Prophylactic Effect of Marinamycin Against Ionizing Radiation

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マリナマイシンによる放射線防禦試験

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マリナマイシン（1957）（以下 M^2 と略称）はX線又は ^{60}Co 照射による Leukopeina に対して白血球数を増加させることについて，数回に亘つて，報告した．このものの，放射線防禦作用について，宮川，朝倉博士等により，^{60}Co 700r，をマウスに全身照射した場合の M^2 の投与時間を検討し，照射後に M^2 200mcg を腹腔内に投与した場合，或程度の防禦作用を認め，従来の作用機序と異る点を指摘している．私共は DD-S 系のマウス群を用いて，X線 100r 又は 200r を全身照射した場合，対照群は白血球減少を示すが，照射2時間前に，M^2 を腹腔内に投与すれば，末梢白血球数は，殆んど減少しないで，快復することが認められた．

2） ^{60}Co 700r を全身照射した場合，対照群は5匹中2匹が生きのこり，M^2 50mcg を2時間前に投与すると5匹中3匹生，24時間後に M^2 で治療すると同様，2日間後に投与すると5匹中4匹が生存する．

3） ^{60}Co 800r を全身照射した場合，対照群は5匹中5匹死亡するが，M^2 50mcg/mice 投与群は，30分後に投与されたものは5匹中5匹死亡するが，24時間後投与されると5匹中2匹たすかり，2～3日後から M^2 で治療されれば，5匹中4匹が健康に生存した．

即ち，^{60}Co 致死量を全身照射した場合でも，M^2 の投与量と投与時間を考慮すれば，かなり高い生存率を示すこととが認められた．

Introduction

It has previously been reported, that Marinamycin[1)2)] possesses an effect to increase white cell count in cases of leukopenia induced by X ray[3)4)5)6)] or Co^{60} irradiation. Shimada et al. have studied and shown that it enhances the phagocytosing function and increases the mobilization capacity of leucocytes of rabbits. Miyagawa and Asakura have studied on the protective activity of Marinamycin against Co^{60} irradiation in mice. The results of their experiments were as follows:

In a control group, 48 adult mice were generally exposed to 700r by Co^{60} irradiation. Its survival rate was 25%. In the second group, 40 mice were exposed to the same dose 1 to 2 hours after intra peritoneal administration of 200 gamma Marinamycin, the survival rate of which was 37.5%.

When it was given 30 minutes after exposure, 11 out of 32 mice remained alive (34.4%). In the next group 200 gamma Marinamycin was given one hour before and two more doses were given 2 and 5 days after exposure. Its survival rate was 12.5%. In the final group, three doses were given 1, 4 and 7 days after exposure. Two out of 16 mice survived (12.5%).

From the results of this experiment, they state that although it is difficult to evaluate the protective effect of this agent on irradiation, it might be possible to postulate some different mechanisms from that of other previously known protective agents, because Marinamycin appeared to have some protective effect even if given posterior to exposure.

We have also carried out experiments in rabbits and confirmed the fact, that Marinamycin has a remarkable leukocyte increasing effect in the peripheral blood and can be used both prophylactically and therapeutically in leukopenia experimentally induced by X ray or Co^{60} irradiation.

We have previously reported on an anti-cancer activity of Marinamycin. During the course of these experiments with Marinamycin, we have isolated a new substance from the same strain of streptomyces, named Marimycin, wheth has a specifically high activity to induce leukocytosis in rabbits, The purpose of this paper is to preseul the experimintal resultsin mice of DD-S strain, in which we have tested the effect of this new substance in parollel with that of Marinamycin.

Methodsand Materials

Adult male mice of DD-s strain, weighing about 20 gm. were used. They were generally exposed to respective doses of 100, 200 and 400 r by X ray irradiation. Leukocytosis inducing effect of Marinamycin was mainly examined in groups irradiated with 100 to 200 r doses. Exposure to higher radiation dose was performed by Co^{60} irradiation and the survival rate of respective groups were compared with each other.

Peripheral blood samples were taken from veins of their tails by puncture with 1/4 size needles. The white cell count was performed by theordinary method. The data shown in table 1 to 4 were obtained from the experiments, in which Marinamycin was used. Other tables are concerned with the effect of the new anti-leukopenic activity (from Experiments. 5.) Marimycin.

Experimental Results

Experiment 1.

Nine mice were generally exposed to 100r by X ray irradiation. They were divided into three groups. In one group, 100 gamma Marinamycin was once given intraperitoneally 6 hours before irradiation. In the second group it was given 2 hours before exposure. The third group served as control. The results are shown in the followingtable.

Table 1.

Group	No. of mouse	Body wt.	Leukocyte count				38days
			1	4	7	8	
Ⅰ.	1	18.1		7,400			18,400
	2	20.0			10,300		15,000
	3	18.9				10,700	9,800
Ⅱ.	4	16.0		3,500			6,600
	5	16.7			9,400		7,100
	6	18.4				8,300	9,200
Ⅲ.	7	15.6		5,700			8,000
	8	17.2			7,000		9,200
	9	16.1				3,900	8,400

Experiment 2.

Nine mice were exposed to 200 r by X ray irradiation. In the first group, 500 gamma Marinamycin was given intraperitoneally 2 hours before, while in the second group 100 gamma was administered in the same way 2 hours before exposure. The results are shown in table 2.

Table 2.

Group	No. of mouse	Body wt.	Leukocyte count			20days
			1	4	6	
Ⅰ.	1	29.3	12,900			11,900
	2	21.3		6,400		14,500
	3	21.6			10,000	
Ⅱ.	4	22.1	7,900	4,100		6,500
	5	21.2				15,800
	6	20.7			5,300	
Ⅲ.	7	24.0	5,080	3,800		10,700
	8	18.6				13,000
	9	15.8			4,500	8,200

In the control group we have seen some leukopenic tendency in two mice, while the mice treated with 500 gamma Marinamycin have shown leukotycosis rather than leukopenia. All mice survived.

Experiment 3.

Mice were exposed to 400 r by X ray irradiation. In the first and second groups, mice were treated in the same way as above. One out of three mice in the control gruop were dead and one mouse was also dead in the first group. All mice in the second group survived.

Experiment 4.

Mice were exposed to 500 r in this experiment. Marinamycin was given in the same way as in experiment 1. and 2. In the control group, one mouse died after 19 days, while in the second group one died after 27 days. Other mice were alive on the 80th. day of experiment.

Experiment 5.

In this experiment, 20 mice were exposed to 700 r by Co^{60} irradiation. They were divided into 4 groups. Lot No. 40 Marimycin was examined in this experiment and given intraperitoneally 2 hours before exposure. In the first, second and third groups, 100, 20 and 10 gamma Marimycin were given respectively. The final group served as control. During the observation of 28 days 4 out of 5 mice in the first group died after 13, 16, 21 and 27 days respectively, while mice treated with 20 gamma were all alive at the final day of observation. Four mice given 10 gamma died after 9, 12, 25 and 25 days respectively. Two mice in the control groups died after 25 days. From these results 20 gamma is thought to be the optimal dose for each mouse to expect protection.

Experiment 6.

Twenty mice were exposed to 700 r in this experiment, which were divided into 4 groups. Lot No. 64 Marimycin was examined. Except for control group, each mouse of three groups was treated with 50 gamma Marimycin. In the first group it was given 2 hours before irradiation, while in the second and third groups it was given after 24 hours and 48 hours respectively. The results are shown in the following table.

Ten mice were alive on the 35th. day and the survival rate of treated mice was 66.7%. The best result was obtained in the third group, in which Marimycin was given 48 hours after exposure.

Table 6.

Group	Treatment	Survivals on the 35th. day
1	50 gamma, 2hours before	3/ 5 (24, 28)
2	50 gamma, 24hours after	3/ 5 (13, 25)
3	50 gamma, 48 hours after	4/ 5 (17)
control		2/ 5 (12, 17, 30)

Experiment 8.

Thirty mice were exposed to 800 r by Co^{60} irradiation. They were divided into 6 groups, one of which served as control. This time we used 25 gamma of lot No. 89 Marimycin throughout the all groups. It was given intraperitoneally 30 minutes, 24 hours, 48 hours and 72 hours respectively after exposure in four groups and in the remaining 5th. group 3 doses of 25 gamma were injected after one, three and five days. The results are shown in the following table.

Table 8.

Group	Treatment	Survivals on the 35th. day
1	25 gamma, 30 min. after	0/ 5 (17, 19, 20, 24, 28)
2	25 gamma, 24 hrs. after	2/ 5 (12, 13, 15)
3	25 gamma, 48 hrs. after	4/ 5 (20)
4	25 gamma, 72 hrs. after	4/ 5 (19)
5	25 gamma, 3 times after 1, 3, 5 days	0/ 5 (9, 10, 10, 10, 13)
control		0/ 5 (10, 11, 11, 11, 13)

It is clear that 10 out of 25 treated mice survived at least 35 days after exposure to 800 r. The reason why mice were all dead in three times treated group is not clear.

Experiment 9.

This experiment was carried out in the almost same way as above, using 50 gamma of lot No. 94 Marimycin instead of 25 gamma of lot No. 89. In this experiment, it was given subcutaneously.

The results are shown in table 9.

Table 9.

Group	Treatment	Survivals on the 35th. day
1	50 gamma, 30 min. after	3/ 5 (21, 25)
2	50 gamma, 24 hrs. after	1/ 5 (6, 6, 11, 11)
3	50 gamma, 48 hrs. after	2/ 5 (6, 20, 24)
4	50 gamma, 72 hrs. after	4/ 5 (15)
5	50 gamma, 3 times after 1, 3, 6 days	3/ 5 (9, 11)
control		0/ 5 (5, 8, 10, 11, 12)

As seen in tables 8. and 9, mice exposed to 800 r (control group) all died within 13 days after irradiation. Mice given 25 to 50 gamma Marimycin within 30 minutes to 72 hours after exposure can tolerate this radiation dose and about half of 50 mice remained alive during the 35-day-course of observation.

Summary

1) Mice of DD-S strain exposed to 100 to 200 r by X ray irradiation usually caused leukopenia within one week, while those given Marinamycin 2 hours before irradiation caused leukocytosis instead of leukopenia.

2) Control mice exposed to 700 r by Co^{60} irradiation could survive for at least 35 days, but its survival rate was 40 to 60%. Mice exposed to 800 r were all dead within 13 days. Its survival rate on the 35th. day was zero. Marimycin, one new anti-leukopenic ogent showed n appreciably beneficial effect to enhance the survival rate of irradiated mice, when given in on adequate dose and time.

In the present stage of preparation, the dosage ranging from 25 to 50 gamma should be thought to be adeouate for an adult mouse and the maximum effect would be expected when given wihein 2 to 3 days after exposure.

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