DYNAMICS OF THE WHITE BLOOD REACTION TO IRRADIATION

I. K. Petrovich and Yu. I. Moskalev

(Presented by Acting Member of the Academy of Medical Sciences of USSR, N. A. Kraevskii)
Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 57, No. 2,
pp. 33-37, February, 1964
Original article submitted January 19, 1963

Numerous investigations [2, 3, 10, 13] have been devoted to the study of leucocyte reactions to different stimuli, including the effects of ionizing radiations. It has been shown in earlier investigations [5, 8] as well as in more recent ones [1, 12]that during the initial period following irradiation, the numbers of neutrophils and lymphocytes in the blood changed to different degrees. Becker and Thom [8] have established that the change in the number of lymphocytes and neutrophils depends on the volume of the tissue irradiated, the localization of the radiation effect, stress reaction and other conditions. However, the peculiarities of the lymphocyte and neutrophil reaction during early stages after irradiation, related to the irradiation dose, have not been studied in detail until now. Moreover, currently, many investigators, when analyzing white blood changes, take into consideration only changes in the total number of leucocytes, and formulate their hypotheses on the relationship between the leucocyte reactions and the strength of the stimulus [2]. But these reactions to irradiation are in fact summation results of opposite processes in the lymphoid and the myeloid branches of hematopoiesis.

In this investigation we have studied the relationship between the early white blood reaction and the irradiation dose. Eight doses were studied.

EXPERIMENTAL METHOD

Experiments were conducted on 70 white rats. Irradiation was done by means of a 12-tube x-ray apparatus with 126 r/min, in doses of 50, 200, 400, 1000, 2000, 4000, 8000, and 12,000 r.

The total number of leucocytes, the leucocyte formulas, and at certain periods the number of erythrocytes were determined after 5, 10, 20, 40, 80, 160, 320, 360, 420, 600, and 1440 min. In surviving rats, blood composition

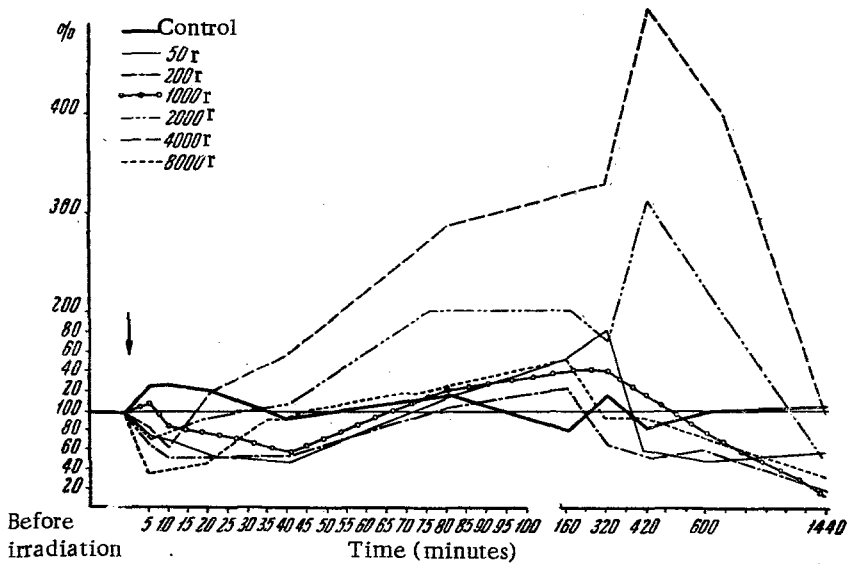

Fig. 1. Change in the total number of leucocytes during 24 hours with different doses of x-irradiation.

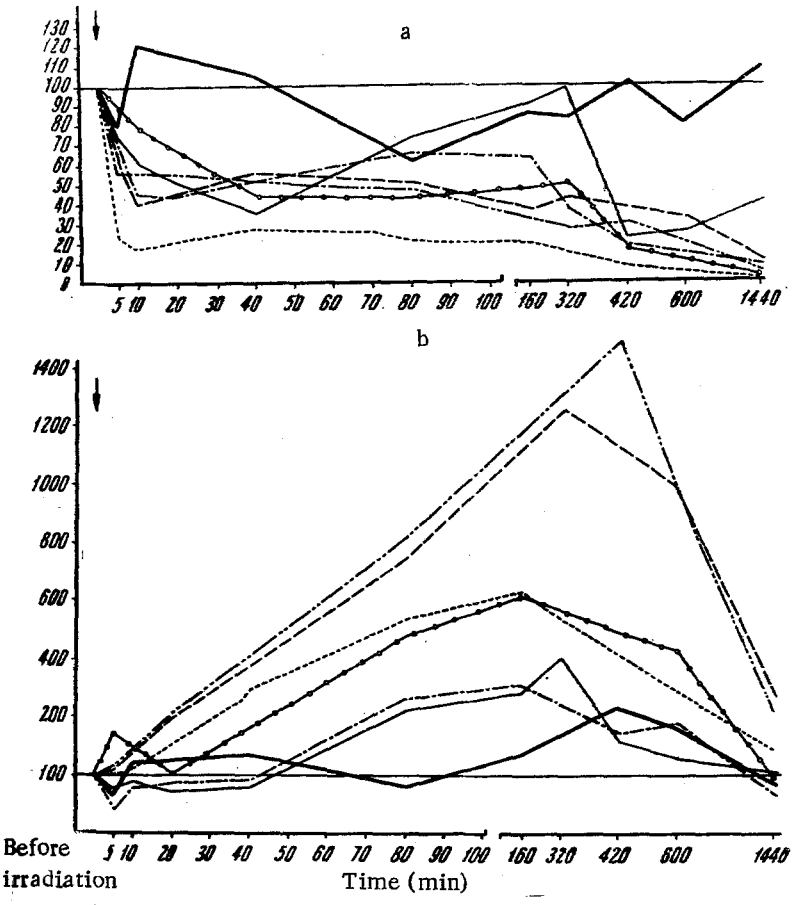

Fig. 2. Changes in the absolute number of lymphocytes (a) and neutrophils (b) during 24 hours with different doses of x-irradiation. Legend as in Fig. 1.

was studied 2-3 times during 24 hours, and after 2, 3 and 5 days, and subsequently at weekly and monthly intervals for a long time. At these periods the absolute numbers of neutrophils, lymphocytes, eosinophils, monocytes and the number of cytolyzed cells were determined. We have also considered the qualitative changes in the cells (pyknosis, chromatinolysis, fragmentation, etc.), and determined the degree of maturity of white blood cells according to the nuclear structure, staining properties of the cytoplasm and chromatin, etc.

EXPERIMENTAL RESULTS

At early periods (5-20 min) in irradiated animals there developed a moderate leucopenia, which later became replaced by leucocytosis, the degree of which depended on the irradiation dose (Fig. 1). Leucocytosis was insignificant at doses of 50-1000 r but it was acute at doses of 2000 and 4000 r. The maximal increase in the number of leucocytes was seen after 4-6 hours following irradiation, while after 1-2 days leucopenia developed in all cases. In control rats there was no significant change in the number of leucocytes.

The number of neutrophils (Fig. 2B) after insignificant variations (during 5-20 min) began to increase after 40-80 min following irradiation and reached its highest level after 4-7 hours. When rats were irradiated with x-rays in a dose of 1000 r the number of neutrophils became increased six times, with 2000 r – 15 times and with 4000 r – 13 times. It is of interest that irradiation of rats with 1000 and 8000 r produced the same change in the number of neutrophils.

Unlike changes seen with neutrophils, the absolute number of lymphocytes (Fig. 2A) after irradiation of rats in doses of 50 r and higher, began to decrease directly following irradiation and remained at a low level throughout the duration of the entire experiment. The decrease in the number of lymphocytes with almost all the doses (except

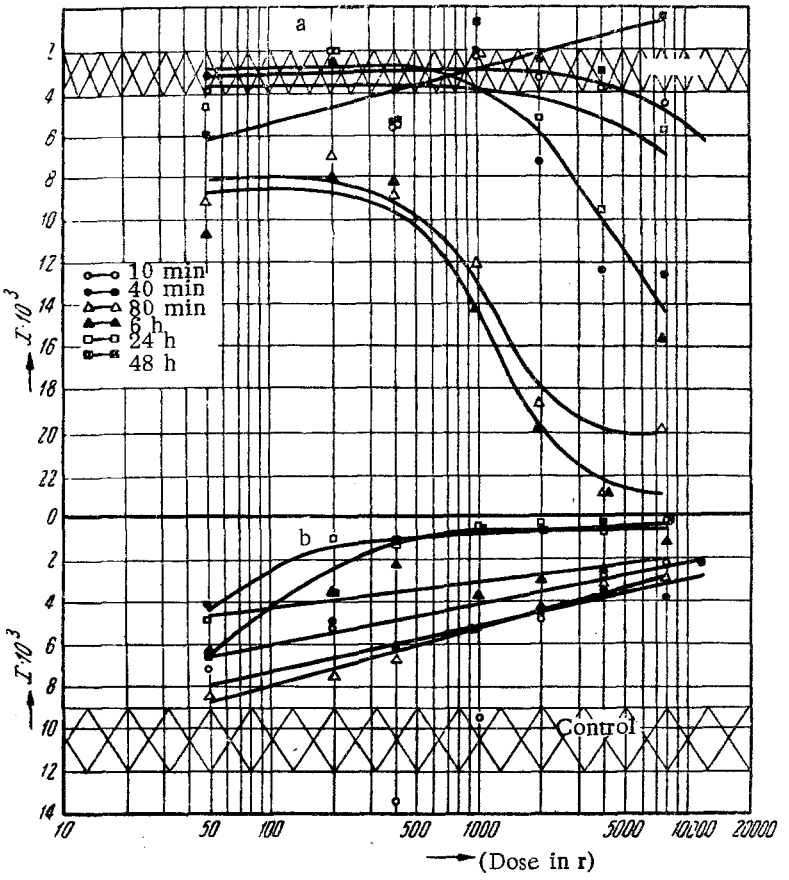

Fig. 3. Dose-effect curves for neutrophils (a) and lymphocytes (b) at different doses of x-irradiation.

8000 r and higher) constituted approximately 40-60% of the original values. Irradiation with 8000 r produced a decrease in the number of lymphocytes to the extent of 80-85%. In control rats the 12 blood samples also revealed significant changes in the leucocyte formula, but the normal rate lymphocyte picture was retained throughout our observations (Figs. 1 and 2). The estimation of the number of cytolyzed cells has shown that it rose from 4-6 to 14-20 per 100 cells after 4-7 hours, i.e., during the period of the highest neutrophil leucocytosis. After 24 and 48 hours cytolysis was practically absent and no destroyed cells were seen in blood smears. During the first 24 hours after irradiation there was no change in the structure of nuclei and the staining properties of the cytoplasm of white blood cells; all the circulating cells were mature. After 48 hours there appeared hypersegmented neutrophils with immature cytoplasm, and in the red blood there was an increase in the number of polychromatophilic erythrocytes and a considerable increase in the number of erythroblasts.

The processes of a regenerative nature were seen in all experimental animals – in surviving ones (doses of 50, 200, and 400 r) as well as in those which died (doses of 1000 r and higher). These processes were noted after 3-5 days and they were characterized by an appearance in the peripheral blood of a considerable number of reticular cells with a fine structure of chromatin fibers and the presence of nucleoli in the nucleus, and also by the appearance of young lymphocytes and lymphoblasts in the blood. After a sharp neutropenia (towards the end of the first and beginning of the second day) there appeared in the peripheral blood after 3-5 days a large number of neutrophils, a considerable number of which were in a state of cytolysis. During this period of time all the neutrophils contained an insignificant amount of chromatin; the nuclei were diffuse and weakly staining. In normal peripheral blood there were 4-6 cytolyzed cells per 100 cells, while during the initial period of the process of regeneration, their number increased 10 to 15 times, reaching 40-60 cytolyzed cells per 100 white blood cells.

The problem of the mechanism of the initial changes in the number of neutrophils is still unsolved [6]. It is hard to say whether neutrophil leucocytosis depends only on the supply of cells from reservoirs (bone marrow, spleen,

liver) or whether there takes place at the same time a more rapid maturation of cells and their rapid emission from the bone marrow due to contraction of blood vessels in hematopoietic organs. Possibly the increase in the number of neutrophils is related to an increased rate of excretion of glucoproteids, which may be assumed to take place in the light of a short-lived eosinophilia which arises after irradiation [7].

The results of our experiments have confirmed the existence of a significant difference in the reaction of the lymphoid and myeloid hematopoiesis at early stages following irradiation. This difference consists not only in the rate of the development of the reaction, but also in the shape of the dose-effect curve, and in the changes of this parameter with time. Figure 3 shows that with an increase of the irradiation dose the lymphocyte content of blood falls; this may be seen almost immediately (after 5-10 min) and it reaches its maximum after 1-2 days following irradiation. The shape of the dose-effect curve for the dose range used (50 to 8000-12,000 r) on the semi-logarithmic scale, as a rule, is either linear or S-shaped. It must be noted that after 1-2 days following irradiation (in doses of 200-8000 r) although the lymphocyte content of blood falls with the increased dose, the slope of the dose-effect curve remains insignificant; this indicates a high sensitivity of the lymphoid hematopoiesis, for which the dose of 200 r is considerably higher than threshold. This dose is high enough in order to produce a severe inhibition of the lymphoid hematopoiesis.

Unlike the lymphocyte reaction to irradiation, the number of neutrophils in the peripheral blood during the first 24 hours does not fall, but increases with the dose and the period of time following irradiation. The dose-effect curve is S-shaped and has a tendency to approach the abscissa with the increase of dose. The change in the dose-effect curve for neutrophils takes place more slowly than with lymphocytes. Thus, after 10 min following irradiation the change in the number of neutrophils may be seen only with irradiation doses of 8000 r and higher, while changes in the number of lymphocytes may be seen as early as 10 min after irradiation with a dose of 50 r. A significant decrease in the number of neutrophils can be seen only 2 days after irradiation, but unlike the lymphocyte reactions, it may be seen only after irradiation with high doses ($\gg$ 1000 r).

With doses lower than 1000 r the number of neutrophils during this period of time remains the same as in the control, or higher than in the control (in doses of 50-400 r). The dose-effect curve during this period is linear.

Thus, at early stages following irradiation the shape of the dose-effect curve may differ for different elements of the peripheral blood and may change with time. The results of our experiments have shown conclusively that the change in the number of leucocytes does not provide a basis for conclusions on the state of hematopoiesis in the irradiated animals and that a differential analysis of the state of the lymphoid and the myeloid hematopoiesis is essential. Special studies are needed to elucidate the mechanisms responsible for the peculiarity of the lymphoid, and especially the myeloid branches of hematopoiesis at early stages following irradiation in different doses.

SUMMARY

A study was made of the early reactions of the lymphoid and myeolid hemopoiesis following a wide range of x-ray doses (from 50 to 12,000 r – a total of 8 doses) in 70 rats. As a result of repeated investigation (12) of the peripheral blood composition (within a 24-hour period) it was established, that the changes of the lymphocyte and neutrophil count differ and it was impossible to assess the changes occurring in the white blood by the total number of leukocytes. The dose-effect curve for lymphocytes and neutrophils differed by the rate of the process and by the shape of the curve these indices show a significant change in time following irradiation. The mechanism of the mentioned changes requires special study.

LITERATURE CITED

1. M. F. Aleksandrova, Med. radiol. (1956), 4, p. 47.
2. P. V. Simonov, Three Phases in Reactions of the Organism to an Increasing Stimulus, Moscow (1962).
3. V. N. Chernigovskii and A. Ya. Yaroshevskii, Problems of the Nerve Regulation of the Blood System, Moscow (1953).
4. Kalcho Tsochev Kalchev, Hygiene of labor in work with radioactive substances and with sources of ionizing radiations, Abstracts, Moscow (1961), p. 53.
5. C. Aubertin and E. Beaujard, C. R. Soc. Biol. (1905), 58, p. 177.
6. Z. M. Bacq and P. Alexander, Fundamentals of Radiobiology, Oxford (1961).
7. M. L. Beaumariage, Arch. int. Physiol. (1955), 63, p. 132.

8. I. Becker and H. I. Thom, Radiobiology, Radiotherapy (1960), 1, p. 110.
9. Z. Dienstbier, F. Hermansky, and J. Camsky, Sang. (1955), 26, p. 321.
10. A. Forsberg, B. Tribukait, and K. Vikterlöf, J. Acta physiol. scand. (1961), 52, p. 1.
11. S. Hajdukovic and L. Stosic, Nature (1957), 179, p. 821.
12. L. O. Jacobson, Radiobiology, Moscow (1960), p. 251.
13. H. M. Patt and A. M. Brues, Radiobiology, Moscow (1960), p. 173.

All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of this issue.
