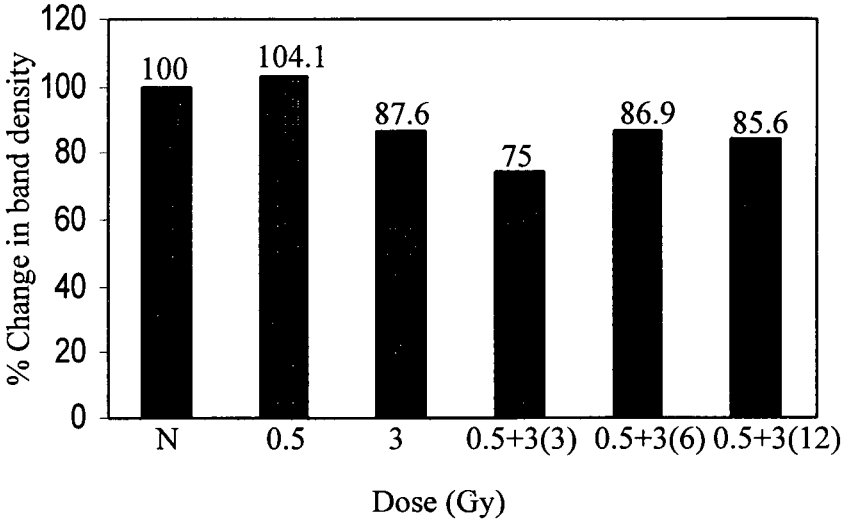

Figure 10: Percentage change in density of glyoxalase I (Gly I) bands in the liver of mice irradiated to conditioning dose (0.5 Gy) followed by exposure to challenging dose (3 Gy) with time interval of 3, 6 and 12h between two exposure. Image Gauge V2.54 (software) was used for densitometry analysis. Values above the bars represents % change in density of glyoxalase I (Gly I) bands in Figure 9. N: unirradiated control.

4.2 Radiation Induced Peroxidative Damage

The oxidative damage due to radiation was measured in terms of lipid peroxidation in microsomal fraction prepared from the liver of mice exposed to different doses (0-7 Gy). The level of lipid peroxidation was monitored as MDA formation. The peroxidative damage increased continuously in dose dependent manner (Table 1). However, the increase was small but consistence upto 5 Gy. At 7 Gy peroxidation was 26% higher compared to unirradiated samples.

Since the dose rate influenced the expression of Gly I, we have also examined the dose rate effect on the peroxidative damage. The animals were irradiated with 3 and 7Gy using three different dose rates: 0.24, 0.06 and 0.015 Gy/sec. The lipid peroxidation was measured in microsomal fractions. The peroxidation was found to increase with decrease in dose rate (Table 2). Remarkable finding is that lipid peroxidation increased to 21% and 30% at the lowest dose rate (0.015 Gy/sec) for 3 and 7 Gy respectively compared to unirradiated control.

The results of split dose effect on lipid peroxidation are shown in Table 3. The total dose of 6 Gy was splitted into two equal parts (3+3 Gy). The exposure between the two fractions was separated by different time intervals (3, 6 and 12h). The peroxidation was measured in the microsomes prepared from the liver of mice after their irradiation with two fractions. The time interval between two irradiation of doses provides opportunity to the cell to repair the damage induced by the first dose. As expected, the levels of peroxidation were lowered incase of split doses than single dose.

As mentioned earlier, adaptive response was studied with 0.5 Gy as conditioning dose and 3 Gy challenging dose. Using the conditioning and challenging dose, the adaptive response in terms of peroxidative damage has been evaluated in the microsomes prepared from liver of irradiated mice. The levels of peroxidative damage was considerably reduced in the animals which were irradiated with conditioning dose first and then followed by challenging dose, compared to irradiated control group of animals (only 3 Gy exposure) (Table 4). Expectedly, an exposure to the conditioning dose (0.5 Gy) alone showed no effect in terms of peroxidative damage.

Table 1: Effect of various doses of γ -rays on lipid peroxidation in the liver of mice

Dose (Gy)	Lipid peroxidation N moles MDA/ mg protein
0	0.665 ± 0.032 (100)
1	0.696 ± 0.053 (104)
3	0.712 ± 0.044 (107)
5	0.731 ± 0.117 (110)
7	0.835 ± 0.044* (126)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein. Dose rate used 0.06 Gy/sec

*Significantly different ($p < 0.01$) against unirradiated control.

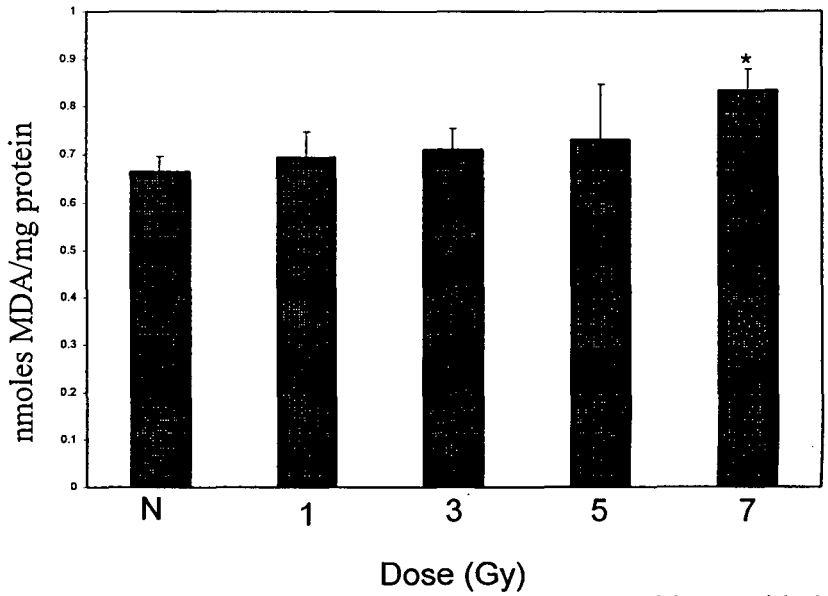

Figure 11: Effect of various doses of the gamma rays on the Lipid peroxidation in the liver of mice. Error bars represent standard deviation * Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

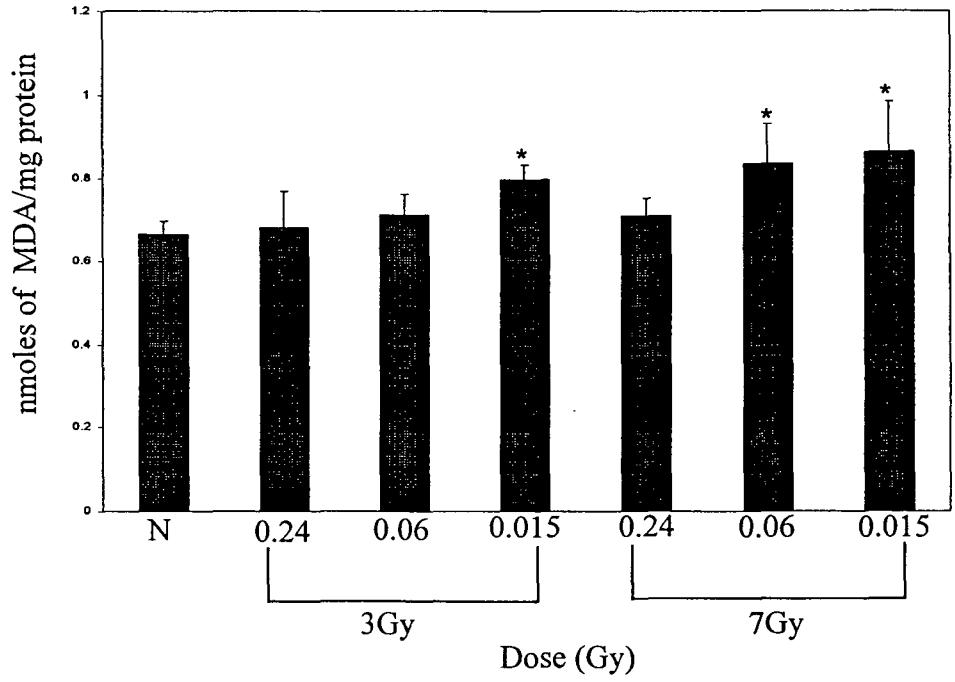

Figure 12: Effect of different dose rates (0.24. 0.06, 0.015 Gy/sec) of the gamma rays on the Lipid peroxidation in the liver of mice. Error bars represent standard deviation * Significantly different ($p < 0.05$) against unirradiated control. N: unirradiated control.

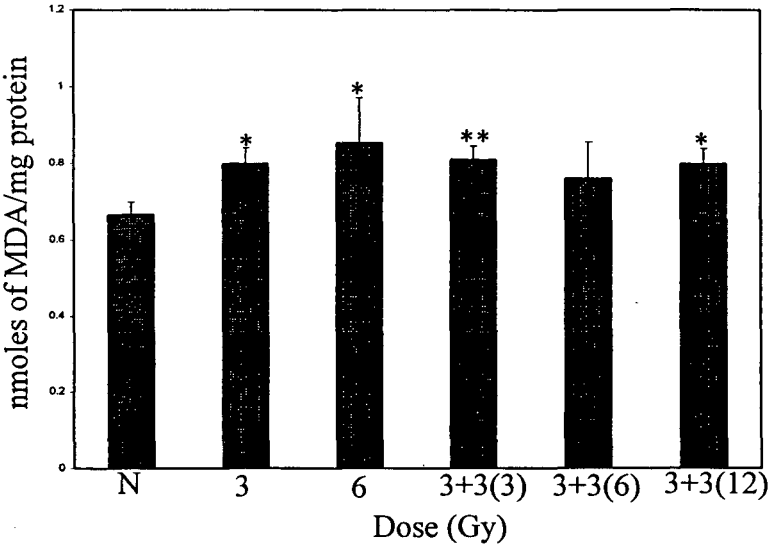

Figure 13: Effect of split doses of gamma rays measured as change in the Lipid peroxidation in the liver of mice. Values in parentheses represents time interval between two equal fractions (3+3 Gy) doses. Error bars represent standard deviation * Significantly different ($p < 0.05$) against unirradiated control. ** Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

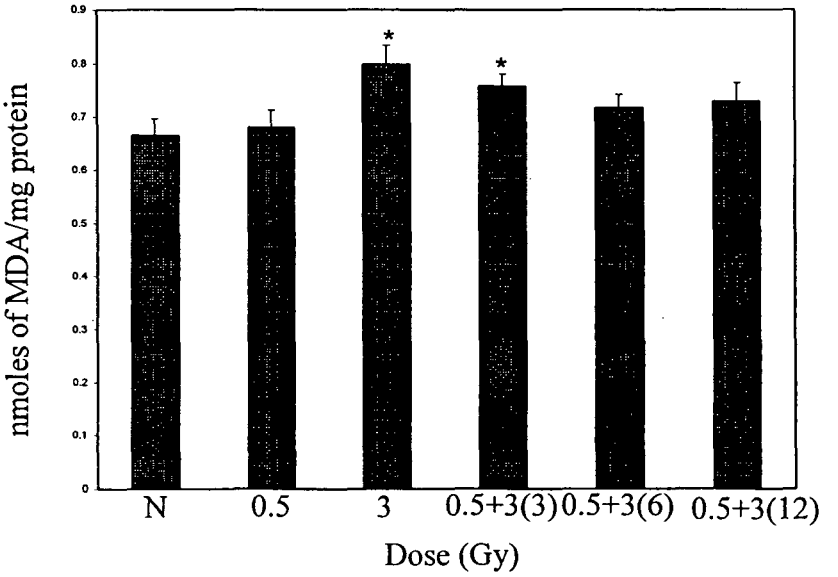

Figure 14: Effect of pre-exposure to the conditioning dose (0.5 Gy) of gamma rays followed by a subsequent higher dose (3 Gy) of radiation on Lipid peroxidation in the liver of mice. Values in parentheses represents time interval between conditioning and challenging doses. Error bars represent standard deviation Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

4.3 Effect of Radiation on Specific activities of glyoxalase I and glyoxalase II

We have also studied the effect of radiations on the specific activities of glyoxalase I and glyoxalase II. Animals were irradiated with different doses (0-7 Gy) at dose rate 0.06 Gy/sec. The specific activities of glyoxalase I and glyoxalase II were observed in the liver of mice at 6h post irradiation (Table 5 & 6). The activity of glyoxalase I increased with dose upto 5 Gy and beyond 5 Gy it declined. On the other hand the glyoxalase II activity progressively declined with dose (Table 6).

To examine the dose rate effect on the specific activity, the animals were irradiated at 3 and 7 Gy with different dose rates (0.24, 0.06 and 0.015 Gy/sec). The specific activity of glyoxalase I measured in the liver of mice. It was found to decrease with decreasing dose rate (Table 7). However, it could be seen that overall level of the specific activity was significantly high particularly with 3 Gy of single dose compared to unirradiated control. Incase of 7 Gy dose and 0.015 Gy/sec dose rate, the specific activity was seen to be lowered than the control value. The similar pattern of change was also observed in the specific activity of glyoxalase II (Table 8).

When 6 Gy was splitted into two equal fractions of 3 Gy, keeping the time intervals of 3, 6 and 12h between the exposures of these two fractions, the specific activity of glyoxalase I was enhanced in the liver compared to the mice irradiated with single dose of 6 Gy (Table 9). Besides this, the progressive decline in the specific activity of glyoxalase II was observed. The decline was increased with increase in time interval between two successive fractionated doses (3+3 Gy). However, the level of specific activity of glyoxalase II was quite higher compared to unirradiated control. With a single dose of 3 and 6 Gy (irradiated control), the specific activity of glyoxalase I was increased by 21% and 12% respectively compared to unirradiated control. Similarly, the increase in the specific activity of glyoxalase II with 3 and 6 Gy was 11% and 85% respectively compared to unirradiated control (Table 10). Importantly, the specific activity of glyoxalase II at 6 Gy as a single dose was quite high compared to Gly I (12%) under similar conditions (Table 9 &10).

In the adaptive response studies, mice were pre-exposed to the conditioning dose of 0.5 Gy followed by irradiation with higher dose of 3 Gy (challenging dose). The time intervals between two exposures were 3, 6 and 12h. The specific activity of glyoxalase I was decreased in the liver at all time intervals. The level of the activity of glyoxalase I was very close to the normal particularly at 3 and 12h (Table 11). In contrast, specific activity of glyoxalase II was relatively higher in the mice which received first the adaptive dose and then the challenging dose (Table 12).

Table 5: Effect of various doses of γ -rays on Specific activity of Glyoxalase I in the liver of mice

Dose (Gy)	Specific activity of Glyoxalase I (units/ mg protein)
0	1.61 ± 0.055 (100)
1	2.10 ± 0.105* (130)
3	2.66 ± 0.060* (165)
5	3.04 ± 0.050* (187)
7	1.69 ± 0.039 (105)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein. Dose rate used 0.06 Gy/sec

*Significantly different ($p < 0.01$) against unirradiated control.

Table 7: Effect of different dose rates of γ -rays on Specific activity of Glyoxalase I in the liver of mice.

Dose (Gy)	Dose rate Gy/sec	Specific activity of Glyoxalase I (units/mg protein)
3Gy	0	1.61 ± 0.055 (100)
	0.24	3.04 ± 0.174* (189)
	0.06	2.66 ± 0.190* (165)
	0.015	1.94 ± 0.100* (120)
7Gy	0.24	2.05 ± 0.145* (127)
	0.06	1.69 ± 0.018 (105)
	0.015	1.55 ± 0.110 (96)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein.
*Significantly different ($p < 0.01$) against unirradiated control.

Table 8: Effect of different dose rates of γ -rays on Specific activity of Glyoxalase II in the liver of mice.

Dose (Gy)	Dose rate Gy/sec	Specific activity of Glyoxalase II (units/ mg protein)
3Gy	0	3.032 ± 0.087 (100)
	0.24	5.532 ± 0.088* (183)
	0.06	5.006± 0. 342* (165)
	0.015	3.373± 0.105* (111)
7Gy	0.24	3.313 ± 0.207* (130)
	0.06	2.845 ± 0.129 (100.4)
	0.015	2.789 ± 0.120* (92)

Each value represents an average of at least three experiments ± standard deviation.The values in parenthese represents the percentage change in the enzyme activity/mg protein.
*Significantly different ($p < 0.01$) against unirradiated control.

Table 9: Effect of split dose of γ -rays measured as change in specific activity of Glyoxalase I in the liver of mice.

Doses (Gy)	Time interval b/w irradiation	Specific activity of Glyoxalase I (units/ mg protein)
0	-	1.61 ± 0.055 (100)
3	-	1.94 ± 0.150* (121)
6	-	1.79 ± 0. 090* (112)
3+3	3	2.05 ± 0.200* (127)
3+3	6	2.12 ± 0. 204* (131)
3+3	12	2.09 ± 0.150* (130)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein. Dose rate used 0.015 Gy/sec.

*Significantly different ($p < 0.01$) against unirradiated control.

Table 10: Effect of split dose of γ -rays measured as change in specific activity of Glyoxalase II in the liver of mice.

Doses (Gy)	Time interval b/w irradiation	Specific activity of Glyoxalase II (units/ mg protein)
0	-	3.038 ± 0.087 (100)
3	-	3.373 ± 0.105* (111)
6	-	5.605 ± 0. .261** (185)
3+3	3	4.750 ± 0.196** (157)
3+3	6	4.305 ± 0. 469** (142)
3+3	12	3.285 ± 0.801 (108)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein. Dose rate used 0.015Gy/sec.

*Significantly different ($p < 0.05$) against unirradiated control.

**Significantly different ($p < 0.01$) against unirradiated control.

Table 11: Effect of pre-exposure to conditioning dose of γ -rays followed by subsequent higher dose of radiation on Glyoxalase I specific activity in the liver of mice

Dose (Gy)	Time interval b/w irradiation (hr)	Specific activity of Glyoxalase I (units/ mg protein)
0	-	1.61 ±0.055 (100)
0.5	-	1.75 ±0.055* (109)
3	-	1.94 ±0.035* (121)
0.5+3	3	1.745 ± 0.045 (108)
0.5+3	6	1.795 ±0.058* (115)
0.5+3	12	1.72 ±0.045 (107)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein. Dose rate used 0.015 Gy/sec.

*Significantly different ($p < 0.05$) against unirradiated control.

Table 12: Effect of pre-exposure to conditioning dose of γ -rays followed by subsequent higher dose of radiation on Glyoxalase II specific activity in the liver of mice

Dose (Gy)	Time interval b/w irradiation (hr)	Specific activity of Glyoxalase II (units/mg protein)
0	-	3.038 ±0.087 (100)
0.5	-	4.939 ±0.284** (163)
3	-	3.373 ±0.105* (111)
0.5+3	3	3.599 ± 0.331* (119)
0.5+3	6	4.987 ±0.714** (164)
0.5+3	12	3.822 ±0.454* (126)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein. Dose rate = 0.015 Gy/sec.

*Significantly different ($p < 0.05$) against unirradiated control.

**Significantly different ($p < 0.01$) against unirradiated control.

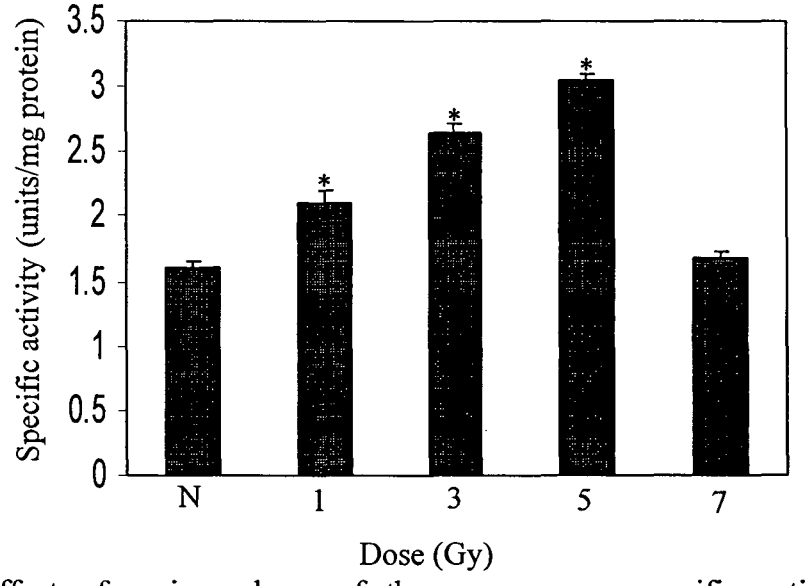

Figure 15: Effect of various doses of the gamma rays specific activity of glyoxalase I (Gly I) in the liver of mice. Error bars represent standard deviation. * Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

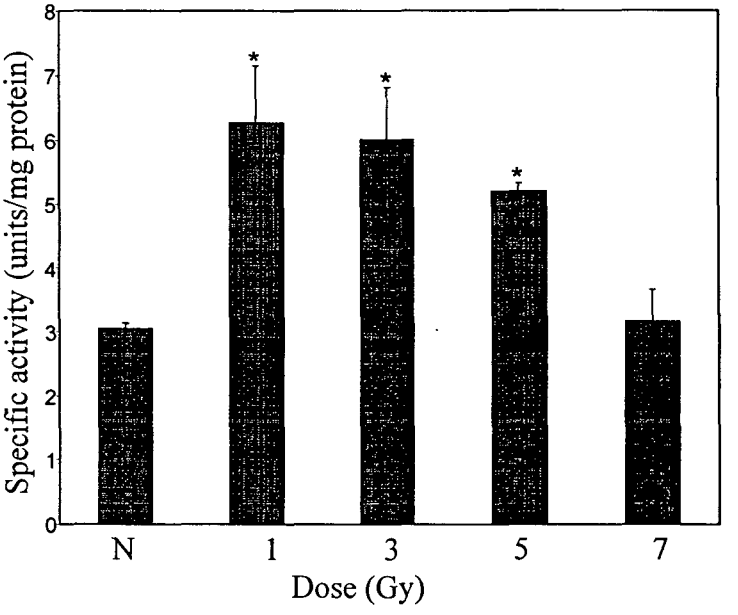

Figure 16: Effect of various doses of the gamma rays specific activity of glyoxalase II (Gly II) in the liver of mice. Error bars represent standard deviation. * Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

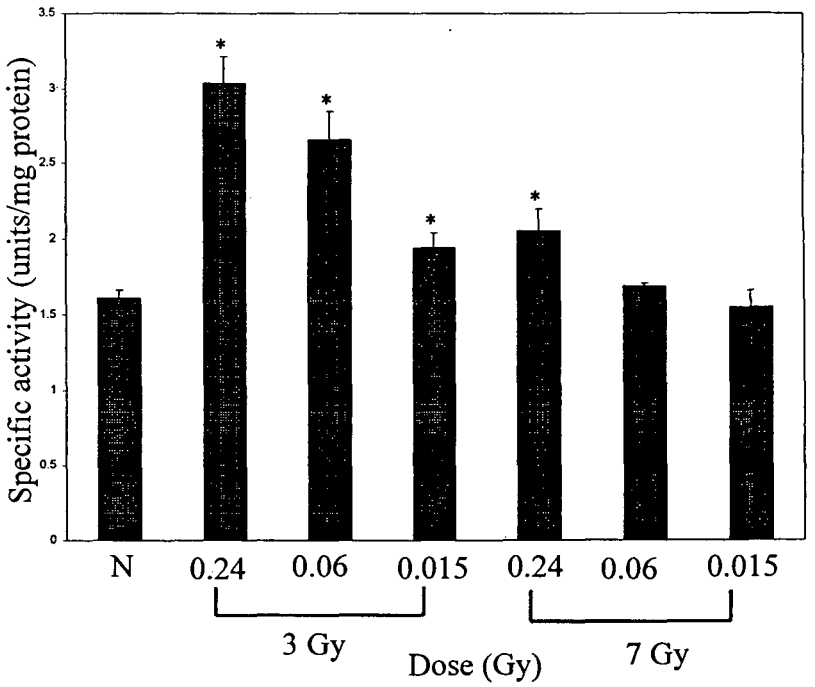

Figure 17 : Effect of different dose rates (0.24, 0.06, 0.15 Gy/sec) of the gamma rays on the Specific activity of glyoxalase I (Gly I) in the liver of mice. Error bars represent standard deviation. * Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

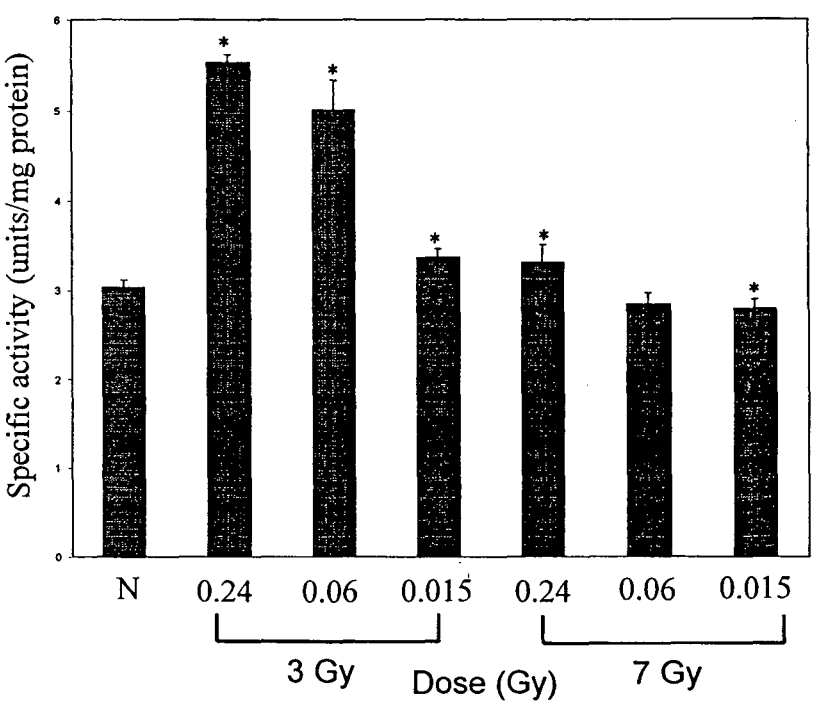

Figure 18: Effect of different dose rates (0.24, 0.06, 0.015 Gy/sec) of the gamma rays on the specific activity of glyoxalase II (Gly II) in the liver of mice. Error bars represent standard deviation. * Significantly different ($p < 0.01$) against unirradiated control.. N: unirradiated control.

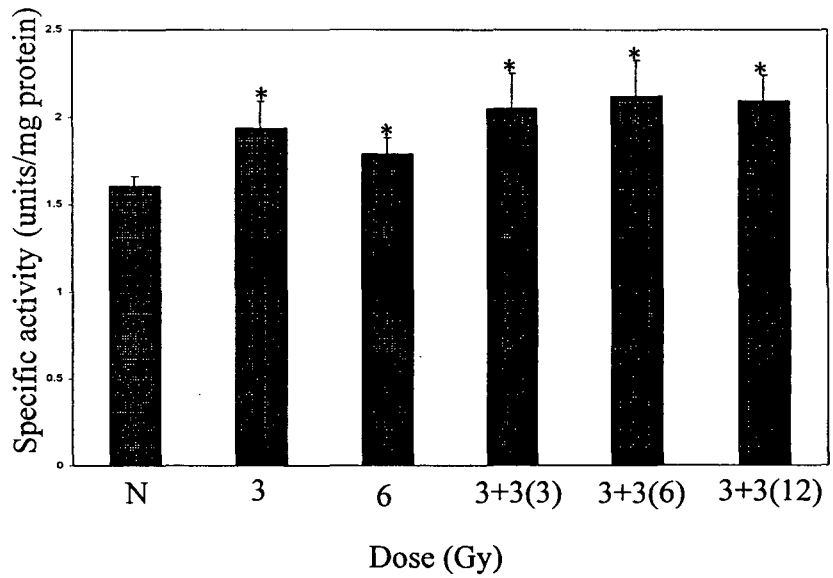

Figure 19: Effect of split doses of gamma rays measured as change in the specific activity of glyoxalase I (Gly I) in the liver of mice. Values in parentheses represents time interval between two equal fractions (3+3 Gy) of doses. Error bars represent standard deviation. * Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

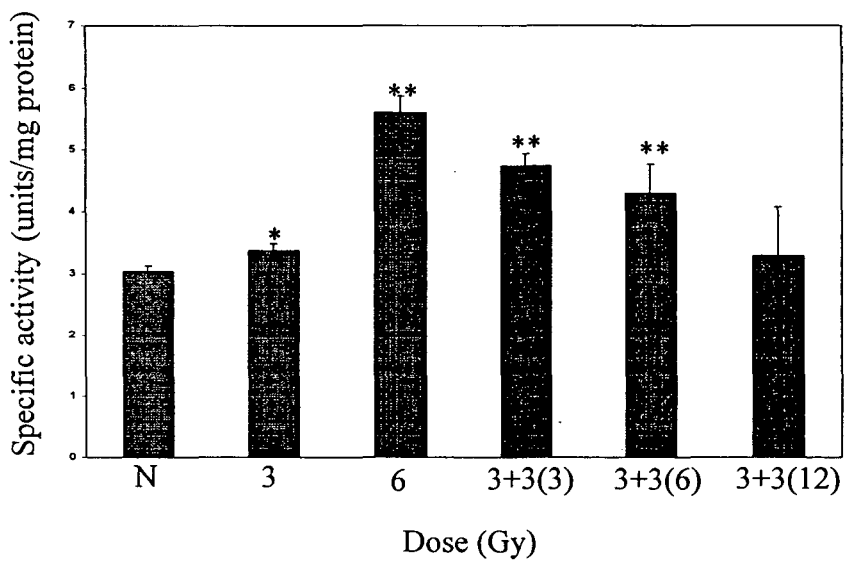

Figure 20: Effect of split doses of gamma rays measured as change in the specific activity of glyoxalase II (Gly II) in the liver of mice. Values in parentheses represents time intervals between two equal fractions (3+3 Gy) of doses. Error bars represent standard deviation * Significantly different ($p < 0.05$) against unirradiated control. **Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

Table 2: Effect of different dose rates of γ -rays on Lipid peroxidation in the liver of mice

Dose (Gy)	Dose rate Gy/sec	Lipid peroxidation n moles of MDA /mg protein
3Gy	0	0.665± 0.032 (100)
	0.24	0.681 ± 0.087 (102)
	0.06	0.712 ±0.049 (107)
	0.015	0.798 ±0.036* (121)
7Gy	0.24	0.711 ±0.042 (107)
	0.06	0.835 ± 0.097* (126)
	0.015	0.864 ±0.121* (130)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein.
*Significantly different ($p < 0.05$) against unirradiated control.

Table 3: Effect of split dose of γ -rays measured as change in Lipid peroxidation in the liver of mice.

Doses (Gy)	Time interval b/w irradiation	Lipid peroxidation n moles of MDA / mg protein.
0	-	0.665± 0.032 (100)
3	-	0.798 ± 0.041* (120)
6	-	0.854 ± 0.117* (128)
3+3	3	0.810 ± 0. 035** (121)
3+3	6	0.760 ± 0.096 (114)
3+3	12	0.797 ± 0.041* (116)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein. Dose rate used 0.015Gy/sec.

*Significantly different ($p < 0.05$) against unirradiated control.

** Significantly different ($p < 0.01$) against unirradiated control.

Table 4: Effect of pre-exposure to conditioning dose of γ -rays followed by subsequent higher dose of radiation on Lipid peroxidation in the liver of mice

Dose (Gy)	Time interval b/w irradiation (hr)	Lipid peroxidation n moles of MDA/mg protein
0	-	0.665 ±0.032 (100)
0.5	-	0.681 ±0.031 (102)
3	-	0.798 ± 0.036* (120)
0.5+3	3	0.757±0.024* (113)
0.5+3	6	0.717 ±0.026 (107)
0.5+3	12	0.729 ±0.035 (109)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein. Dose rate used 0.015 Gy/sec.

*Significantly different ($p < 0.01$) against unirradiated control.

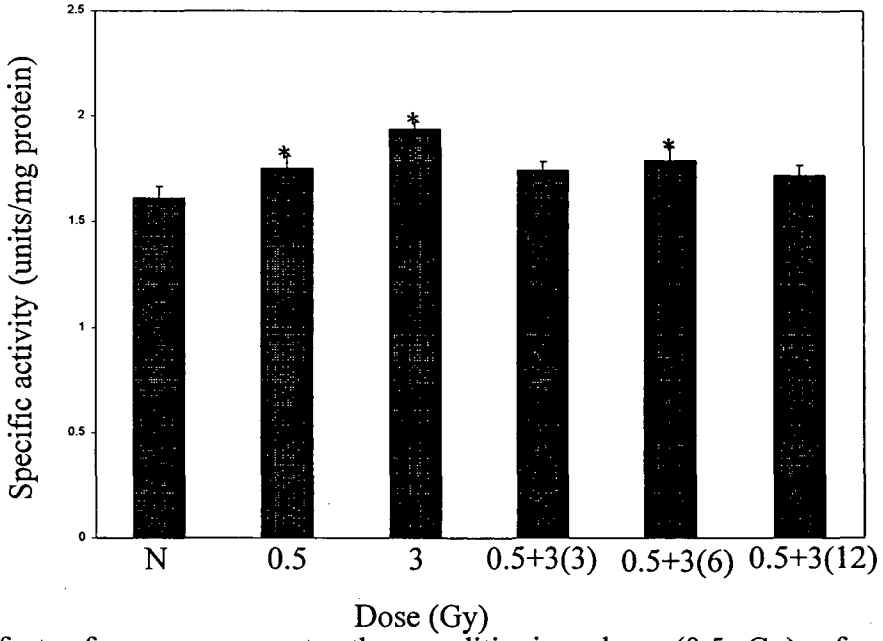

Figure 21: Effect of pre-exposure to the conditioning dose (0.5 Gy) of gamma rays followed by a subsequent higher dose (3 Gy) of radiation on glyoxalase I (Gly I) specific activity in the liver of mice. Values in parentheses represents time interval between conditioning and challenging doses. Error bars represent standard deviation * Significantly different ($p < 0.05$) against unirradiated control. N: unirradiated control.

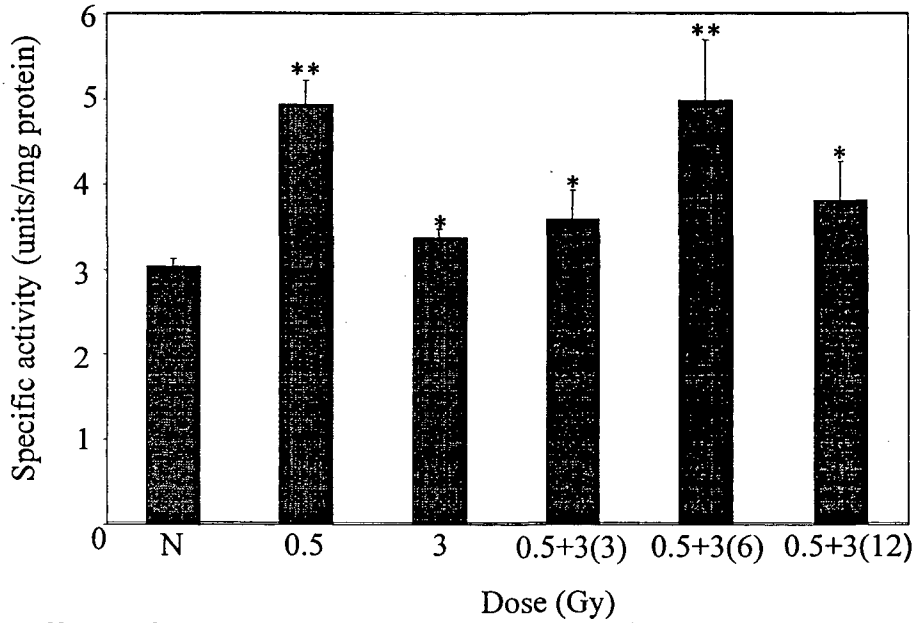

Figure 22: Effect of pre-exposure to the conditioning dose of gamma rays followed by a subsequent higher dose of radiation on glyoxalase II (Gly II) activity in the liver of mice. Values in parentheses represents time interval between conditioning and challenging doses. Error bars represent standard deviation. *Significantly different ($p < 0.05$) against unirradiated control. ** Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

4.4 Effect of Radiation on Antioxidant Enzyme System

SOD, GST, and catalase are important member of antioxidant defense system. The effect of different doses (0-7 Gy) on these enzymes has been studied in the liver of mice. The specific activities of SOD and GST were found to increase with the dose upto 5 Gy and then declined beyond 5 Gy (Table 13). However, there was continuous decline in the catalase activity with increase in dose of radiation.

For the adaptive response studies, the specific activity of antioxidant enzymes i.e. SOD, GST and Catalase was measured in the liver of the mice 6h after irradiation with 3 and 7 Gy using different dose rate such as: 0.25, 0.06 and 0.015 Gy/sec (Table 14). The specific activities of SOD and GST declined progressively while the activity of catalase increased with decreasing dose rate.

The effect of split dose on the antioxidant enzymes is shown in Table 15. Total dose of 6 Gy was fractionated into two i.e. 3+3 Gy. The time intervals between the exposures of two fractions were 3, 6 and 12h. It was found that the specific activities of SOD and GST were enhanced due to fractionation of the dose. Maximum increase was seen at 6h interval between two exposures. Overall response of Catalase was found to be inhibitory (Table 15).

The response of antioxidant enzymes was also examined in the liver of mice which were pre-exposed to an adaptive dose of 0.5 Gy and subsequently irradiated with challenging dose of 3 Gy (Table 16). The specific activities of SOD and GST were enhanced in the group of animals which were first irradiated with 0.5 Gy and then with 3 Gy. The mode and magnitude depends on the time interval between the exposure of conditioning and challenging dose of radiation. However, catalase showed inhibitory response. The decline in the activity was small but consistence.

Table 13: Effect of various doses of γ -rays on Specific activity of different antioxidants enzymes in the liver of mice.

Doses (Gy)	Specific activity (units/mg protein) SOD	GST	CATALASE
0	7.02 ± 0.540 (100)	1.74 ± 0.117 (100)	33.42± 1.99 (100)
1	7.79 ± 0.545 (111)	1.97 ±0.211 (113)	31.90 ± 3.87 (95.5)
3	8.05 ± 0.343* (116)	2.05 ± 0.153* (118)	29.66 ±1.60 (89)
5	8.51 ±0.523* (121)	2.29 ±0.157* (131)	26.78 ±2.77* (80.1)
7	7.85 ± 0.976 (112)	1.99 ±0.192 (114)	25.14 ±3.40* (75)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein. Dose rate used 0.06 Gy/sec

*Significantly different ($p < 0.05$) against unirradiated control.

Table 14: Effect of different dose rates of γ -rays on Specific activity of different antioxidants in the liver of mice.

Dose (Gy)	Dose rate Gy/sec	Specific activity (units/mg protein)		
		SOD	GST	CATALASE
3Gy	0	7.02 ± 0.540 (100)	1.74± 0.177 (100)	33.42 ± 1.999 (100)
	0.24	8.33 ± 0.530* (118)	2.102 ± 0.302 (121)	24.08 ±3.701* (72)
	0.06	8.08 ± 0. 810 (115)	2.05 ±0.208 (118)	29.66 ± 2.99 (89)
	0.015	7.87 ± 0.159* (112)	2.02 ± 0.237 (116)	37.51 ±3.91 (112)
7Gy	0.24	8. 15 ± 0.502* (116)	2.12 ± 0.163* (122)	23.13 ± 2.09** (69)
	0.06	7.85 ± 0.588 (112)	1.99 ± 0.098* (114)	25.14 ±3.201* (72)
	0. 015	7.38 ± 0.190 (105)	1.705 ±0.173 (98)	34.91 ±4.80 (104)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein.
*Significantly different ($p < 0.05$) against unirradiated control.
** Significantly different ($p < 0.01$) against unirradiated control.

Table 15: Effect of split dose of γ -rays measured as change in activity of different antioxidants in the liver of mice.

Dose (Gy)	Time interval b/w irradiation (hr.)	Specific activity (units/ mg protein)		
		SOD	GST	CATALASE
0	-	7.02 ± 0.540 (100)	1.74 ± 0.177 (100)	33.42 ± 1.999 (100)
3	-	7.87 ± 0.159* (112)	2.02 ± 0.203 (116)	37.13 ± 4.120 (112)
6	-	7.55 ± 0.098 (107)	1.90 ± 0.210 (109)	35.83 ± 5.033 (106)
3+3	3	8.93 ± 0.999* (127)	2.13 ± 0.245* (122)	31.08 ± 3.213 (93)
3+3	6	9.22 ± 1.151* (131)	2.24± 0.501 (128)	27.21 ± 1.700* (81.5)
3+3	12	7.80 ± 0.802 (111)	2.15 ± 0.205* (123)	29.03 ± 3.78 (87)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein. Dose rate used 0.015 Gy/sec.

*Significantly different ($p < 0.05$) against unirradiated control

Table 16: Effect of pre-exposure to conditioning dose of γ -rays followed by subsequent higher dose of radiation on specific activity of different antioxidants in liver of mice

Dose (Gy)	Time interval b/w irradiation (hr)	Specific activity (units/mg protein)		
		SOD	GST	CATALASE
0	-	7.02 ± 0.540 (100)	1.74 ±0.177 (100)	33.42 ±1.99 (100)
0.5	-	7.40 ± 0.163 (105)	1.87 ±0.065 (107)	34.14 ±2.065 (102)
3	-	7.87 ±0.159* (112)	2.02 ±0.037* (116)	37.50 ±3.020 (112)
0.5 +3	3	8.46 ±0.158* (120)	2.15 ±0.085** (123)	31.80± 3.903 (95)
0.5+3	6	8.77 ±0.132** (125)	2.26 ±0.071** (129)	29.34 ±2.103 (88)
0.5+3	12	8.34 ±0.172* (118)	2.15 ±0.056** (124)	27.01 ±3.891 (81)

Each value represents an average of at least three experiments ± standard deviation. The values in parentheses represents the percentage change in the enzyme activity/mg protein. Dose rate used 0.015 Gy/sec.

*Significantly different ($p < 0.05$) against unirradiated control.

** Significantly different ($p < 0.01$) against unirradiated control.

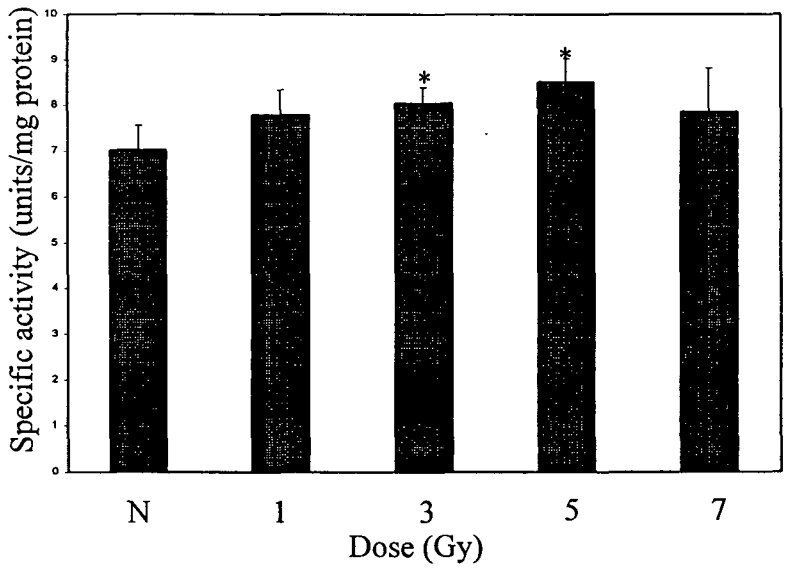

Figure 23: Effect of various doses of the gamma rays on the specific activity of Superoxide dismutase in the liver of mice. Error bars represent standard deviation* Significantly different ($p < 0.05$) against unirradiated control. N: unirradiated control.

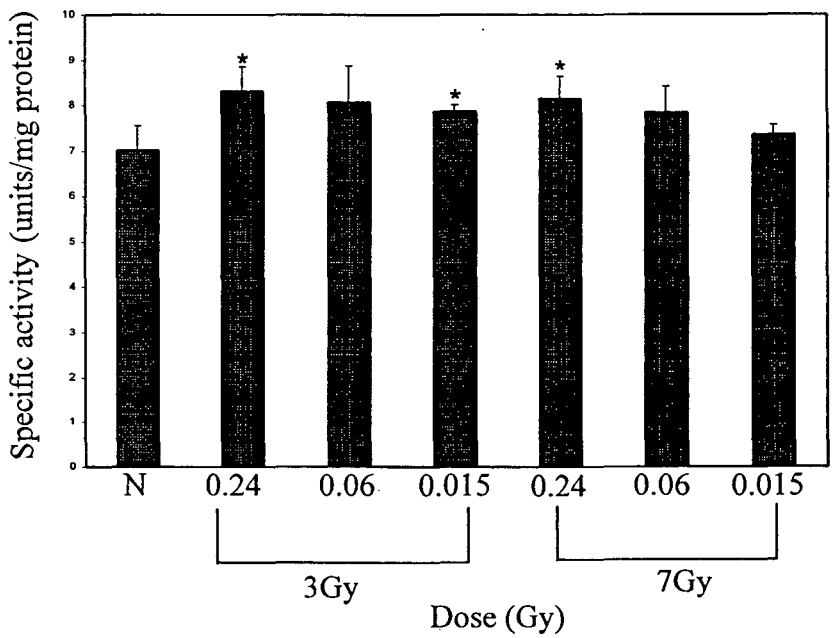

Figure 24 : Effect of different dose rates (0.24. 0.06, 0.015 Gy/sec) of the gamma rays on the specific activity of Superoxide dismutase in the liver of mice. Error bars represent standard deviation * Significantly different ($p < 0.05$) against unirradiated control.** Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated contol.

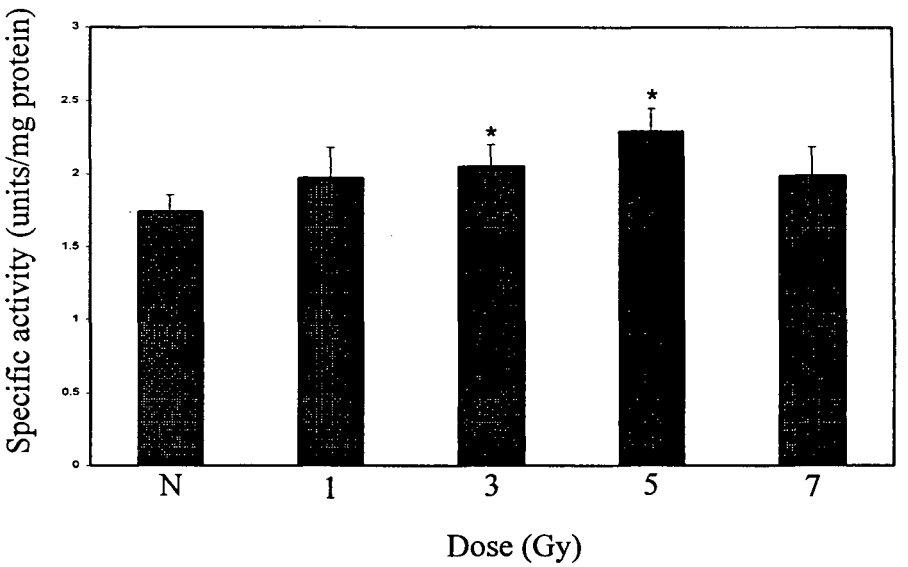

Figure 25: Effect of various doses of the gamma rays on the specific activity of Glutathione- S-transferase in the liver of mice. Error bars represent standard deviation. * Significantly different ($p < 0.05$) against unirradiated control. N: unirradiated control.

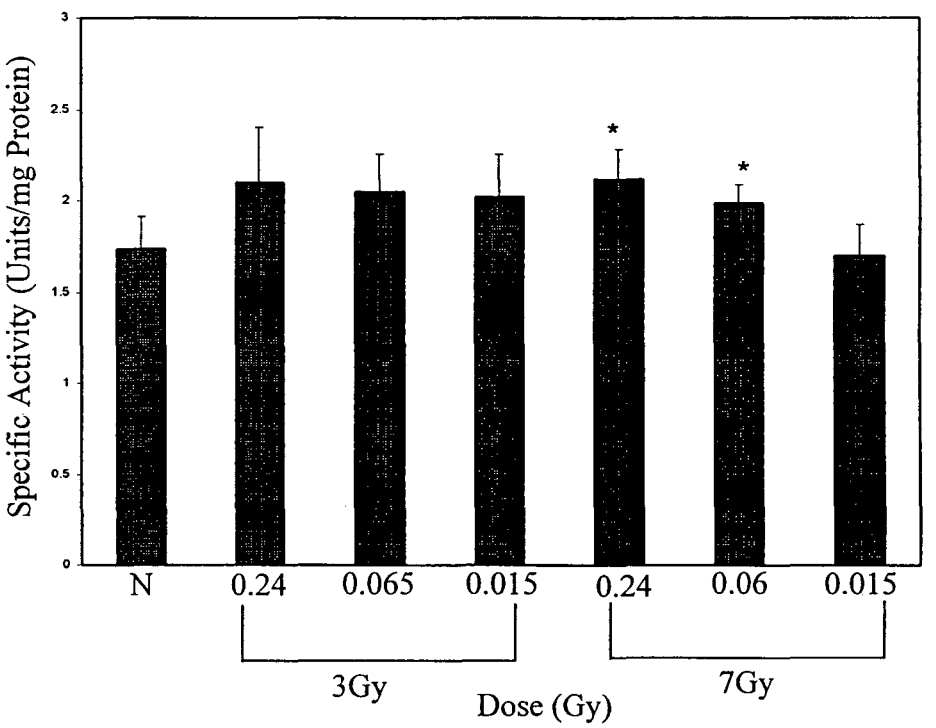

Figure 26: Effect of different dose rates (0.24. 0.06, 0.015 Gy/sec) of the gamma rays on the specific activity of Glutathione-S- transferase in the liver of mice. Error bars represent standard deviation * Significantly different ($p < 0.05$) against unirradiated control. N: unirradiated control.

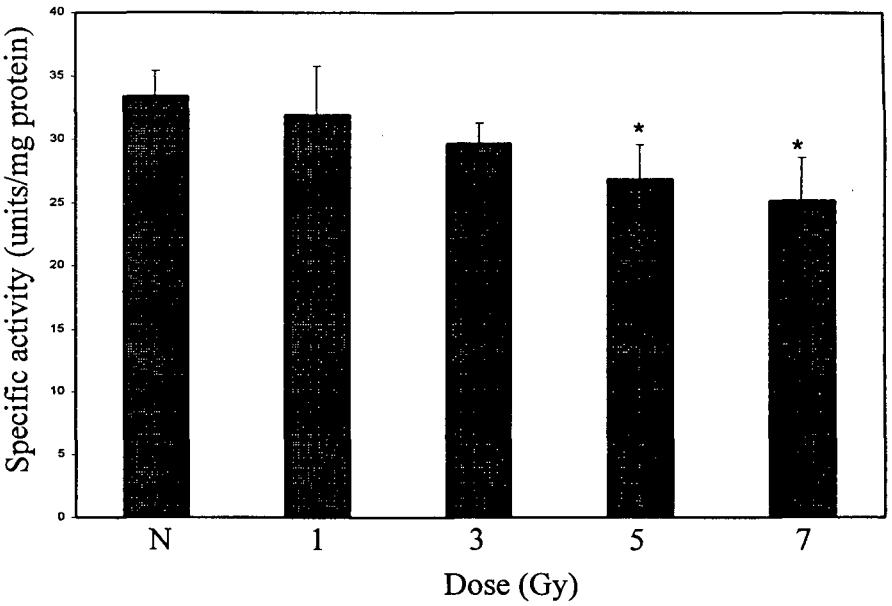

Figure 27: Effect of various doses of the gamma rays on the specific activity of Catalase in the liver of mice. Error bars represent standard deviation * Significantly different ($p < 0.05$) against unirradiated control. N: unirradiated control.

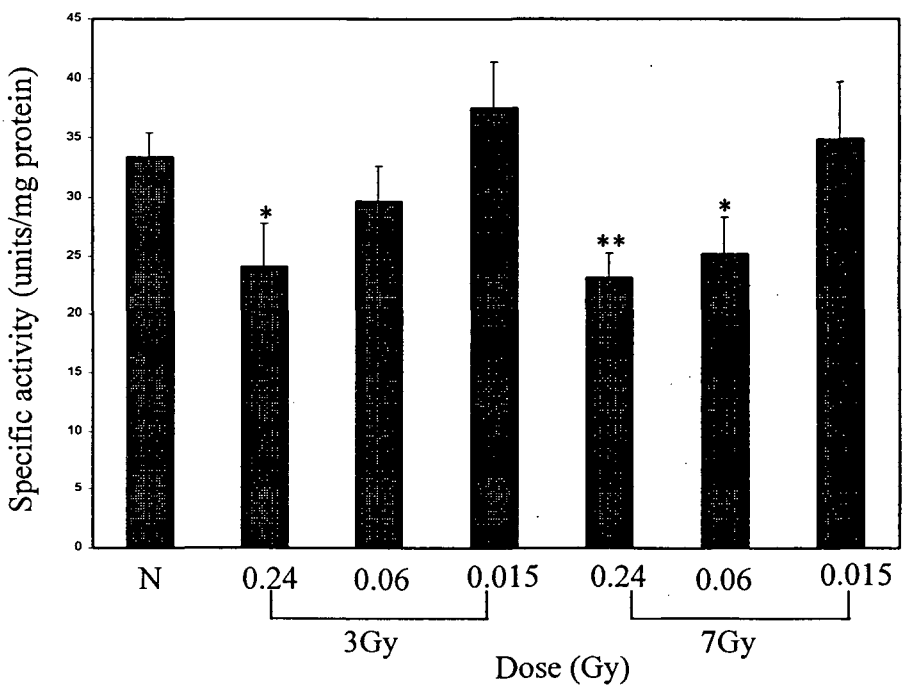

Figure 28: Effect of different dose rates (0.24. 0.06, 0.015 Gy/sec) of the gamma rays on the specific activity of Catalase in the liver of mice. Error bars represent standard deviation * Significantly different ($p < 0.05$) against unirradiated control.** Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

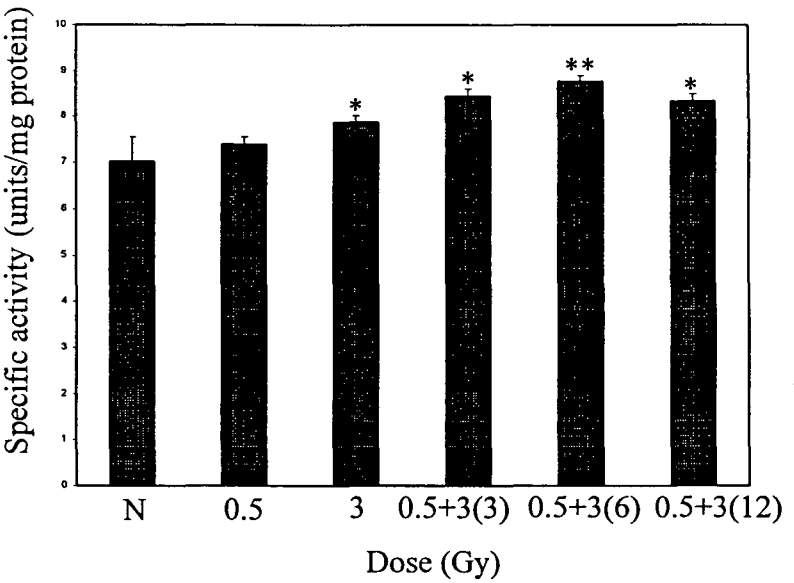

Figure 29: Effect of pre-exposure to the conditioning dose of gamma rays followed by a subsequent higher dose of radiation on Superoxide dismutase specific activity in the liver of mice. Values in parentheses represents time interval between conditioning and challenging doses. Error bars represent standard deviation * Significantly different ($p < 0.05$) against unirradiated control. ** Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

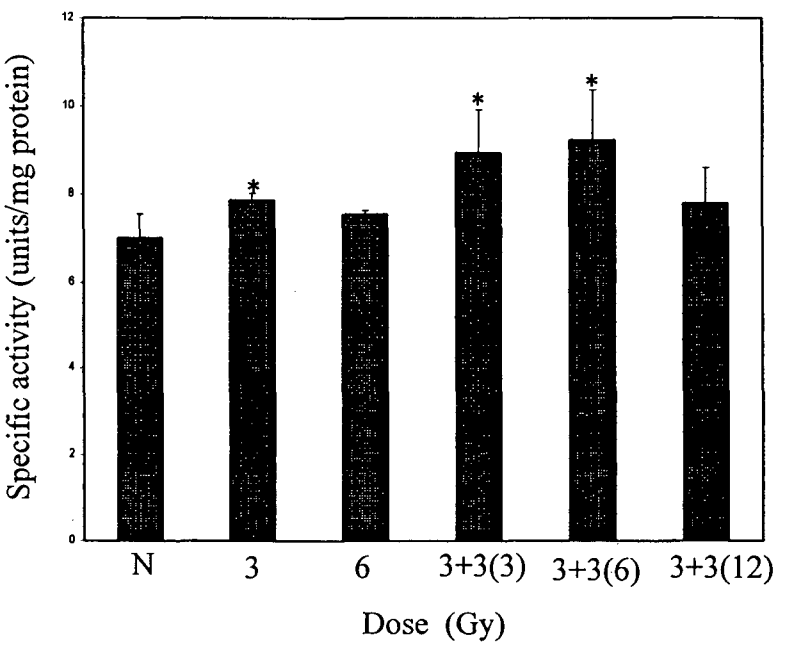

Figure 30: Effect of split doses of gamma rays measured as change in Superoxide dismutase specific activity in the liver of mice. Values in parentheses represents time interval between two equal fractions (3+3 Gy) of doses. Error bars represent standard deviation * Significantly different ($p < 0.05$) against unirradiated control. N: unirradiated control.

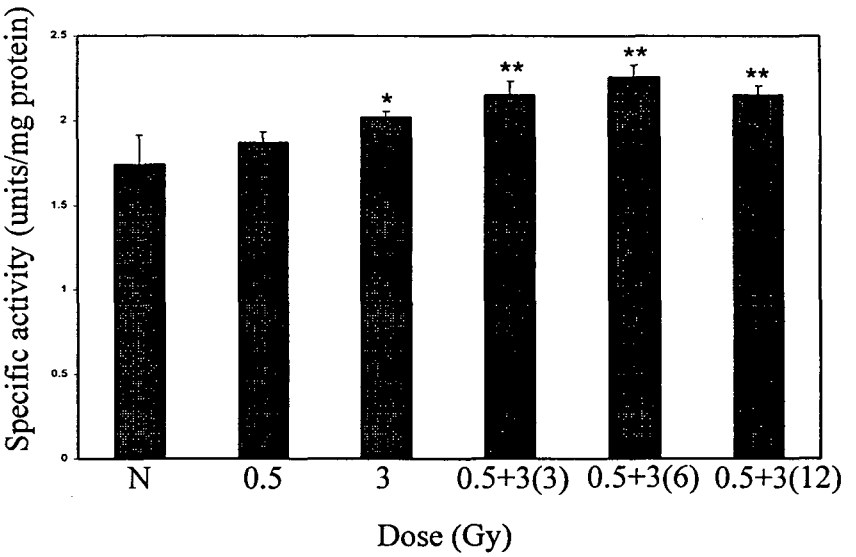

Figure 31: Effect of pre-exposure to the conditioning dose of gamma rays followed by a subsequent higher dose of radiation on the specific activity of Glutathoine-S-transferase in the liver of mice. Values in parentheses represents time interval between conditioning and challenging doses. Error bars represent standard deviation *Significantly different ($p < 0.05$) against unirradiated control. **Significantly different ($p < 0.01$) against unirradiated control. N: unirradiated control.

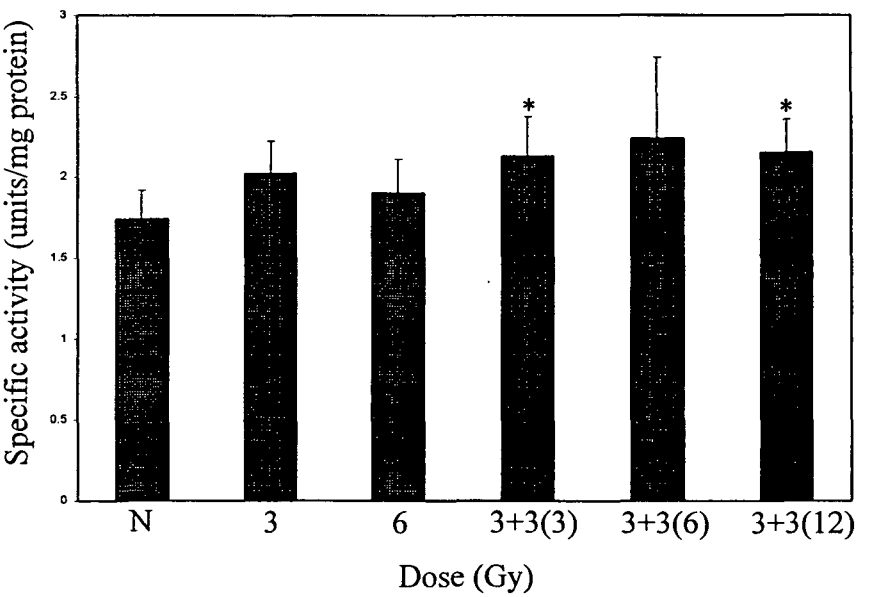

Figure 32: Effect of split doses of gamma rays measured as change in the specific activity of Glutathione-S-transferase in the liver of mice. Values in parentheses represents time interval between two equal fractions (3+3 Gy) of doses. Error bars represent standard deviation * Significantly different ($p < 0.05$) against unirradiated control. N: unirradiated control.

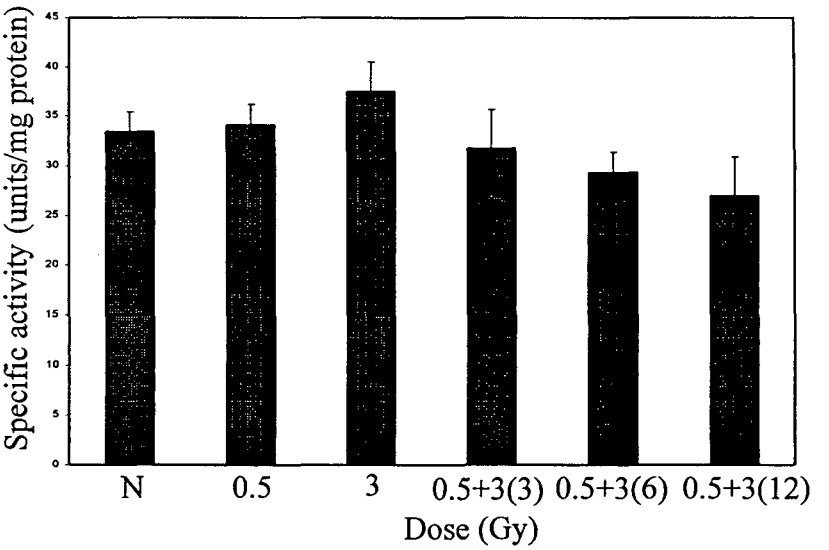

Figure 33: Effect of pre-exposure to the conditioning dose (0.5 Gy) of gamma rays followed by a subsequent higher dose (3 Gy) of radiation on the specific activity of Catalase in the liver of mice. Values in parentheses represents time interval between conditioning and challenging doses. Error bars represent standard deviation. No significant change found. N: unirradiated control.

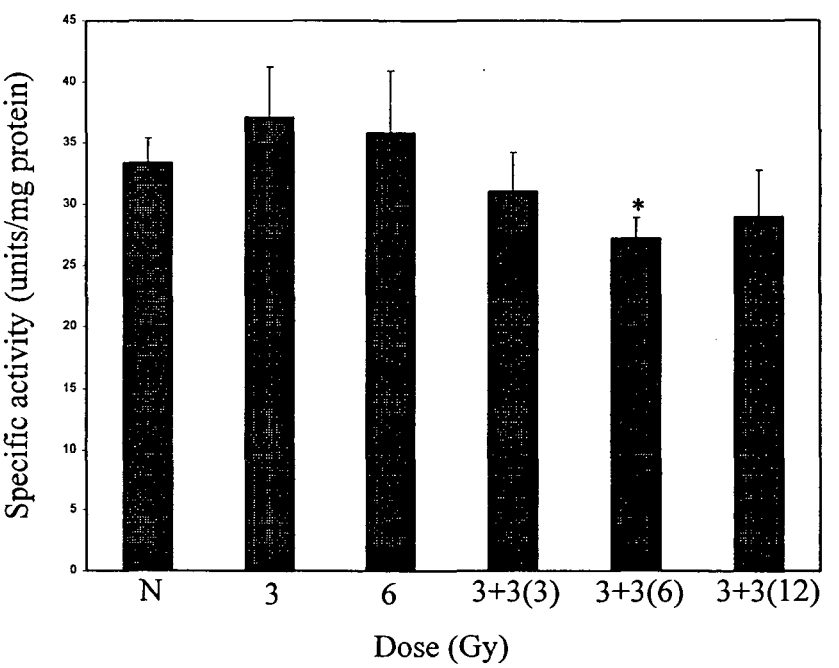

Figure 34 : Effect of split doses of gamma rays measured as change in the specific activity of Catalase in the liver of mice. Values in parentheses represents time interval between two equal fractions (3+3 Gy) doses. Error bars represent standard deviation. *Significantly different ($p < 0.05$) against unirradiated control. N: unirradiated control.