# RADIOPROTECTIVE EFFECT OF MPG ON THE PERIPHERAL BLOOD OF MAMMALS - A REVIEW*

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## SUMMARY

*The present review is mainly based on the experimental work regarding the effect of different doses of gamma rays on mouse blood with or without prior injection of MPG (2-mercaptopropionyl-glycine). It is found that MPG has no noticeable effect on the various peripheral blood components at an early post-irradiation phase. However, it definitely provides a significant protection to them at a later phase when the recovery is expected. It also provides an early and fast recovery after low dose exposure of gamma rays when compared with non-drug treated irradiated animals acting as control. Clinical studies carried out in cancer patients also indicate that the drug provides effective protection against the side effects of radiation.*

## Introduction

Since the discovery of chemical radiation protection, an intense search was on with a view to find a substance, which would give an effective protection with a minimum toxicity. Various chemical compounds have been shown to protect against ionizing radiation, but in comparison to others, MPG has better properties. MPG (2-mercaptopropionyl glycine) is a synthetic-SH compound. It is a condensation product of thiolactic acid with glycine ($CH_3$-CH-CONH-$CH_2$-COOH). It is commercially available in the name of Tiola (Japan) and widely used as a detoxicating agent. It is orally effective and is protective at a very low optimum dose of 20 mg/Kg body weight.

The radioprotective action of MPG was first reported by Koyama et al in 1965[1]. They reported an increase in the survival time of mice exposed to daily doses of x-rays after the injection of MPG. However, they could not establish it as a radioprotector. In 1970, Sugahara and his co-workers[2] showed MPG to be a potent radioprotector, effective in a very low, non-toxic dose by oral or parenteral route in the mouse as well as in man. Dose reduction factor (DRF) based on LD/50/30 was 1.4 at the dose of 20 mg/Kg body weight or 0.5 mg/mouse. When the drug dose was increased to 1.0 mg/animal (approximately 40 mg/Kg body weight), the DRF was reduced to 1.22. Thus it is clear that MPG is a better protector at a lower dose than at a higher dose[3]. The LD 50 toxic dose for mice has been reported to be 1400 mg/Kg body weight[4]. Toshioka et al[5], using $S^{35}$ labelled Tiola in rats showed that MPG reached a maximum blood level of about 14 Ug/ml in 15 minutes after i.p. injection, and this formed about 70% of the total dose of MPG. At one hour after injection, the radioactivity was reduced to half and after that it gradually decreased. The present review is based on our personal experience and the available reports on the radioprotective effect of MPG on peripheral blood components.

## Material and methods

Experimental studies were performed on inbred strain of Swiss albino mice. The animals were bred and maintained on standard mice-feed and water ad libitum. The animals were broadly divided into two groups. One group of animals were injected with MPG in the dose of 20 mg/Kg body weight (MPG dissolved in double distilled water and pH of the solution adjusted to 6.4 with 0.1 N. NaOH) and another group of animals received equal amount of double distilled water in the similar fashion. Fifteen to thirty minutes after injections, all the animals were exposed to different doses of gamma rays (2.5, 5.0, 7.5, and 10.0 Gy in case of adult mice; 0.5, 1.5, 2.5 Gy in case of pregnant mice; and 1.0 and 3.0 Gy in case of young ones) from a $^{60}Co$ gamma source at the dose rate of 0.5 Gy/minute. Peripheral blood studies

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were conducted using routine methods at various post-irradiation time intervals i.e. 1/8 to 14 days (adult mice), 1 to 6 weeks (in-utero treated mice), and 2 to 6 weeks (young ones).

## I. Observations on adult mice

After 2.5 Gy exposure, in the control group, there was a gradual and continuous decrease in the erythrocyte count, haemoglobin and haematocrit values. However, in the MPG treated group, the values slowly started increasing from the fifth day onwards. After 5.0 and 7.5 Gy exposure, all the three parameters showed a continuous decline and the minimum values were recorded on 14th day. In the drug protected group, the values were significantly higher than in the control group at later intervals[6,7]. The erythrocyte and related parameters showed a similar pattern of changes after 10.0 Gy exposure group as in the case of lower exposure groups (i.e. 5.0 and 7.5 Gy), but at this dose level, there was no significant difference between the two groups. At this dose, all animals died within 10 days in control group, while in MPG treated group a few animals survived upto 12 days.

The values were significantly higher in the MPG treated animals than in the controls in the lower dose groups i.e. 2.5, 5.0 and 7.5 Gy at later autopsy time intervals. Patkau et al[8] also reported that superoxide dismutase did not significantly affect the initial phase of erythrocyte depletion but hastened the recovery. Uma Devi and Kumar[6] have reported that recovery was faster and complete in MPG pretreated animals as compared to non drug treated control animals after lower dose (2.5 Gy) of gamma radiation.

The pattern of total leucocyte, lymphocyte and neutrophil changes was similar in both sexes, but the female mice were more radiosensitive haematologically that male[9]. Recovery in leucocyte counts was observed in both sexes after 3 days in the 2.5 Gy and after 7 days in the 5.0 Gy group. Both male and female mice showed similar response to MPG treatment and at 14 days, the leucocyte counts in the experimental animals in both the exposure groups were significantly higher than in the control. Thus, these studies indicate that MPG did not have any noticeable effect on the radiation induced initial blood cell depletion, but the drug provided significant protection at later intervals, when the replenishment from the proliferative compartment is expected, indicating protection of the haematopoietic function. The drug significantly brought down the number of degenerating cells in the peripheral blood even at the early post-irradiation intervals, indicating a protection against the direct cell killing by radiation[10]. However, Ghosh and Pant[11] failed to detect leucocyte protection by MPG. This lack of protection may be due to the high dose radiation used by them. MPG protection of the blood cells has been further demonstrated in the studies on thymus by Kumar et al[12]. They reported that the maximum weight loss in the thymus corresponded to the depopulation of both medulla and cortex due to severe cell death and a very low mitotic activity. The recovery of organ weight resulted from active regeneration of the tissues, indicated by an increased number of mitotic figures. MPG reduced the early cell killing effect of gamma rays and also hastened recovery by accelerating mitotic activity both in medulla and cortex and reducing cell death, especially in cortex at later intervals.

Recently, Kumar and Uma Devi[13] reported that the total plasma proteins level was lower on the 5th and 7th day in the 5 and 10 Gy exposure groups respectively. No significant changes were observed either in the drug protected or in the non-drug protected groups after 2.5 Gy exposure. A significant elevation in the plasma protein level was found at later intervals i.e. from 5th day onwards in the 5.0 Gy and 10 Gy dose groups receiving MPG before irradiation in comparison to the corresponding non-drug protected group. To some extent, MPG also checked the radiation induced rise in cholesterol level in the blood plasma[14]. The time and and extent of radiation induced changes and the protection afforded by the drugs on the different peripheral blood component depend upon the exposure dose; the higher the dose the more the damage and the lesser the protection. The drug also provides effective protection to the blood components after fractionated doses of gamma rays[15]

## II. Observation on in-utero treated mice

Radioprotective effect of MPG was also evaluated in young mice which were irradiated at different days of foetal life (14-1/4 post conception days (P.C.) 16-1/4 p.c. and 18-1/4 p.c.) with different doses of

gamma rays i.e. 0.5, 1.5 and 2.5 Gy.

It was found that the neonatal abnormalities were reduced by MPG. The peripheral blood studies showed a significant elevation in various blood components in one and two weeks old animals exposed on 14-1/4 and 16-1/4 days of foetal life, while those irradiated at later embryonic live (18-1/4 p.c.) after prior administration of MPG showed higher values after one week postpartum [16,17]. The maximum depression in leucocyte count was observed at 18-1/4 p.c./1 week and 18-1/4 p.c./2 week animals as compared to earlier gestations [18]. It is possible that 14-1/4 p.c. animals had a relatively longer intrauterine life to recover than 16-1/4 or 18-1/4 p.c. irradiated animals. Thus it may be concluded that the drug protected peripheral blood constituents of post-natally developing mice by boosting up haemopoiesis and helping in rapid replenishment of blood cells by indirect protection provided to the neonates.

### III. Observation on developing mice

The effect of this drug on the peripheral blood cells was also studied in post-natal developing Swiss albino mice. For this purpose, one and two weeks old mice were exposed to 1.0 and 3.0 Gy of gamma rays in presence or absence of MPG. It was found that MPG provides significant protection to the peripheral blood commonents against radiation induced changes [19]. It was also observed that 1.0 Gy did not produce any noticeable change in erythrocytes and haemoglobin level during the study i.e. upto six weeks, whereas after 3.0 Gy, significant depletion was noted in these parameters as compared to normal [20]. Goyal et al[17] irradiated animals in uterus and reported that erythrocyte count decreased at early post-natal age with 50 and 150 R gamma rays. They also reported that the depression in various blood components was dose dependent. Thus it may be concluded that animals irradiated in uterus are more sensitive to radiation as compared to young ones.

### Clinical studies

A few clinical studies have been conducted by various investigators in cancer patients receiving radiotherapy. Tanaka (1972)[21] studied the radioprotective effect of MPG in patients with neoplastic diseases undergoing radiotherapy. He reported that leucocytes in the MPG treated group recovered earlier than control. Over 2000 R, the increase in cells with aberrations was remarkably depressed in the drug treated group as compared to the control. Recently, Kumar et al[22] and Nawalloha et al[23] observed the effect of this drug in patients with cancer of head and neck undergoing radiotherapy. They observed that MPG checked radiation induced side effects and maintained a higher haemoglobin level during and after radiotherapy in comparison to untreated patients.

### Mechanism of Protection

The ~~effect~~ mechanism by which MPG protects against radiation effects is not yet clearly understood. Preliminary studies indicate that the drug inhibits mitosis temporarily[24] and brings about metabolic inhibition[25]. The drug may also combine with the cellular sulphydryl and disulphides and form mixed disulphides, which act as a shielding around the target molecule against radiation. In the process of disulphide formation, NPSH (non-protein sulphydryl) is released which helps in scavenging the free radicals produced by irradiation. Glutathione has been shown to reduce the radiation induced formation of toxic lipid peroxides by converting them to non-toxic hydroxy acids. On the basis of mechanical simulation techniques, Brouch et al[26] reported that amino group of the radioprotector (cysteamine) may form a bridge between the phosphate groups of the DNA backbone, which in turn develops resistance against radiation. Thus MPG may also protect the DNA molecule in this way which in turn protects the chromosome and reduces abnormal mitosis. MPG seems to protect the chromosomes of the dividing cells by increasing the radioresistance of the biochemical molecules and/or by facilitating the enzymatic repair of the DNA lesions, as a result of which divisional abnormalities and the consequent cell death during tissue recovery are reduced. A large number of abnormal cells in the peripheral blood and mitosis-linked abnormalities of the intestinal epithelium[27] were also observed in control animals as compared to MPG treated animals.

Further studies, with the aim to under-

tand the mechanism of drug protection, as well as the application of this drug in clinical field are in progress.


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## LABORATORY BULLETIN

# Reference value in Haematology (Adult Male)

| | Conventional units | SI Units |
|---|---|---|
| Hb (d) | 14.0 - 18.0 grams/100 ml | 2.17 - 2.79 mmol/l |
| PCV (a) | 40 - 54% | 0.4 - 0.54 |
| **Indices** | | |
| MCH (d) | 27 - 31 picogram | 0.42 - 0.48 fmol |
| MCV | 80 - 105 cu. micra. | 80 - 105 fl |
| MCHC (a) | 32 - 36% | 0.32 - 0.36 |
| **Cell counts** | | |
| Erythrocytes | 4.6 - 6.2 million/c.mm. | $4.6 - 6.2 \times 10^{12}$/l |
| Leukocytes (T) | 4500 - 11,000/c.mm. | $4.5 - 11.0 \times 10^{9}$/l |
| Platelets | 150,000 - 350,000/c.mm. | $150 - 350 \times 10^{9}$/l |
| Reticulocytes (b) | 0.5 - 1.5% of erythrocytes | $25 - 75 \times 10^{9}$/l |
| Erythrocyte sedimentation rate | 0 - 10 mm in 1 hr. | 0 - 10 mm h. |
| S. Iron | 75 - 175 mcg/100 ml | 13 - 31 umol/l. |
| Total iron binding capacity | 250 - 410 mcg/100 ml | 45 - 73 umol/l. |
| Saturation (a) | 20 - 55% | 0.20 - 0.55 |
| S. Ferritin | 20 - 200 ngm/ml | 20 - 200 ug/l. |
| S. Folate | 2.3 ng/ml | 5.2 nmol/l |
| Erythrocyte folate | 140 ng/ml | 318 nmol/l |
| S. Vit. $B_{12}$ | 180 - 900 pg/ml | 133 - 664 pmol/l |
| S. Haptoglobin (d) | 100 - 700 mg/100 ml | 16 - 31 umol/l |
| Methaemoglobin (e) | 0 - 130 mg/100 ml | 4.7 - 20 umol/l |
| Plasma haemoglobin | 0 - 5 mgm/100 ml | 0 - 0.8 umol/l |
| RBC osmotic fragility | begins at .45 - .39% NaCl | Begins in 77-67 mmol/l NaCl |
| | complete in .33 - .30% Nacl | Completes in 56 - 51 mmol/l NaCl |
| Red cell protoporphyrin | 27 - 61 mcg/100 ml packed RBC | 0.48 - 1.09 umol/l packed RBC |
| Fibrinogen (c) | 200 - 400 mg/100 ml | 5.9 - 11.7 umol/l |

**Notes :**

a) Percentage is expressed as a decimal fraction.

b) Percentage may be expressed as a decimal fraction, however, the absolute value is more meaningful. There no reason other than custom for expressing reticulocyte count in percentage rather than in absolute number.

c) mol. wt. of fibrinogen = 341,000 Daltons

d) mol. wt. of Hb. = 64,500 daltons.
Conventional g/dl is retained by many, as there is disagreement as to whether the monomer or tetraner of Hb should be used in the conversion. The tetraner is used in this table.

e) Mol. wt. of methaemaglobin = 64,500 daltons. See 'd' above.