ASSESSMENT OF THE BIOLOGICAL EFFECTS OF "STRANGE" RADIATION

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The results from studies of electrical explosions of foils made from super-pure materials in water pointed to the emergence of new chemical elements. An additional finding was the discharge of a "strange" radiation accompanying transformation of chemical elements [1]. Identical phenomena were revealed during tests of voltage limiters (VL) for breakdown.

The intensive research work focusing on effects of "strange" radiation, and the use of voltage limiters for industrial purposes require that appropriate safety regulations would be developed and applied in practice. However, currently, the mechanism involved in the interaction between "strange" radiation and a substance or a biological entity remains unclear.

The two principal questions brought up in connection with the assessment of the biological effects of "strange" radiation:

- 1. Is exposure to "strange" radiation safe for the personnel engaged in the studies?
- 2. Can biological systems serve as detectors of "strange" radiation?

Therefore, the aim of research was to investigate the biological effects of the "strange" radiation.

Project tasks :

- 1. Estimate the key hematological parameters after exposure of experimental animals to "strange" radiation resulting from explosion of foils in water or aqueous solutions.
- 2. Study the genotoxic effects of the "strange" radiation.

MATERIAL AND METHODS Experimental exposure

Pilot studies were performed at the RECOM RRC "Kurchatov Institute" in April-May of 2004.

Animals used in the experiment were female mice of C57Bl/6 line aged 80 days with body weight 16-18 g.

The animals were exposed to radiation discharged during explosions of Ti foils in water and aqueous solutions [1].

The cages with animals were placed at 1 m from the epicenter of the explosion. As can be seen from Table 1, explosions were carried out on the 19th (3 explosions), 20th (4 explosions) and 22nd (3 explosions) of April, 2004 (explosions ##1373-1382, respectively).

The animals were assigned to 4 experimental groups, each of 17-20 mice. The animals received experimental exposure within 1, 2 and 3 days of the experiment (Table 1). In total, the experimental groups were exposed to 3, 7 and 10 explosions, respectively.

Table 1

Group	Number of exposure	Total number of	Number of animals	
	days	explosions	per group	
Control	-	-	20	
B1	1	3	20	
B2	2	7	17	
B3	3	10	19	

Schematic presentation of the experiment

HEMATOLOGICAL STUDIES

In order to identify the basic reactions of the hemopoietic system, the following parameters were estimated using conventional methods: number of nucleated cells in the bone marrow, number of leucocytes in the peripheral blood, cell composition of the bone marrow, the rate of bone marrow erythrocytes at different levels of maturation (polychromatopile to oxiphile), and cell composition of the peripheral blood.

In the other experiment the number of CFUs was evaluated after acute whole body gammairradiation at a dose of 6 Gy.

Table 2

Schematic presentation of the experiment. Estimation of the CFUs after acute gamma irradiation

Group	Number of	Total number of explosions	Number of animals
	exposure days		per group

Control +	-	-	20
6 Gy			
B1 + 6 Gy	1	3	20
B2 + 6 Gy	2	7	17
B3 + 6 Gy	3	10	20
6 Gy + B3	3	10	20

Study of genotoxic effects of "strange" irradiation

To assess the potential genotoxic effect of "strange" radiation, the rates of bone marrow polichromatophile erythrocytes with the micronuclei were studied using the bone marrow slides from the hematology experiment. In addition we studied genotoxic effects of combined exposure to "strange" radiation acute gamma-irradiation at a dose of 2 Gy, which was given on the next day.

Table 3

Schematic presentation	of the exp	periment.	Study	of ger	notoxic	effect
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Group	Number of exposure	Total number of	Number of animals	
	days	explosions	per group	
Control	-	-	20	
B1	1	3	20	
B2	2	7	17	
B3	3	10	19	
Control + 2 Gy	-	-	10	
B3 + 2 Gy	3	10	10	

RESULTS AND DISCUSSION

Studies of the effects of "strange" radiation on the number of nucleated cells in the bone marrow

The number of nucleated cells in the bone marrow counted for the control group of C57B1/6 mice amounted to 38.6 + 1.6 mln/femur (Fig. 1). In spite of the fact that exposure to "strange" radiation of experimental group B1 on the 19th of April, 2004, and of experimental group B2 on the 19th and 20th of April, 2004, brought about a certain increase in this value up to 42.3 + 1.9 and 42.4 + 2.1, respectively, these changes, however, did not reach statistical significance. Exposure of the experimental animals to 10 explosions carried out within 3 days led to a further increase in this value. In this group the number of nucleated cells in the bone marrow was 45.1 + 1.7 mln./femur which exceeded significantly (by 17%) the respective values obtained for the control group ($t_{Student's} = 2.79$, P = 0.008).





Thus, it has been shown by our experiment that exposure of animals comprising group B3 to explosions of Ti foils in water and aqueous solutions results in an increased number of nucleated cells in the bone marrow of C57B1/6 mice. Such changes were accompanied by an increase in the percentage and count of dividing cells in bone marrow.

It can be assumed that these changes are induced by the following mechanisms: stimulation of stem cell division in the bone marrow, accelerated division of cells in a proliferating cell population within an organ, delayed maturation, decreased cell cycle time, decreased frequency of apoptotic cell

loss, disturbances of cell migration from the bone marrow to the blood, or by a combination of two or several of the above-listed mechanisms.

Studies of the population of bone marrow stem cells.

To understand the biological mechanisms of bone marrow changes we studied the reaction of bone marrow stem cells to "strange" radiation. In that experiment animals were exposed to acute whole body gamma-irradiation at a dose of 6 Gy after exposure to "strange" radiation. The spleen colony-forming units were analyzed at day 9 after gamma-irradiation. No significant changes were revealed in bone marrow stem sells in that experiment (fig.2.). So, we can conclude that "strange" radiation did not influence bone marrow stem cells.



Fig. 2. Number of CFUs in different experimental groups after gamma-irradiation of mice at a dose of 6 Gy.

Simultaneously, in that experiment we studied the repopulation of bone marrow cells at day 9 after gamma-irradiation. It allowed us to reveal a decrease in bone marrow repopulation in the group of animals exposed to gamma-irradiation followed by exposure to "strange" radiation for 3 days (fig. 3).



Fig. 3. Bone marrow cell numbers in different experimental groups after gammairradiation of mice at a dose of 6 Gy

The ratio of polychromatophil-to-oxiphil bone marrow erythrocytes in different experimental groups

To study the involvement of changes in cell on the increase in total bone marrow cell count, we analyzed the ratio of erythrocytes at different levels of maturation (polychromatophil-to-oxyphil). We did not reveal statistically significant changes in the analyzed parameter, but we noted that the PCE/OPE ratio increased with time of exposure (fig. 4). And after subsequent gamma-irradiation at adose of 2 Gy we revealed a significant increase in the PCE/OPE ratio as compared with the reaction of bone marrow cell to gamma-irradiation in control group. These two facts allow us to suppose that "strange" radiation leads to increased levels of immature erythrocytes in the bone marrow.



Fug. 4. The ratio of polychromatophil-to-oxiphil bone marrow erythrocytes in different experimental groups

Studies of the genotoxic effects of "strange" radiation

To evaluate the genotoxic effect of "strange" radiation, the rates of micronuclei in the bone marrow erythrocytes were analyzed. We did not reveal any statistically significant changes of the parameter. But in the experiment with combined exposure to "strange" radiation and subsequent gamma-irradiation, the rate of micronuclei was 1.5 times lower in comparison with the effect of only gamma-radiation. This type of reaction is called "adaptive response" in radiobiology (fig. 5). It may be attributed to the activation of protein synthesis in cells and activation of DNA repair mechanisms.



Fig. 5. Rates of micronuclei in polychromatophil bone marrow erythrocytes for different experimental groups

Studies of the effects of "strange" radiation on the peripheral blood leucocyte counts

It can be seen based on the analysis of leucocyte counts in the peripheral blood of mice exposed to "strange" radiation, that none of the experimental groups showed statistically significant changes in the parameters studied.

It is known that the occurrence of changes in peripheral blood cell counts lags behind the reaction manifested by changes in bone marrow cell counts by about 5 days. It can be suggested, therefore, that an increase in bone marrow cell counts would have been manifested by increased numbers in leucocytes of peripheral blood, if the exposure time had been prolonged.

Analyses of the peripheral blood cell composition

The studies of the peripheral blood cell composition is a routine procedure applied for diagnosing various pathologic conditions in humans and animals.

In our experiment the rates of peripheral blood neutrophils observed for the control group amounted to 15%.

The experimental exposures led to a consistent increase in peripheral blood neutrophil counts. Thus, within a day after exposure to 3 explosions, the rate of neutrophils in the peripheral blood was 18 %, within 2 days it was 22%, and at day 3 a statistically significant increase in the rate up to 25.5% was registered (fig. 6).

The regression analysis of the relationship between the rate of neutrophils and exposure time allowed estimation of a statistically significant (F=13.53; P = 0.0005) effect of the factor on the parameter studied.

The increase in the rate of neutrophils occurred primarily at the expense of mature segmented neutrophils, whereas the number of stab cells did not differ significantly from that found for controls.



Fig. 6. The relationship between the rates of peripheral blood neutrophils and exposure time

Along with the changes in the contents of neutrophils in the peripheral blood, the rate of peripheral blood lymphocytes was found to be reduced. A statistically significant decrease in this parameter to 68% was observed at day 3 of exposure.

No statistically significant fluctuations in the rates of peripheral blood eosinophils and monocytes were observed in the experimental animals, however, attention is attracted to a reduction in the rates of monocytes with increasing exposure time. It can be suggested that after the analysis of the total sample is completed, such changes may reach statistical significance.

The increase in the contents of neutrophils in the peripheral blood can be the result of several reasons: 1) An increased bone marrow neutrophil production (which is unlikely in our situation as there was not enough time for the manifestation of such effect); 2) Incoming of neutrophils from the place of deposition; 3) Decrease in neutrophil apoptosis rate.

Thus, it can be concluded based on the results of our experiments that:

1. "strange" radiation stimulates proliferation of bone marrow cells with or without delay in maturation;

2. it induces changes resulting in increased resistance to genotoxic exposures (gamma-irradiation and others);

3. "strange" radiation aggravates the clinical course of acute radiation disease if it is applied after gamma-irradiation.

4. it leads to changes of cells composition in the blood.

CONCLUSIONS

The studies performed allowed drawing the following conclusions:

- 1. "Strange" radiation resulting from explosion of Ti foils in water and aqueous solutions has a capacity for producing biological effects.
- 2. Biological effect of "strange" radiation is manifested by increase in the number of nucleated cells in the bone marrow.
- 3. "strange" radiation leads to an increase in dividing cells in bone marrow.
- 4. "strange" radiation resulting from 10 explosions carried out within 3 days after exposure to gamma-radiation (6 Gy) leads to a decrease in bone marrow repopulation.
- 5. Assessment of the rate of micronuclei in bone marrow erythrocytes did not reveal any genotoxic effect of "strange" radiation.
- 6. Exposure of mice to "strange" radiation leads to 1.5-fold decrease in genotoxic effect resulting from additional gamma-irradiation (2 Gy). Such reaction may be described as an adaptive response.
- 7. Exposure to "strange" radiation can bring about an increase in the proportion of neutrophils in the peripheral blood of experimental animals.

- 8. It can be suggested by the results of the test exposures that "strange" radiation can affect human health.
- 9. It has been shown by these preliminary studies that in order to gain an insight into the biological effects of "strange" radiation further investigation would be necessary.