# Bioresonance Therapy Corrects the Immunodeficiency of Chernobyl mice

Sakharov D., Savtsova Z., Indyk V., Kovbasyuk S., Voicikova I., Zaritskaia M., Ortovsky A., Serkiz Ja, Kavetsky R. E. Institute of Experimental Pathology, Oncology, and Radiobiology of the National Academy of Science of the Ukraine

Lednyickzy G. Hippocampus Research Facilites, Central Division, 1031 Budapest, Nánási út 67, Hungary, Tel/fax: 36-1-188-6865

## Abstract:

The investigation was carried out over seven months using 057BI mice which were held for 6 months in the ambient conditions of the radiation zone of the Chemobyl accident. Total dose of external gamma-noise during the whole period of time in Chemobyl zone was less then 0.1 Gy. In these experimental animals, damage of immune homeostasis and formation of secondary immunodeficiency was observed.

To correct the immune system status, the endogenous modified electromagnetic fields of the radiated Chemobyl mice was applied. The treatment led to normalization of thymus and peripheral lymph nodes weight and cell numbers indices as well as the peripheral lymph nodes' cellular composition. The resistance of the animals to an experimentally induced viral infection returned to the level of the control animals of the same strain Keywords: mice, gamma-noise, immunodeficience, electromagnetic field, radiation

### Introduction:

Described here is the influence of ELF-ELI-EMF on the functioning of the immune system and how the observed results can be explained, taking into account that the sources of ELF-ELI EMF are the bodies of mice experienced with the action of radionucleids. It. is suspected that the harmonious oscillations correspond to the physiological (nonpathological) processes in living and disharmonious oscillations correspond to pathological processes. Three basic assumtions were made before the experiment began: 1) each mouse's EMF is the sum of its harmonious and disharmonious oscillations; 2) both oscillations have certain intervals or frequencies that differ from each other; 3) if the H+Di treatment mode (Harmonious and inverted Disharmonious oscillations) is used for a chosen frequency which is suspected to belong to the physiological interval of oscillations and Ai - (for inverting all endogenous frequencies) then a considerable improvement in the condition of the mice's organism should be produced.

The study of the influence of continuous low intensity irradiation (the so called Chernobyl factor) on the immune system reveals a wide variety of immunomodifying effectts of this type of irradiation. Quantitative (the number of cells and cellular composition of central and peripheral organs of the immunity system and the blood) [1-4] and qualitative (functional activity of various immununocompetent cell populations and the rate of cooperative immune reactions) [2,5] indices of the immune system are changed substantially. Regulative interactions (the level of some cytokines, a- and g-interferon, IL-1, IL-2, TNF, and antigen-specific T-suppression) were altered [6-8]. Proliferation and activation of processes stimulation of some other indices [4,6,7] is possible in an early stage of radiation damage when the nonspecific adaptive reactions dominate alterations in the immune system [3].

The immune system homeostasis disturbances develop with the time of irradiation to form a secondary immune deficiency. This is revealed in experimental [2,5,7] and wild [9] rodents and cattle [10] in the radiation zone created after the meltdown at the Chernobyl nuclear power plant. The preferential disturbance of the activity of natural killer cells and T-lymphocyte-dependent reactions is a peculiarity of such immune deficiencies [2,3,5,7,8]. The obtained results show that in a post-Chernobyl situation, the correction of the immune imbalance by using conventional immunostimulators may be not only unexpedint but also harmful.

The correction should be mostly aimed at the enhancement of the immune system adaptability, rehabilitation of the normal maturation and differentiation of the lymphocytes and optimization of the immune cell interactions [11,12]. New methods of immune correction are offered to counter the effects of the continuous low-intensity radiation [11,12]. A new helpful method in the correction of immunodeficiency could be Bioresonance therapy.

Materials and methods:

### Animals:

Nine months old male mice of line C57BI, are under investigation. At the age of 2.5 months mice were conveyed from the vivarium of the R. E. Kavetsky Instnute of Experimental Pathology, Oncology, and Radiobiology of the Natl Acad. Sci. of Ukraine to the institute's experimental base, situated in the town of Chernobyl. Hence, they were maintamed under conditions of low-dose ionizing radiation for 6.5 months. The animals were influenced by both background  $\gamma$ -irradiation and the action of radionucleids being, administered, generally with water and food. The animal's diet was composed of the food-stuffs produced in the contaminated zone. The spectral content of radionucleids, its activity, distribution wrthm various organs and tissues were precisely described previously [14]. Total dose of external  $\gamma$ -noise was less then 0 I Gray (Gy) during the whole period of time in the Chernobyl zone. The activity of radionucleides in food stuff was in rate of 224 Becquerel (Bq) per day The contribution of  $\gamma$ -component in total dose of radiation was not counted.

## Apparatus:

The BICOM device (Brügemann, Germany) was used to acquire the ELF-ELI-EMF of the untreated Chernobyl animals (group 1, as denoted below), modify such EMF and transduce this field into the mice of the remaining. experimental groups. The device is equipped with 10Hz - 150kHz frequency band-pass filter. The device also makes it possible to process with a more narrow frequency band or a fixed frequency. The device can amplify 0.05-64 times the received ELF-ELI-EMF. Moreover, a special separator makes it possible to filter out harmonious oscillations (H) from disharmonious (D) within the total spectrum of oscillations (A). The mice's electromagnesic oscillations can be inverted (the "Ai" function), then transmitted to the treated mice as a mirror image, phase-shifted through 180°. The same can be done with the separated (D) part of oscillations (the "Di" function) [13]. On the surface of "output" electrodes the power density was less than 1 mW/m<sup>2</sup>.

Experimental procedures:

Let the group consisting of "Kiev control" nues be group 0.

Group 1: Mice from Chernobyl that did not interact with Bioresonance Treatment (BRT). Group 2. Animals continuously treated by BRT programs using the electromagnetic fields of radiated Chernobyl mice after they were transported to Kiev: I) Ai program, at amplification 0.8, run speed of bandpass (18 sec.), interval 5 min

II) H+Di program, at amplification (0.1+0.5) center frequency - 870Hz, wobbling, interval 5 min.

Group 3: Animals continuously treated by BRT programs using the electromagnetic fields of radiated Chernobyl mice after they were transported to Kiev

I) Ai program, run speed of bandpass (18 sec.), interval 5 min.

II) Ai program, at amplificanon 18, center frequency – 1.15kHz, wobbling, interval 5 min.

To avoid a misunderstanding of "run speeds of bandpass", in this case accepted EMP comes up and down through bandpass consequently following from frequency with constant chosen speed. BRT is administered once a day, five times on every second day in the morning (11 a.m.) to mice in experimental groups 2 and 3.

The immunological assays were conducted 1 week after the last BRT. Organ indices (the weight of thymus, spleen, and peripheral lymph nodes as well as mean number of lymphocytes in those organs) are estimated by a commonly used method [1].

The lymph-node cellular composition is analyzed by Shtokunger and Kelner's staining [15].

Delay Type Hypersensinvity Reaction (DTHR) is tested by the routine method [16] in modification [5]. The natural citotoxity or functional activity of NK cells is checked radiologically and derived from the destruction of the K562 target-cells labeled with 14-hydrolyzed protein [5]. For the purpose of estimating the resistance to a viral infection (B-immunity), the animals are infected intrinsically with an allantois culture of the A/PR8/34 influenza virus at a 100 EID dosage. The dynamic of mortality is determined until the 21st day after infection [17] At the same time, all changes in morphology and weight parameters of the lung of the survived mice are measured under the microscope [5]. Antigen-antibody reactivity (serum antihaemagglutinin titres) was examined using the standard method of haemagglutination reaction.

Results and Discussion:

Indices m the thymus and lymph nodes for group 1 mice (untreated, Chernobyl mice) differ much from that of the Kiev control group (Tab 1). The indices for Macronucleus T-lymphocytes, and Blasts and large lymphocytes in group 1 mice differ greatly from the Kiev control group (Tab. 2). The same can be said regarding the rate of functional activity of NK cells, mass of lungs and antibody production against flu viruses (Tab 3, 4). Thus the

antiviral resistance of group 1 mice was decreased considerably though the dose of infection was  $0.01 \text{ LD}_{50}$  of the dose admimstered for mice of the Kiev control group, nevertheless the mortality of the group 1 animals was 20% (2 mice in 10). The results show the physiological condition and immunological status of group 1 mice was much deteriorated.

In lung tissue, the large foci of haemorragic pneumoma was observed three weeks after infection. The weight of the lung on average was 15.9 +- 3.0 mg/g, the weight increase index was I 94 In contrast, among the control animals from Kiev (group 0), no deaths resulting from infection were observed. No macroscopic changes in the morphology of the lung tissue were found.

The treatment mode for group 2 animals resulted in the rehabilitation of the immune systém; the weight of the thymus and peripheral lymph nodes returned to a normal level. Normalization of the lymph-node's cellular composition, regarding the component of mature T-celll, was found (Tab. 1, 2). As compared with group 1, B-lymphocytes in spleen are the highest in the group 2 (Tab. 1). The indicators of effected activity of different immunocompetent cell's populations are altered: NK cell's activity remained at the level of untreated mice. Application of treatment programs caused an increase of the resistance of organisms to the infection: 100% of the treated mice survived. Microscopic alterations appeared on the 21st day after infecting like a single small foci of pneumonia (1-2 foci pres mouse). The relative weight of lung was 9.7 +- 0.8 mg/g (P1-2=0.1;P2-4=0.1). The weight increase index was 1.18.

The increase of the organ indices took place after the treatment of animals with programs for group 3. In comparison with group 2, the effects of treatment are more pronounced in thymus and less in spleen and lymph-node (Tab. 1.). Lymph-node cellular composition is also significantly distinguished and exhibits the lowest content of the mature T-cells, while content of blasts and large lymphocytes was increased as well as the number of B-cells (Tab. 2.). The rate of the NK-cells activity was remarkably lower than in the other groups. The mortality in the acute period of inflammation was 20%, as in the case of the untreated Chernobyl mice. The relative weight of the lung by the 21st day after infection was 15.2 +- 2.9 mg/g, the weight increase index was 1.85.

The 6.5 month placement of the male mice line C57BI in the radiological conditions of the Chernobyl zone caused alterations in a variety of immunological determinants. Spectrum, peculiarities and nature of the alterations are consistent with those revealed previously in the mice of other lines and in rats kept under analogous conditions [1-3,5].

The status of immune systém of animals may be defined as immunodeficiency [2,5]. Regardless of the program, essential alterations in a variety of immunological indices strictly connected to the current state of the immune systém were observed. But only by applying the programs selected for group 2, immunocorrecting effects manifested themselves integrally in the increase of the anitiviral resistance of animals.

The distant damages of immune systém caused by pronolonged low-intensity radiation are substantially connected with the depression of T-cell differentiation (mostly of T-helper cells) [18].

One of the characteristics of that depression is redistribution in the ratio of mature and immature immunocompetent cell towards a prevalence of the immature cell population [1,3]. The scheme of treatment in group 2 appeared to cause a predominant optimization of T-cell differentiation and maturation. This may explain the intensified migration of cells towards peripheral immune organs and subsequent increase of the cell number in spleen and lymph-nodes. The radio of mature/immature cells in effector populations was optimized and the efficacy of cooperative immune reactions (antibody's production) grows. Since spontaneous antigen-un-specific separation is known to decrease after prolonged habitation under radiation conditions this does not contradict the proposed mechanism of BRT. As this takes place, the damage in activity of supperesser-cells is observed at the level of mature T-suppressers [5]. The increase of antigen-un-specific supperession may well cause a decrease in the level of immunological reaction when using program 2, and improvement of differentiation and T-cell maturation involves the supperesser subpopulation as well. However, this aspect of the program requires special examination.

The following part of the discussion covers the immunological effects of ELF-ELI EMF that are observed in group 3 mice. The program appers to stimulate the proliferation of lymphoid cells but not differentiation or maturation. This is suggested by the increase in the cell number, primarily of lymph-nodes. The unbalance of functions of mature and immature lymphoid cells of different populations being kept (or increased) may cause a significant decrease in the natural cytotoxity and the lack of pronounced defects in program 3 concerning resistance to viral infection.

Finally, several explicit conclusions are drawn: Endogenous ELF-ELI EMF can influence the functioning of the immune systém and correct a condition of immunodeficiency created by exposure to continuous, low-intensity radiantion. Indirectly, shown is that intervals of frequencies around 870Hz posses physiological oscillations. A clarification of these results would require a more sophisticated theoretical as well as practical approach.

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Table 1. The Alteration of the Mass and the Number of Lymphocytes of Peripheral Lymphoid Organs							
		Relative Mass (mg), M+m			Number of lymphocytes, x10 <sup>5</sup> , M+m		
Group	Treatment	Thymus	Spleen	Lymph nodes	Thymus	Spleen	Lymph nodes
0	Kiev control	1.9 ± 0.2	4.1 ± 0.5	1.2 ± 0.1	93.5 ± 12.0	81.6 ± 7.5	24.5 ± 4.2
1	Chernobyl control	1.4 ± 0.1	$4.9 \pm 0.4$	0.9 ± 0.1	48.0 ± 13.8	73.3 ± 8.1	10.6 ± 4.0
2	Ai; H+Di870	2.1 ± 0.1	6.1 ± 0.5	1.3 ± 0.1	82.5 ± 7.0	130.0 ± 7.2	24.0 ± 3.0
3	Ai1.15kHz	3.1 ± 0.2	5.3 ± 0.4	1.3 ± 0.2	110.0 ± 8.0	90.0 ± 6.3	17.1 ± 4.0
P(1-0)		0.1>P>0.05		0.1>P>0.05	0.1>P>0.05		0.1>P>0.05
P(2-1)		< 0.05		< 0.05	0.1>P>0.05	< 0.05	< 0.05
P(2-0)			< 0.05			< 0.05	
P(3-1)		< 0.05		< 0.05	< 0.05		
P(3-2)		< 0.01			< 0.05	< 0.05	
P(3-0)		< 0.05					

M = mean

m = std.deviation

Table 2. Alteration in the cell composition of Peripheral Lymph Nodes							
	Content, % (M+m)						
Group	Treatment	Macronucleus T-lymphocytes Spleen	Micronucleus T-lymphocytes	Blasts and large lymphocytes	Other cellular elements		
0	Kiev control	62.9 ± 0.9	31.5 ± 0.9	4.0 ± 0.3	0.7 ± 0.2		
1	Chernobyl control	1.4 ± 0.1	0.9 ± 0.1	2.9 ± 0.2	0.8 ± 0.01		
2	Ai; H+Di870	2.1 ± 0.1	1.3 ± 0.1	2.6 ± 0.1	0.8 ± 0.1		
3	Ai1.15kHz	3.1 ± 0.2	1.3 ± 0.2	4.8 ± 0.2	0.6 ± 0.1		
P(1-0)		< 0.05	< 0.05	< 0.05			
P(2-1)		< 0.05	< 0.05	< 0.05			
P(2-0)		> 0.1	0.05				
P(3-1)		< 0.05	< 0.05	< 0.05			
P(3-2)		< 0.01	< 0.05	< 0.05			
P(3-0)		< 0.05	< 0.05	0.1>P>0.05			

M = mean

m = std.deviation

Table 3. The Alterations in the Activity of NK cytolysis and cooperative immune reactions				
Group	Treatment	NK cytotox., %		
0	Kiev control	17.6 ± 1.1		
1	Chernobyl control	10.9 ± 0.4		
2	Ai; H+Di870	10.6 ± 0.8		
3	Ai1.15kHz	7.7 ± 0.4		
P(1-0)		< 0.05		
P(2-1)				
P(2-0)		< 0.05		
P(3-1)		< 0.05		
P(3-2)		< 0.05		
P(3-0)		< 0.05		

Table 4. Results of infecting mice with Influenza virus:						
Index:	Group 0 Kiev Control	Group 1 Chernobyl Control	Group 2 Ai;H+Di870	Group 3 Ai1.15kHz		
Lethality(%)	0	20	0	20		
Mass of lungs(mg)	8.2 ± 0.4	15.9 ± 3.0	9.7 ± 0.8	15.2 ± 2.9		
Weight increase index	1.00	1.94	1.18	1.85		