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Using the Biomaterial of the Radiobiological Human Tissue Repository for Solving the Problems of Radiobiology

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Banks of human biological material are the most important source of information and research resources for biological and medical studies of the effects of different types of ionizing radiation. The work described in this presentation was performed at the Radiobiological Human Tissue Repository (RHTR) of the Southern Urals Biophysics Institute of the FMBA of Russia, that was established in the framework of the USA-Russian collaborative project.
The objectives of RHTR are:

- **establishment, maintenance and development** of the Repository as it is a unique informational and research source for studying medical-biological effects of radiation on different levels of integration with the help of latest technologies.

- Along with the other types of biomaterial (autopsy, surgical/biopsy tissues, sputum cells and supernatant, saliva, etc.), the RHTR comprises a constantly replenished bank of blood and its components: whole blood, red blood cells, buffy coat, lymphocytes, B-lymphocytes, serum, plasma, DNA.

- A total of ~ **126 000** samples of blood and its components from **6370** individuals exposed to long term radiation and controls was accumulated.
Accumulated doses of external and internal radiation:

- Prolonged combined exposure was in occupational conditions on Mayak PA radiochemical and plutonium plants.
- External γ-doses accumulated during a working career ranged within 0.01-4.9 Gy, and Pu body burden – within 0.03-10.9 kBq.
- Age of individuals was from ~60 to ~80 years.
- Control group included Ozyorsk population who were not exposed to occupational radiation, matched by age and gender.
- The samples of blood serum stored in freezers under low temperature for 2-5 years were used in this research.
- The electronic database contains complete information for the biomaterial stored and for each registrant, including medical and dosimetry data.
Experimental Methods

Enzyme multiplied immunoassay was used to study the regulatory proteins levels. The biomaterial was obtained from nuclear workers occupationally exposed to different doses of radiation (480 individuals) and from the control residents (215 individuals).

The levels of 50 serum proteins regulating immune homeostasis was studied: growth factors, multifunctional interleukins, pro- and anti-inflammatory cytokines, and their receptors.
Results

- Significant differences in the levels of the proteins regulating immune status in blood were observed in comparison to the control group.

- Significant correlations were observed between the type of exposure, accumulated dose of external γ-exposure, Pu body burden, and the level of the following proteins in blood: growth factors EGF, TGF-β1, Ang-1, PDGF; interleukins IL-15, IL-17, IL-18, IFN-γ, IL-1β.

- These findings give evidence of radiation-induced deviations of protein status: overall, the exposed workers had significant increases of regulatory proteins and their receptors compared to population reference levels (2-5 times and more).
Examples: EGF and Ang-1 Levels

The level of EGF
(2.1-76)
Pg/ml

<table>
<thead>
<tr>
<th></th>
<th>Control 129.20</th>
<th>Workers 102.10</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>±6.89</td>
<td>±6.90*</td>
</tr>
</tbody>
</table>

The level of Ang-1
(14272-65570)
Pg/ml

<table>
<thead>
<tr>
<th></th>
<th>Control 45647±2483</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Workers 55784±4262*</td>
</tr>
</tbody>
</table>

* indicates a significant difference from control.
CORRELATION BETWEEN THE EGF LEVEL AND ACCUMULATED DOSE OF $\gamma$-EXPOSURE

$r = 0.24; p = 0.02; n = 88; y = 124 - 24x$
CORRELATION BETWEEN THE EGF LEVEL AND PU BODY BURDEN

\[ r = -0.3; \quad p = 0.0009; \quad n = 135; \quad y = 135 - 13x \]
CORRELATION BETWEEN THE TGF LEVEL AND ACCUMULATED DOSE OF $\gamma$-EXPOSURE

$r = -0.43; p = 0.00000003; n = 156; y = 52138 - 16636\times x$

Accumulated from exposure, Gy

Concentration TGF-beta1 in serum, pg/ml
CORRELATION BETWEEN THE TGF LEVEL AND PU BODY BURDEN

\[ r = -0.21; \ p = 0.005; \ n = 18; \ y = 16678 - 1554x \]

**Graph:**
- **X-axis:** \(^{239}\text{Pu body burden, кБк}\)
- **Y-axis:** Concentration of TGF-beta 1 in blood, pg/ml

**Equation:**
\[ y = 16678 - 1554x \]
CORRELATION BETWEEN THE PDGF LEVEL AND PU BODY BURDEN

\[ r = 0.82; \quad p = 0.0001; \quad n = 16; \quad y = 4814 + 4342 \times x \]

- Concentration PDGF in serum, kBq
- $^{239}$Pu body burden, kBq
CORRELATION BETWEEN THE IL-1 LEVEL AND ACCUMULATED DOSE OF γ-EXPOSURE

$r = 0.44; p = 0.003; n = 45; y = 40+45*x$

Accumulated dose from external gamma-rays, Gy

Concentration of IL-1 beta in blood serum, pg/mL
Conclusions

• Among a group of subjects with a prolonged period of occupational exposure, the following proteins have strong potential for use as markers of radiation-induced changes:
  - **Growth factors**: epidermal (EGF), transforming (TGF-β1), platelet-derived (PDGF), Ang-1, fibroblastic (FGF)
  - **Multifunctional interleukins**: (IL-17A, IL-18, IL-15) and cytokines IL-1β, INF-γ.
  - Average decreases of the level of EGF, IL-18, and increases of FGF, IFN-γ, IL-15, Ang-1 were observed.
  - Linear correlations were seen between the level of proteins and Y-dose for IL-17A, IFN-Y, and IL-1β, but with inverse effects for EGF, and TGF-β1.
  - Linear correlations were seen between the level of proteins and Pu body burden for PDGF, and IL-1β, but with inverse effects for EGF, and TGF-β1.
Future Directions

We will investigate the relationships of these protein and receptor changes on chronic disease development in this unique cohort and its biorepository.

Such examples will include malignant transformation of cells, accelerated growth of malignant tumors, development of cardiovascular diseases and other serious somatic pathology.

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THANK YOU FOR ATTENTION!