

# A HISTOCHEMICAL STUDY OF THE NUCLEIC ACID CONTENT IN MONKEYS WITH ACUTE RADIATION SICKNESS

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Translated from *Byulleten' Éksperimental'noi Biologii i Meditsiny*, Vol. 51,

No. 1, pp. 31-36, January, 1961

Original article submitted March 17, 1960

Our previous investigations [3,4,5] on rats, rabbits, and cats demonstrated substantial change in nucleic acid content attending acute radiation sickness induced by the action of x-rays. We observed that the character and degree of the change in the nucleic acid content differed considerably in the different organs of the irradiated animals. The data we obtained tallied with the observations of other researchers [2,6,7,9,10,11,12].

We then decided to trace the nucleic acid changes in more highly organized animals — monkeys — and to compare them with the results obtained with the other animals. We found no information concerning change effected by irradiation in the nucleic acid content of monkeys in the literature available to us.

## Experimental Methods

We had at our disposal material from seven monkeys (*Macaca rhesus*), five of which had undergone total irradiation by x-rays in a dose of 700 r,\* and two of which served as the control. The animals were sacrificed at the end of the latent period (third day), and at the height of the radiation sickness (sixth and eighth days after irradiation).

A typical picture of acute radiation sickness with the characteristic peripheral blood changes was observed in all the irradiated animals. According to A. S. Petrova's data [8], a decrease in the number of leukocytes and lymphocytes occurs in experimental monkeys under these conditions on the third day, later followed, on the sixth to eighth day, by a progressive decrease in the number of all the formal blood elements, and by qualitative changes of the latter (hypersegmentation of neutrophil nuclei, thrombocyte pyknosis, etc.).

The nucleic acid content was determined in the lymph nodes, duodenum, pancreas, liver, spinal cord, and brain. Pieces of the experimental organs were fixed in Carnoy's solution and embedded in paraffin. The DNA content was determined by Feulgen's test, RNA, by Brachet's method. The general picture of change in the irradiated tissues was examined after staining with hematoxylin-eosin. The brain and spinal cord sections were stained by Nissl's method.

## Experimental Results

The investigations showed that substantial disturbances in the nucleic acid content of the experimental organs occurred in the monkeys under conditions of acute radiation sickness, as well as structural changes demonstrated by the usual histological methods.

\*Conditions of irradiation: two-tube apparatus, voltage 180 kw; current force 15 ma; dose power 21.8 r/min; filter, 0.5 mm Cu. The experiments were performed at the Institute of Experimental Pathology and Therapy's base in Sukhumi.

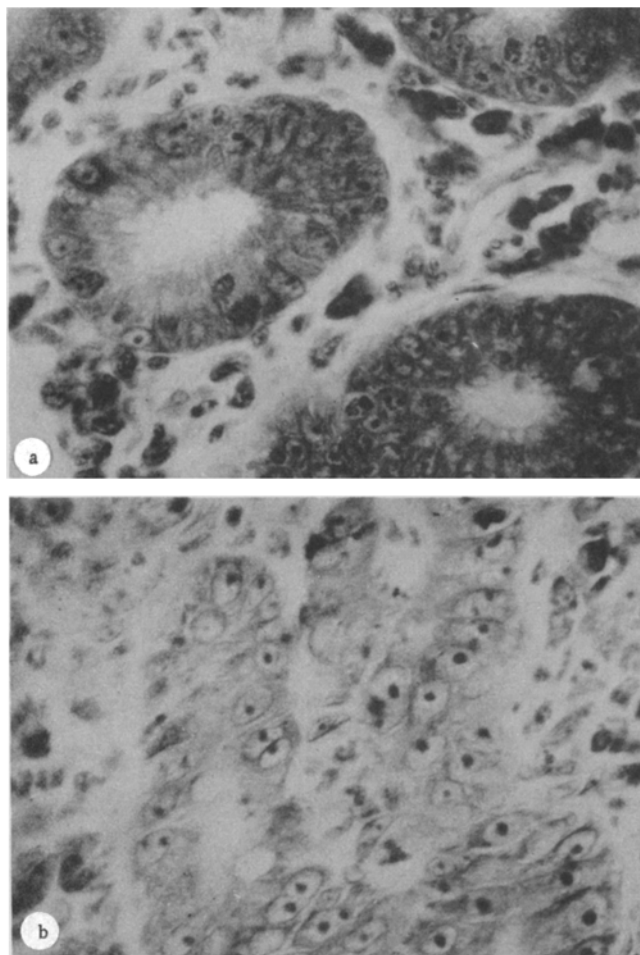


Fig. 1. Decrease in the RNA content of the protoplasm and increase of RNA content in the nucleoli of the mucous membrane of a monkey's duodenum: a) control; b) 6 days after irradiation. Stained with methyl-green pyronine. Magnification: ocular, 10  $\times$ ; objective, 40  $\times$ .

On the third and sixth days, we observed a considerable decrease in the amount of lymphoid elements in the lymph nodes, which caused the tissue to acquire a homogeneous structure made up largely of reticular cells. The secondary nodes had become smaller and were difficult to detect. Bits of destroyed lymphocytes could be seen here and there.

Along with the picture showing the almost total lack of lymphoid elements in the node, we observed a definite decrease in the DNA and RNA content of the remaining undestroyed lymphocytes. A small amount of the nucleic acids was found in the reticular cells; the degree to which these cells stained with pyronine and sulfuric acid fuchsin was essentially the same as in the case of the analogous formations in the control preparations. The few fragments of destroyed lymphocytes observed at this time stained intensively; our previous investigations on other animals showed that these fragments are present in large numbers during the first few hours after irradiation.

The changes observed on the eighth day after irradiation were of a similar nature.

On the third, sixth, and eighth days, we observed slight structural changes in the duodenum, consisting of compression of the epithelium of the villi and crypts, and swollen and irregularly shaped nuclei. The nucleoli were greatly enlarged. The changes in the nucleic acid content of the epithelium of the crypts and villi were analogous to the changes observed in the lymph nodes. Besides the decrease in the nucleic acid content of the cell protoplasm, however, we observed a large nucleic acid content in the nucleoli (Figs. 1a, b).

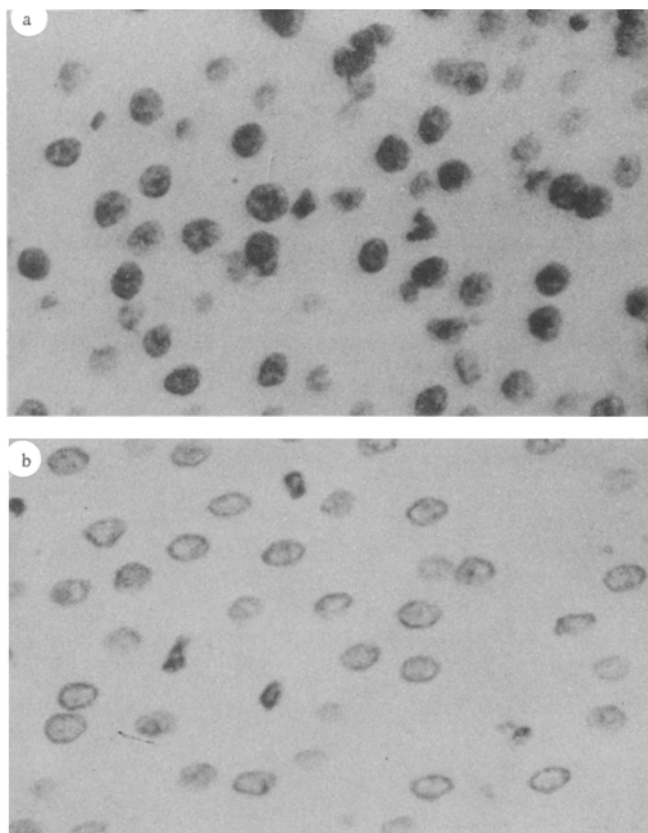


Fig. 2. Decrease in the DNA content of hepatic cell nuclei: a) control; b) 8 days after irradiation. Feulgen's reaction. Magnification: ocular, 10  $\times$ ; objective, 40  $\times$ .

In the liver and pancreas, we observed at these stages of the sickness a decrease in the DNA content and an increase in the RNA content. These changes were especially marked in the liver where, on the sixth and eighth days, it was barely possible to detect DNA in the cell nuclei by means of the method employed (Figs. 2a,b).

The change in the nucleic acids of the nerve cells of the spinal cord and brain chiefly concerned the RNA content.

During the experimental period, a large number of pyramidal cells intensely stained by pyronine were found in the cerebral cortex. These cells were usually wrinkled and irregularly shaped. A small number of these cells, intensely stained by pyronine, were also found in the control preparations, but the number found in the irradiated animals was much greater. In this case, the intense stain of the cells observed under conditions of irradiation could be due to disturbance of the chemical nature of RNA, rather than to an increase in the content of this compound.

We observed a decrease in the RNA content of the nerve cells in the anterior cornua of the spinal cord. The protoplasm of most of the cells contained rather small lumps, weakly stained by pyronine; in the control preparations, these lumps were large and intensely stained and analogous in shape, size, and distribution to the tigroid substance demonstrated by Nissl's method (Figs. 3a,b). The protoplasm of some cells was totally devoid of lumps, and homogeneous in structure. Although the protoplasm of these cells contained only a tiny amount of RNA, a large quantity of this compound was found in the enlarged nucleoli.

Under conditions of irradiation, the decreased RNA content of the cell protoplasm attended by a simultaneous increase in the RNA content of the nucleoli was observed in other organs also, particularly in the duodenum.

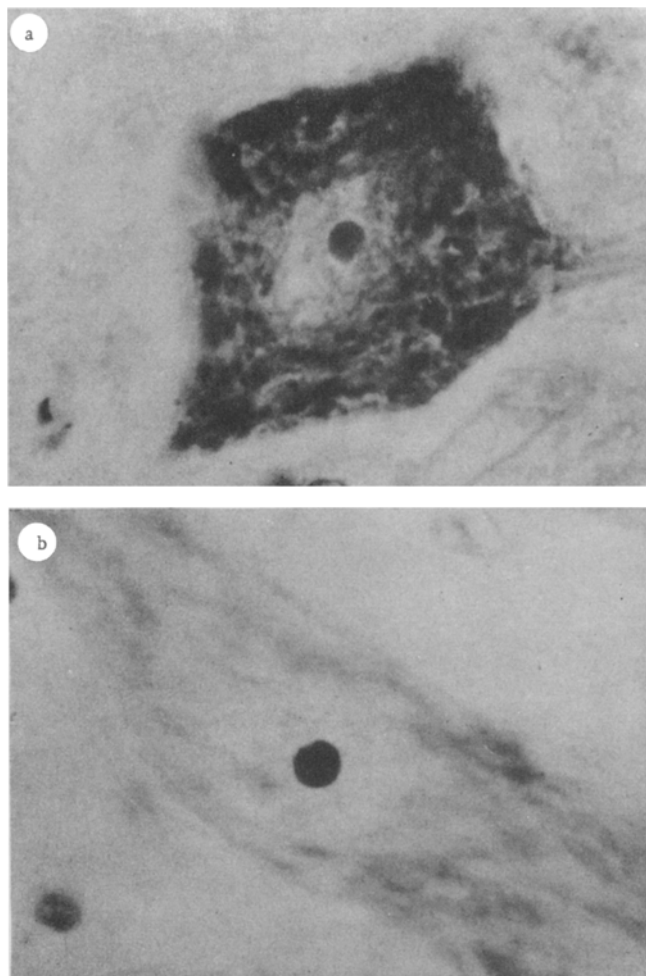


Fig. 3. Decrease in the RNA content of the protoplasm, and increase in its content in the nucleoli of the spinal cord nerve cells: a) control; b) 6 days after irradiation. Stained with methyl-green pyronine. Magnification: ocular, 10  $\times$ ; objective, 90  $\times$ .

Moreover, we observed similar phenomena in our previous investigations conducted on other animals (rats, cats).

In the light of the available literature data [1] describing analogous change in the nucleic acid content under conditions of protein deficiency, this phenomenon would seem to be due to disturbances in the protein metabolism. In our opinion, the increased RNA content of the cell nucleoli of the irradiated animals could be explained as compensation for the considerable decrease in the content of this acid in the protoplasm, intended to sustain the vital activity of the cells.

In the nuclei of the nerve cells of the spinal cord and brain, no substantial changes of any kind were observed in the DNA content at the experimental intervals after irradiation.

When the results obtained from studying the nucleic acid content in the organs of irradiated monkeys were compared with the data from the investigations previously conducted on other animals (rats, rabbits, cats), the picture of these changes was found to be basically the same in the different animals. In the monkeys, as in the other animals studied, the disturbances observed in the nucleic acid content of the experimental organs were not all of the same type, but differed in nature and degree of expression, which seems to be due to peculiarities of metabolism.

A few differences were noted, however.

The changes in the content of nucleic acids observed under conditions of irradiation were more pronounced in the monkeys than in the rats, rabbits, and cats. We also noted some difference in the degree of these changes in the different organs of the irradiated animals. In the monkeys, the changes in nucleic acid content were most pronounced in the liver, lymph nodes, and duodenal mucous membrane, while the most outstanding disturbances noted in the rats, rabbits, and cats occurred in the lymph nodes and duodenal mucous membrane; the changes in the liver were less pronounced in the latter three animals.

These investigations have covered only certain periods in radiation sickness. Because of the similarity of the results obtained in the experiments on monkeys with those obtained with the other animals, however, it seems possible to assume that the character of the changes in acute radiation sickness is identical. The slight differences remarked in the monkeys do not alter the general picture of similarity of disturbances in irradiated animals.

On the basis of the results obtained, we propose that the nature and degree of the changes in nucleic acid content which occur under conditions of irradiation are not determined by the type of animal. The character of the radiation injury evidently plays an essential role in these changes.

#### SUMMARY

The nucleic acid content was studied in various organs of Macaca rhesus monkeys subjected to x-ray irradiation (700 r). Changes in the nucleic acid concentration were similar to analogous disturbances in other animals, examined previously (rats, rabbits, and cats). The character and the extent of these changes differed in various organs.

The results obtained lead to a suggestion that the changes in the nucleic acid content occurring during acute radiation sickness follow the same pattern in different species of animals.

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\*Original Russian pagination. See C. B. translation.