

STUDIES ON X-RAY EFFECTS.

II. STIMULATIVE ACTION ON THE LYMPHOCYTES.*

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Taylor, Witherbee, and Murphy¹ have reported on the destructive action of x-rays on the circulating lymphocytes, confirming and extending the earlier work on this subject. It was noted by Murphy in his studies on x-ray effects² that while large doses destroyed, a small dose of x-rays would bring about a stimulation of the lymphocytes. This observation was later applied experimentally.³ In the earlier experiments the older type of x-ray tube was used, and it was practically impossible to establish a standard and uniform dose. With the introduction of the Coolidge tube the difficulty was eliminated to a large extent, and there was an opportunity to check this observation and extend it.

Mice have not been used here as in the previous experiments for the reason that blood counts could not be made on these animals more frequently than once a week without causing too marked a fluctuation.

EXPERIMENTAL.

Brown rabbits of the same relative size were used in the nine experiments. All the animals were kept in separate cages. Several blood counts were made on these normal rabbits, and they were then exposed to the rays of a Coolidge tube. A dose of low penetration

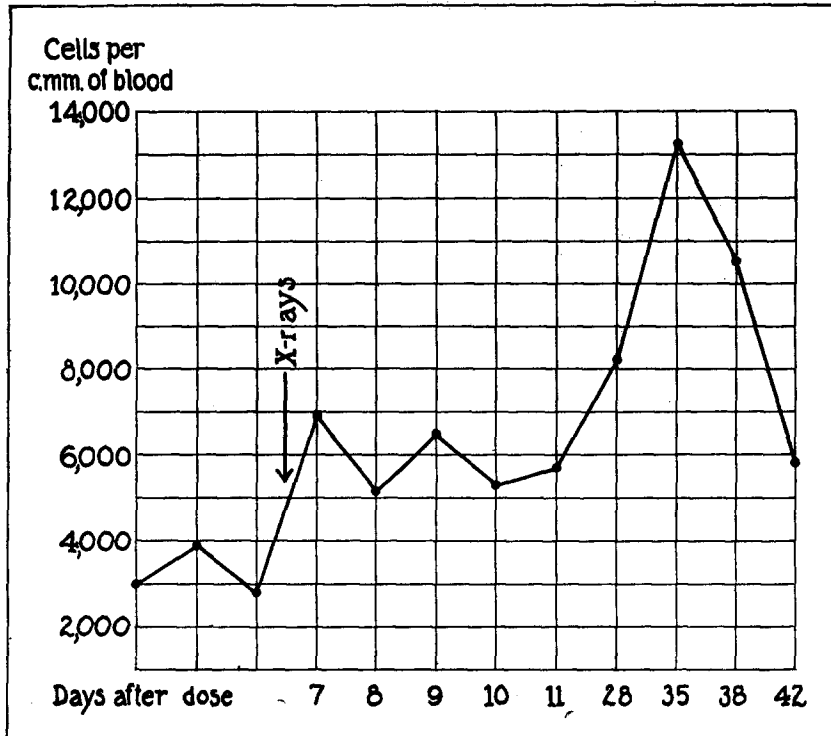
*This investigation was carried out by means of funds from the Rutherford Donation.

¹ Taylor, H. D., Witherbee, W. D., and Murphy, Jas. B., *J. Exp. Med.*, 1919, **xxix**, 53.

² Murphy, Jas. B., unpublished observation.

³ Murphy, Jas. B., and Morton, J. J., *J. Exp. Med.*, 1915, **xxii**, 800.

was applied to the dorsal area: the spark-gap measured $\frac{7}{8}$ inch, the milliamperage was 25, the distance from the target to the back 8 inches, and the time of exposure 20 minutes. The temperature 8 inches from the target was 31°C. In almost every case a blood count made 48 hours after exposure showed a slight drop in the lymphocytes.

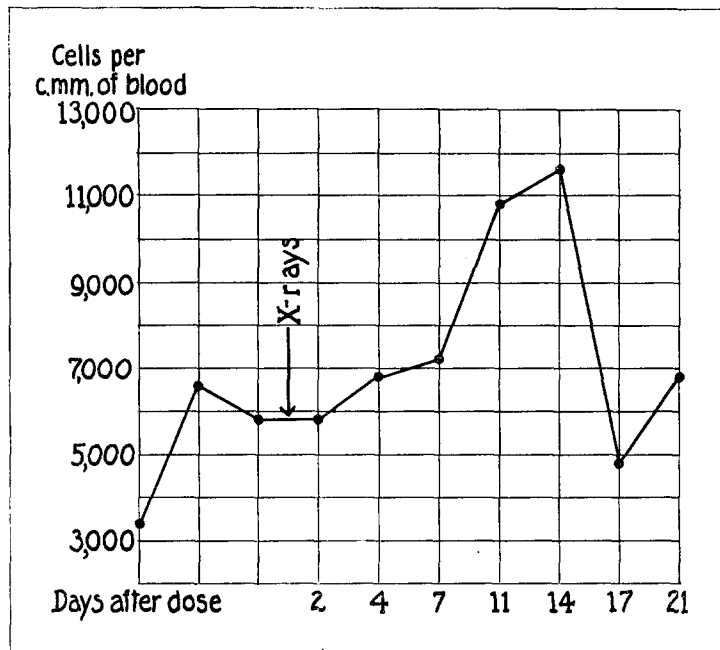


TEXT-FIG. 1. Effect of unfiltered x-rays of low penetration on the lymphocytes of a rabbit.

Experiment 1 (Text-Fig. 1).—Three consecutive counts were made, the lymphocytes numbering 3,000, 4,000, and 2,800 respectively, and the rabbit was then exposed to x-rays. The count made 1 week later showed a stimulation in the cells, the number of lymphocytes being 6,900. Counts made at intervals showed slight fluctuation until the 28th day, when the count reached 8,300. On the 35th day there were 13,300 absolute lymphocytes. In the course of the 7 days following the last count the number of cells fell to 5,800.

Experiment 2 (Text-Fig. 2).—Considerable fluctuation was shown in the normal counts. The first count 48 hours after exposure to x-rays showed little or no change, and for 7 days afterwards there was little increase, the lymphocytes numbering 5,000, 6,000, and 7,000 respectively, but on the 11th day the number had risen to 10,500, and on the 14th day to 11,600. They fell to 4,800 on the 17th day and in the last count rose to 6,900.

Experiment 3.—The standardizing counts were 6,900, 6,370, and 7,000. 48 hours after exposure to the x-rays a slight fall was noted, the number of cells



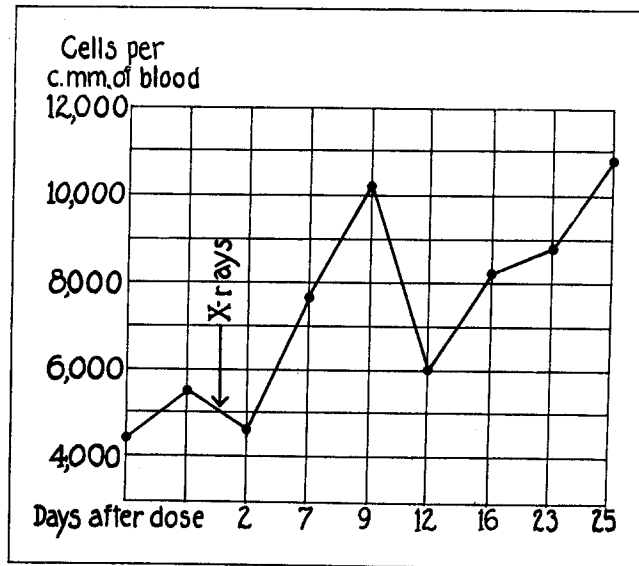
TEXT-FIG. 2. Effect of unfiltered x-rays of low penetration on the lymphocytes of a rabbit.

at that time being 5,100. On the 4th day there were 5,300 lymphocytes, and 6 days afterwards 10,240, showing marked stimulation. On the 8th day the number had decreased to 6,100 and on the 15th day to 4,000. After that there was a slight rise (6,500) on the 18th day and also on the 23rd, followed by another fall to 3,410. A rise to 8,200 was noted on the 64th day, and the last count, at which the number of cells was 6,300, was made on the 70th day after exposure.

Experiment 4.—4,300, 4,670, and 4,900 were the lymphocyte counts of a rabbit before x-ray treatment. 48 hours after exposure there was a slight increase (6,600). The 4th day the number of cells was 4,500, the 6th day 6,500, and the

15th day, when the last count was made, 5,979. This rabbit did not show a stimulation as did those of the preceding experiments.

Experiment 5 (Text-Fig. 3).—At the two counts preceding x-ray treatment there were 4,560 and 5,300 lymphocytes respectively. 48 hours after exposure to the x-rays there was a slight fall to 4,600, and on the 7th day a rise (7,600) which continued to the 9th day and at that time reached 10,200. On the 12th day there was a slight decrease, but from that time to the 25th day, on which the last count was made, there was a continued rise in the number of lymphocytes, which finally reached 10,900.



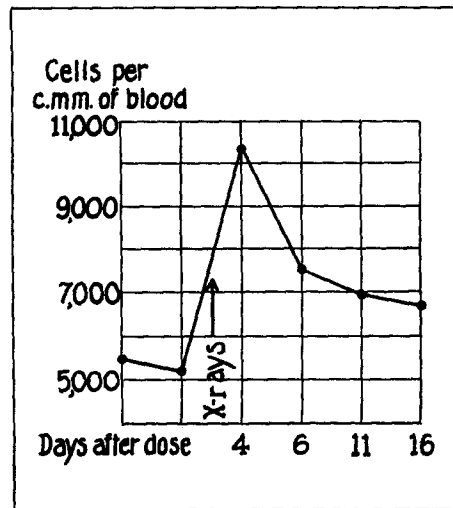
TEXT-FIG. 3. Effect of unfiltered x-rays of low penetration on the lymphocytes of a rabbit.

Experiment 6.—The normal counts showed a great fluctuation, the lymphocytes numbering 3,500 at the first count, at the second 6,100, and at the third 6,290. 48 hours after x-ray exposure they fell to 3,600, rose on the 4th day to 6,400, and fell again on the 6th day to 3,800. The cell counts, which were made at intervals until the 25th day after exposure, continued to rise and fall, never reaching a point beyond 5,900 or below 3,000.

Experiment 7.—The normal counts in this instance were 4,900 and 5,800. 48 hours after exposure to the x-rays the cells numbered 5,600, the 4th day 5,400, the 6th 5,700, and the 8th 6,180, showing a slight increase. On the 13th day the number of cells was 5,600, and on the 20th day, when the last count was made, 5,750.

Experiment 8.—2,270, 5,340, and 2,267 were the three consecutive lymphocyte counts made before x-ray treatment on a rabbit. 48 hours after treatment there was little or no change, the cells numbering 2,200. On the 4th day there was a rise to 4,800, on the 6th to 7,900. 11 days after exposure the number was 6,560. Subsequently there was a decline to 3,900 (13th day) and finally, on the 25th day, to 3,000.

Experiment 9 (Text-Fig. 4).—Two normal counts, the first 5,500, the second 5,100, were followed by the short x-ray dose. The 48 hour count was not made on this rabbit. The first count following exposure was made 4 days afterwards, the lymphocytes then numbering 10,390. On the 6th day there was a fall to 7,500, on the 11th day to 6,900, and on the 16th day to 6,700.

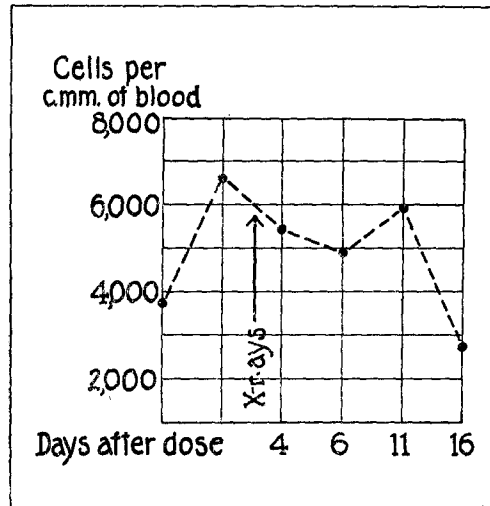


TEXT-FIG. 4. Effect of unfiltered x-rays of low penetration on the lymphocytes of a rabbit.

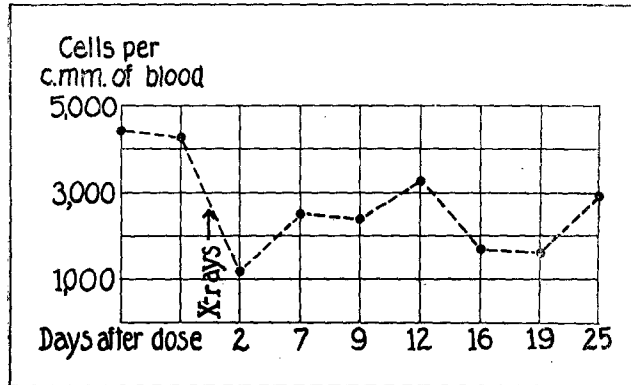
A comparable dose of filtered x-rays⁴ was used also on a smaller number of brown rabbits (spark-gap 6 inches, milliamperage 5, distance from the target to the back 10 inches, time 26 minutes and 57 seconds). The rays were filtered through 3 mm. of aluminum. The animals were exposed in the same way (over the dorsal area) and kept under the same conditions as those of the preceding experiments.

⁴ Remer, J., and Witherbee, W. D., *Am. J. Roentgen.*, 1917, iv, 303.

Experiment 1, a (Text-Fig. 5).—In the two counts preceding x-ray exposure the number of lymphocytes was not very constant, being 3,700 at the first and 6,600 at the second. After the filtered x-ray dose (6 inch spark-gap, milliamper-



TEXT-FIG. 5. Effect of filtered x-rays on the lymphocytes of a rabbit.



TEXT-FIG. 6. Effect of filtered x-rays on the lymphocytes of a rabbit.

age 5, distance from the target to the back 10 inches, and time 26 minutes and 57 seconds) the count showed little change, 5,470 in the first count, which was made on the 4th day, 4,900 on the 6th day, 5,900 on the 11th day, and 2,733 on the 16th day.

Experiment 2, a (Text-Fig. 6).—The lymphocytes numbered 4,400 and 4,350 at the two counts preceding x-ray treatment. 48 hours after exposure there was a marked fall (1,230), and on the 7th day a slight rise (2,500). On the 9th day the count was similar to that of the 7th; on the 12th it was 3,500, on the 16th 1,700, on the 19th 1,720, and on the 25th 2,981. There is no sign of stimulation in Experiment 2, a.

DISCUSSION.

It is of interest to note in these experiments that the x-ray dose used was of low penetration, the spark-gap being under an inch. The use of a larger spark-gap with apparently the same dose of x-rays did not give a stimulation. This suggests that the effect on the lymphoid organs is not the result of a direct action of the rays but is secondary to changes brought about either in the circulating blood or in the superficial tissues. The amount of x-rays penetrating to the deeper structures with this dose must be infinitesimal.

Another question arises as to the nature of the energy generated by the x-ray tube operated on so small a spark-gap. This point has not yet been taken up, but it is conceivable that other factors than the pure x-rays may play a part.

The results obtained in this small series of animals would not in themselves be accepted as conclusive evidence but are of interest principally as a parallel to our histological studies.⁵ It is conceivable that a marked stimulation may be taking place in the lymphoid organs without a proportionate number of these cells being thrown into the circulation. The question in itself offers an interesting problem of just what determines the number of cells in the circulation. It is well known that individuals with normal counts react differently in the number of cells thrown into the circulation in response to infections. So here, even with marked stimulation taking place in the lymphoid tissue of the glands and spleen, in only a part of the animals perhaps could we expect this stimulation to be evidenced by an increase in the number of lymphocytes in the circulating blood.

⁵ Nakahara, W., *J. Exp. Med.*, 1919, xxix, 83.

SUMMARY.

This study consists of blood counts on nine rabbits after an exposure to x-rays of a $\frac{7}{8}$ inch spark-gap, milliamperage 25, distance from the target 8 inches, and time of exposure 20 minutes.

In seven of the nine animals there resulted an increase of the circulating lymphocytes. In five of these the increase was marked and in two others definite but not striking.

Of the two animals which showed no stimulation one showed marked fluctuation of counts both before and after x-rays and the other little or no change.

The higher penetrating dose (6 inch spark-gap, milliamperage 5, distance from the target 10 inches, time 26 minutes and 57 seconds) given to two animals produced no appreciable stimulation.