Resting tremors were induced in mice with harmine (15 mg/kg). The severity of tremors were scored as intensive, severe, moderate or simple presence designated by 4, 3, 2 and 1 pluses, respectively. The tremors were scored at 5, 7, 15, 20, 30, 45 and 60 min after harmine administration. The cumulative scores of tremors were plotted against time. Levo-propranolol (20 and 40 mg/kg), dl-propranolol (20 mg/kg) were administered to mice 30 min prior to i.p. injections of harmine alone or in combination with quipazine (20 mg/kg). To verify that harmine does indeed act through 5-HT receptors, cyproheptadine (0.5 mg/kg) was used. To show whether propranolol exerts its effects on the harmine-induced tremors as a result of its anti-serotonergic or anti-adrenergic activity, another beta-blocker, practolol (20 mg/kg) and an alpha-blocker, phenoxybenzamine (15 mg/kg) were employed. Any change in animal behaviour such as stereotypic cage biting response and vocalization, brought about by harmine, quipazine or their combination were noted.

Harmine-induced tremors were noticeable within minutes of its administration. The peak effect was apparent in 5–7 min and lasted up to 15–20 min. Quipazine prolonged the duration of harmine-induced tremors. These animals showed severe head and body movements, repetitive vocalization, and stereotypic cage biting behaviour. The peak effect, i.e. maximum intensity of tremors appeared much faster in combination treatment (figure). Both l- and dl-propranolol significantly antagonized harmine effects, the higher dose showing complete inhibition of tremors. Quipazine-induced prolongation of the harmine effect was also blocked by prior treatment of mice with l-propranolol (figure). Cyproheptadine significantly blocked the effects of harmine. Similarly, it also reduced the severity and the duration of tremors that induced by combined treatment of harmine and quipazine. However, phenoxybenzamine and practolol did not modify the harmine-induced tremors (figure).

The structural similarities between harmine and serotonin would suggest that a modification of serotonin function in the central nervous system may be involved in eliciting the pharmacologic effects of harmine[2]. Moreover, cyproheptadine, a 5-HT antagonist, blocked the effects of harmine. The exaggerated effects of harmine in quipazine-pretreated mice could very well be explained on the basis of their common site of pharmacologic action, namely, the serotonergic receptors.

There is convincing evidence in the literature supporting the idea that l-propranolol competitively antagonizes the effects of 5-HT on smooth muscles. These effects are demonstrated to be independent of the general membrane stabilizing properties of beta-adrenergic blocking agents[7]. Green and Grahame-Smith[4] using 5-HT precursors have recently demonstrated that l- and dl-propranolol in intact animals inhibited central serotonin function. It is also known that propranolol abolishes the stimulant effects of 5-HT on transmission in the cat superior cervical ganglion[7]. The present observations that l-propranolol significantly antagonized the harmine-induced tremors in mice could, therefore, be attributed to its anti-serotonergic property. Furthermore, neither practolol, a relatively specific beta-blocker, nor phenoxybenzamine had any effect on harmine-induced behavioural changes in mice.

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Modification of radiation-induced spleen weight changes in mice by 2-mercaptopropionylglycine

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Summary. It was found that the MPG partially protects the spleen against weight loss due to radiation, and exaggerates the compensatory reaction in the tissue during recovery. It is also concluded that MPG protects the stem cells in the spleen, which helps to restore the peripheral blood by extramedullary erythropoiesis.

2-Mercaptopropionylglycine[1] (MPG), an artifically synthesized thiol compound is effectively radioprotective at a very low optimum dose as compared to other radioprotectors[2-3]. Recently MPG was tested on various tissues of Swiss albino mice after external irradiation in this laboratory[4-9].

Material and methods. Male Swiss albino mice of 6–9 weeks with an average weight of 24 g were selected from an inbred colony maintained on Standard mice feed (procured from the Hindustan Lever Ltd, Delhi) and water ad libitum. 3 doses of gamma rays were used for the experiment. Each dose was given to an experimental (MPG treated) and a control (untreated) group with an equal number of animals. MPG (2-mercaptopropionylglycine, received from Santen Pharmaceutical, Osaka, Japan, was dissolved in distilled water so as to give a concentration of 1 mg/ml and pH was adjusted at 6.4 with 0.1 NaoH) was injected i.p. with 20 mg/kg b.wt. in the experimental animals and control groups were given an equal volume of distilled water in the same manner. After 15–30 min of this treatment, the animals were exposed to 500, 1000 and 1500 R at the dose rate of 24 R/min.

Animals were sacrificed by cervical dislocation at 12, 24, 72 h and 5, 7, 14, 21 and 28 days after irradiation. At least 4 animals were used at each interval and the wet weight of the spleen from each animal was determined separately. The results were calculated as the mean tissue weight ± SE and were plotted against autopsy interval on a linear graph.

Results. Control. The pattern of weight changes is similar in all the control groups up to 7 days after irradiation. But reduction in weight is dose dependent, i.e. the higher the dose (1500 R) the greater is the weight loss (table, figure). The maximum weight loss is observed at 7 days. The spleen in 1000 R group becomes more than 3 times heavier than that of the 500 R group at 21 days after exposure. After 21 days weight decreases but does not return to the normal level by 28 days. However, in the 500 R group the weight gain is slow and does not reach normal value even at 28 days. Supralethally (1500 R) irradiated

Changes in spleen weight[a] of Swiss albino mice exposed to various doses of gamma rays in the presence and absence of MPG

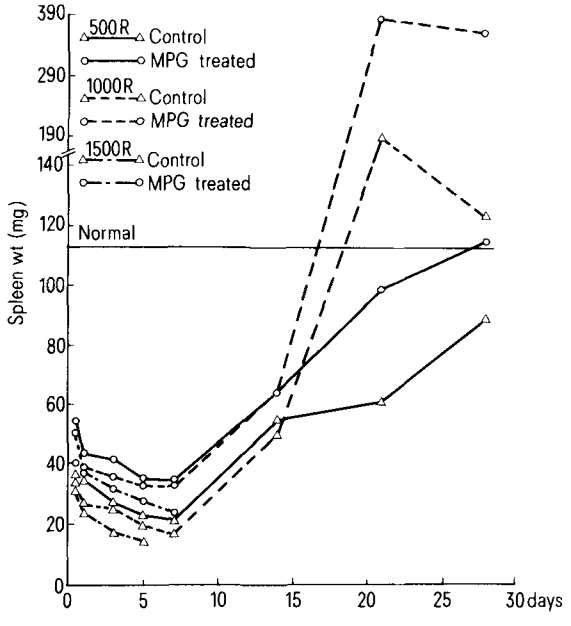
Intervals doses	12 h	24 h	72 h	5 days	7 days	14 days	21 days	28 days
Control 500 R	37.0±4.4	35.0±0.7	27.0±2.4	23.0±2.5	22.3±1.1	55.0±3.1	61.3±6.9	88.7±11.8
Experimental	55.0±7.4 NS	43.6±0.8 p<0.002	42.0±6.8 NS	34.6±4.3 NS	34.0±3.7 p<0.05	64.3±2.9 NS	99.0±4.6 p<0.02	115.0± 3.1 NS
Control 1000 R	33.3±2.9	27.6±2.5	26.0±2.5	20.6±0.8	16.6±1.5	50.0±2.8	190.3±6.7	123.5±12.2
Experimental	51.0±2.8 p<0.02	39.0±2.5 p<0.05	36.0±2.5 p<0.05	33.0±4.4 p<0.05	33.6±0.8 p<0.001	64.3±2.7 p<0.05	390.0±6.4 p<0.001	336.6±10.1 p<0.001
Control 1500 R	32.3±3.5	23.6±1.8	17.3±2.3	14.6±2.3	[b]	[b]	[b]	[b]
Experimental	41.0±2.5 NS	38.6±0.4 p<0.002	32.0±1.2 p<0.01	28.0±2.8 p<0.05	24.0±4.3	[b]	[b]	[b]

[a] Each value is the mean of 4 readings ± SE. [b] No survivors. Spleen weight in normal unirradiated animals = 113.6±3.9.

animals did not survive beyond 5 days and therefore no further study was possible.

Experimental. In all the exposed groups the pattern of spleen weight loss is similar to that of the control animals upto 7 days, i.e., loss of weight increases from sublethal to supralethal. But the weight reduction is much less as compared to control groups at each respective interval. After day 7 there is a gradual increase in weight in the 500 R irradiated animals which reaches normal level by 28 days. However, in 1000 R exposure group the organ shows 3.5- and 3-fold increases in weight as compared to unirradiated normal animals and 2- and 3-fold increases as comapred to the control group with the same dose at 21 days and 28 days respectively (table, figure).

Discussion. One of the earliest manifestations of the radiation effect is a reduction in organ weight, which reflects the histopathological changes in the tissues. Weight change varies with dose in a relatively simple manner; those of the more radiosensitive organs could be described by a linear change in the logarithm of the weight against dose[10-12]. The spleen, however, showed a more complicated and unusual response in the present observations. Although maximum reduction takes place at 7 days in both control and experimental animals in all exposure doses, at 21 days tissue weight in 1000 R irradiated unprotected animals becomes 3 times greater than in the control of 500 R and 1½ times than in the unirradiated normal. This increase in weight is followed by a decrease to near normal value at 28 days. In the protected irradiated animals the initial weight loss is significantly lower than in the control animals in most of the intervals upto 7 days in sublethally to supralethally irradiated animals. In 1000 R-MPG treated animals the weight becomes more than 2½ times the control with the same dose and 4 times greater than 500 R-MPG protected animals at this interval (table, figure). In the histopathological study white pulp and red pulp decrease in mass while areas of the extramedullary erythropoiesis increase strikingly more in protected and less in unprotected animals[13]. It is well established fact that red pulp contains undifferentiated or 'stem' cells, and ectopic erythropoiesis was observed in these areas after treatment with glutathione[14] and MEG[15]. Similarly in the present study in 1000 R irradiated animals, when the bone marrow becomes totally aplastic and proliferative capacity of other blood forming organs is reduced or nullified, pretreatment by MPG results in accelerated production of 'stem' cell types, differentiating into erythroblasts and myeloblasts in these regenerating areas to compensate the peripheral blood-cell loss. Thus MPG protects these (blast?) cells and causes an accelerated regeneration which may result in an increase in weight of the tissue.



Spleen weight changes in MPG treated and untreated Swiss albino mice exposed to 60 Co gamma radiation.

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