THE EFFECT OF IONIZING RADIATION ON PHAGOCYTOSIS AND THE BACTERICIDAL POWER OF THE BLOOD

I. The Effect of Radiation on Migration of Leucocytes*

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The generally held view that infection plays a prominent role in the morbidity and mortality that follow exposure to ionizing radiation is supported by many observations. Several investigators have demonstrated that animals exposed to total body radiation exhibited increased susceptibility to pathogenic and presumably non-pathogenic microorganisms (1–4). This increase in susceptibility is not surprising in view of the debilitating effect of irradiation. Exposure to radiation may result in bacteriemia (5), leading to death of the irradiated animal. The irradiated host is either unable or is much less able to prevent proliferation and multiplication of the bacteria and the ensuing bacterial invasion, and to remove the microorganisms.

Under normal circumstances a host may protect itself against the invading microorganisms by several mechanisms, two of which are phagocytosis and the bactericidal action of the blood. Experimental evaluation of the effect of ionizing radiation on these two defense mechanisms is presented in this and in the following report.

In this study the phenomenon of phagocytosis by granulocytes was considered as being governed by at least four factors: production of the leucocytes, and their ability respectively to migrate, to ingest and then to digest the microorganisms. It is assumed that optimum defense requires that all four factors operate at maximum capacity.

The effect of x-radiation on the hematopoietic tissues and their cellular components has been studied intensively. It is generally accepted that lymphocytes are radiosensitive, whereas neutrophils are relatively radio-resistant. A marked fall in lymphocytes has been observed in mice, rats, rabbits, and swine 15 minutes to several hours after exposure to various doses of radiation (6–9). In rats the decrease in lymphocytes appeared to be related to their life span in the absence of further cell proliferation, and their regeneration was first apparent on the 12th postirradiation day with the

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development of the mast cells (10). On the other hand, it has been shown that the effect of radiation on neutrophils and their mechanism of formation is a more gradual process (6, 7, 11–13).

The migration of leucocytes is considered an important factor in the quantitative evaluation of phagocytosis. Studies with virulent and avirulent strains of *Mycobacterium tuberculosis* and *Brucella abortus* revealed that whereas virulent strains inhibited the migration of leucocytes *in vitro*, the avirulent strains did not (14–16). Certain fatty acids isolated from tubercle bacilli have been found to exert an effect similar to that produced by *M. tuberculosis* (17). Inhibition of leucocyte migration induced by certain bacteria has been ascribed to their characteristic endotoxins (18, 19). The implied relationship between leucocyte migration and bacterial virulence, together with the demonstration that certain bacteria exert a chemotactic effect on granulocytes (20) suggests that the ability of leucocytes to migrate may be directly related to the effectiveness of phagocytosis.

With these considerations in mind, a study was undertaken on the effect of radiation on migration of rat and rabbit leucocytes at various postirradiation periods. This is one part of a study of leucocyte function of irradiated animals. A subsequent report (21) will deal with effects of ionizing radiation on ingestion and digestion of bacteria.

Materials and Methods

Experimental Animals.—200 to 250 gm. male and female Sprague-Dawley rats and 3 to 4 kg. male and female rabbits were used throughout this study. The animals were obtained from a commercial breeder and appeared to be free of detectable infections. They were fed Ralston Purina laboratory chow. Water was provided in special bottles, which were sterilized prior to use.

Irradiation[1].—When irradiated, each animal was housed in a cardboard container placed on a rotating platform. A Keleket deep therapy x-ray machine was used and the radiation factors were as follows: 200 KVP[3], 20 ma., filter 0.25 mm. Cu and 1.0 mm. Al, HVL[2] 0.75 mm. Cu, distance from target to skin 75 cm., output approximately 21 r per minute. All x-ray doses were measured in air.

Preparation of Glassware[4].—Glassware used for migration experiments was subjected to special washing procedures. All capillary pipettes, test tubes, and syringes were washed by boiling in a 1 per cent solution of a general cleaning compound (A. H. Thomas and Co.). They were then placed in hot running water for 1 hour, rinsed in several changes of distilled water, and dried in a hot air oven at 160 to 180°C. The test tubes were subsequently coated with silicone (dri film, E. H. Sargent and Co.), rinsed in hot water for 1 hour and in distilled water for ½ hour, and transferred to a 3 per cent NH_4OH solution to neutralize any residual acidity. The tubes were then carried through several changes of distilled water, plugged, and auto-

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[1] The authors gratefully acknowledge the cooperation of Dr. Hugh M. Wilson of the Mallinckrodt Institute of Radiology at Washington University, in making available the facilities for irradiation of animals.

[2] HVL, half value layer.

[3] KVP, kilovolt peak.

[4] The authors are indebted to Dr. S. S. Elberg and Miss P. Schneider for valuable suggestions regarding preparation of glassware.

claved. The capillary pipettes and the syringes, not coated with silicone, were sterilized in the hot air oven. Regular 3 by 1 inch microscope slides and 24 by 24 mm. or 25 by 50 mm. coverslips were immersed in a 5 per cent concentrated HNO_3-95 per cent H_2SO_4 solution for 1 hour, rinsed thoroughly in distilled water, and placed in a 1 per cent solution of the general cleaning compound for 20 minutes to counteract the acid. They were again rinsed in distilled water and stored in 95 per cent ethyl alcohol.

Assembling the Cell for Observation of Leucocyte Migration[5].—Whatman 50 filter paper was cut approximately into 1 inch squares and autoclaved in a Petri dish. After trying a considerable number of compounds, a high melting point paraffin, MX170 wax, code 8788, manufactured by the Shell Oil Company was found satisfactory. The slides and coverslips, sterilized prior to use by flaming off alcohol, were separated by the thickness of two pieces of filter paper. Holding the cell together with the fingers, one edge was sealed by immersing it into the paraffin bath (temperature 92°C.) and then placing that edge on an iced surface. The filter paper was then removed and two other edges were sealed in the same manner. The cells were stored aseptically at about 4°C. and were used not later than 1 week after preparation.

Determination of Migration.—Cardiac blood removed aseptically from etherized rats, or marginal ear vein blood from rabbits, was placed into silicone-coated tubes, which contained a known amount of sterile heparin of predetermined concentration. If whole blood was used for determination of leucocyte migration, the blood was kept in an iced bath for 10 minutes, and then left at room temperature for 20 minutes before being pipetted into the assembled cells. The subsequent operations were conducted at 2–4°C. In order to avoid hemolysis, the rat blood was centrifuged at 1,000 R.P.M. for 3 minutes. The buffy coat and plasma layers were then removed with a capillary pipette and placed into the prepared cells. The openings of these cells were then sealed with paraffin and the cells were centrifuged at 1,000 R.P.M. for 5 minutes to separate the blood cells from plasma.

All glass chambers were examined prior to and 15 minutes after incubation at 37°C. to record any false migration of the blood components. Measurement of migration was made microscopically (100 magnification) at various time intervals by means of a calibrated mechanical stage. The extent of migration was measured from the outer edge of the buffy coat to the end of "mass migration"—a solid line of leucocytes across the microscope field; scattered white cells were not included since their number would depend on the concentration of leucocytes in the chamber.

Titration of Heparin.—Blood, added to varying dilutions of heparin, was placed in the cells in the manner described above. These preparations were examined for clotting and migration. The concentration of heparin which allowed the plasma to clot after incubation for 15 to 20 minutes and was found to give maximum migration of the leucocytes was used in subsequent experiments. The concentrations of heparin used were, for rat blood 0.07 mg. per ml., and for rabbit blood 0.02 mg. per ml.

Total and Differential Counts of White Blood Cells.—Standard methods were used to determine the total and differential W.B.C. counts. In most cases 100 blood cells were counted, but when the W.B.C. count was extremely low only 50 or even 25 cells were counted.

RESULTS

Migration of Leucocytes from Normal, Irradiated, and Infected Rats.—After several preliminary experiments, a group of 9 rats was used to determine the rate and extent of migration of leucocytes from normal animals. Ten cells

[5] The authors gladly acknowledge valuable suggestions made by Dr. M. Smith regarding preparation of cells for leucocyte migration.

were prepared for each animal with 2 ml. of cardiac blood. Since rat red blood corpuscles lysed readily at 37°C., only the buffy coat and plasma layers were used in this series. The cells were examined prior to and 6, 12, and 24 hours after incubation at 37°C. The results, presented in Table I, indicate that after 24 hours of incubation the extent of migration ranged from 0.57 to 1.11 mm. Because of this rather wide variation, a statistical evaluation of migration was carried out in replicate cells prepared from the same sample of blood; readings obtained were consistently uniform, clustering close to the mean.

Since each rat was used as its own control, it was important to establish the effect of multiple bleeding on leucocyte migration. Results of such an experi-

TABLE I

Migration of Leucocytes from Normal Rats

Rat No.	No. of cells	Average migration			
		6 hrs.	12 hrs.	24 hrs.	± S.E.*
		mm.	*mm.*	*mm.*	*mm.*
1	10	0.81	0.97	1.08	±0.05
2	10	0.88	1.00	1.11	±0.04
3	10	0.70	0.80	0.96	±0.02
4	8	0.54	0.56	0.61	±0.02
5	9	0.50	0.60	0.64	±0.04
6	10	0.60	0.72	0.84	±0.04
7	10	0.50	0.58	0.64	±0.03
8	7	0.47	0.54	0.57	±0.04
9	7	0.51	0.61	0.61	±0.04

* S.E., standard error of the mean.

ment, collected in Table II, show no difference in the rate or extent of migration when an animal was allowed to rest 5 days between two successive bleed ings.

Comparison of the rate and extent of migration of leucocytes from normal animals and from the same animals 1 and 5 days after irradiation are presented in Table III. Leucocytes from normal rats migrated in a similar manner to those obtained from the same animals 1 day after irradiation. However, leucocytes removed 5 days after irradiation showed a pronounced decrease in both the rate and the extent of migration. The mean distance traversed by leucocytes at each period of observation is plotted for the respective groups in Fig. 1.

The following experiment was carried out to determine whether infection of normal or irradiated animals had any effect on migration of their leucocytes.

20 rats were exposed to 600 r total body radiation and divided into two equal groups: one group received an intraperitoneal injection of 8×10^7 *Micrococcus aureus* in 0.2 ml. of

culture, while the other group served as radiation controls. Another 10 rats, which were not irradiated, were also injected with the same concentration of *M. aureus* and thus served as the untreated infected controls. All these animals were bled 5 days prior to any treatment to determine the extent of normal leucocyte migration. After a 5 day rest, various groups

TABLE II

Effect of Multiple Bleeding on Leucocyte Migration in Rats after a 5 Day Rest

Rat No.	No. of cells	Average migration after 24 hrs. of incubation	
		1st bleeding; normal rats	2nd bleeding; normal rats rested 5 days after 1st bleeding
		mm.	*mm.*
1	10	0.93	0.98
2	10	0.64	0.75
3	10	0.57	0.70
4	10	0.87	0.73
5	10	0.85	0.74
6	10	0.83	0.77
7	10	0.90	0.90
8	10	0.83	0.77
Average	10	0.83	0.79
Standard deviation		0.14	0.07
(Standard error of the mean)		0.05	0.02

TABLE III

Migration of Leucocytes from Normal and Irradiated Rats

Condition	No. of rats	No. of cells	Average migration ± S.E.* at indicated time of determination		
			6 hrs.	12 hrs.	24 hrs.
			mm.	*mm.*	*mm.*
Normal	9	87	0.61 ± 0.05	0.71 ± 0.06	0.78 ± 0.07
600 r—1 day	9	78	0.57 ± 0.09	0.65 ± 0.05	0.74 ± 0.05
600 r—5 days	8	40	0.25 ± 0.02	0.32 ± 0.03	0.38 ± 0.03

* S.E., standard error of the mean.

were treated as indicated above and were bled 1 day after infection; *i.e.* on the 2nd post-irradiation day.

The results (Table IV) indicate that infection did not influence the extent of migration; the slight decrease in migration observed in the irradiated infected group was similar to that observed in the irradiated controls.

It can therefore be concluded (Table III) that 600 r had no effect on leucocyte migration when measured 1 day after the animal's exposure to this dose of

radiation. A slight but significant decrease in migration appeared 2 days after irradiation (Table IV); this decrease became more pronounced when measured 5 days after exposure (Table III).

The Effect of Radiation on the Migration of Rabbit Leucocytes.—Because only a limited number of cardiac bleedings could be performed on the same rat, rabbits were used to study leucocyte migration at prolonged postirradiation periods.

3, 5, and 4 rabbits were irradiated with 100 r, 500 r, and 800 r respectively. Prior to irradiation and on various postirradiation days each rabbit was bled from the marginal ear

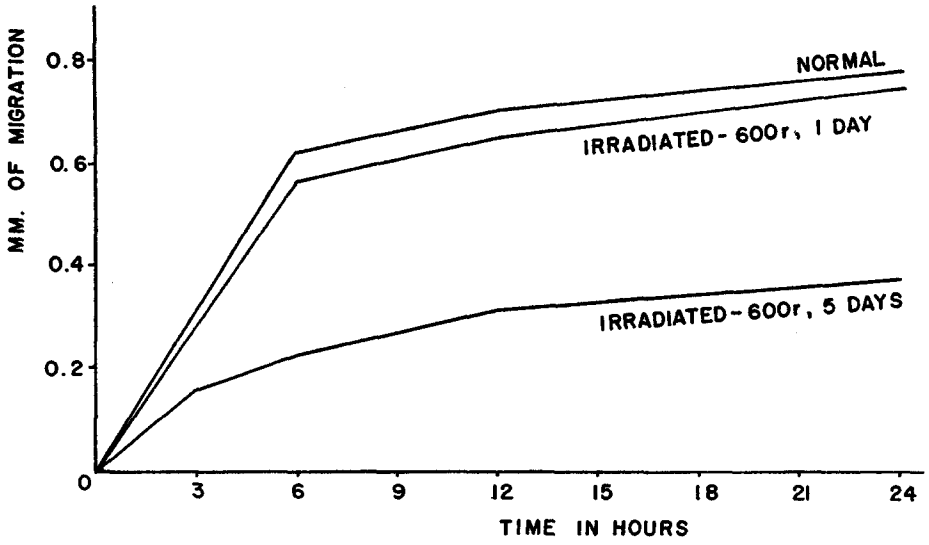

FIG. 1. The mean migration of leucocytes from normal and irradiated rats.

vein and the migration of leucocytes was determined 3, 6, 12, and 24 hours after incubation at 37°C. The results are presented in Figs. 2, 3, and 4.

While exposure to 100 r had no effect on leucocyte migration, a definite decrease in migration occurred in the 500 r and 800 r groups. The maximum effect was observed in the 3 to 5 day interval after irradiation, when the extent of migration dropped from an average of 1.20 mm. to 0.52 mm. for the 500 r group, and from 1.20 to 0.44 mm. for the 800 r group. This was followed by a rise observed 6 to 8 days after irradiation. At this time the values were 1.44 mm. and 1.50 mm. for the 500 r and 800 r groups respectively, not significantly different from the values of normal animals. Although a slight increase in lymphocytes and granulocytes was also noted 6 to 8 days after irradiation, their concentration was still far below normal. A second depression in the extent of migration

occurred in both groups 10 to 13 days after irradiation: a drop to 0.86 mm. for the 500 r group and to 0.66 mm. for the 800 r group. This was followed by a gradual recovery; the normal migration values were reached by the 17th to 21st day and were maintained for as long as 50 to 60 days after irradiation, when the experiment was discontinued.

Total and differential W.B.C. counts were made on each of the samples used for leucocyte migration, in order to determine the existence of any correlation between the observed effects of radiation on the blood picture and on the ability of white blood cells to migrate. The results (Figs. 5 and 6) indicate that a marked lymphopenia observed in both the 500 r and the 800 r groups 1 day

TABLE IV

Effect of Radiation (600 r/2 days) and/or Infection on Migration of Rat Leucocytes

Condition	No. of animals	No. of cells	Mean migration ± S.E.* in 24 hrs.
			mm.
Irradiated			
5 days before irradiation	9‡	90	0.87 ± 0.06
2 days after irradiation	9	90	0.66 ± 0.06
Infected			
5 days before infection	10	100	0.91 ± 0.05
1 day after infection	10	100	0.86 ± 0.06
Irradiated and infected			
5 days before irradiation	10	100	0.80 ± 0.04
1 day after infection *or* 2 days after irradiation	10	100	0.56 ± 0.04

* S.E., standard error of the mean.

‡ One animal succumbed during cardiac puncture.

after irradiation became most pronounced on the 3rd day after exposure. A slight but significant increase in the number of lymphocytes occurred in these two irradiated groups during the 5th to 7th day, decreasing again on the 10th to 13th day, after which time a gradual recovery was indicated. A similar picture was observed for the granulocytes, with the exception that on the 1st day after irradiation there was a slight but definite granulocytosis. The fluctuation in the concentration of granulocytes observed 20 to 60 days after exposure to 800 r had no apparent effect on the extent of migration.

The Effect of Total W.B.C. Counts on the Extent of Migration.—To determine whether the decreased leucocyte migration depended on leucopenia as a concentration effect, the extent of migration was established for different concentrations of W.B.C.

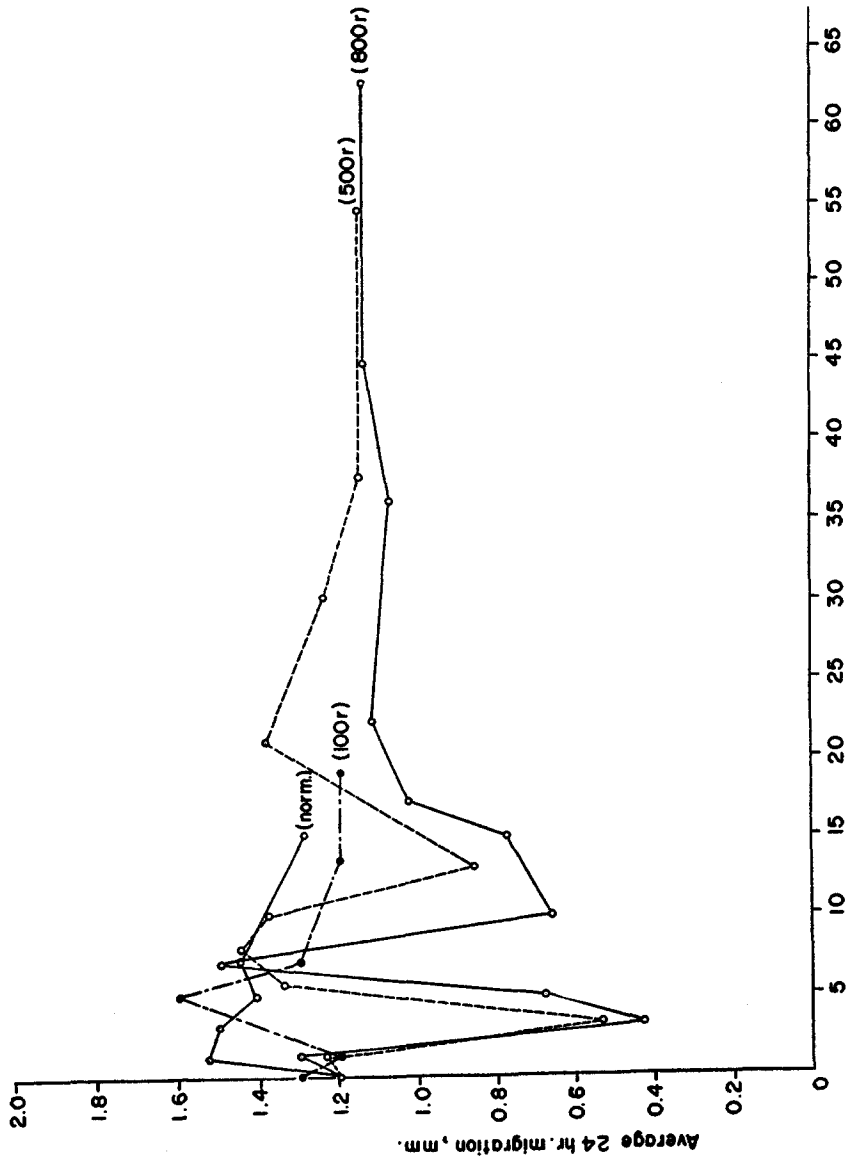

FIG. 2. Effect of radiation on migration of rabbit W.B.C.

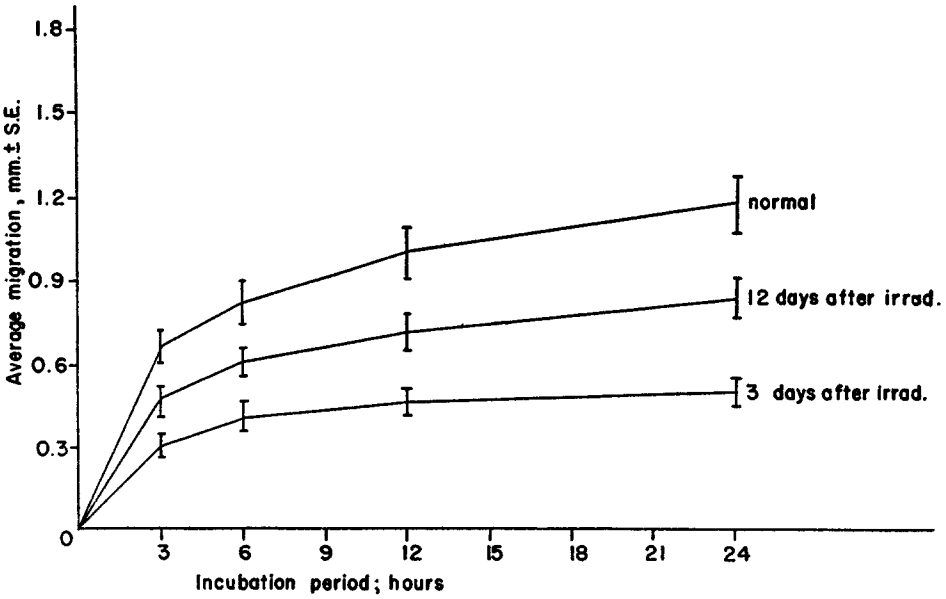

FIG. 3. Effect of radiation (500 r) on migration of rabbit leucocytes.

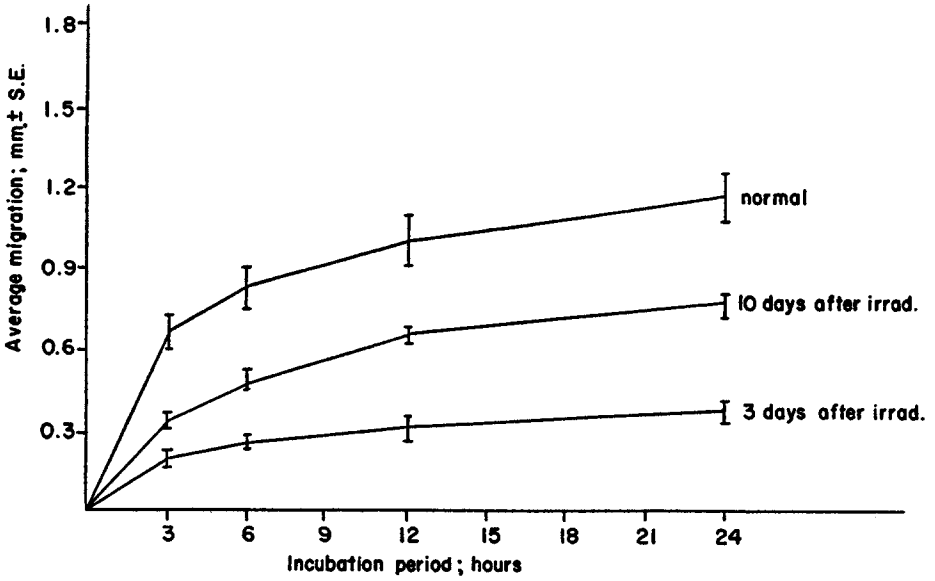

FIG. 4. Effect of radiation (800 r) on migration of rabbit leucocytes.

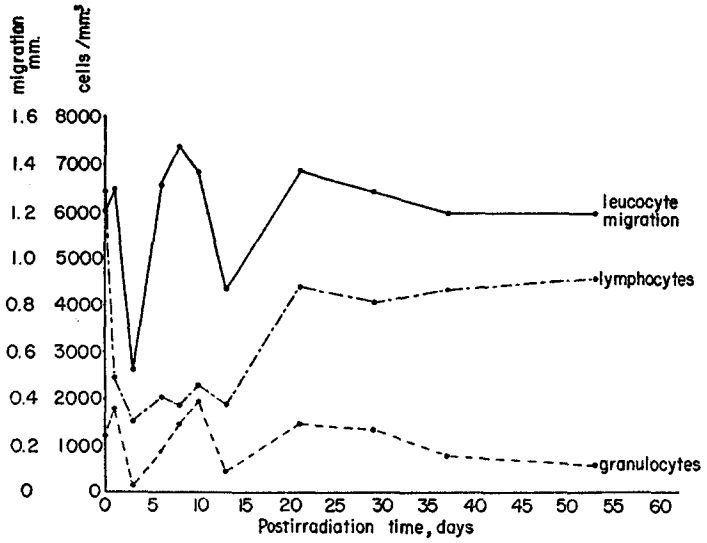

FIG. 5. The effect of radiation (500 r) on differential W.B.C. count and migration of rabbit leucocytes.

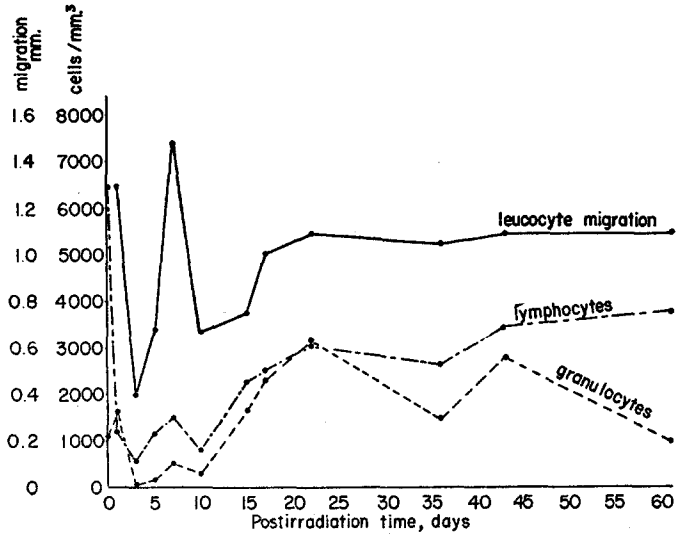

FIG. 6. The effect of radiation (800 r) on differential W.B.C. count and migration of rabbit leucocytes.

For this purpose two rabbits were bled from the marginal ear vein and a total W.B.C. count was made of each sample, which was subsequently diluted 10- and 100-fold. Aliquots of the undiluted and the two diluted blood samples were each placed into ten cells and readings were made prior to and 3, 6, 12, and 24 hours after incubation at 37°C.

The results are seen in Fig. 7 and Table V .It can be noted that a W.B.C. count of 680 cells/mm.^{3} did not alter the rate or extent of migration. This corresponds to the lowest total W.B.C. count encountered in any of the experi-

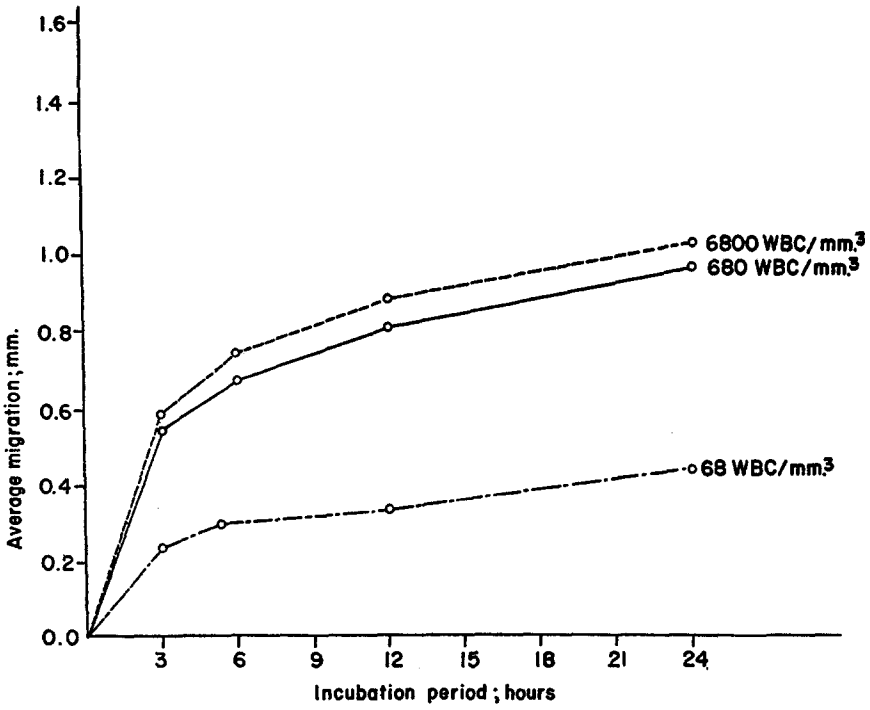

FIG. 7. Effect of concentration of W.B.C. on their migration.

ments. However, a definite decrease in the extent of migration was observed with samples having 60 to 80 cells/mm.^{3} This was probably due to the inability to determine the distinct line of mass migration rather than to the loss of ability of these white blood cells to traverse across a plasma medium.

The Influence of Plasma Factors from Normal and Irradiated Rabbits on Leucocyte Migration.—The following experiment was carried out in an attempt to determine whether one or more plasma factors in the irradiated animals may account for the decrease in migration of leucocytes. The procedure is outlined diagrammatically in Fig. 8.

The migration of leucocytes of 10 rabbits was determined, and after a 3 to 5 day rest period, 5 of the animals were exposed to 800 r total body radiation while the remaining five served as controls. 1, 3, 5, and 7 days after irradiation blood was obtained from each of the 10 animals. Aliquots of each sample of blood were centrifuged, and the cells washed three times with sterile Locke's solution. The cells from the control animals were mixed with the

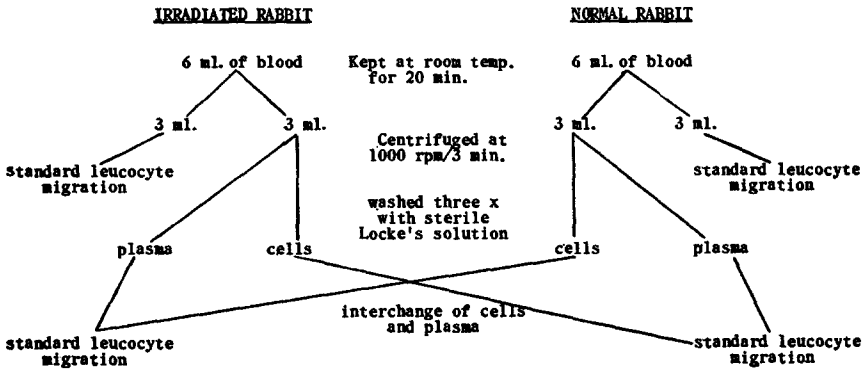

FIG. 8. Diagrammatic outline of experiment dealing with influence of plasma factor(s) from normal and irradiated animals on leucocyte migration.

TABLE V

The Effect of W.B.C. Concentration on Leucocyte Migration

Rabbit No.	W.B.C./ mm.³	Average migration ± S.E.* at indicated incubation period			
		3 hrs.	6 hrs.	12 hrs.	24 hrs.
		mm.	*mm.*	*mm.*	*mm.*
A	6800	0.59 ± 0.03	0.76 ± 0.02	0.88 ± 0.06	1.02 ± 0.04
	680	0.57 ± 0.05	0.68 ± 0.05	0.80 ± 0.07	0.95 ± 0.06
	68	0.22 ± 0.03	0.30 ± 0.03	0.34 ± 0.04	0.42 ± 0.03
B	8300	0.92 ± 0.04	1.04 ± 0.04	1.14 ± 0.04	1.20 ± 0.03
	830	0.76 ± 0.02	0.92 ± 0.02	1.04 ± 0.04	1.12 ± 0.03
	83	0.24 ± 0.02	0.34 ± 0.02	0.36 ± 0.02	0.36 ± 0.02

* S.E., standard error of the mean.

plasma of the irradiated rabbits, and the cells from the irradiated animals were mixed with the plasma of the control animals. Aliquots of these mixtures, as well as the whole blood of each animal, were each distributed into 10 prepared cells, which were centrifuged and incubated at 37°C. Blood from a normal rabbit was treated in a similar manner and its washed cells recombined with its own plasma to control the effects of manipulation. The migration of leucocytes in this sample, measured in the usual manner, was comparable with that of unmanipulated normal blood.

The results of the cross-mixing of plasma and blood cells from the control and the irradiated animals (Table VI and Fig. 9) indicate that the effect of

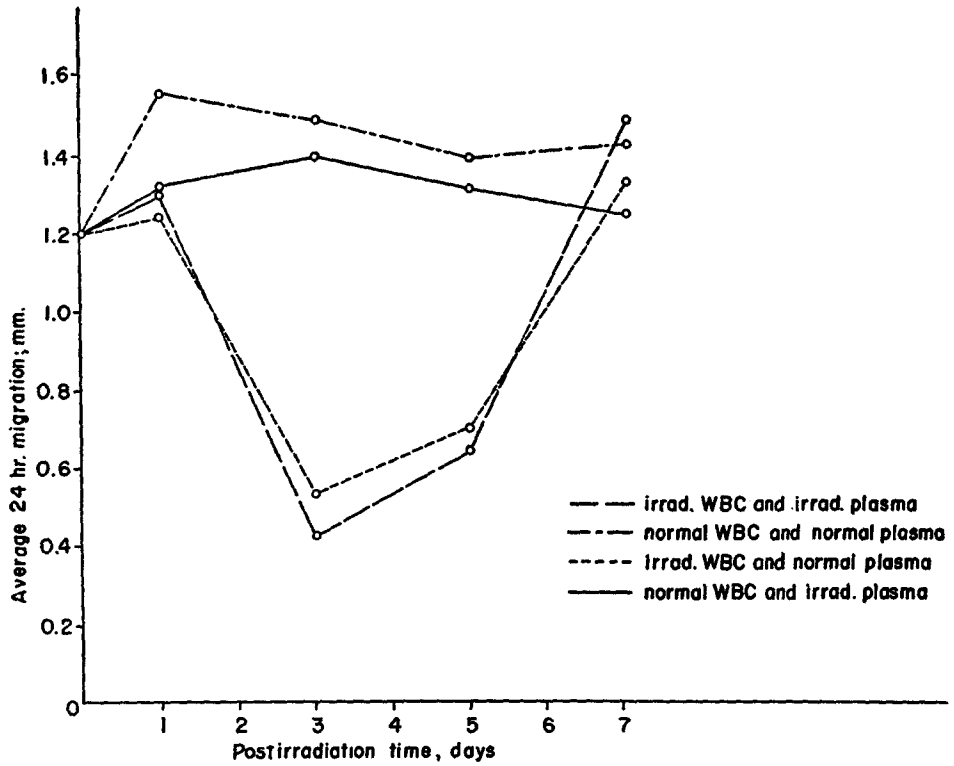

FIG. 9. Effect of plasma from irradiated rabbits (800 r) on migration of normal rabbit leucocytes.

TABLE VI

Effect of Plasma on Migration of Leucocytes from Normal and Irradiated Rabbits

Group	Incubation period	Average migration of leucocytes at the indicated post-irradiation time				
		0 day	1 day	3 days	5 days	7 days
	hrs.	*mm.*	*mm.*	*mm.*	*mm.*	*mm.*
Normal blood	3	0.65	0.82	0.91	0.93	0.82
	6	0.80	1.22	1.21	1.25	1.22
	12	1.03	1.41	1.46	1.36	1.41
	24	1.20	1.56	1.51	1.40	1.45
Normal leucocytes + irradiated plasma	3		0.85	0.90	0.88	0.81
	6		1.04	1.13	1.13	1.04
	12		1.23	1.31	1.26	1.15
	24		1.32	1.41	1.32	1.23
Irradiated blood	3		0.79	0.28	0.29	0.62
	6		1.00	0.35	0.41	0.81
	12		1.16	0.41	0.58	1.19
	24		1.30	0.44	0.68	1.50
Irradiated leucocytes + normal plasma	3		0.63	0.33	0.35	0.56
	6		0.90	0.46	0.52	0.83
	12		1.05	0.53	0.65	1.10
	24		1.24	0.56	0.72	1.34

radiation in reducing leucocyte migration is exerted directly on the leucocytes rather than indirectly *via* plasma.

DISCUSSION

The cumulative results with rats and rabbits exposed to either non-lethal or lethal doses of x-radiation show that the biological effects caused by this ionizing agent are dependent on radiation dose and postirradiation time. X-radiation appears to have a primary and a secondary effect on the ability of leucocytes to migrate. The consideration of postirradiation time made it possible to observe the two distinct responses of the leucocytes. It was noted that their migration is normal 1 day after irradiation. At this time the only observed effect of radiation is a leucopenia, principally a lymphopenia. The radiosensitivity of lymphocytes is, therefore, the first observed component of the radiation syndrome. This observation was made with rabbits irradiated with either 100 r, 500 r, or 800 r, and with rats exposed to 600 r total body radiation. On the 2nd postirradiation day, the ability of leucocytes to migrate is slightly but significantly decreased, while the blood picture remains essentially the same as during the first postirradiation day. This gradual manifestation of radiation damage becomes more pronounced on the 3rd to 5th day after irradiation, when there is a definite decrease in leucocyte migration and a concomitant drop in concentration of both lymphocytes and granulocytes. The effect on leucocyte migration is temporary, since 6 to 8 days after irradiation leucocytes from the irradiated animals are found to migrate to the same extent as do the leucocytes from the non-irradiated animals. By this time a complete turnover of leucocytes has occurred (22) and the newly formed cells are able to migrate in a normal manner.

The secondary phase of radiation injury, due to the continued destruction of the hematopoietic system by the initial dose of radiation, is manifested in the depression in the migration of leucocytes on the 10th to 13th postirradiation day. It is noteworthy that another two peak phenomenon has been reported in relation to the susceptibility of rats to high doses of radiation (23), when the peaks of increased mortality were observed on the 5th and the 11th postirradiation days.

Recovery of production and migration of leucocytes begins after the 5th postirradiation day, and is an inverse function of radiation dose. Rabbits exposed to 100 r showed a normal total W.B.C. count by the 5th day. No effect on leucocyte migration was seen in these animals. A recovery of W.B.C. counts of rabbits exposed to 500 r was noted on the 21st postirradiation day, when the cells also showed normal rate and extent of migration. A similar picture was observed with the animals exposed to 800 r.

The decrease in leucocyte migration could not be attributed to the presence of factors in the plasma of the irradiated animals. Similarly, the influence of

concentration of W.B.C. *in vitro* on the rate or extent of leucocyte migration was insufficient to account for the effect of radiation. However, the above considerations did not indicate the possible influence that each type of white blood cell may have had on the migration. A positive correlation between migration and concentration of granulocytes is clearly indicated in rabbits exposed to 500 r and 800 r. In referring to "leucocyte" migration, both the lymphocytes and the granulocytes were involved in the measurement of the extent of migration. The technique used did not allow a clear differentiation between the cell types. The extent of migration of granulocytes alone, obtained from peritoneal washings, has been found to be similar to that of cells from whole blood, which contained 80 per cent lymphocytes. This observation suggests that the proportions of white cell types may not greatly influence the rate or extent of migration.

The effect of radiation on leucocyte migration may also depend in part on the age of the surviving leucocytes. Aging may account for the decreased leucocyte migration noted on the 3rd to 5th postirradiation day and the subsequent return of migration to normal on the 6th to 8th postirradiation day, when a turnover of granulocytes has taken place. The second depression in migration during the 10th to 13th postirradiation day and the gradual return to normal values, however, cannot be explained on this basis. These two depressions in migration—one immediate in onset and subsiding rapidly, the other appearing later and disappearing more gradually—would seem to depend on two different mechanisms.

The results reported herein indicate that radiation depresses at least two phases of phagocytosis: the formation of leucocytes and their ability to migrate. The fact that the maximum effect on both phenomena occurred on the 3rd to 5th postirradiation day may explain in part the observed increase in susceptibility of irradiated rats to *Micrococcus aureus*, which occurred at the same postirradiation period.

The next phase of this investigation was directed to a determination of the effects of radiation on the ability of granulocytes to ingest and digest bacteria and on the bactericidal power of the blood. The results are reported in the second paper of this series.

SUMMARY

Exposure of rats to 600 r total body radiation did not influence either the rate or the extent of migration of their leucocytes 1 day after irradiation, but did decrease their migration on the 2nd and the 5th postirradiation day. Migration of rat leucocytes was not altered by infection of the animal with *M. aureus*.

Leucocytes of rabbits irradiated with 100 r showed a normal rate and extent of migration. However rabbits exposed to 500 r or 800 r showed depression of

leucocyte migration at two postirradiation intervals, on the 3rd to 5th and the 10th to 13th days after irradiation, with normal activity intervening. By the 21st postirradiation day the ability of leucocytes to migrate returned to normal. The effect of radiation on total and differential W.B.C. counts and the relationship of this effect to migration is discussed.

The decrease in leucocyte migration could not be ascribed either to leucopenia or to plasma factors.

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