# THE EFFECT OF NANOSECOND ULTRAWIDEBAND ELECTROMAGNETIC RADIATION ON XENOGENEIC ERYTHROCYTES

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Experiments were the intention to investigate the effect of short-pulse ultrawideband electromagnetic radiation on biological objects. An insulated rod antenna excited by a high-current electron beam (E ~ 0.5 to 1.0 MeV, I ~ 4 to 10 kA, $\tau \approx 15$ ns) served as a radiation source. The objects to be irradiated, i.e., erythrocytes of both donors and diabetics, were put in the regions with field intensity varying from 100 to 1000 V/cm.

The effect of radiation on the lifetime and shape of erythrocytes on the permeability of erythrocyte membranes for the penetrating nonelectrolyte (1 M glycerin) and the state of intracorpuscular hemoglobin was investigated by the methods of small-angle light scattering, UV spectrometry and phase-contrast microscopy.

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## 1 INTRODUCTION

Recently electromagnetic situation of environment promptly varies. It is caused by appearance of new factors of environment - electromagnetic fields (EMF) of anthropogenic parentage, which exert essential influence on the man as well as on bioecosystems. The biological response can be influenced by the following EMF parameters: intensity, frequency of radiance, duration of irradiation, modulation of a signal, combination of frequencies, periodicity of activity [1]. The biological effect of EMF can be caused by thermal [2] and non-thermal mechanisms [3], resonance phenomena [4] and other processes.

One of types of pulsed EMF is ultrarwideband (**UWB**) radiation, which because of the presence of a wide frequency spectrum can render biological action at the expense of several mechanisms.

In this paper the experiments on action of power short-pulse UWB-radiation on donor blood erythrocytes are described.

## 2 METHOD OF PREPARATION OF OBJECTS FOR EXAMINATIONS

The effect of radiation on the lifetime and shape of erythrocytes, on the permeability of erythrocyte membranes for the penetrating nonelectrolyte (1M glycerin) and the state of intracorpuscular hemoglobin was investigated by the methods of small-angle light scattering, UV spectrometry and phase-contrast microscopy.

The permeability of erythrocyte membranes was estimated by kinetics of penetration into cells of glycerinum of 1 M water solution. The essence of the estimation consists in the following.

In [5] developed were the physical and mathematical models of the processes of matter transmission through cellular membranes during a hypotonic hemolysis in an aqueous solution of penetrating matter. There was obtained the algorithm of erythrocyte permeability coefficient calculated for electrically neutral matters, in particular, cryogenic protectors, by the form of the dependence of the intensity of light scattered by erythrocyte suspension (proportional to quantity of scattering particles, their volume and refractive index) on the time during hypotonic hemolysis.

To measure the intensity of the light scattered with a dilute erythrocyte suspension at an angle 9° to the direction of the incident beams we used the device designed and fabricated at the research-and-production firm "CRYOCON" together with a department of the Science and Technical Complex at the Cryobiology and Cryomedicine Institute of the National Academy of Sciences of Ukraine [6]. The device comprises: cell-chamber with a system of primary transformation of a signal, device for maintenance of temperature of the cell-chamber, system for measurement data recording. The photometric cell has a volume of 3 ml. The device contains the collimated light source (arsenide-gallium diode such as AL107B) with a spectral characteristic ranging from 950 nm up to 1100 nm and the photoreceiver (photodiode FK-25K with an integrated sensitivity of $8\text{-}10^{-3}$ mA/lux) which is fixed at an angle 9° to a direction of a light beam, which falls onto the cell suspension. The wavelength of the incident light is chosen in the infrared spectrum, far from the range in which the spectral bands of a light absorption by a hemoglobin (blue) takes place. The signal from the photoreceiver goes at the input of the current-to-frequency convertor (by the method of capacity charge compensation), in which the analogue signal is converted to normalized – by magnitude and duration – pulse sequence for its further digital processing. The input pulses from the transformer were counted by the 16-digit counter of the data. The signal, converted to the digital form, is output on the PC display as a diagram "intensity of scattered light-time".

We compared the experimentally measured intensity of the light scattered with erythrocyte suspension at an angle 9° to the direction of an incident beam with the theoretical value obtained by simulation of the process of a hypotonic hemolysis in an aqueous solution of the penetrating material. It has given the possibility to find the permeability coefficients of erythrocyte membranes for neutral materials, for example, cryoprotectors. The physical and mathematical model of hypotonic hemolysis connects, in particular, the time, at which 50% of a hemoglobin initially contained in the erythrocyte is extracted, with a permeability coefficient of the nonelectrolyte dissolved in the cell medium.

The state of an intracorpuscular hemoglobin is the important performance of a blood describing ability of erythrocytes to transfer oxygen, and also relation in the blood functionally high-grade and old cells.

The hemoglobin in erythrocytes can be in different forms. Basic of them, always presented in the blood, are: oxyhemoglobin - hemoglobin boundedowith oxygen (HbO2), deoxyhemoglobin - hemoglobin not bounded with oxygenium (Hb) and methemoglobin (Hi) - hemoglobin, not capable to bind oxygen, in which iron is in a trivalent state. The biochemical systems working in an organism, sustain equilibrium of various forms of hemoglobin.

The content of the mentioned forms of the hemoglobin in the blood was measured by the method of a differential spectrophotometry The calculation of HbO2, Hb and Hi was performed taking into account that the spectrum of the total hemoglobin is a superposition of spectra of its various forms, previously having convinced in absence in a peaks' spectrum of sulfhemoglobin and carbonhemoglobin. The absorption spectra were recorded on the spectrophotometer Pye Unicam SP 8000.

## 3 EXPERIMENTAL PART

Experiments on studying the influence of short-pulse ultrawideband electromagnetic radiation on erythrocytes of donor blood were carried out at the high-current accelerator of relativistic electrons " TEMP-A" the parameters of which could be varying in limits: I = (4÷10) kA, E = (0.5÷1.0) MV, at a current beam duration of 15 ns. UWB irradiation was realized by direct excitation of an insulated rod antenna with high-current beam. The layout of experiments is shown in Fig. 1.

The chosen values of the field intensity in the "working zone" were ≈2000, 500 and 50 V/cm (position I, II and Ш, Fig. 1, respectively).

The state of erythrocytes before and after irradiation was defined by the following parameters: cell membrane permeability for nonelectrolyte, state of intracorpuscular hemoglobin, keeping and form of cells. In the experiment the small doses of blood (5 ml) were placed in predetermined points and were exposed to action of different quantity of EMF pulses (1, 5, 10) with an interval between pulses 90 -100 sec at a temperature 16÷20°C. The examination of erythrocyte state was carried out in 3-5 hours after irradiation.

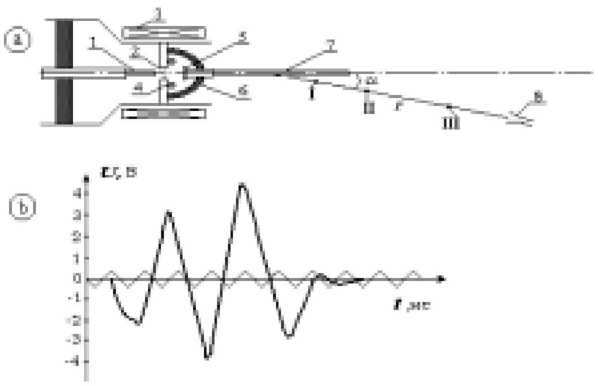

*Fig. 1. a) high- current accelerator: 1 - cathode; 2 - anode; 3 - magnetic field; 4 - Rogovsky coil; 5 - collector; 6-insulator; 7-irradiation antenna; 8 - receiving antenna;* ***I, II, III****–places of blood samples b) form of a radiated* ***UWB****-signal.*

## 4 DISCUSSION OF RESULTS

The data of phase-contrast microscopy have shown that erythrocytes after action of pulsed EMF with different intensity were weakly distinct from the control sample. In this study the definite tendency in changing the permeability coefficient of erythrocytes under irradiation applied is not revealed. In Fig. 2 the dependence of a permeability coefficient on the number of pulses of EMF with different intensity for one donor is presented.

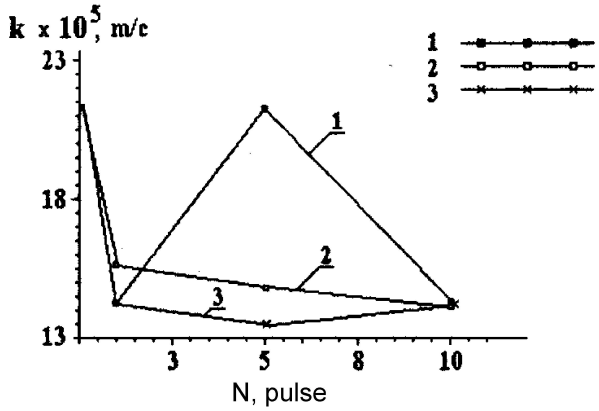


*Fig. 2. Erythrocyte permeability coefficient versus number of pulses at different field intensity: I - 2000 V/cm; 2 - 500 V/cm; 3 - 50 V/cm.*

In Fig. 3 (a, b, c) the similar dependencies for three other donors are shown, where the curves 1 and 2 concern to integral plasma and plasma washed from a blood, respectively. In Fig. 3d the erythrocyte permeability coefficient as a function of the number of pulses with an intensity ~ 2000 V/cm for two patients suffering from sugar diabetes is presented. The results obtained are determined, in all probability, both by the individual character of a permeability of different donors, and by conditions under which the cells were irradiated, for example, the composition of medium.

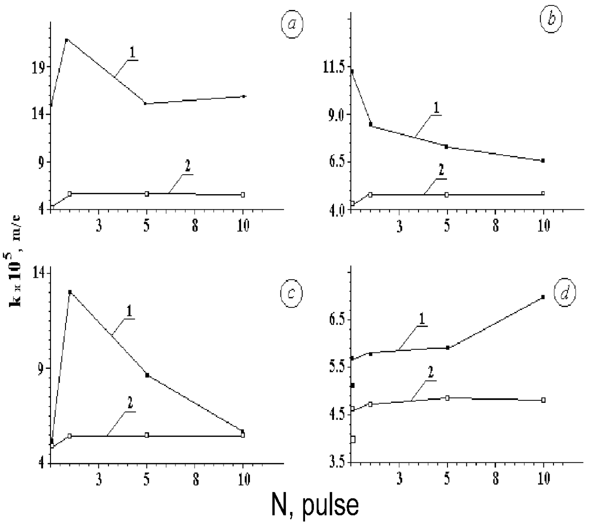


*Fig. 3. Coefficient of erythrocyte permeability for different donors at a field intensity of 2000 V/cm: a, b, c- three different donors (1 - integral blood, 2 - washed), d —for two patients suffering from sugar diabetes.*

In Fig. 4 given are the data on pulsed EMF influence on the forms of hemoglobin (solid lines) which testify that the content oxy- and deoxyhemoglobin after irradiation practically do not vary, and the content of a methemoglobin tends to drop. Such a result most likely is caused not by influence on metabolic processes in erythrocytes, but by destruction of some part of old cells distinguishing, as is known, by the increased content of methemoglobin. As one more test for a state of erythrocyte membranes the potassium ferrocyanide influence on cells was used. In norm this low-molecular substance does not penetrate in erythrocytes. However, in the case of some influences which do not lead to hemolysis but break the membrane permeability, ferrocyanide penetrates into cells and oxidizes the hemoglobin iron up to 3 valence, forming methemoglobin. However, the investigations we have performed indicate (Fig. 4, dotted curves) that the influence exerted by such fields does not lead to similar disturbance of a permeability.

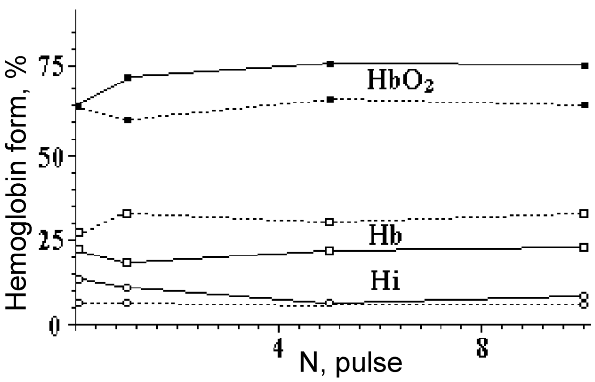


*Fig. 4. Hemoglobin form versus quantity of pulses: solid line — hemoglobin, dotted — the methemoglobin.*

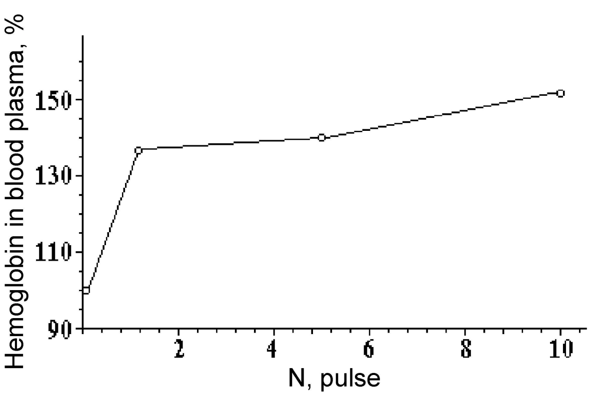


*Fig. 5. Hemoglobin content versus quantity of* ***UWB*** *pulses.*

The evaluation of the hemoglobin content in blood plasma shows that the hemoglobin content in plasma after irradiating is increasing (Fig. 5).

## 5 CONCLUSION

As a result of our studies on UWB short-pulse influence on erythrocytes of donor blood it has been revealed the following: the coefficient of erythrocyte permeability, apparently, is determined by the individual characteristics of donors; the hemoglobin content in plasma is increasing; the biological effect reveals at fields higher than in the case of narrow-band microwave radiation of microsecond duration. The authors are grateful to V.Burlova for helping in the preparation of this paper.

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