# Effect of Low-Dose Irradiation on Immunological Reactivity

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A decrease in induced production of immune and leukocytic interferons by peripheral blood cells of patients exposed to doses of 10-30 rem is associated with increased production of IgA and acute phase proteins and decreased levels of complement C3 component and $\alpha_2$-macroglobulin.

**Key Words:** *irradiation; interferon production; immune proteins; acute phase proteins*

Ionizing irradiation of different intensity causes various immunomodulating effects. Interleukins-6 and -1, tumor necrosis factor-α, and immune interferon (IFN) stimulate differentiation of immunocompetent cells and intensify the production of acute inflammation phase proteins in the liver [2,3,5,7,8]. Leukocytic and immune IFN activate natural killer cells as the main factor of antitumor defense and through Fc-receptor stimulate phagocytes. T-helpers are activated and the subpopulation composition of T-lymphocytes is imbalanced in subjects exposed to low-dose ionizing radiation in the Chernobyl disaster zones.

The purpose of this study was to examine the effects of low-dose ionizing radiation on induced production of immune and leukocytic IFN by peripheral blood cells and on serum levels of immune proteins and acute phase proteins in Chernobyl cleanup workers.

## MATERIALS AND METHODS

Sixty-one subjects who participated in liquidation of the Chernobyl disaster in 1986 were examined in November, 1994. They aged 32-60 years. The mean age of subjects exposed to a dose of under 1 rem (group 1) was 47±3.8 years, to a dose of 1-10 rem (group 2) 41±1.7 years, to doses of 10-20 rem (group 3) 49±2.7 years, and to doses of 20-30 rem (group 4) 51±2.8 years. The mean age of healthy donors was 46±3.6 years.

Production of leukocytic and immune IFN by blood cells and serum IFN were estimated. Leukocytic IFN production was induced by Newcastle disease virus. The virus was passed in the allantoic fluid of 9-10-day chick embryos. The infective titer of the resultant virus-containing material was $10^{-7}$-$10^{-9}$ 50% infective dose ($ID_{50}$).

For inducing IFN production, 100 μl whole blood, 800 μl RPMI-1640 medium with glutamine (0.3 mg/ml), gentamicin (0.08 mg/ml), and 10% fetal calf serum were placed in a flask under sterile conditions. Then 100 μl of Newcastle disease virus (10-50 $ID_{50}$) was added for inducing leukocytic IFN production or phytohemagglutinin in the final concentration of 5 μg/ml for inducing immune IFN production. The production of leukocytic IFN was induced for 24 h, of immune IFN for 72 h.

After 24 h, the resultant leukocytic IFN was treated with 1 N HCl (pH 2.0-2.2) for 72 h at 4°C in order to inactivate Newcastle disease virus, after which pH was adjusted to 7.0-7.2 and the preparation was titered in M-19 cells. The titer of resultant immune IFN was determined in M-19 cells without pretreatment. Mouse encephalomyocarditis virus was the indicator during titration of IFN preparations in human embryonal cells M-19. The value inverse to IFN dilution inhibiting the destruction of fibroblast

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**TABLE 1.** Effect of Radiation Dose on Induced Production of Leukocytic and Immune IFN by Blood Cells (*M±m*)

| Radiation dose, rem | Mean IFN titers, units/ml | |
|---|---|---|
| | leukocytic | immune |
| Group 1, up to 1 (*n*=6) | 104±39 | 56±18 |
| Group 2, 1-10 (*n*=8) | 82±14 | 68±12 |
| Group 3, 10-20 (*n*=7) | 78±13 | 41±7 |
| Group 4, 20-30 (*n*=8) | 72±8.6 | 36±9 |
| Control, intact donors (*n*=10) | 307±92 | 89±6 |

monolayer by 50% and expressed in units/ml was the unit of IFN activity. Statistical analysis was carried out using Student's *t* test [1].

Immune (IgA, IgM, IgG, C3 and C4 components of the complement) and inflammation proteins ($\alpha_2$-macroglobulin, haptoglobin, transferrin, α-1-antitrypsin, and ceruloplasmin) were measured in the sera of exposed subjects in a Beckman ICS II immunochemical analyzer. This analyzer is based on the nephelometric method and is equipped with a microcomputer for light signal processing and detection of the immune precipitation reaction occurring in the buffer during the antibody-antigen reaction.

## RESULTS

Stimulated production of leukocytic and immune IFN decreased proportionally to the radiation dose, and the differences were statistically significant only

**TABLE 2.** Serum Proteins in Irradiated Patients

| Radiation dose, rem | Number of patients | | No. | Serum proteins | | |
|---|---|---|---|---|---|---|
| | total | with shifts | | class | concentration, g/liter | shift, % |
| Group 1, up to 1 | 9 | 4 | 1 | IgA | 4.26 (0.8—3.8) | 10 |
| | | | | Transferrin | 4.08 (2.04—3.6) | 13 |
| | | | 2 | C3 | 0.78 (0.85—1.9) | -8 |
| | | | 3 | C3 | 0.79 (0.85—1.9) | -7 |
| | | | 4 | Transferrin | 0.62 (2.0—3.6) | -1 |
| Group 2, 1-10 | 30 | 6 | 5 | Transferrin | 3.79 (2.0—3.6) | -3 |
| | | | 6 | Haptoglobin | 1.63 (0.27—1.39) | 17 |
| | | | | IgA | 4.06 (0.8—3.8) | 5 |
| | | | 7 | Transferrin | 3.73 (2.04—3.6) | 4 |
| | | | 8 | Transferrin | 3.66 (2.0—3.6) | 2 |
| | | | | α-1-Antitrypsin | 2.19 (0.9—2.13) | 3 |
| | | | 9 | $\alpha_2$-Macroglobulin | 1.03 (1.2—2.69) | -14 |
| | | | 10 | C3 | 0.79 (0.85—1.9) | -7 |
| Group 3, 10-20 | 12 | 3 | 11 | $\alpha_2$-Macroglobulin | 0.95 (1.2—2.69) | -21 |
| | | | 12 | $\alpha_2$-Macroglobulin | 1.12 (1.2—2.69) | -7 |
| | | | | Haptoglobin | 1.67 (0.27—1.39) | 20 |
| | | | | Transferrin | 3.90 (2.0—3.6) | 8 |
| | | | 13 | Transferrin | 7.21 (2.0—3.6) | 100 |
| | | | | α-1-Antitrypsin | 2.78 (0.9—2.13) | 31 |
| | | | 14 | α-1-Antitrypsin | 2.39 (0.9—2.13) | 12 |
| Group 4, 20-30 | 10 | 5 | 15 | $\alpha_2$-Macroglobulin | 1.05 (1.2—2.69) | -13 |
| | | | | Haptoglobin | 1.49 (0.27—1.39) | 7 |
| | | | 16 | C4 | 0.81 (0.15—0.45) | 67 |
| | | | | Transferrin | 4.36 (2.0—3.6) | 29 |
| | | | | α-1-Antitrypsin | 2.17 (0.9—2.13) | 2 |
| | | | | Ceruloplasmin | 0.51 (0.18—0.45) | 10 |
| | | | 17 | C3 | 0.83 (0.85—1.9) | -2 |
| | | | 18 | Haptoglobin | 1.67 (0.27—1.39) | 20 |

**Note.** Minimal and maximum levels of serum proteins in health are given in parentheses.

in groups 3 and 4 (Table 1) for the production of both immune ($p<0.01$) and leukocytic IFN ($p<0.05$) in comparison with the control (donors). Titers of serum IFN in exposed patients of all groups did not differ from the control and were 12.4±3 units/ml. These results indicate sensitivity of stimulated IFN production to low-dose radiation.

Table 2 shows that the production of IgA and C3 component of the complement are often altered in irradiated subjects. The level of IgA increased and that of C3 decreased. In patients exposed to the highest dose (group 4), IgG and IgM approached the lower threshold normal value, while the level of the complement C4 component increased. Of the inflammation proteins, the concentrations of transferrin, haptoglobin, α-1-antitrypsin, and ceruloplasmin increased. The levels of the antiprotease proteins α-1-antitrypsin and $\alpha_2$-macroglobulin changed in different directions: the level of the former increased and that of the latter dropped. Probably, such an effect of low-dose exposure is due to decreased production of immune IFN, because immune IFN increases the production of $\alpha_2$-macroglobulin and decreases the synthesis of α-1-antitrypsin [6]. Interactions of $\alpha_2$-macroglobulin with interleukin-6 stimulating the production of acute phase proteins in the liver are no less probable [4]. The increase in the content of transferrin, a $Fe^{2+}$ carrier in irradiated subjects, may reflect the effect of radiation on the erythroid stem of bone marrow and activation of cytopoiesis.

Thus, our study revealed that exposure to 10-30 rem decreases induced production of immune and leukocytic IFN and C3 component of the complement and increases the production of acute phase proteins, except $\alpha_2$-macroglobulin. These changes can be interpreted as a result of direct action of radiation on the cells producing these factors and of mediated reactions to radiation injury.

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