

# ***DETERMINATION OF ANTIOXIDANT ACTIVITY OF BLOOD SERUM IN CHRONIC BODY IRRADIATION OF ELECTROMAGNETIC RADIATION OF 460 MHZ***

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**Abstract** — In today's world of electromagnetic radiation (EMR) in the radio and microwave bands have become an integral part of human activity, therefore, to study their effects on the redox homeostasis of the organism is not of secondary importance. This work was carried out in order to identify changes in the total oxidant and antioxidant activity in plasma and erythrocytes under the influence of chronic exposure to EMR 460 MHz. White rats were irradiated for 1 month daily for 20 minutes at power flux density - 30 mW/cm<sup>2</sup>. Total oxidant and antioxidant activity of plasma and erythrocytes was determined. The data obtained proves that exposure of rats relatively high intensity results in a lower total plasma antioxidant activity (relative to control) for minor fluctuations oxidant activity. A significant increase in antioxidant activity when irradiated EMP 460 MHz is observed in erythrocytes. Decrease in the total antioxidant activity of plasma and persistent increase in the level of free radical oxidation products allows talking about the imbalance in the antioxidant defense system of the blood and the body as a whole, which is an unfavorable factor in the pathological process and requires effective measures of metabolic correction.

**Keywords**— *electromagnetic radiation, the total antioxidants, total oxidants, free radicals, plasma, erythrocytes*

## **I. Introduction**

Most living organisms belonging to complex ecological systems, the ability to live can not do without the loss of oxygen, through which can perform a variety of metabolic functions. Redox processes in the body are highly reactive with the participation of reactive oxygen type (ROT). In case of violation of metabolism of ROT in various pathological conditions and uncontrolled increase in the formation of free radicals in the body. Lipid peroxidation (LPO) is enhanced at the so-called oxidative stress, the consequence of which is a violation of the properties of biological membranes and cell function [1]

Excessive formation of free radicals can damage nucleic acids, proteins, primarily lipids of cell membranes, which are stimulated by lipid peroxidation (LPO), that result in a violation of the barrier properties of the membranes and change their structure, impaired permeability, which leads to

damage or destruction of cells. [2, 3]. In recent publications it was that EMR 460 MHz at the irradiation of total-body, cause changes in balance of redoxreactions in the different structures of cerebrum, blood and eyes [4, 5, 6, 7, 8].

It is known that inhibition of universal erythrocytes mechanisms in the process of lipid peroxidation of free radicals provides by antioxidant system (AOS). Thus, in the course of evolution two interrelated and interdependent systems - oxidant and antioxidant were formed in the body. The simultaneous study of both systems is a good tool for identifying the specifics of exposure to a particular environmental factor [9]. Taking into account foregoing, in this article we studied the changes of general oxidant and antioxidant activity in blood of rats chronically undergoing 460 Mhz by an electromagnetic radiation.

## **II. Methods**

Studies were conducted on white Wistar rats weighing 250-300 g with in the conditions normal vivarium contained. The experiment was conducted in accordance with ethical standards and rules of working with laboratory animals. Animals were divided into experimental and control groups. The experimental group of animals irradiated decimeter radiation (460 MHz) using the "Volna- 2" (made in Russia) in a metal cylindrical chamber. Irradiation was carried out daily for 20 minutes during 1 month, it was implemented in the exposure mode of a relatively high intensity at a power density of 30 mW/ cm<sup>2</sup>. The total blood was collected by decapitation to rats which were separated from plasma and erythrocyte solution 1.34% (w/obbem) sodium oxalate was used as an anticoagulant.

Total oxidant (TOA) and total antioxidant (TAA) activity of plasma and erythrocytes were determined by common method [10]. The principle of the method. RVA estimated by the accumulation in the reaction mixture, the final product of peroxidation - malonic dialdehyde (MDA) by the oxidation of

the substrate (Tween-80).

2 mL of Tween-80 (1%) was poured to the dark glass vial with stopper. 0.2 ml of plasma was added to the sample. Sample was incubated at 40° C for 48 hours, after 40% trichloroacetic acid (TCA) was added to 1 ml of sample and left at room temperature for 60 min. Then, centrifugation is performed at 8000 rev/min for 15 min. Supernatant - 2 ml, mixed with 2 ml of 0.25% thiobarbituric acid (TBA) and boiled for 15 minutes. Thus, a complex trimetinovogo having a pink color was formed as a result of reaction between MDA and thiobarbituric acid. Samples were cooled and registered by spectrophotometer "Spekol-221" of 532 nm in 10 mm cell. Sample containing biological material instead of the water and the treated as well as the test sample was used as control.

Calculation

$$TOA(\%) = \frac{E_o - E_c}{E_o} \times 100$$

here  $E_0$  and the  $E_C$  - extinction, respectively the experimental and control samples.

Principle of method of determination of total antioxidant (TAA) activity is based on determination of MDA, as an oxidant of Tween-80 at inhibition ascorbate or  $Fe^{3+}$ . Plasma is deleted from hemolysate of red blood cells. 0,4 ml of physiological solution (0,9% NaCl) was added to the sediment medix and centrifuged for 5-10 mines at 3000 rev/min. Liquid is poured out and add physiological solution was added. This procedure is repeated twice. Then 0,9 mls of the distilled water was added to the hemolysate consisting of 0,1 ml of the washed erythrocytes. Then, 2 mL of Tween-80, 0.2 mL of ferrous sulfate solution (1 mM  $Fe_2(SO_4)_3$ ), 0.2 ml of a solution of ascorbic acid (10 mM ascorbic acid) 0,1 ml of the hemolysate or 0.2 mL of plasma were added to the dark glass vial. In the control sample instead of the biological material appropriate amount of distilled water was used. After incubation at 48 h and 40°C, 1 ml TCA was added to the 2 ml mixture, the mixture was centrifuged at 8000 rev/min for 15 min. 2 ml TBA was added to 1 ml supernatant and boiled for 15 min. The procedure was followed with cooling. Optical density of samples was registered by spectrophotometer "Spekol-221". Measurements for the absorbance of the upper phase conducted at a wavelength of 532 nm against distilled water.

Calculation of results is carried out according to the formula

$$TAA(\%) = \frac{E_c - E_o}{E_c} \times 100$$

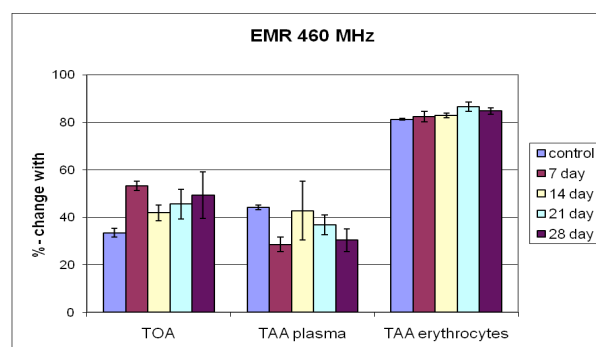
where  $E_0$  and the  $E_C$  - extinction, respectively the experimental and control samples.

Statistical analysis was made using program Microsoft Excel and significance of means' difference was evaluated using paired Student T-test.

### III. Results and discussion

Research results on determination of general oxidant and

antioxidant activity in plasma and erythrocytes are presented for control and radiation-exposed rats from relatively high intensity of irradiation (30 mBT/of  $cm^2$ ) (Fig. ). As shown, the level of total oxidant activity irradiated rats ( $53,24 \pm 1,9$  (7 days);  $41,84 \pm 3,3$  (14 days);  $45,52 \pm 6,2$  (21 days);  $49,2 \pm 98$  (28 days)) sequentially higher than the control rats ( $33,45 \pm 1,8$ ). Also in fig. shows that total plasma antioxidant activity is reduced in the continuation of irradiation ( $28,51 \pm 3,1$  (7 days);  $42,74 \pm 12,4$  (14 days);  $36,9 \pm 4,2$  (21 days);  $30,3 \pm 4,7$  (28 days) than the control rats ( $44,21 \pm 0,98$ ). The experiment produced interesting data on erythrocytes. As shown on a picture, level of total antioxidants in red blood cells rises for the radiation-exposed rats to 82,4% at a 7 day of irradiation ( $p > 0,05$ ), 82,8% at a 14 days ( $p > 0,05$ ), 86,47% at a 21 days ( $p = 0,05$ ), 84,8% at a 28 days ( $p > 0,05$ ), as compared to control rats (81,2%).



**Fig.** Activity common oxidants and antioxidants in plasma and erythrocytes of rats exposed to radiation of 460 MHz for 1 month 20 min daily at a power density of 30 mW /  $cm^2$  (irradiation intensity is relatively high) each represents the mean  $\pm$  SE ( $n = 5$  rats). \*  $P < 0.05$  versus the irradiated control group.

In recent years the world has conducted intensive studies of mechanisms of action of ROS, and based on the results of these studies, the search for substances with antioxidant activity and are useful primarily for the treatment of various diseases associated with free radical damage to cells and subcellular structures.

Activation of lipid peroxidation - a universal consequence of the influence on the living system of extreme variety of agents, including electromagnetic radiation - result of increased oxidative metabolism of complex organic structures. With a significant increase in the content of lipid peroxidation products endogenous antioxidant system becomes unable to maintain the balance of proantioksidantov that leads to the development of oxidative stress - one of the universal mechanisms of tissue damage. Given that the rate of free radical processes also depends on the degree of antioxidant protection of the organism, we determined the level of total antioxidant activity.

Thus, in accordance to literature in animals because of lower total antioxidant activity the free radicals are enhanced in the plasma. [11, 12]. Normally the oxidative stress induced

aging erythrocytes, which is known by a lowering of the cell volume, the loss of the cell surface and decrease deformability violation asymmetric distribution of membrane phospholipids. Grown old erythrocytes derived from the blood of the reticuloendothelial system, particularly absorbed by macrophages.

It is known that at different oxidation effects on erythrocytes observed oxidation and denaturation of hemoglobin (the formation of the so-called Heinz bodies), accompanied by the release of heme / hemin (Ferriprotoporphyrin IX) [13]. It is well known that the exogenous hemin easily integrated into the membrane, destabilizing it and causing hemolysis. Induced oxidation of erythrocyte dehydration is the result of increasing the concentration of intracellular  $Ca^{2+}$  and activation of the Gardos channel [14, 15]. And there is evidence that the products of lipid peroxidation activate phospholipase that initiate the cascade of cyclo- and lipoxygenase reaction from membrane phospholipids, eicosanoids in determining the state of the cell membrane receptor system in close connection with the system of cyclic nucleids. Strengthening of lipid peroxidation in cell membranes alters the conductivity of calcium channels and calcium reduces the ability of phospholipase -binding cell membrane [16].

Thus, high intensity microwaves' causes significant violation to oxidative processes in blood. This study testify the idea of the implementation of the biological effect of non-ionizing EMR nature by free radical processes.

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