Low-intensity EMR effects
Effectiveness of experimental electromagnetic therapy
Specific broadband-spectrum stochastic EMR: influence on cardiomyocytes and neurons
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Dear Reader!

Our journal is 5 years old! Let’s make the first summary of our history!

It took us a relatively short time to get recognition by the world scientific community: we present in CARDIOMETRY hot topics only. Most papers herein are devoted to a new science: cardiometry. Contemporary history does not offer any precedent for a situation when a new science based on laws, axioms and logical analysis, imported from natural sciences, is established. But the mission of our journal is beyond the scope of simple popularization of fresh concepts or innovations: feedback to the scientific community is of great importance to us, since the developers of cardiometry have not been able to predict responding to their revolutionary ideas. And now, five years later, we come to a better understanding what has been generally accepted by scientists and researchers.

First we arrive to a conclusion that the existing system of education of medical staff has not kept pace with current progress in science at all. While scientific information doubles every five years, medicine seems to be governed by a rigid set of obsolete rules having been applied for many, many centuries. Some skeptical scientists may say this evaluation of the current situation is too much critical, but it is an adequate assessment thereof. So, conventional cardiology treats the heart performance isolated from the functioning of the organism as a whole, not to mention the multitude of interrelations between the heart performance and hemodynamics! Why so? Looking more closely into hemodynamics, we can state: an essential feature of the heart design is just to provide the proper hemodynamics that is often overlooked by conventional cardiology, and if hemodynamics is present in some theories, then it is done in a quite contradictory manner.

Similar to other organs in a human body, the performance of the heart depends on metabolic processes. Cardiology is disregarding this fact at all. The law of naturally designed mechanism of the heart muscle contraction, provided by the strictly specified sequence of activity of the Na, Ca and K ions, is interpreted by the existing concepts in the most arbitrary way, allowing for occasional ion channel sequence alterations, that absolutely contradicts to the basic principles of the heart performance.

We can cite a lot of further examples to illustrate a very complicated situation in conventional cardiology. We intentionally avoid publishing of those papers which relay on “bare statistics” under ambiguous interpretations. Our responsibility is to release only such research articles, which are prepared according to the generally accepted natural science fundamentals.

Our intention is to address a wider range of topical issues: cardiometry in sports activity to treat the healthy heart performance and doping problems in athletes; cardiometric rheology with its unique data obtained in current research; neurocardiometry as a basis for phsycoanalysis, including pioneering concepts to explain complex interconnecting mechanisms responsible for the performance both of neurons and cardiomyocytes; pediatric cardiometry to cover the proper conditioning of children; other associated themes to discover new mechanisms of the heart performance in a human.

We think it would be reasonable to prepare some on-line education courses for our public. It is not improbable that our Readers will find soon some other sections in our journal to attract more interested people therewith.

Let’s introduce the key point of the latest issue: resonance in bio-systems. Some intimate natural mechanisms governing the life performance in complex bio-systems have been discovered and described in our topical article herein. Results obtained by the originators of the discovery hold the greatest promise for finding most advanced, high-efficacy, treatment technologies. This issue contains also brief information about the last International Nobel Congress and decisions made by the Nobel dynasty representatives. And we write again about the problem of the existence of a smooth nonstationary vortex solution of the 3D NS equation included by Clay Mathematics Institute into the list of the seven fundamental Millennium Prize problems. Last issue of our journal offered the solution of the above equation found by Russian scientist S. Chefranov. It should be emphasized that the problem has been solved by him in the course of his investigations of blood flow regime in a human body, which is known as the elevated fluidity mode: it is just the mode that is the theoretical groundwork for cardiometry.

And now let’s open the journal! Your feedback will be very much appreciated by us!

We believe our readership is growing in synchronism with development of our journal, so that we can celebrate our birthday together: we wish health, private success and new achievements in science, business or education to everybody!

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EDITORIAL

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Mechanisms of electromagnetic influences and effects on membrane systems in neurons and cardiomyocytes

Valery Orlov, Mikhail Rudenko, Alla Shikhlyarova, Alexander Sukhov, Evgeniya Kirichenko, Svetlana Filippova, Vladimir Zernov, Dmitry Makedonsky, Konstantin Mamberger, Sergey Rudenko

The aim of our studies was to discover responses of the membrane systems in neurons and cardiomyocytes as well as mechanisms of influences and effects produced by the broadband-spectrum stochastic electromagnetic radiation (BBSS EMR) on them according to data on membrane potential (MP) levels and action potential (AP) parameters.

The state of anti-oxidation system in rats under chemically induced cancerogenesis and influence by complexly modulated ultra-low frequency magnetic field (ULF MF)

Oleg I. Kit, Alla I. Shikhlyarova, Elena M. Frantsiyants, Natalia D. Cheryarina, Lyudmila D. Tkalya

At present, control of properties of biological membranes as targets of magnetic field applications within a living cell is the subject of considerable discussion in the scientific community. Experimental studies carried out by us demonstrate that ULF MF has an access to the most important mechanisms of tumor growth. The most likely mechanism among them is that responsible for the processes of free-radicals oxidation.

The pattern of the immune processes during activation therapy with the use of microwave electromagnetic radiation

Galina V. Zhukova, Alla I. Shikhlyarova, Tatiana A. Barteneva, Marina I. Bragina, Tatiana N. Gudtskova, Vladimir M. Petrosyan, Elena A. Shirnina, Elena Yu. Zlatnik, Elena P. Korobeinikova

The mechanisms of anti-tumor action of low intensity factors of activation therapy remain little investigated. The aim of the present paper is to study the changes in the immune system organs, the state of blood leucocytes and tumor area under effective activation therapy in tumor-bearing animals using EMR microwave range. The research is relevant due to the immune processes influence on cardiovascular system performance.

Morphologic correlates of the multimodal electromagnetic exposures efficiency (as exemplified by the experiment tumor growth)

Tatiana A. Barteneva, Alla I. Shikhlyarova, Galina Zhukova, Tatiana P. Protasova, Marina I. Bragina, Elena A. Shirnina

The aim of the present paper is to study the morpho-functional changes in the tumor tissue and peripheral zone in experimental animals under the influence of extremely high-frequency electromagnetic radiation (EHF EMR). Combined effect of EHF EMR and ultra-low-frequency magnetic field (ULF MF) without the use of special anti-tumor agents is to be studied as well. The experiments have been performed on 56 outbred male rats with inoculated sarcoma 45.

Pathogenetic substantiation for anti-tumor effectiveness of experimental electromagnetic therapy

Oleg I. Kit, Ekaterina F. Komarova, Alla I. Shikhlyarova, Elena P. Korobeinikova, Ludmila V. Vanzha, Viktoria V. Pozdnyakova, Andrey V. Dashkov, Elena M. Frantsiyants

The objectives to be pursued by our studies are to assess some effects made by experimental actions of ultra-low frequencies electromagnetic field (ULF EMF) in combination with the SCENAR therapy on morphological changes in lung tissues and the regulation structures considering tumor growth dynamics.

Use of physical factors of electromagnetic nature for decreasing complications in respiratory and cardiovascular systems in patients after surgical treatment of lung cancer

Oleg I. Kit, Alla I. Shikhlyarova, Igor N. Turkin, Tamara G. Aiparetova, Sergey P. Pyltsin, Yury N. Lazutin, Anna V. Chubaryan, Igor A. Leiman

Surgical treatment of lung cancer is associated with a high risk of postoperative complications in cardiovascular and respiratory systems. Studied is the ability to prevent the postoperative complications in respiratory and cardiovascular system when exposed to ultra low-frequency magnetic field in the given category of patients.
Influence of postoperative magneto therapy on homeostasis central regulation efficiency


The topical issue is the study of mechanisms providing a stable performance of the brain as the highest section of nervous system which largely predetermine the oncology patients life quality.

The aim of the present research is to study the dynamics of bioelectric markers of the CNS functional state in lung cancer patients during an early postoperative period for the purpose of evaluation of the conducted magneto-therapy adaptive and correcting effects.

Neurophysiological status and adaptation responses upon application of electromagnetic fields in complex treatment of patients with malignant gliomas of the brain


The aim of this paper is to discover some advantages and benefits of the complex treatment of patients with malignant gliomas of the brain with the use of electromagnetic field therapy.

Our examination focused on patients suffering from malignant gliomas of the brain. One cohort of them received surgery and chemoradiotherapy (CHRT) without electromagnetic field exposure of the brain, while the other group was subjected to the same therapy, but accompanied by the electromagnetic field treatment of the brain.

Xenon effect on electrophysiological markers in oncology patients with post-castration syndrome during early post-operative period: pilot studies


Breast cancer (BC) takes a leading position among all malignant neoplasms in women. One of the critical aspects of the radical treatment of BC is the development of post-castration syndrome (PCS) in patients of reproductive age.
Professor Alla I. Shikhlyarova, Dr.Sci.Biol., Head of Experimental Research Lab at the Rostov-on-Don Oncology Research Institute, Ministry of Healthcare of the Russian Federation, Rostov-on-Don, Russia. She is a co-developer of the internationally recognized theory of activation therapy.
Visible effects of an invisible factor: semi-centennial experience in research on electromagnetic field effects produced on organism

Some introductory remarks on the current issue topics
Alla I. Shikhlyarova

Plenty of medical and biological problems requiring novel solutions in cardiology are inseparably linked to the core laws and regularities found in response patterns of an organism treated as an open, integrated, complexly organized system, influenced by factors at the global ecosystem level. It is fantastically difficult to imagine all the hierarchy of the myriad interactions in a human organism in order to identify or at least localize the control panel responsible for every control function and the entire performance of the organism, including all its subsystems, as well as to provide an adequate assessment of actual capabilities, options and possibilities required for the proper bio-adaptive regulation and the rise in the organism resistance to steadily growing pathogenic effects, actions and influences.

However, the third millennium has inherited a rich unique experience in the proper understanding the laws of the existence of bio-systems, which are exposed to natural factors like sunlight, water, air and electromagnetic field environment, treating the bio-systems in the context of their evolution on the Earth. As to electromagnetic fields (EMF), or electromagnetic radiation (EMR), we will try to catch some fine threads to bind together the EMR actions and the operation of the molecular, cellular, systemic and integral mechanisms in a living organism that may have the promise of finding some pioneering natural ways for health improvement and longevity. An interdisciplinary friendship between magnetobiology and oncology, as one of the critical disciplines in medicine, which needs development of new fundamental scientific approaches to enhance anti-tumor safety, elevate the resistance to a malignant process and reduce the damaging side effect of aggressive anti-cancer therapy, may serve as an exemplary case to illustrate our scientific philosophy.

Adhering to the philosophy of inter- or multi-disciplinary research, a pioneering approach is offered by us on the basis of an outstanding discovery of the law of development of qualitatively differing, general non-specific adaptation reactions of the organism (L.Kh.Garkavi, M.A. Ukolova, E.B. Kvakina, Registered Scientific Discovery No. 158, 1975). Referring to the research of Hans Selye, who discovered the reaction of stress to the effects of excessive force, the Russian scientists have succeeded in developing an original concept of quantitative and qualitative response patterns, provided by a human organism, and creating a periodic system of integral adaptation reactions of the anti-stress nature, which differ in quantity and quality, to various actions and exposures. Interactions between magnetic field, electromagnetic radiation and the human organism demonstrate that all the phenomena obey this law, and this conception can be effectively utilized as a beneficial factor in the cancer therapy.

The present issue of our journal CARDIOMETRY has collected only a small part of the experimental and clinical materials representing one of the priority areas of scientific investigations undertaken by Rostov Research Institute of Oncology (RRIO, Rostov-on-Don, Russia). 55 years have passed since the time of publication of the first in the USSR article on the anti-tumor effect produced by an action of magnetic field (MF) on growth of sarcoma 45, which was discovered by Professor Maria A. Ukolova in 1960, up to the experimental and clinical developments in the recent years. This time span is considered to be rather short, considering science progress in general, but it has given us findings and results showing considerable promise. The idea of the existence of a periodic system of the adaptation reactions in a human organism (the regular periodic occurrence of like reactions of different types at different reactivity levels) made it possible to develop and apply in practice the principles, regimes and algorithms for dosing of some sorts of the electromagnetic field exposures. This fruitful soil has always been the focus of attention of the RRIO management, and the research work focusing on this concept has been carefully supported by them. In this connection, it should be mentioned that at the present time, Professor Oleg Kit, General Director of FSBI RRIO, Corresponding Member of the Russian Academy of Sciences, makes great efforts to upgrade test equipment and train personnel of the unique profile R & D lab at RRIO, responsible for MF anti-tumor effect investigations as well as promote the unique theory of
adaptation reactions and the priorities, given by the Rostov school of tumor magnetotherapy, both in Russia and abroad.

According to the range of electromagnetic oscillations, falling into the areas of our studies on responses of the living systems, we separate the following exclusive trends in our research work: they are effects made by magnetic field radiation and those produced by millimeter waves. At the same time, the respective reactions of bio-systems, from viruses to the biosphere, thanks to their own oscillatory properties of the endogenous origin, show many, fundamentally similar, types of their response to exogenous physical actions of the electromagnetic nature. Special attention should be drawn to the similarity of a wide range of rhythms of the external environment to the auto-oscillation processes found in the living systems.

In the organism, there are some regions of resonance frequencies in the UV, radio and low frequency range available, beginning with the protein synthesis process, enzyme-substrate relations, conjugation of chromosomes, i.e. from the smallest bio-vibrators at the nano-level, and ending with the frequencies of the brain and the heart performance as well as the circadian rhythm patterns. And if we recognize the fact that the auto-oscillations are one of the fundamental properties of living matter, then it follows that a possibility of information exchange & control realized on the basis of resonance interactions, considering the fact that the organism is an open complex oscillation self-regulating system, is not without strong appeal.

Feasibility of applications of magnetic fields (MF) for the therapy purposes has been proven since time immemorial, but we should note that in this case practice has always outpaced the theory, and it can be exemplified by the so-called diseases of modern life, namely, the cardiovascular and oncology diseases. The fundamental concepts of biorhythmogenesis (auto-oscillation activity), field interactions and the role of frequencies as a sort of communication language and a key parameter to establish the code and information links to the environment, may seem to be a good distance in the future.

If we address an exemplary case from practice, illustrating blood circulation in an organism exposed to constant MF, that has been the subject of interest for talented neurosurgeon R.P. Kikut, who has investigated the treatment of vascular aneurysms, it is possible to show that, when an electromotive force is induced in the region of a positive potential, some conditions facilitating thrombosis formation are established therein, and at the same time, in the negative potential region found are conditions promoting thrombolysis, respectively. In an intact blood vessel, there is no thrombus formation available at all, even under exposure of MF of 200–300 mT that leads us to a conclusion that turbulence has played a part in the blood flow in an aneurysm during the thrombus formation.

It took G.M. Poyedintsev and O.K. Voronova a very long time to make a discovery and formulate their full mathematical description of a complete set of revolution laws of hemodynamics related to the specificity of the natural blood flow structure. The scientists have established that blood circulates through the blood vessels in a specific mode of elevated fluidity. This so-called “third mode” of fluid flow, as opposed to the laminar and turbulent ones, has low friction due to the specific blood cell pattern of circumferential-layer type, formed by alternate circumferential layers of blood cells and plasma. In addition, detected has been the vortex-type and pulsating motion of aquatic systems in an organism, under which the aqua-clathratation of non-polar ions, molecules and active forms of oxygen occurs, creating a negative reduction potential, as shown by V.I. Slesarev.

Apparently, the presence of charged cellular elements in the multi-layered structure and the pulsatile motion in blood circulation is responsible for the appearance of a stochastically organized oscillation activity in the ultra-low frequency range in a pathogenic region (like aneurysms, or a cluster of proliferating malignant cells). Such mechanism of the rhythmogenesis is capable of collaboratively fine tuning to external electromagnetic radiation, i.e. it is possible to make a biological process controllable (formation of a thrombus or its destruction, death or proliferation of tumor cells).

Thus, our approach to oncological aspects of the use of electromagnetic fields (EMF) is based on the fundamentals of the organism adaptation reaction theory. We believe that by virtue of the non-specific complex nature of development of the anti-stressor type adaptive reactions, any regulatory or acceptor system will be collaboratively fine-tuned and timed in that region of the biological rhythms, which are adequate to the involved system. This universal capability of the bio-systems of any level to provide wave interactions opens unlimited possibilities for researches on some
mechanisms, which are responsible for control of the resistance to formation of malignant neoplasms and which are capable of optimizing the quality of life for cancer patients.

First of all, let us pay attention to high sensitivity to EMF exhibited by the CNS structures, in particular, the brain as the highest-level integrative system, designed to control the organism performance. An EMF exposure, addressing the brain as a generator of its own oscillatory activity, induces generation of certain patterns of some integral reactions, the nature of which may significantly influence further events related to malignant transformations.

In order to obtain a deeper understanding of the specific features of the brain rhythmogenesis and trace a contribution of EMF exposure to development of the generalized response by the human organism, it would be reasonable to address a fresh analysis of the CNS living cell model as an exemplary case, which covers gentle responses by the cell membrane able to perceive the wave-nature factors. The paper presented by Valery Orlov et al. in the current issue is distinguished by some exclusive lines of attack: firstly, it refers to specific conditions of preservation and prolonged (up to 3 days) maintenance of the normal life performance of neurons in the isolated central nervous system after a microsurgical preparation in the Roman snail Helix pomatia. Secondly, another exclusive line of the study is that superfine nanometer-dimensioned microelectrodes of special design have been used in the experiments, which are absolutely unique in their application capabilities, for continuous recording of intracellular potentials of a neuron. Thirdly, the experimental studies were carried out by utilizing EMR with a rhythmic wide-range spectrum of stochastically radiated frequencies from 100 Hz to 1 GHz at a power of only 0.1 µW as an acting factor. It is important that this EMR was delivered in rhythms close to the endogenous frequencies of the brain, and it led to changes in the amplitude, the duration of the action potential and the membrane potential of a regulatory response, following the pattern typical of the known adaptation reaction of training. At the same time, in view of the common electrogenic nature, the similarity of electrophysiological and ionic mechanisms operating in neurons and cardiomyocytes is quite obvious, that has made possible to refine a better explanation of the nature of the neuronal cell auto-regulation of the cardiovascular system phase activity.

Let us note that, actually, the effects made by EMR on the CNS neurons, analyzed in the offered experimental study, have been realized directly. However, here another point arises: how evident are mediated effects thereof? Is the influence of ultra-low frequency mag-
netic fields (ULF MF) accessible to the mechanisms of the regulation of free radical oxidation processes, the immune homeostasis and, what is most intriguing, to the mechanisms of tumor growth? It is precisely these problems that are discussed in the research works, in which the central (on a head) ULF MF action was utilized in animals with tumors of extra-cerebral localization upon transplantation of tumor cells under the femur skin, into the lung, as well as the subcutaneous injection of a carcinogen.

Nowadays, the issue of changes in properties of the bio-membranes, as the principle point of applications of the magnetic field effects on a living cell, is widely discussed. The most sensitive membrane indicators are products of free radical oxidation associated with energy metabolism. An investigation into the ULF MF effects on the antioxidant system parameters in plasma and blood elements in rats during chemical carcinogenesis is treated by a paper by Elena Frantsiyants et al. presented further herein. It should be emphasized that the process of carcinogenesis is a long period, which may take many months from the time of introduction of a carcinogen through the appearance of a visually distinct tumor, its further progression up to the possible lethal outcome of a tumor-bearing animal.

In this respect, the latency period of tumor growth till the time, when the tumor manifests itself, is of great importance, since it is in this time that a serious reformatting in the organism milieu takes place. It is because in this period of time a program of further tumor development is embedded in the program of the organism performance that is decisive for subsequent events, which might occur in the affected organism. Therefore, it is reasonable to use just this period to influence the key links in the framework of the organism protection and identify the respective “bifurcation points of decision-making” for the organism performance, and, in particular, for the anti-oxidation system state. Such “bifurcation points” in carcinogenesis have been found to be week 1, week 6 and week 9. It has been our success to reveal strong biochemical arguments to confirm the pronounced protective effect made by the ULF MF central action. A statistically significant prolongation of the latency period in carcinogenesis correlates with the restoration of the level of the anti-oxidation system performance in blood in animals-tumor bearers. It has been established that just the immune-competent cells represent an important link in resetting of the peroxide homeostasis that has been manifested in an elevation of the activity of the enzyme-antioxidants cascade (catalase, SOD, TPA) and an accumulation of non-enzymatic antioxidants (vitamin A) in blood lymphocytes.

The nature of the immune processes under anti-tumor effects of electromagnetic influences was investigated by Galina Zhukova and her colleagues. With the use of low intensity EHF EMF under poly-frequency modulation in experiments carried out in albino rats with inoculated tumors of sarcoma 45 and Pliss lymphosarcoma, pronounced favorable changes in various parts of the animal immune system have been noted. Particularly highlighted should be such factors of the produced anti-tumor effect as an identification and presentation of tumor antigens, an induction of differentiation and proliferation of helper and cytotoxic lymphocytic clones, elimination of tumor cells by apoptosis, activation of the cytotoxic effect by NK cells, macrophages and other cells of the immune system. It is the opinion of the scientists that these mechanisms of the tumor-specific immunity are contributors under the effect of frequency-modulated EHF EMF to regression of the tumors of particularly large size, timing and a deferred onset of the tumor resorption effect without signs of deep intoxication in rodents. The factual results have been found to be in correspondence with the most successful variants of immunotherapy of different malignant tumors.

The logical extension of the topics covering the intimate molecular and cellular mechanisms of the electromagnetic field effects has been supported by new data obtained by Tatiana Barteneva and her co-authors. She applied statistical approaches to the study of histological structures of inoculated tumors and their peritumoral zone under the influence of experimental magnetotherapy. Demonstrated has been a quantitatively significant increase in the numbers of lymphocytes, plasmatic cells, macrophages and tissue basophils in the tumor tissue and the perifocal area that has been especially evident under combined application of EHF EMF and ULF MF. The behavior of cell systems in the immediate surroundings of the tumor, where the mobilization of local immune mechanisms apparently takes place, attracts particular interest of many scientists.

Indeed, the attention of researchers can be concentrated on reactions produced by cellular systems located around the tumor. At the same time, it may be the case that the EMR action is focused not imme-
An adequate selection of the intensity and especially the frequency of exposure can greatly contribute to optimization of the organism state at every stage of the anti-tumor treatment under the clinical conditions.

According to data presented in an article by Ekaterina Komarova et al. herein, it is precisely the ULF MF central action that is capable of organizing certain neuro-hormonal changes in the lung, which may alter the behavior of foreign invading tumor cells. The scenario of the events in the lung tissue from the initial formation of duct-like structures and large conglomerates of the inoculated tumor to the stage of activation of the lymphocytic-macrophagal subsystem, histiocytes and fibroblasts, which suppress proliferative activity of tumor cells and induce tumor cell destruction and lysis, has been illustrated by the above researchers in their strictly defined step-by-step description of the processes. Over the course of the morphological resettings in the rat lung, the hormonal status of the lung and the pituitary gland tissues have been studied, that has provided the proper interpretation of the role of central and local mechanisms of the “hormonal” environment regulation responsible for production of an anti-tumor effect. It has been found that the levels of prolactin and progesterone concentration play a special role in the realization of tumor regression, and the above levels, possibly, determine the transition of some tumor cells from proliferation into differentiation.

Thus, on the basis of fundamental research, we are led to an interpretation of the signal information role of electromagnetic influences aimed at the trigger mechanisms of the regulation of the internal environment and targeted at an increase in the anti-stressor potential and the non-specific resistance in the organism. An adequate selection of the intensity and especially the frequency of exposure can greatly contribute to optimization of the organism state at every stage of the anti-tumor treatment under the clinical conditions.

The experience, demonstrated in the articles presenting the ULF MF applications at the stages of the specialized anti-tumor treatment of cancer having different localization, opens us wide possibilities of the accompanying activation magnetotherapy. In the research work presented by Tamara Airapetova and co-authors, an analysis of direct results of lung cancer surgical treatment supported by the central ULF MF exposure in the modes of activation therapy within the early post-operative period has been completed. Noted should be a considerable decrease in post-operative complications and mortality. It has been a success to avoid the development of some severe stressor manifestations in the respiratory and cardiovascular systems, like pulmonary thromboembolism, acute myocardial infarction and stroke. The researchers’ analysis of electrophysiological, hematological, immune, biochemical and hormonal markers has shown that the effectiveness of rehabilitation and improving the quality of patient life are associated with the adaptive response mechanisms and an increase in their non-specific anti-tumor resistance.

The foundation for such changes in the organism is laid first and foremost at the level of the central nervous system. Returning to the assumption that the CNS undertakes the control, trigger function in the formation of the adaptation reactions and the realization of the magnetotherapeutic effects, it is appropriate to supply actual data on the CNS condition in the same cohort of the lung cancer patients. The work presented by Tatiana Protasova and her co-authors shows a significant improvement in the values of spatial synchronization in different frequency bands in the areas of the right and left hemispheres as well as the central cortex. A correlation has been established between an increase in the compensatory-restorative processes of bioelectrical activity and the normalization of the galvanic skin resistance in both pairs of the reference measuring points of the hypothalamus. These studies allow us to present a complete picture of how the higher vegetative regulation is optimized under the effects produced by weak electromagnetic signals adequate to the natural rhythms generated by the brain.

The effects of the accompanying ULF MF therapy can best be appreciated from the results obtained in patients with malignant cerebral gliomas. In the studies on the performance status conducted by Dmitry Atmachidi and his co-authors, the following results are present-
ed: firstly, the total length of survival life and a recurrence-free two-year survival rate demonstrate a significantly reliable increase. Secondly, the pronounced positive dynamics of the performance status, a reduction in neurological toxicity with respect to cerebral symptoms, motor-, speech- and sensitivity-related disorders as well as abnormalities in the higher nervous activity are recorded. All the observed effects have been obtained under the conditions of development of adaptation reactions of the anti-stressor type, which have been much less frequent found in patients received chemoradiotherapy without accompanied magnetotherapy.

There is one more important peculiarity of the electromagnetic field effects available: it is a change induced in tumor cell membrane permeability. However, as opposed to the ULF MF exposure applied to the brain with an algorithm of frequencies close to the endogenous rhythms in the brain, in order to increase the concentration of the chemotherapeutic agent in the tumor, another frequency approach is needed. A new electromagnetic exposure technique in the mode of scanning frequencies within the range of 1–100 Hz in combination with a constant MF application has been tested in the experiment on tumors in animals with monitoring of luminescent measurements of the level of cytostatics in tumor cells. The obtained data has shown an increase in accumulation of medication substances by 2–7 times!

Thus, it becomes clear that the mechanisms of achieving the desired therapeutic effect of an electromagnetic field action cover an intricate multi-level system organization from its subcellular level up to the integrative one. Finally, a complex response to elevate the non-specific resistance is provided. It is precisely the non-specific type of the events in cells, tissues, organs and systems that is responsible for health or pathology. This can be seen by getting acquainted with a thrilling research work performed by Julia Arapova, Natalia Popova and their co-authors, where an analysis of the cardiovascular system performance, the brain bioelectrical activity and the psychological state under the influence of xenon as a factor of other nature is given. At the same time, pathological changes in the organism due to progression of breast cancer and the subsequent mastectomy are aggravated by development of an early post-castration syndrome after ovariectomy in reproductive females aged under 45. The studies conducted with the use of the most advanced medical instrumentation, including the unique device CARDIOCODE (designed & developed by Prof. M.Yu. Rudenko and his R & D team, Russian New University, Taganrog, Russia) have made it possible to detect some marked effects of the cardioprotective action of xenon in the above cohort of the patients, including normalization of their myocardial metabolic and hemodynamic parameters, an increase in power of the EEG alpha activity and harmonization of their psycho-emotional state. The realization of the non-specific anti-stress effect of xenon is evident, since in this case all the governing regulatory mechanisms of the regulation, self-organization and recovery are involved.

This approach can be treated as applicable to every area of medicine, especially in the current situation, when the clear-cut trend of personalized health care is prevailing. The offered combination of various factors of the specific and non-specific action will apparently be able to involve all the existing organism mechanisms in the most efficient way and ensure the effectiveness of therapy of various diseases, especially in children.

As to the non-specific effects produced by electromagnetic factors, one more convincing example of the effectiveness of the selection of an effect combining the wave specifics and non-specifics should be mentioned herein. In a report presented by Elena Sheiko and her co-authors, electromagnetic radiation in the red region of the optical spectrum is described. The treatment of complicated forms of hemangiomas in children from their birth to 1 year is discussed therein. With the use of instrumental and morphological methods of scientific investigations, changes in the volume and the structure of the tumor at stages of its regression have been traced. At the same time, some structural markers of sclerosis of hemangioma in the blood serum, which have an important prognostic significance, have been identified. The high clinical effect has been produced by a targeted red light radiation induction of the processes of vascular sclerosis in a hemangioma, activation of cellular elements in connective tissue, granulation and epithelization of the wound. These local specific effects have been found within the framework of the general non-specific anti-stressor reactions, and this sort of synergy has contributed to acceleration in recovery and improvement in the quality of life of infants.

When presenting to you these outlines of our experimental and clinical developments concerning the
influence and effects of electromagnetic fields, first it is necessary to note that the old problem of oncology, as well as cardiology and other areas of medicine, that refers to the relations constructed between the tumor (or other pathological process) and the organism, may be solved from the standpoint of the concept of the oscillation processes. Following this way, we can see that it is really amazing to observe that the regulation centers become highly sensitive to electromagnetic signals, which, actually, penetrate and fill our habitat, when the collaborative adjustment, tuning and timing are realized at the level of the very fine oscillatory interactions. The brain and the heart, all the cellular and molecular structures up to and including elementary particles have the properties of auto-rhythmicity and oscillatory activity, as an essential feature of the unity of matter and field structures. The offered studies represent research works which have been completed at the interface between sciences and should be regarded as the multi-disciplinary ones, and, therefore, all the articles presented in this issue of our e-journal CARDIOMETRY are of great consequence not only in terms of increasing the effectiveness of treatment. They primarily deal with designing a new paradigm both in biology and medicine that refers to finding a possibility to control the organism state on the basis of the laws of the adaptation responses and an increase in the non-specific and the anti-tumor resistance. The fundamental scientific developments of the mechanisms, principles, algorithms and evaluation criteria for activation magnetotherapy, discovered by the Rostov school of researchers, offer wide opportunities and prospects for an introduction of these technologies in the health care practice, improvement in the life quality and longevity in general.
Generalization of the Millennium Prize problem to the Navier-Stokes (NS) equation for compressible medium

S. Chefranov, Member of Editorial Board

The foundation for all CFD design and calculations of parameters to describe motion for air- and spacecrafts of every type, marine surface vessels and submarines as well as other natural and engineering systems, including blood circulation in a living organism, as well as a better understanding of fundamental astrophysical and geophysical phenomena in near and far space, in the oceans and the atmosphere on the Earth and other planets, having their own atmospheres, is formed by the Navier-Stokes (NS) equation.

The Navier-Stokes (NS) equation and the continuity equations have a tensor representation as follows:

for the viscous (with the first and second coefficients of viscosity \( \eta \) and \( \zeta \), correspondingly) compressible medium, which has a distribution of fields of density \( \rho \) and pressure \( p \) in motion at a velocity of \( u_i \), \( i = 1, 2, 3 \) as given below:

\[
\frac{\partial u_i}{\partial t} + u_j \frac{\partial u_i}{\partial x_j} = \frac{\eta}{\rho} \nabla u_i - \frac{1}{\rho} \frac{\partial}{\partial x_i} \left( p - \zeta \frac{\partial u_i}{\partial x_i} \right) \Delta = \frac{\partial^2}{\partial x_j \partial x_j} \rho \frac{\partial u_i}{\partial x_j} + \frac{\partial\rho}{\partial x_j} \nabla u_i = 0
\]

The Navier-Stokes equation is known for about two hundred years owing to research papers by C.L.M.H. Navier, 1827; S.D. Poisson, 1831; B. De Saint-Venant, 1843; and G.G. Stokes, 1845. However, till now the equation system (1) remains unsolved due to the absence of a general nonstationary solution, which should remain smooth on a bounded time interval and which should explicitly depend on arbitrary smooth initial conditions.

Known is only the stationary one-dimensional solution of the equation system (1), which has been completed by the equation of state of medium to relate the density and pressure field in (1) (R.Becker, 1922; L.H. Thomas, 1944; N. E. Kochin, I.A. Kibel, N.V.Rose, 1963; J. Iannelli, 2013; B.M. Johnson, 2014).

Moreover, all the attempts to find a numerical solution of the Navier-Stokes equation even with the use of most advanced PC equipment have failed so far, so that it is still impossible to produce a reliable prediction based thereon even for a relatively short, bounded time spans (an exemplary case: weather forecasting).

In this case, the fundamental reason of the absence of predictability based on the Navier – Stokes equation has not been identified. Actually, so far, considering a three-dimensional case, the theorem on the existence of the smooth solution on a bounded time interval still requires a proof. But the latter does not exclude that on a bounded time interval the solution may lose smoothness (its derivatives become singular), so that, in doing so, it seems that it is in principle impossible to produce an accurate prediction based on the equation in question.

In this connection, the problem of the existence of a smooth solution of the Navier – Stokes equation on any arbitrarily large intervals of time was included by Clay Mathematics Institute into the list of the seven fundamental Millennium Prize Problems (MPP).

However, at the same time, in the formulation of this MPP it is proposed to reduce considerations only to such solutions of the system (1), which have the zero-divergence of the velocity field at a constant value of the medium density. It is connected not only to the fact that the full system (1) is conventionally regarded as much more complex than the mentioned approximation of incompressible medium (for motion velocities much less than the sound velocity in the medium under consideration).

Indeed, it is generally assumed that the full system (1) a priori cannot have smooth solutions on a bounded time interval due to the appearance of singularity in processes like shock and air pressure waves created by an object traveling through the medium at a speed great that the sound speed (for instance, shock waves when an airplane is breaking the sound barrier). Therefore, it is suggested that the generalization of the MPP to the full system (1) a priori has a Negative solution only. But the obtained analytical solution of the system (1) gives us the Positive solution for the MPP generalization!
S. Chefranov & A. Chefranov: the Navier – Stokes equation has been solved!
HAPPY Birthday
Open-Access e-Journal CARDIOMETRY is 5 years old!
Mechanisms of electromagnetic influences and effects on membrane systems in neurons and cardiomyocytes

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Aim
The aim of our studies was to discover responses of the membrane systems in neurons and cardiomyocytes as well as mechanisms of influences and effects produced by the broadband-spectrum stochastic electromagnetic radiation (BBSS EMR) on them according to data on membrane potential (MP) levels and action potential (AP) parameters obtained by us.

Materials and methods
Neurons from the isolated central neural system (CNS) of the snail Helix pomatia were selected to serve as a model for our experiments. We applied an electrophysiological technique implying recording of intracellular potentials of a neuron. The presented research work is of fundamental nature and reveals some intimate mechanisms of actions made by electromagnetic radiation (EMR) on the cytoplasm membrane of a neuron and its channels that is applicable to cardiomyocytes. An absolutely exclusive distinctive feature of our work is that the specific BBSS EMR used in our experiments has unique characteristics and offers broadband stochastic radiated frequencies in the range from 10^6 Hz to 10^10 Hz at an integral radiation power of only 0.1 μW. Another distinguishing feature of the applied EMR parameters is that the stochastically organized EMR is supplied under such action rhythms, which are close to the natural alpha-, theta- and delta-rhythms of an EEG as well to the natural background base resonance frequency of the Earth, which has been physically measured to be 7.83 Hz, that is evolutionary significant for biological systems.

Results and discussion
We are pioneers in the world science who succeeded in obtaining objective evidence for the effects made by the broadband-spectrum stochastic electromagnetic radiation (BBSS EMR) on intracellularly recorded electrophysiological properties of a CNS neuron. The used BBSS EMR was demonstrated to have the effects on the amplitude, the duration of an action potential and a membrane potential, following the pattern of a regulatory response to weak stimulation, similar to the reaction of training. The effects produced by BBSS EMR on the CNS neurons in the snail Helix pomatia are applicable to cardiomyocytes, since the electrophysiological and ionic mechanisms of firing an action potential and creating the required level of a membrane potential in these cells in the organism are identical; however they differ in their time parameters.

Conclusions
The regulation of the functional state of a neuron in the neuron network with the use of a non-invasive remote method of a control action has been objectively shown herein. According to our philosophy, a neuron is considered as an analogue of the formation of a transmembrane potential by a cardiomyocyte and at the same time as the basic model of the neural cellular auto-regulation of the phase-structured performance of the cardiovascular system. Adhering to this interpretation, a fine tuning instrument to control the functional state of a neuron, CNS, a cardiomyocyte or other cell systems in the organism has been discovered. It offers widest possibilities of a targeted fine-tuning regulation of the control systems in a human organism.

Keywords
Electromagnetic radiation, Cardiomyocyte, Neuron, Action potential, Membrane potential, Ion channels, Endogenous pacemaker activity

Imprint
Introduction

At present, a multitude of methods and techniques are known which are capable of providing a targeted control over the functional state and individual sub-systems in a human. They can include both contact and contact-free methods to exert an influence on the organism as follows: manual, chemical and verbal methods as well as actions produced by fields of different physical nature. Among the fields, the electromagnetic fields, which belong to the family of the contact-free control methods, are the special case. The range of their actual values stretches from extremely low frequencies (ELF) of $3 \times 10^{-3}$ Hz, which are referred to as decameter waves with a wave length from 100 Mm in accordance with their accepted international classification, up to and including gamma-rays, having a frequency of $3 \times 10^{23}$ Hz with a wave length of $3 \times 10^{-18}$ m, which are comparable to the size of an atom and which possess the highest energy within the energy range from 12.4 eV to 5000 keV. A large portion of the said range is used in medicine for the therapy and diagnostics purposes. As a rule, medical instrumentation designed for the therapy and diagnostics practice comprises generators or detectors operating within a narrow band of electromagnetic waves. Electromagnetic radiation (EMR) of different frequencies, even within a narrow band, is perceived or sensed by a human organism in various ways, and for this purpose the organism is “equipped” with the respective EMR sensors, namely, the receptors. The thermoreceptors residing in the skin are sensitive to energy in the infrared region of the EMR spectrum, and the light receptors in the eyes operate as sensors receiving the EMR visible region. The above spectrum regions are found very close to each other.

But a large portion of the EMR range cannot objectively be perceived by a human, despite the fact that the unperceivable EMR regions sometimes may produce essential effects on the organism, up to fatal outcomes. Effects of this sort are exerted by some EMR regions like gamma radiation, microwave and UHF electromagnetic radiation, when power flux densities reach extremely high values. In contrast thereto, the absence of EMR, as it is the case in experiments modeling the disappearance of the Earth’s natural magnetic field, is another matter. The peculiarities of the influence exerted by magnetic field on the human organism show fundamental differences from any other action provided by chemical exposure, heat, radiological impact or electricity. For example, musculature and circulation circuit in the human body are capable of partially shunting hazardous currents, or, to take one more example: ionizing radiation can be absorbed to some extent by the body surface, while magnetic field can freely penetrate the entire living organism. The Earth’s magnetic field patterns act in the ultra-low frequency range, and therefore they meet the basic physiological rhythms in a human organism: the heart and brain rhythmic patterns as well as the respiration rhythm. As opposed to other physical actions and factors, a human individual is not able to sense magnetic fields, but however the human organism always responds to a magnetic field exposure by initiating primarily certain functional changes in the performance of the nervous and cardiovascular system as well as the brain activity. And it is Dr. Kyoichi Nakagawa’s opinion that magnetic field deficiency syndrome occurs due to a decrease in the Earth’s magnetic field so that it is responsible for appearance of many abnormalities like sleep disorders, appetite abnormalities, decreased immunity performance, increased susceptibility to diseases, including joint- and skin diseases, urogenital disorders, nervousness and generalized weakness. The concept offered by K. Nakagawa was given the name “magnetic field deficiency syndrome”. Actually, a magnetic field deficiency can be created by artificial means: an exemplary illustration is a space flight; another example of the said conditions is a submarine, where magnetic field shielding effects are used. In individuals, who have experienced a long-term magnetic field deficiency, observed are considerable deviations of the functional parameters from their normal levels, metabolic deceleration, a decrease in the total number of leukocytes; prenosological and premorbid states are found in them, too. Every human organism has its own integrative electromagnetic field surrounding the body, which interacts with external electromagnetic fields in the environment. The modern knowledge and expertise in the area of electromagnetic fields and radiation allow stating that one of the most advanced fundamental line in research and development of medical instrumentation, theoretical & practical biophysics and bioinformatics is an investigation of the life performance and the associated information processes in a human organism, so that, following this way, research on endogenous electromagnetic fields (EMF) of biological entities and their interplays should be considered as a highest priority task [1].
Recent scientific explorations have reinforced the conceptual idea that information exchange among separate units within the nervous system, i.e. among the neurons, in an organism is provided not only by chemical and electrical signals, but also via electromagnetic fields generated by the neurons, their dendrites and axons, through which the excitation current flows.

When designing most advanced medical equipment for therapy & diagnostics, it is of great importance to properly assess the patient’s own integrative electromagnetic field along with generators and conductors creating EMF, responses and properties of the systems responsible for the regulation in the organism: principally it should be applied to the constituents of the nervous system, the neurons and their networks as well as some other cell structures, for instance, cardiomyocytes, which reside in the heart; and such assessment should be completed by an estimation of their interactions with the physiotherapeutic EMR sources applied.

As to the two above cell populations, i.e. the neurons and the cardiomyocytes, which represent one of the most important regulatory operation systems in the organism, they share a common property: it is their pacemaking activity. Pacemaker (origin: pace as a rate of activity, tempo + maker) is a natural heart beat controller governing the proper heart rhythm. It might be a single cell or a group of the cells, which are capable of generating rhythmic excitation pulses propagating through the neighboring cells. It should be noticed that the pacemaking activity of the neurons is found occasionally, whereas pacemaking of the cardiomyocytes is their permanent mode of operation designed to provide the normal performance of the organism throughout its entire biological life span. Variations in frequencies and shapes of potentials in the pacemaking activity are interconnected with the respective changes in the functional state of the organism in general. Abnormalities in the cardiomyocyte pacemaking rhythmicity result in pathological outcomes in the organism. Migration of the supraventricular pacemaker, as an example, takes place, when the natural cardiac pacemaker locus shifts in a gradual manner between the sinoatrial node and the atrioventricular node. The shifting of the pacemaker can be detected on an ECG by tracing the same ECG lead record with assessing changes in the P-wave configurations, amplitudes and polarity as well as alterations in the P-P and P-Q time intervals.

Based on the above, actually, development of a contact-free physiotherapy instrumentation designed for generation of superweak EMR in a wide band of frequencies, which are objectively not sensed by a human individual, but which are adequate to the endogenous resonance frequencies produced by the cells, organs and systems in the human organism and which are capable of normalizing their performance, appear to have considerable promise [2].

Materials and methods

Bioelectrical activity of the neurons in mollusks and warm-blooded animals is mainly provided by the activity of sodium, calcium and potassium ion channels, which are built and operate according to the same general principles [3–8]. It enable us to use the neurons of mollusks as eligible models [9–18], in parallel with the neurons of warm-blooded animals, for experimental research on the performance of the ion channels and the mechanisms of influences on them made by various physical factors, chemical substances and pharmaceutical drugs either of already known nature or being designed.

Our studies were carried out on the large identified (100–200 µm) neurons of the parietal ganglia (LPaG and RPaG) (Fig. 1) and the adjacent non-identified neurons in the subesophageal ganglion complex of the isolated CNS in the mollusk Helix pomatia. The neurons in the ganglia of this mollusk kind have a pigmented edge in the region of the axon hollock and are clear-
ly visible under a binocular magnifying glass. Prior
to the studies, we had succeeded in developing our
own original technique of preparation of the required
Helix pomatia test samples [19]. For the studies de-
scribed herein we used only a snail organism section
comprising the CNS. From the mollusk body we iso-
lated the circumoesophageal ganglion complex and
fixed the latter onto a silicon slide with brackets inside
a recording non-magnetic chamber designed by us for
the experimental purposes. The recording chamber
had a volume of 0.5 cm$^3$ and contained physiological
saline composed as follows (in mmol/L): NaCl – 50;
KCl – 2; CaCl$_2$ – 4; MgCl$_2$ – 1.5; Tris-ОН – 10; pH –
7.5. From the dorsal side of the subesophageal gan-
glion complex we removed the thick neural sheath
made of connective tissue and the arachnoid sheath
in the projection of the neurons under studies. The
experimental equipment system designed and manu-
factured by us, the scope of which incorporated the
recording chamber and its auxiliaries, has made it
possible to carry out chronic experiments on freshly
prepared single neurons taken from the CNS of the
snail Helix pomatia with continuous long-term re-
cording of electrophysiological potentials up to three 24-hour days.

The physiological saline supplying system and the
system of discharge of waste fluid were designed for
creating and maintaining of a stream of the saline flow-
ing through the recording chamber at a constant flow
rate. The material used for the chamber was capable
of withstanding steam-and-pressure sterilization, and
the chamber design was intended for multiple uses.

To record electrophysiological characteristics, utilized
were special superfine microelectrodes (SME) filled
with 2.5 moles of KCl, with a tip diameter under 0.05
μm, having a resistance up to 5 GΩ. The bio-potentials
were recorded and measured by the unipolar method
with the use of an indifferent electrode being removed
from the neuron to a distance $r_{ie} >> \phi_n$, where: $r_{ie}$ is
the distance between the indifferent electrode and the
neuron, and $\phi_n$ is a diameter of the neuron soma. This
determines the position of the zero line on a recording.

In our experimental studies, biological potentials
were recorded with a 12 digit analog-to-digital con-
verter (ADC) at a time quantization of 40 milliseconds. A special preamplifier developed by us for input
currents to 3 fA, an input resistance of 100 GΩ and a
bandwidth from 0 to 10 kHz, provided the proper op-
eration with our SME and the adequacy in recording
of amplitude-frequency responses of the intracellular
potentials measured. Following that way, we investi-
gated the dynamics of changes in a resting potential
(RP) and pulse activity (PA) as well as parameters of
an action potential (AP). The original software devel-
oped by the Neurocybernetics Research Institution at
the Academy of Biology and Biotechnology, Southern
Federal University, Rostov-on-Don, Russia, allowed
us obtaining the proper visualization of the intracel-
lular potentials captured from a single neuron, their
recording and measuring accompanied by production

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Fig. 2. Legend: A. Disintegration of the neuron membrane due to insertion of the conventional ME: strong response to the ME
invasion into the neuron as frequent multiple action potentials (AP) and prolonged endogenous pacemaking generation of them. Z
is the time of the ME invasion into the neuron and “the capture” of the electrode by the membrane. B. Disintegration of the neuron
membrane with the use of a pulse electromagnetic micromanipulator system designed by Ivlev S.A. at al. [24]. C. Disintegration
of the neuron membrane due to insertion of a superfine microelectrode applied by us in our experimental studies. The picture
exhibits two APs only, which are produced due to the SME invasion into the neuron; subsequently the resting functional state of
the silent neuron is found, and the stable level of its membrane resting potential (MRP) is noted. Arrows in the picture identify
individual steps of advance of the microelectrode tip being inserted into the membrane. Calibration for A, B and C: 10 mV/1 s
of slides. The BBSS EMR generator employed in the experiments had the following parameters: the frequency range from 10^4 Hz to 10^8 GHz; an integral radiation power of 0.1 µW; the regimes of rhythmic modulation of the radiated frequency spectrum were as given below: regime 1: m_1 Hz and m_2 Hz; and regime 2: m_3 Hz and m_4 Hz, respectively.

Results and discussion

The chronic experiments offered stabilization of the functional state of the neurons. Our research work was carried out based on “the Rule of Initial State” formulated by S.M. Leites, the former Soviet pathophysiologist and endocrinologist. Delivery of stimuli to the neurons was provided upon expiration of 4–5 hours after completion of the sample preparation. According to our long-term observations, a stable background state in neurons is reached upon expiration of 3–4 hours from the time of sample preparation. Sometimes the stabilization process takes a shorter time because it depends on the initial functional state of the biological object and specificity of the technique applied to prepare the CNS samples as well as procedures of insertion of a microelectrode into the neuron. When a tip of the conventional microelectrode (ME) is contacting the neuron surface, and when the ME is being moved towards the center of the neuron, the neuron membrane is being bent and stretched. At that time, when, upon application of a pressure produced by the ME tip, the membrane surface is being disintegrated, an opening is appearing in the membrane letting the ME tip be inserted into the neuron, so that drastic vibration of the neuron membrane takes place, and, as a consequence, mechanical activation of the ion channels in the neuron membrane occurs, and pacemaking action potentials are fired (see Figure 2 herein).

It is explained by the fact that, when a microelectrode is penetrating the membrane, between them a gap, a large pore opening, is appearing, through which an ion exchange between the intercellular cytoplasm fluid and the external solution washing the neuron takes place. By this means, the mechanism of a long-term operation of the mechanically sensitive ion channels is initiated. The pore opening appeared in such a manner around the microelectrode is quickly shrinking, and finally the contacting surfaces are being sealed by the membrane and securely kept thereby. In Figure 2, fragment A, this process is designated by line Z. In some cases, under fluctuations in the functional state (FS) of the neuron during many hours’ intracellular recording, or upon exposure to some chemical substances, the membrane may release the captured conventional-type microelectrode, so that the latter may even leave the neuron body. In our test studies, we used our original SME with a high electrical resistance. Researchers, who deal with recording of intracellular activity, know that this sort of technique is always associated with overcoming of a number of drastic engineering challenges. In case, when the diameter of a channel conductor is reduced to 0,05 µm, we face a sharp increase in the resistance of the microelectrode. As a consequence, sensitivity of the high-ohmic input of the amplifier to external electromagnetic noise, and first and foremost, to network interference, is raised. The input members (an object under studies, the microelectrode and the
conductors connecting an object with the amplifier input) become extremely sensitive both to static charges and their changes. The input in the circuit responds even to some slight fluctuations in humidity of the ambient air in the testing room, and it might be illustrated by the case, for example, when other two or three colleagues enter the testing room with one operating researcher already present there-in: their overall breathing changes the actual ambient air humidity. In this connection, the question arises of whether it is generally reasonable to apply the SME technique because of a plethora of complicated tasks to be solved. We say definitely: yes: the application of the technique is quite justified. Our superfine micro-electrode (SME) enters freely, without any hindrance or friction, the neuron cytoplasm, as if SME were abundantly lubricated (see Figure 2, fragment B). The process of inserting SME into the neuron does not produce any violent reaction by the neuron: in this case the inserted micro-electrode is "not detected" or "not seen" by the neuron. There is another noticeable matter: the very small diameter of the tip makes possible to exclude electrolyte infiltrations from the SME body into the internal fluid of the neuron, so that the natural functional state of the neuron is retained unchanged for several days. To illustrate this fact, we present Figure 3 herein, which gives fragment A to exhibit a recording of the potentials of the LPaG2 neuron upon the 43-hour residence of the SME in the neuron soma. We can trace the natural intracellular activity pattern: excitatory postsynaptic potentials (EPSP), endogenous slow-wave potentials and endogenous pacemaker action potentials.

The background state of the LPaGx neuron in the prepared isolated CNS specimen, upon expiration of more than 2 days (52 hours) after preparation, as illustrated in Figure 3, fragment B, that represents the norm, is characterized by a specific language of communication of the neuron with the other neuron units within the network: it comprises slow and fast endogenous waves of de- and hyperpolarization of the cytoplasm membrane of the neuron, the stability of the AP parameters, the constant duration and some minor amplitude fluctuations, a wide range of frequencies in the generation of APs initiated by mixed endogenous pacemaker activity and EPSP, and some MP variations within the physiological limits. In the first set of the experiments we considered whether pathology in the shape of an action potential is available or not as well as the dynamics of the synaptic potentials arrived at the neuron under studies in a distant post-pulse period, at an interval within 60 minutes upon the EMR exposure. An action on the neurons by BBSS EMR was produced in the standard first regime of the generator operation. The time of delivery of the stimulus was 5 minutes. The mode of the rhythmic modulation of the radiated flux covered m₁ Hz and m₂ Hz. The spacing between the EMR source and the neuron was 50 mm. The axis of the EMR source was perpendicular to the surface of the ganglia and the neuron. Utilized was an integral radiation power of 0,1 µW. We traced the post-firing activity of the neuron on the BBSS EMR action within an interval of 1 hour, and there were no significant variations in the bio-potentials of the examined neuron detected. In Figure 4 shown is a recording segment to illustrate the intracellular activity of the Helix pomatia CNS LPaG1 neuron in 35 minutes after the BBSS EMR exposure. The state of the membrane potential, the endogenous pacemaker potentials, the amplitude, the shapes and durations of AP as well as the excitatory postsynaptic potentials (EPSP), arriving at the neuron, enable us to conclude that the functional state of the neuron has not experienced any pathological alterations. The functional state of the neuron has been found to be in correspondence with the respective physiological norm, as it has been the case before the BBSS EMR exposure.

In the second set of the experiments we estimated the effects made by electromagnetic radiation produced by our generator in another regime of operation, which
differed from the first one in the modulation frequency of the radiated electromagnetic flux, utilizing the following frequencies: \( m_3 \) Hz and \( m_4 \) Hz. Under the conditions, the neuron operating in the pacemaking mode was exposed to the above specific EMR [20]. The dynamics of the background-active non-identified neuron RPaG in the snail Helix pomatia CNS is presented in Figure 5, curve 1, to portray the initial background functional state of the neuron in question.

Upon completion of the electromagnetic exposure, the firing rate decreased from 14 to 9 pulses as indicated in Figure 5, curve 2, illustrating the neuron FS after the EMR exposure, and the level of the membrane potential was getting more negative: from minus 67±2 mV to minus 75±3 mV that indicated that there was hyperpolarization of the neuron membrane available. Upon the BBSS EMR action on the neuron, there was a change of the critical level of the membrane depolarization denoted by Threshold 2 referred to Threshold 1 of the initial state. Figure 5 herein depicts some fragments of the recordings of the neuron activity before the EMR exposure and after it. The event of the BBSS EMR delivery starting is intentionally ignored herein due to large artifacts interfering with a signal to be recorded.

The third set of the experiments was designed to deliver the BBSS EMR stimulation against the background of the pathological activity pattern of the non-identified neuron LPaGx. It was decided to test the influence made by BBSS EMR on the pathological activity of the neuron, which usually appeared upon an action of some chemical substances resulting in over-excitation of a cell. An example of production of a pathological activity pattern occurred in our experiment is given in Figure 6, fragment 1 herein, where a degradation of the neuron performance upon action of a chemical factor is demonstrated. We can find therein a steady-state pathological condition represented by fluctuating-amplitude spikes reaching values from 8 mV to 19 mV.

Some measures taken by us to eliminate the artifacts induced by BBSS EMR allowed us minimizing the signal distortions and improving in the quality of recordings of the potentials during the EMR exposure sessions. That has made possible to trace the bio-potentials immediately during the course of the EMR action. Figure 6, fragment 2, exhibits the intracellular potentials of a neuron recorded during the BBSS EMR exposure session. The given set of the experiments was dedicated to the application of the BBSS EMR stimulating signal at a signal modulation frequency of \( m_1 \) Hz. An exposure time from switching-on of the BBSS EMR generator was limited to 30 seconds. The other parameters remained unchanged. Upon completion of the electromagnetic exposure, the amplitude of some large spikes reached 21 mV; the value of the membrane potential increased and became more electronegative, and the spiking pattern was found to be more rhythmic that was a marker of a relative stabilization and improvement in the functional state of the neuron under study.

**Conclusion**

Our experimental studies aimed at discovery of influences and effects produced by BBSS EMR under the above specific parameters on the intracellular potentials of the central nervous system neurons within the neuronal network in the circumoesophageal ganglion complex in the snail Helix pomatia, and, following our philosophy, on the intracellular potentials of the cardiomyocytes, have demonstrated the presence...
of responses given by the neurons and changes of their functional state. The detected responses have been found to be reversible, and they should be classified as reactions of training according to the Kvakina-Garkavi-Ukolova scale for assessing the activity level in biological systems. This is the evidence that the neurons and the entire nervous system recognize and sense the specific BBSS EMR as a soft regulatory and hyperpolarizing action ranked by us at this stage of the studies as producing a calming effect and allowing the neuron not consume, but store up its internal energy.

Below we would like to outline some considerations relating to the mechanisms of neuronal responses to the above EMR exposure. According to the findings in the reference literature [21], this role is assigned to the pacemaker potential-dependent membrane channels of hyperpolarization, which also can be found in different structures in the brain, including the cortex region, according to current knowledge. Specifically, based on the data in the reference literature, the appearance of rhythmic burst discharges of the neurons in the thalamus is treated to be an endogenous intrinsic property, arising due to joint activation of the potential-dependent K⁺ channels of hyperpolarization and the low-threshold Ca²⁺ and Na⁺ currents initiating the burst discharge. So, mentioned should be some research papers which outline activation of the h-channels (hyperpolarization-activated cyclic nucleotide-gated ion channels) that results in local generation of intrinsic pacemaker currents, and then oscillations of this sort can propagate both into the other cortical and subcortical structural regions in the brain. This activation of the pacemaking h-channels, as we may think, and as it is evidenced by the reference publications, is initiated as a genetically determined adaptive response of the neurons to an excessive hyperpolarization of the neuronal membrane due to a reduction in tonus of the reticular formation in the brain in order to provide a restoration by them of their initial level of the resting potential with elevating their excitability level towards the spindle termination. This assumption is supported by researches which have established that the dendritic h-channels play a critically important role in the regulation of excitability of the cortical cells. At the same time, there is another way of looking at this mechanism as discussed below. Electrical potential is an intrinsic property of all excitable tissues (the nervous, muscle fiber and gland tissues). An essential feature of the excitability is its close interplay with the specific sensitivity of the cell membranes and their property to respond to stimuli by certain specific changes in the ionic permeability and the membrane potential. In this case, the excitability should not be confused with the excitation, since the excitation is a response by a biological cell to a stimulus, when the living system experiences a conversion from the state of its physiological resting into the specific activity state peculiar to a given cell or tissue. If a cell is placed in an electrical field, it becomes polarized, i.e in one region of the membrane the sign of the resting potential is identical to that of the field strength, while the opposite membrane region has the same potential, but with the opposite sign. What this means is, that, on the one hand, the membrane potential in one half of the cell has increased (the membrane has been hyperpolarized), while, on the other hand, the potential in the other half of the cell in question has decreased (the membrane has been depolarized). An excitation appears as soon as depolarization of the membranes reaches or exceeds a threshold level under an action of a current applied from the outside. This process is also referred to as stimulation. Following this way of
the interpretation, we suggest that one of the methods of an artificial change in the level of excitability for different cells, individual structures in the brain and the general performance as well as formation of a predominance in the model experiment is polarization of some brain tissue regions produced by an electromagnetic field applied from the outside.

In this connection, the following points can be highlighted:

a) EMR under the specific parameters, as indicated above, being a stimulus can be exactly graduated in strength, time and pattern effects produced on a living tissue;
b) EMR under the above specific parameters and the current induced by the latter are found to be close to the natural mechanisms responsible for generation and propagation of excitation in living tissues;
c) EMR under the above specific parameters is remote and contact-free in its effects.

The effects produced by BBSS EMR on the Helix pomatia CNS neurons are also applicable to the performance of cardiomyocytes. Similar to AP of a neuron, in case of cardiomyocytes, the EMR influence will be manifested in an increase in growth rate of the rapid depolarization phase. This is encouraged by opening of a greater number of the influx sodium and calcium ion channels, and, as a consequence, an increase in the amplitude of the action potential and a rise in the initial phase repolarization rate with a steeper fall that is provided by opening of a greater number of the fast potential-dependent potassium ion channels and a faster efflux of the K⁺ from the cell. The ionic balance between the Ca²⁺ entering the cell and the K⁺ exiting the latter keeps the potential of the membrane at the level corresponding to the plateau portion of the action potential of the cardiomyocyte. The effect made by BBSS EMR is manifested in an increase in steepness of the phase of the rapid end repolarization of the membrane due to activation of most fast potassium channels, determining the outward potassium current, as well as due to an increase in the inward calcium and chlorine currents that results in a shift of the phase of the recovery of the membrane resting potential towards more negative values (hyperpolarization). In consequence of the above, the slow repolarization phase, the plateau portion, should become somewhat shorter, and a pulse-to-pulse interval between the two successive action potentials of the cardiomyocyte should be longer that has been evidenced by our model studies. Hyperpolarization of the cells under the BBSS EMR exposure can be explained by an enhancing of contribution of the electrogenic component of the sodium-potassium pump to the MP value and either by a decrease in the passive permeability to sodium and calcium ions, or an increase in the permeability to potassium ions. Changes in the total ion currents, the AP and PA parameters of the neurons, exposed to BBSS EMR, are essentially attributed to the respective variations in the MP value and a marginal direct influence on the potential-controlled ion channels. Besides, a minor depolarization of the neurons upon the BBSS EMR exposure and the stabilization of the frequency of electrical activity may indicate that there is a slight activating effect of the ionic channels available. A quite interesting effect produced by the above specific BBSS EMR is initiation of delayed-rectifier currents. The basis for effects of this sort might be activation of the processes of phosphorylation – dephosphorylation of the channel proteins and the stabilization of the membranes that facilitates faster conformational mutual transitions of the ion channels between the states (closed – open – inactivated). The cause of the prolonged changes detected in the neurons might be an EMR effect exerted on the expression of the ion channels and embedding of them in the plasma membrane. The membrandotropic activity of BBSS EMR traced in our experimental study sets was reported to be considerably less as compared to that revealed by us in the mollusk neurons for anesthetics and anti-arrhythmia drugs, applied immediately onto the neuron membrane [22, 23]. Many anti-hypoxants produce their pharmacological effects at least at three levels: at the neuronal, vascular and metabolic ones. They may promise a wide spectrum of pharmacological activity offering effects of anti-hypoxia, stress protection, nootropy, anticonvulsant and anxiolytic actions, inhibition of the free-radicals processes of lipid oxidation, a rise in the resistance of the organism to exposures of various damaging factors as well as to oxygen-dependent pathological conditions (shock, hypoxia and ischemia, brain circulation disorders, alcohol and anti-psychotic intoxication, including neuroleptic intoxication). By this means we can conclude that BBSS EMR under the above specific parameters, similar to anti-hypoxants, by changing MPs and ion currents of the cardiomyocytes and neurons modulates their functional state. BBSS EMR has both “calming” effects and activating influences on the performance (electrical activity) of the neurons that is similar to the effects produced by anti-hypoxia medication.
Based on the evidence obtained in our experiments, we may conclude the following:

1. We have conducted pioneering research on influences and effects made by some rhythmic patterns, similar to those found in an electroencephalogram, or the natural resonance frequencies of the Earth of the rhythmic broad-band stochastic electromagnetic field patterns, on intracellular potentials of a neuron within the isolated central nervous system. Treating the neuron from the isolated CNS in the snail Helix pomatia as an exemplary case, we have demonstrated the responses by the neurons to the BBSS EMR exposure and the BBSS EMR action mechanisms, based on the data on the membrane potential levels and the action potential parameters measured.

2. The rhythmic BBSS EMR initiates reversible hyperpolarization of the cytoplasm membrane of the neuron with maintaining it at the level of the residual hyperpolarization under the action of the field. The recovery of the membrane resting potential is accompanied by an enhanced operation of the Na+/K+-ATPase to remove the sodium ions influxed in the first phase of depolarization of AP. Besides, an accelerated restoration of the concentration of the calcium ions owing to operation of the 3Na+/1Ca2+ antiporter and the Ca2+ ATPase takes place.

3. The influence of the rhythmic-pattern BBSS EMR raises the rate of growth of the rapid depolarization phase of the action potential. This is promoted by opening of a greater number of the influx sodium and calcium ion channels, and, as a consequence, an increase in the AP amplitude and a rise in the repolarization rate with a steeper fall provided by opening of a greater number of the potassium ion channels and a faster efflux of the K+ from the cell.

4. The effect produced by BBSS EMR is manifested in a greater steepness of the rapid end phase of polarization because of faster closing of the sodium channels and activation of the fast potassium channels, governing the outward potassium current, as well as in an increase in the inward calcium and chlorine ion currents, which lead to the shift of the membrane resting potential recovery phase towards more negative values (residual hyperpolarization) and maintenance of the membrane potential at this level as in the course of the specific EMR action.

5. The regulation of the functional state of a neuron in the neuron network with the use of the non-invasive remote method of control has been objectively shown herein. The neuron is treated by us as an analogue for the formation of the transmembrane potential of the cardiomyocyte and at the same time as the basic model of the neuronal cellular auto-regulation of the phase-structured performance of the cardiovascular system. In doing so, a fine-tuning instrument to control the neuron, CNS and other cell systems in the organism has been discovered. EMR of this sort, being a stimulus, can be exactly graduated in strength, time and effects produced on a living tissue. Besides, the employed BBSS EMR and the current induced by the latter have been found to be close to the natural mechanisms of generation and propagation of excitation in living tissues.

6. A favorable effect made by the specific BBSS EMR therapy produced on the functional state of a neuron has been demonstrated. It finds its reflection in the “calming” dynamics traced in the neuron membrane potential changes and the hyperpolarizing influence. An approximation of the results obtained at the cellular level makes possible to assume that there is a possibility available to control the performance of the autonomous highly specialized nervous system of the heart, the integrated system, CNS and development of physiological reactions at the level of the human organism.

The results of our research offer widest possibilities of a targeted fine tuning and regulation of the control systems in a human organism.

We have studied one of the aspects of the mechanism of the targeted cardiac regulation that has been analyzed on the basis of the cardiac cycle phase analysis in the context of cardiometry, but it might involve new challenges in science related to further investigations of much finer subcellular mechanisms responsible for the regulation in the high-specialized cell systems in the organism.

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Statement on ethical issues
Research involving people and/or animals is in full compliance with current national and international ethical standards.

Conflict of interest
None declared.
Author contributions
The authors read the ICMJE criteria for authorship and approved the final manuscript.

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The state of anti-oxidation system in rats under chemically induced cancerogenesis and influence by complexly modulated ultra-low frequency magnetic field (ULF MF)

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Abstract
At present, control of properties of biological membranes as targets of magnetic field applications within a living cell is the subject of considerable discussion in the scientific community. Experimental studies carried out by us demonstrate that ULF MF has an access to the most important mechanisms of tumor growth. The most likely mechanism among them is that responsible for the processes of free-radicals oxidation. Therefore, our research work was aimed at studies of influences of ULF MF on indicators of the anti-oxidation system in blood plasma and blood cells in rats in the chemically induced cancerogenesis. Our experimental studies were performed in 45 outbred albino male rats having a body mass from 160 to 180 g.

The state of the anti-peroxidation protection of cell membranes was assessed by indicators of the fermentation subsystem of AOS (catalase, TPA, SOD), and the state of the non-fermentive subsystem of AOS was estimated by indicators of the concentrations of vitamins A and E in blood serum. A prolonged latency period in the chemically induced cancerogenesis, obtained in our experiments, is treated by us as a positive effect produced by the ULF MF therapy that was found to be in correlation with the maintenance or restoration of the level of the functional activity of the anti-oxidation system in blood of the rats. Moreover, we have detected that it is precisely these immune competent cells which are the main point of applications in responses by the anti-oxidation system to ULF MF exposure that confirms our assumptions that the cancerogenesis process can be controlled by ULF MF.

Keywords
Chemically induced cancerogenesis, ULF MF, Anti-oxidation system in blood

Imprint

Introduction
Free radicals are those atoms, molecules or some fragments of the latter, an excessive amount of which leads to development of oxidative stress that is judged to be the cause of many tumor-related, neurodegenerative and inflammation diseases as well as aging in an organism. Environmental pollution by industries, tobacco smoke air pollution, ionized radiation, unfavorable ultrasound influences and magnetic fields are major exogenous sources supplying free radicals. Biological alterations and modifications, which may occur upon effects made by electromagnetic fields, were studied with regard to sources both of permanent and varying low frequency magnetic fields. A lot of research work have demonstrated that the permanent magnetic fields induce apoptosis, depending on time of their influence [1], along with an increase in concentration of malondialdehyde (MDA) and activity of catalase in rat tissues [2].

The permanent magnetic fields may reduce activity of glutathione peroxidase (GPX) in the rat kidneys and elevate activity of superoxide dismutase (SOD) in the rat liver [3]. At the same time, it is assumed that low frequency magnetic fields having biotropic parameters may play a decisive role in the cell life by regulating reactions of lipid peroxidation [4]. An exposure to low frequency magnetic fields promotes development of adaptive mechanisms responsible for maintenance of the proper oxidation & reduction balance in the organism [5, 6]. It has been detected, when weak magnetic interactions can radically alter properties of chemical and bio-chemical systems and the manner in which they can do it [4, 7]. At the present time, there is a broad discussion conducted on the issue of changes in the properties of bio-membranes as the primary point of an application of effects produced by magnetic fields on a living cell.
Use of ULF MF as a wave-type factor, covering not only the polyfrequency regime of action within the range 0.03–9 Hz, but also frequency modulation by signals of different shapes (sinusoidal, rectangular and saw-type waveforms) increases the informative value of the action by adding complexity to the signal structural pattern. The relevant evidence data obtained by Shikhlyarova et al. [8] in their research work have made it possible to reveal a high biotropic capability of the parameters of the frequency modulation in relation to tumor growth and the course of cancerogenesis.

Despite the low intensity (3.2 mT) and the low frequency level (0.3–9 Hz), the effects made by the above mentioned factor bear witness to the fact that we deal with a phenomenon of an anti-tumor effect produced by the action of ULF MF, provided that a sinusoidal signal is filled with rectangular-shaped pulses: we have discovered reductions in tumor growth rates, regression of tumor conglomerates, prolongation of latency periods in cancerogenesis and an increase of life spans in the animals [9].

The obtained effects demonstrate that ULF MF has an access to the most critical mechanisms responsible for tumor growth. It should be noted that the most likely mechanisms governing the tumor growth are the processes of free-radicals oxidation (FRO), and research on this subject was carried by us herein in the context of dynamics of chemically induced cancerogenesis [10].

Taking into account the regular, stage-related features of re-arranging of the bio-chemical processes in chemically induced cancerogenesis [11], we formulated the aim of our research studies as an investigation of a possibility to model conditions of an ULF MF action for correction and prevention of disorders in lipid peroxidation and anti-oxidant systems (LPO-AOS) in the organism at the corresponding stages (1–9 weeks) exhibiting pronounced reduced protective anti-oxidative properties of the organism.

Therefore, the aim of this paper was to closely study the action provided by ULF MF on indicators and parameters of the anti-oxidation system in blood plasma and blood elements in rats under the chemically induced cancerogenesis.

**Materials and methods**

Our experimental studies were performed in the autumn/winter period in 45 outbred albino male rats having a body mass from 160 to 180 g.

In our research work, we used models of tumors, which were experimentally produced upon subcutaneous injection of the rat Sarcoma 45 cell line suspension and induction by chemical cancerogene 3,4 benzo[a]pyrene with a dose of 5 mg as 1% solution (0.5 ml) in peroxidized olive oil [12, 13].

Volumes of experimentally developed tumors were calculated by the Schreck's formula applicable to an ellipsoid as follows: \( V_{\text{ell}} = \frac{\text{ab} \cdot \text{c}}{6} \), where \( V \) is a volume of a tumor, and \( a, b, c \) are height, length and width of the tumor, respectively. Effects of the electromagnetic influence on cancerogenesis dynamics was evaluated with chronometric examination of the latency periods of tumor progression from the time of cancerogenic cell line injection till the time of palpatory detection of a tumor node. Levels of differences in latency period times between the reference group and the experimental treatment group was determined by computing the Iball's index showing the relation between the number of tumors and the latency time.

In addition to recording of tumor sizes and identification of the latency period, a comparative assessment of life spans in animals involved in the experiments against those in the reference group was carried out. The MF exposures were started as soon as each tumor reached a volume about 1 cm³ (1000 mg).

The procedure of the application of the varying magnetic field (VMF) was as follows: before the exposure, a rat was placed into a plexiglass chamber designed taking into account the specific rat-hole reflex. A magnetic field inductor was installed overhead, in the area of the rat hypothalamus projection. The MF exposure sessions were carried out every morning. In the experimental studies used was modulation of rectangular pulses at a frequency of 300 Hz by a low frequency (0.03 – 0.3 – 3 – 9 Hz) field with sinusoidal shape pulses. Exposure times were different: for a frequency of 0.03 Hz the exposure time was 5 minutes, and for the other frequencies it was prescribed to be 1 minute. The treatment sessions were performed on the regular basis, every day, in week 1, 5 and 9.

In order to assess the state of the anti-peroxidation protection of cell membranes, the respective data on the AOS fermentation by identifying activity of catalase [14] and the total peroxidase activity in the blood serum by A.A.Pokrovsky method [15] modified by V.V.Vnukov [16] were under study; our investigations addressed also the activity of superoxide dismutase (SOD) based on Tetrazolium-Based Superoxide Dis-
mutase Staining Assay in the system of aerobic oxidation of xanthine by xanthine oxidase (XO) [17]. The state of the non-fermentative subsystem of AOS was estimated by indicative values of the simultaneous fluorimetric determination of concentrations of vitamins A and E in serum with the use of the method by Ceresauskene R.Ch., et al. [18]. Statistics data were processed with the use of Software SPSS 11.5 for Windows. To assess significance in differences between indicators in the respective groups, preliminarily we identified correspondence of the obtained sampling to the normal law of distribution. An estimation of reliability of the data was completed with the Student’s t criterion. The level P<0.05 was accepted to be significant.

Results and discussion

It was found that even in chemically induced cancerogenesis week 1 a reduced concentration level of vitamin E in lymphocytes in blood in rats was observed that was 31.9 times less as referred to that recorded for the intact animals. The above mentioned level in erythrocytes and plasma showed a decrease by a factor of 2.9 and a factor of 13.6, respectively, against the respective indicators in the intact rats. Similarly, a decrease in the concentration level of vitamin A was recorded: that identified in lymphocytes was 61.9 times less, and that indicator recorded in erythrocytes was 2 times less; the same indicator in plasma was reduced by a factor of 5.5 (see Table 1 herein). An estimation of indices of the vitamin E and A exchangeability between the cells and plasma in blood demonstrated the following: the coefficient of the vitamin E relation between lymphocytes and plasma was halved, and that between erythrocytes and plasma was found to be 4.1 times higher as compared with the respective values in healthy rats. A similar variety of trends in the concentration fluctuations was typical for vitamin A, too: the coefficient of the relation of its concentrations between lymphocytes and plasma was 12.3 times less, and that of the relationship between erythrocytes and plasma was detected to be 6.7 higher.

In the above mentioned period of time, it was found that there were varied trends in changes in the relations of the vitamin E and A concentrations in lymphocytes, erythrocytes and plasma in blood in the rats. So, the indices in plasma and erythrocytes were 2.4 and 1.5 times less than the respective values in healthy animals, and as for the index for lymphocytes, it was 2 times higher than the normal levels.

At the same time, the performance of the AOS fermentation subsystem of blood in rats demonstrated changes in its data as indicated below (see Table 2 herein). The total peroxidase activity (TPA) was traced to be statistically significantly reduced in lymphocytes by a factor of 6.3 as compared with that parameter in the intact animals, and the latter in plasma was recorded to be 3.8 times higher, respectively. As that took place, we noted that activity of superoxide dismutase (SOD) was lower and showed the following levels: in lymphocytes it was 17.5 times less, and in lysate it was found to be 1.6 times lower as compared with the respective indicators in healthy rats. The activity of catalase was reduced as indicated below: in lymphocytes it was 7.2 times less, in erythrocytes it was 1.3 times lower, and in plasma it was recorded to be 1.3 time less in reference to the intact rats.

The coefficient to express the relationship SOD/TPA, characterizing the actual status of the natural anti-oxidative fermentation cascade, in lymphocytes was identified to be 2.8 times less referred to the values in the intact animals.

By this means the obtained evidence data show that we deal with a suppression of both subsystems of the anti-oxidant protection of the lymphocytes mem-

<table>
<thead>
<tr>
<th>Groups of animals</th>
<th>Vitamin E</th>
<th>Vitamin A</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>PLT un/ml</td>
<td>RBC un/ml</td>
</tr>
<tr>
<td>Reference group (n=6)</td>
<td>0.11±0.004</td>
<td>1.95±0.06</td>
</tr>
<tr>
<td>Test group</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 1 (n=6)</td>
<td>0.15±0.01</td>
<td>0.2±0.03</td>
</tr>
<tr>
<td>Week 9 (n=6)</td>
<td>0.35±0.02</td>
<td>1.5±0.2</td>
</tr>
<tr>
<td>Intact animals (n=6)</td>
<td>1.5±0.15</td>
<td>2.8±0.2</td>
</tr>
</tbody>
</table>

Notes: 1 – statistically significantly reliable in relation to intact animals; 2 – statistically significantly reliable in relation to the previous

Table 1. Effects produced by ULF MF on the state of the non-fermentation
branes as major immune competent cell fraction in rats already in week 1 of the chemically induced cancerogenesis.

Upon analyzing the data on the anti-oxidative activity in rats in week 1 and 9 of the combined effects exerted by 3,4 benz[a]pyrene and the experimental treatment by applying ULF MF, we have obtained some factual results as given below herein. As can be seen from Table 1 herein, the trends of the effects upon the ULF MF treatment in chemically induced cancerogenesis week 1 widely varied for the vitamin E and A concentrations in plasma and blood elements in rats as opposed to the reference group. So, no magnetic field effects on the levels of concentrations of the above vitamins in plasma were detected. The concentrations of vitamins E and A in erythrocytes were found to be decreased by a factor of 4.7 and by a factor of 2.7, respectively. And in regard to lymphocytes, the concentrations of vitamins E and A were identified to be 6.9 and 9.2 higher, respectively, than those in the reference group, but at the same time they were 4.7 and 6.7 times lower as compared with those in the intact animals.

Taking into account the synergy of the E & A vitamin action as well the value of the E/A coefficient, affecting development of the malignant process [11], we investigated the level of the coefficient in rats of the test group within the specified period of time. It was found that the effect made by ULF MF on the E/A coefficient, in comparison with the reference group, was available for lymphocytes only, and, due to the ULF MF effects, the said coefficient value was maintained at those levels which were typical for intact animals.

A careful analysis of the coefficient of the ratio of the vitamins between the blood cells and plasma, on the one hand, is an indication of the demand by the cells for these substances, and, on the other hand, it shows fluctuations in permeability and elasticity of membranes [19].

For the comparison purposes, we should note that on day 7 of cancerogenesis the relation of the E & A vitamins between erythrocytes and plasma exceeded the background values by 4.1 and 6.7 times, respectively, and, to the contrary, the values of the coefficient of the relations between vitamin E and vitamin A, between lymphocytes and blood plasma were reduced by 2.1 and 12.3 times, respectively, as compared with the intact animals.

Judging by the obtained data, considering the same time interval of the experimental studies, it may be noted that the specific ULF MF was capable of preventing some deviations from the relation between the cells and the blood plasma for vitamin E and even of substantially improving the similar relation for vitamin A. The coefficient of the relation of the vitamin E concentrations between lymphocytes and plasma increased by 2.7 times, as against the reference group, and by 1.4 times, as against the intact rat group. As for vitamin A, it was found that an increase in the said parameters by 4.9 and 2.5 times, respectively, was found typical as compared with the animals groups under comparative study. The coefficient of relation between the levels of concentrations of vitamins E & A between erythrocytes and plasma demonstrated a decrease.

A more pronounced effect was produced by ULF MF on the anti-oxidative ferments in blood of rats upon expiration of the 7 cancerogenesis progression days. It was detected that ULF MF completely prevented any changes in the activity of the ferment in erythrocytes, while the activity of SOD was 1.5 times less only, as against that recorded in the intact rats. TPA in lymphocytes in rats in the reference group decreased by 8 times, as compared to the indices identified for the intact rats, and, in contrast therewith, upon the

<table>
<thead>
<tr>
<th>subsystem of anti-oxidation system in blood in rats (M±m)</th>
<th>Coefficient E/A</th>
<th>Coefficient vit. E</th>
<th>Coefficient vit. A</th>
</tr>
</thead>
<tbody>
<tr>
<td>PLT</td>
<td>RBC</td>
<td>Lym.</td>
<td>PLT</td>
</tr>
<tr>
<td>2.9±0.3</td>
<td>4.5±0.4</td>
<td>6.1±0.4</td>
<td>8.3±0.9</td>
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<tr>
<td>2.0±0.2</td>
<td>3.0±0.08</td>
<td>4.0±0.5</td>
<td>1.5±0.2</td>
</tr>
<tr>
<td>2.3±0.1</td>
<td>2.1±0.2</td>
<td>2.5±0.09</td>
<td>4.0±0.3</td>
</tr>
<tr>
<td>7.1±1.1</td>
<td>6.7±0.4</td>
<td>3.0±0.2</td>
<td>2.0±0.2</td>
</tr>
</tbody>
</table>

period of the studies; ³ – statistically significantly reliable in relation to the non-treated animals, not exposed to ULF MF (p≤0.05)
Table 2. Effects produced by ULF MF on activity of the fermentation subsystem of the blood anti-oxidation system in rats (M±m)

<table>
<thead>
<tr>
<th>Groups of animals</th>
<th>TPA</th>
<th>SOD</th>
<th>Catalase</th>
<th>Coeff. SOD/TPA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Plasma un/mg</td>
<td>Lymph. un/mg</td>
<td>Lysat un/mg</td>
<td>Plasma un/mg</td>
</tr>
<tr>
<td>Reference group</td>
<td>22,2±3,1 1</td>
<td>0,04±0,006 1</td>
<td>139,0±7,6 1</td>
<td>0,08±0,007 1</td>
</tr>
<tr>
<td>(n=6)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 1 (n=6)</td>
<td>17,0±1,18 1</td>
<td>0,51±0,04 1</td>
<td>192,1±8,1 1</td>
<td>0,9±0,04 1 1</td>
</tr>
<tr>
<td>Week 9 (n=6)</td>
<td>9,9±0,8 1,2,3</td>
<td>0,05±0,005 1,2</td>
<td>185,9±7,2 1,3</td>
<td>0,4±0,02 1,2,3</td>
</tr>
<tr>
<td>Intact animals</td>
<td>5,9±0,6</td>
<td>0,25±0,05</td>
<td>226,4±12,3</td>
<td>1,4±0,1</td>
</tr>
</tbody>
</table>

Notes: 1 – statistically significantly reliable in relation to intact animals; 2 – statistically significantly reliable in relation to the previous period of the studies; 3 – statistically significantly reliable in relation to the non-treated animals, not exposed to ULF MF (p≤0,05)

ULF MF treatment, TPA demonstrated its increase by 2 times (see Table 2 herein). We also established a marked protector effect upon the MF applications on the activity of catalase in erythrocytes and lymphocytes in blood of rats. The related indices in the test group showed no differences from those identified in the intact rats, whereas activity of catalase in erythrocytes and lymphocytes in the reference group was reduced by 25,2 % and by 7,2 times, respectively.

But it should be mentioned that by influencing the activity of some individual ferments, ULF MF did not show any normalizing effect on their physiological cascade. No differences were found in the SOD/TPA coefficient value in lymphocytes of immune competent cells (see Table 2 herein).

When assessing effects made by ULF MF on cancerogenesis day 7 as a whole, we should notice the following regularity: the treatment completed by ULF MF, being not capable of completely restoring the initial level of the concentrations of vitamins A and E (in absolute values) in plasma and blood elements, identical to those in the intact animals, did promote prevention of disorders in permeability and elasticity of the cell membranes of lymphocytes, as compared with damaging effects due to cancerogenesis alone, that was confirmed by our analysis of the relevant coefficients. We made a correlation between the E/A coefficients in lymphocytes and the vitamin E/A relationship in lymphocytes vs. plasma of blood and the duration of the tumor progression latency period that demonstrated that there was a strong correlation available. The respective correlation coefficients were recorded to be 0,9, 0,86 and 0,7, accordingly (P < 0,05).

It is evident that the state of the cell membranes of lymphocytes responsible for the anti-tumor resistance of the organism is directly linked to the mechanisms governing tumor process progression, among them the duration of the latency period in tumor growing.

Following this way, we should mention that the fact of growing and developing of the induced sarcoma depends most likely on lowering of the E & A vitamins concentrations, on an imbalance in the anti-oxidative fermentation system and some disorders in the processes of detoxication of xenobiotics and the lymphocyte cell respiration that is evidenced by a decrease in activity of catalase, because it is just catalase that is assigned a decisive role in energy exchange of the cells according to the latest research findings [20].

A distinct protecting effect produced by ULF MF revealed in week 1 of cancerogenesis has demonstrated the capability not only of avoiding a deep decline in catalase concentrations in blood lymphocytes in tested animals, which was observed in the reference group animals, but also of doubling SPA of immune competent cells in blood, as compared with the healthy animals, and of increasing the latter by 12,8 times in comparison with the reference values.

Considering week 9 of the chemically induced cancerogenesis, accompanied by the ULF MF treatment, the indicators of the non-fermentation subsystem of the anti-oxidative protection of rat blood lymphocytes remained by and large at those values, which were typical for the previous period of the studies (see Table 1 herein). But for the rats, exposed to the ULF MF treatment, peculiar was an enhanced positive dynamics in accumulation of vitamin E in plasma and erythro—
cytes, as against the previous period (week 1), so that its concentrations were found to be 2.5 and 7.5 times higher, respectively. At the same time, the concentrations of the same vitamin in blood lymphocytes, being kept at a level 7.4 times higher than the reference one, did not show any changes.

The concentration of vitamin A was identified to be statistically significantly increased in relation to the respective indicator in the previous period of the experimental studies. As to plasma and erythrocytes in blood, despite the fact of growing of the absolute concentration levels of vitamins E & A, the coefficients under studies indicated further deterioration in permeability, elasticity and oxidation of the erythrocyte membranes. Upon the ULF MF exposure, the coefficients expressing the vitamin E exchange between blood cells and plasma within the test period experienced the following variability: the mentioned indicator for erythrocytes increased by 2.6 times, as compared with the previous period, and by 2 times, as referred to its level in the intact rodents. The coefficient of the lymphocyte/plasma relationship for vitamin E showed no statistically significant difference as against the similar indices in the intact animals. Discussing the vitamin A issue, it should be noted that the coefficient of the lymphocyte/plasma relationship remained at that level as it was the case in the previous period of the experiment, while the same indicative values for erythrocytes became 4.8 times higher with the reference to the period in question.

Upon completion of the repeat course of the magnetotherapy (week 9 of cancerogenesis), the total peroxidase activity in blood plasma in test group rats demonstrated a decrease by 2.2 times, as against the reference data, and an increase by 1.7 times only, as compared with the respective parameters in the intact animals. As to the lymphocyte data thereon, the values declined by 10, 2 times, when compared with the analogue parameters in the previous period of the experimental studies, and they were recorded to be 5 times less, as against the same parameters in the healthy rats (see Table 2 herein).

Activity of SOD in lymphocytes was found to be declined by 2.3 times, and that in plasma was recorded to be unchanged, as compared with week 1 of the combined exposure to 3,4 benzo[a]pyrene and ULF MF. For week 9 of the experimental studies, a decline in the activity of catalase in plasma by 1.6 times, as against the relevant reference values, was identified as typical. In erythrocytes and lymphocytes we observed similar trends in the changes: the parameters decreased by 1.5 and 1.8 times, respectively, compared with the previous period of the studies. In our opinion, an important point is that we record an increase in the coefficient of the SOD/TPA relationship in blood lymphocytes by 3.8 times, as against to the relevant reference values, and by 1.4 times, as compared with the values in the healthy animals.

Conclusions

By this means, notwithstanding the fact that the progressing dynamics of cancerogenesis implies alterations in the fermentation subsystem, which aggravate an imbalance in the SOD/TPA cascade in lymphocytes and involve the subsequent drop in the catalase levels in all blood test samples under studies, it becomes evident that re-setting of the anti-oxidative systems in an organism plays a key role exactly in the first week of chemically induced cancerogenesis.

The experimental anti-cancerogenic ULF MF therapy, conducted with the use of the sinusoidal-shaped pulse modulation in week 1 of cancerogenesis, has produced the protective effect on activity of catalase, SOD and TPA in blood lymphocytes. The adding treatment with ULF MF in week 9 of cancerogenesis has led to a manifestation of activity of the physiological anti-oxidative fermentation cascade in the above cells. At the same time, the concentrations of the non-fermentative anti-oxidants in lymphocytes have been maintained at a higher level, that is confirmed by the fact that there is an apparent trend in accumulation of vitamin A available.

A prolongation of the latency period in cancerogeneses, which is judged by us to be a positive effect produced by the completed ULF MF therapy, correlates with the maintenance or the restoration of the level of the functional activity of the blood anti-oxidative system in rats. In addition, it should be noted that it is precisely the immune competent cells that is the key point of applications in the responses by the anti-oxidation system to the ULF MF exposure that confirms our assumptions that the cancerogenesis process can be controlled by ULF MF.

Statement on ethical issues

Research involving people and/or animals is in full compliance with current national and international ethical standards.
Conflict of interest
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References
The pattern of the immune processes during activation therapy with the use of microwave electromagnetic radiation

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Abstract
The mechanisms of anti-tumor action of low intensity factors of activation therapy remain little investigated. The aim of the present paper is to study the changes in the immune system organs, the state of blood leukocytes and tumor area under effective activation therapy in tumor-bearing animals using EMR microwave range. The research is relevant due to the immune processes influence on cardiovascular system performance.

Materials and methods
In experiments on 77 albino outbred male rats with inoculated sarcoma-45 and Pliss lymphosarcoma tumors low intensity EMR of microwave and EHF ranges with bioeffective frequencies has been used as an activation therapy factor. The structural-functional changes in the immune system organs, some indices of blood leukocyte activity, composition of immune cells infiltrating tumors when inhibiting their growth and regression have been studied. Besides, the signs of intercellular interactions with participation of leukocytes in the tumor area have been considered.

Results
The pronounced anti-tumor effects in the studied EMRs are related to their anti-stressor action and, as a rule, accompanied by an increase in lymphoproliferative activity and intercellular interactions in thymus and spleen. Under the effect of microwave resonance radiation (1 GHz) noted are changes in blood lymphocytes activity, which precede the tumor regression beginning, as well as numerous contacts between malignant cells and leukocytes in regressing tumors. Peculiarities of large tumors regression and changes in thymus and state of blood lymphocytes, NK cells, monocytes and neutrophils influenced by modulated EHF EMR are described.

Conclusions
Immune mechanisms of anti-tumor effects in the studied EMR microwave ranges as activation therapy factors may be related to the following basic processes: increase in tumor cell differentiation under the effect of the immune cell factors, development of the antigen presentation processes and effective destruction of tumor cells by apoptosis induction, cytotoxic action of natural killer cells, macrophages and other immune system cells and their factors.

Keywords
Activation therapy, Microwave range electromagnetic radiation, Inoculated tumor, Anti-tumor effect, Immune processes, Thymus, Leukocytes

Imprint

Introduction
Malignant process causes the multilevel system changes in the organism. The intensity of these changes depends on the stage of development and localization of a tumor. At the same time the earliest disorders occur in the neuroendocrine and immune systems later on influencing the performance of other organs and systems. A close relation between the cardiovascular and neuroendocrine systems is well-known. In recent years the immune processes’ influence on the cardiovascular system performance and pathology development becomes also evident [1, 2]. Thus, the type of changes in different parts of the immune system under malignant processes and anti-tumor actions may be very important for dynamics of state of cardiovascular and other systems in the tumor-bearing organism.

It has been demonstrated before that the activation electromagnetotherapy contributes to improvement
in the regulatory system state and effective mobilization of anti-tumor resistance mechanisms [3–5]. At the same time the irreversible malignant cell injury, often observed in the experiment, may above all be caused by the immune system factors. It denotes the necessity of investigation of the immune processes, which assist the anti-tumor effects of the activation electromagnetotherapy.

The aim of the present paper is to study the changes in the immune system organs, the state of blood leukocytes and tumor area under effective activation therapy in tumor-bearing animals using microwave electromagnetic radiation (EMR).

Materials and methods

Examination has been carried out in 77 albino out-bred male rats weighing 180-300g with inoculated sarcoma 45 (23), Pliss lymphosarcoma (47) as well as in animals without tumors (7). The used tumors differed in histological structure, development rate and invasion depth. The EMR of centimeter and millimeter (extremely high frequency, EHF) ranges have been applied. The experiments have been conducted in accordance with international ethical standards. The electromagnetotherapy course duration is 3.5–4 weeks. The actions have been carried out in accordance with the activation therapy principles [3–5].

In the experiments in 23 rats with inoculated sarcoma 45 applied has been resonance radiation (RR), low intensity (less than 10 mW/cm²) EMR of centimeter range, the frequency (1 GHz) of which coincides with the frequency of water-containing media self-radiation, generated by the EMR of millimeter range [6]. “Aquaton” device has been used for that. A head and tumor area have been sequentially exposed (double exposure) for 3–10 min.

Low intensity (not more than 10 mW/cm²) modulated by EHF EMR with 42.2 GHz frequency served as an additional factor. For modulation the 7.8 Hz frequency signal of Schumann resonance spectrum or the sequence of signals with pairwise multiple bioeffective frequencies of 1.7, 3.4, 7.8 and 15.6 Hz have been used [7]. The occipital region of head (hypothalamus projection area) has been influenced with changes during 15–30 min. depending on blood leukogram and animal’s behaviour in the chamber. For that purpose the “Yav 1” device for EHF therapy with an additional ferrite isolator and the modulator developed in Rostov Research Institute of Radio Communication on the basis of the special form signal generator G6–37 have been used. The electromagnetotherapy course started with minimal exposure 3 days before tumor inoculation.

During the research studies the change in tumor size has been defined by calculating the volume according to Schreck formula for ellipsoids. The animals state has been evaluated according to dynamics in type and tension of general non-specific adaptation reactions (AR) of the organism, which is determined using the hematologic indicators, namely blood leukogram parameters, calculated for 200 cells [3, 8]. During the stages of the experiment in the peripheral blood lymphocytes activity of basic enzymes dehydrogenases, namely, succinate dehydrogenase (SDH) and alpha-glycerophosphate dehydrogenase (α-GPDH), which participate in significant energetic processes of cell metabolism [9], has been studied. The functional activity of natural killers (NK-cells) has been evaluated using the cytotoxic test [10]. Herewith the lymphocytes isolated in a ficoll-verografin density gradient (ρ 1,077–1,078) of rats blood have been used as effectors, and the cell culture of human erythromyeloleukosis K562, obtained by Research Institute of Clinical Immunology RAMS (Moscow), has been used as target. Besides, evaluated have been the indicators characterizing activity of the phagocytic part of the immune system, namely, the phagocytic activity (PA – percentage of phagocytic cells), the phagocytic number (PN – average quantity of test objects absorbed per one phagocyte) and the intensity of oxygen dependent reactions of “respiratory explosion” which is evaluated in rats blood neutrophils and monocytes with the help of spontaneous and stimulated (by zymosan or latex) test of nitro blue tetrazolium reduction to diphormazan (NBT test) with calculation of NBT stim./NBT spont. coefficient [11]. We would like to emphasize that the above indicators have been used with the aim of indirect, approximate evaluation of the blood neutrophils and monocytes functional state.

At the end of the experiments the animals were over-narcotized with ether; organs and blood were investigated. During morphologic and morphometric investigation of the immune system organs and tumor tissue the method of Brache staining modified by R.A. Simakova to identify nucleoproteins as well as the hematoxylin-eosin staining have been used. At the same time in thymus determined have been the indicators reflecting the lymphoproliferative processes activity, i.e. lobule size, width of cortical and medullary sub-
stances, as well as the stromal-parenchymatous coefficient. The prevalence of contacts between tissue basophils and thymocytes has been evaluated. In spleen defined have been the relative size of germinal centers and relative number of complexes (associates) of macrophages and lymphocytes.

The anti-tumor efficiency of activation electromagnetotherapy has been evaluated with the help of the following indicators: percentage of tumor growth inhibition, relative number of regression cases, intensity of regression, intensity of degenerative-dystrophic and necrotic changes in tumor cells, qualitative and quantitative composition of immune cells infiltrating the tumor. In the studies with resonance radiation (RR) the electron microscopic investigation of tumor tissue has been carried out with the help of the electron microscope Philips EM208.

For statistical processing of the obtained results the Student’s t- criterion and Wilcoxon–Mann–Whitney criterion have been applied.

Results and discussions

The examined microwave range EMR had an evident anti-tumor effect on a number of animals, accompanied by rather significant change in central and peripheral parts of the immune system.

Multilevel immunotropic effects in case of resonance radiation exposure on rats with sarcoma-45

Thus, the RR exposure proved to be effective for 77 % of animals. Besides, complete or practically complete (85–95 %) regression of sarcoma-45 has been observed in a third of animals. The remaining 6 rats showed in equal shares a partial regression of tumor (two times) or 40 % inhibition of its growth. It is necessary to note that an anti-tumor effect of the given factor has been related to its anti-stressor action, i.e. development of anti-stressor AR of calm and elevated activation without evident signs of tension during the last stages of the experiment, when the AR of stress...
prevailed in the reference group of animals (70–80 % cases).

It should be noted that we have already considered the problem of greater efficiency of RR with double localization in comparison with the influence of this factor in case of its localization only on the head or tumor area as well as the problem of firstly detected possibility for low intensity EMRs to lead to an increase in a degree of differentiation of tumor cells, thus reducing their life cycle demonstrated exactly for RR [12, 13].

In the present research we are interested, first of all, in the changes occurring in the immune system of the tumor-bearing rats and accompanying the anti-tumor effect of RR. The results, obtained during studying micro pictures of thymus and spleen under the RR effective influence, corresponded to the information in literature concerning changes in the immune system organs in case of tumor regressions under the effect of activation therapy with ultra-low frequency EMRs [3, 4, 12]. As opposed to hypoplasia and degenerative disorders, typical for thymus and spleen under tumor stress (Fig.1A and 2A), in the cases of effective influence of resonance EMR in these organs high lymphoproliferative activity (Fig. 1B and 2B) and evident signs of intercellular interaction activation with participation of tissue basophils in thymus (Fig. 1C) and macrophages in red pulp of spleen (Fig. 2C) have been observed.

Changes in the immune system of the animals with the most evident effect of RR, i.e. the tumor regression, have been also observed in blood lymphocytes (Fig. 3). When examining the activity in basic enzyme-dehydrogenases of SDH and α-GPDH tricarboxylic acid cycle in mitochondria of these cells 10 days after starting the exposure, increase in SDH activity has been detected. The SDH activity increased by 1.4 times (p < 0.05, Fig. 3А) as compared with the data of the reference group. It has shown the activation of metabolic processes in blood lymphocytes under the effect of RR during the stages preceding tumor regression. After finishing the exposure sessions in animals with regressing tumors marked has been almost a 1.5 time increase in ratio between SDH and α-GPDH activity indicators as compared with rats in the reference group (р < 0,05, Fig. 3В).

Thus, the anti-tumor effect of RR is evidently related to the changes in central (thymus) and peripheral (spleen, blood) parts of the immune system. An effective influence of RR on central and peripheral parts of neuroendocrine and immune systems ended in mobilization of effector anti-tumor mechanisms and directly in the area of malignant process localization. In vessels, located close to tumor cells, the lymphocytes

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**Fig. 3.** Activity in enzyme-dehydrogenases of peripheral blood lymphocytes in animals in case of regression of sarcoma-45 under the resonance radiation (RR) effect. A. Increase in the SDH activity 10 days after starting the RR exposure. B. Rise in ratio between SDH and α-GPDH activity indicators in the end of the exposure course.

**Fig. 4.** Increase in infiltration of sarcoma-45 deep layers by the immune system cells in case of inhibition of tumor growth under the resonance radiation effect. Quantity of lymphocytes and plasmocytes in 10⁶ tumor cells
migrating through the tissue have been frequently observed. As a result of morphometric study of sarcoma-45 tissue with inhibition of growth demonstrated has been a significant (2.9 times, р < 0,05) increase in a number of lymphocytes and plasmocytes, infiltrating deep layers of tumor (Fig. 4).

In cases of effective RR exposure the immune cells infiltrating the tumor have been observed in the capsule, directly under it in the area of mature connective tissue, and in the form of lymphoplasmacytic barrier in the area of young connective tissue (width 145–165 µm) of the tumor boundary. At the same time immune cell infiltration under partial regression has been more intensive than under inhibition. The qualitative composition of cells also increased. In addition to lymphocytes and plasmocytes (Fig. 5A) the tissue basophils and numerous macrophages have also been observed (Fig. 5B).

In our opinion, the noted rise in a number of lymphoid and macrophage elements, as well as other immune system cells in the tumor area, is a result of the intercellular interactions activation with the participation of leukocytes and tumor cells under the influence of RR, which has led to malignant cells elimination by means of their direct destruction and increase in the degree of their differentiation, the signs of which have been described by us earlier [12, 13].

Thus, when investigating the tumor tissue under electron microscope numerous contacts between the activated lymphocytes cytoplasmic outgrowths and
the tumor cells surface (Fig. 6A) are observed. Besides, in some cases several lymphocytes contacted with one sarcoma-45 cell. Moreover, frequently observed are simultaneous contacts of the immune system cells with each other (lymphocyte-lymphocyte, lymphocyte-macrophage) and with the tumor cells. Thus, in the electron-diffraction pattern herein (Fig. 6B) we see the monocyte contacting simultaneously with several tumor cells and forming a cytoplasmic bridge with the lymphocyte.

The detected at the electron microscopic examination numerous intercellular contacts allow assuming the presence of the factors in immune cells which lead to the differentiation degree increase and, as a consequence, to decrease of the tumor cell life cycle. It corresponds to modern literature data which proves the leading role of lymphocytes (especially, T-lymphocytes) in the regulation of cellular growth and differentiation of not only immune but any somatic cells in the organism [14]. Quite probable are the other effector mechanisms of tumor cells elimination under the influence of RR, which are related to different variants of cytotoxicity in lymphocytes, macrophages and other immune system cells, as well as to production of cytokines by these cells and any other active factors directly or indirectly damaging the tumor cells [15].

Changes in the immune system and dynamics of regression of Pliss lymphosarcoma of different size under the effect of modulated EHF EMR

Another investigated acting factor, namely, the modulated EHF EMR, turned out less effective compared with the RR in rats with formed sarcoma-45 of 0.7–1.0 cm³ volume, than the RR [7]. Noted has been an unstable 43% inhibition of tumor growth (p<0.05) during the 2-nd week of exposures as well as 1.2–1.3 times increase in number and area of segments of degeneratively changed sarcoma-45 cells after finishing the exposures (p<0.05) as compared with the indices in the reference group of animals. Such an effect correlates to the improvement in the AR type and the lymphoproliferative process activation in the immune system organs. However, the EHF EMR effect in the studied mode of application turned out to be insufficient for mobilization of the effector anti-tumor immune mechanisms and tumor cells destruction.

It has been a success to obtain a significantly more evident effect of modulated EHF EMR on the mech-
anism of anti-tumor resistance with a part of male rats with inoculated Pliss lymphosarcoma in case of starting the exposure 3 days before the tumor inoculation. As it has already been mentioned, two modes of low frequency modulation, namely mono-frequency modulation by signal 7.8 Hz (group 1, n = 18) and poly-frequency modulation with the use of a sequence of signals of paired multiple frequencies of 1.7–3.4–7.8–15.6 Hz (group 2, n = 18) have been used in the experiment. Besides, conventionally used dilution (to 20 %) has been 10 times increased (to 2 %) in order to prolong the period of detectable involvement of the tumor to obtain more accurate evaluation of the acting factors influence. It is known that Pliss lymphosarcoma is characterized by rapid growth and active infiltration of the neighbouring tissues, as well as by practical absence of cases of spontaneous regression even in very small tumors (not exceeding 0.03 cm³ in volume, the so-called “pea”). We supposed that the low frequency modulation of the high-frequency signal could slow down the tumor formation and invasion processes that would lead to an increase in animals’ life expectancy. However, the result of this experiment turned out to be different and was related to not known earlier delayed reactions of the tumor-bearing organism on the modulated EHF EMR, which manifested themselves in complete or partial regression of the tumor.

The majority of animals in two main groups under exposure during the first five days after detectable involvement of the tumor had the same rate of the tumor growth as the reference group. Some cases in group 2 (EHF EMR with poly-frequency modulation) were an exception. In that group noted was presence of the smallest tumors not exceeding 1.3 cm³ volume whereas with all other animals the lymphosarcoma volume was not less than 2.4 cm³. More evident effect of the EHF EMR with poly-frequency modulation manifested itself on day 6 after inoculation, when 3 animals (17 %) had regression of the tumors with size up to 2 cm³. Since then, the EHF EMR effect has been noted in other animals: on the eighth day 4 males (22 %) from group 2 where the EHF EMR with mono-frequency modulation (7.8 Hz) had been used showed complete or 80 % (and more) evident regression of 4.9–6.1 cm³ tumors. Thus, a partial, evident or complete regression of tumors has been observed in approximately one third (6–7 of 18) male rats in each group where the EMR has been applied. At the same time, a partial, evident and complete regression of large tumors (4.9–13.5 cm³) occurred quite rapidly (4–7 days), and no signs of intoxication were noted. In other animals in main groups the tumor growth rate did not differ from the malignant process development in male rats in the reference group.

Visually different peculiarities of changes in tumors in case of their complete and partial regression under
the effect of EHF EMR are demonstrated in Fig. 7. For cases of partial regression of Pliss lymphosarcoma with a size of more than 10 cm (group 2) typical was formation of “botryoidal” structure of regressing tumor, as well as formation of a capsule, which clearly marks the boundary between the tumor and the neighbouring tissue. Thus, mobility of formation and absence of its invasion into skin was provided (Fig. 7B). In cases of complete regression of Pliss lymphosarcoma some kind of tumor “drying” occurred, at the same time a little invasion to the skin in a form of local injure (wound) in microvessels of skin outer layers has been observed (Fig. 7C).

As in case with the RR application, the anti-tumor effect of the modulated EHF EMR manifested itself only against the background of anti-stressor action of these factors. 2 weeks after the detectable appearance of the tumor in all the reference group tumor-bearing rats the AR stress has been noted, whereas in the main groups the anti-stressor AR have been observed in more than a half of the cases.

Since then, the animals in main groups 1 and 2 have been divided into 3 subgroups, namely A, B, and C-subgroups depending on the EHF EMR effect intensity with different mode of modulation. 6 male rats with complete tumor regression have been included in subgroup A – 3 animals from group 1 and 3 animals from group 2. Subgroup B consisted of 7 animals, of which 3 female rats with evident tumor regression were from group 1 and 4 male rats with
partial lymphosarcoma regression with a size of more than 10 cm³ were from group 2. Subgroup C included 6 rats from group 1 and group 2. Those rats had no reduction in tumor size compared with the reference values. They were more active than the animals in the reference group having developed during the last 1.5 weeks of the experiment not only the AR stress, but also tensed anti-stressor ARs as well as absence of evident hypoplasia in thymus (Fig. 8).

In our opinion, rats with an evident EHF EMR effect, included into subgroup A and B, differed in degree of mobilization, type and phases of the immune anti-tumor processes development. In subgroup A the realization of anti-tumor effect modulated by EHF EMR, evidently, has been carried out to the full extent compared with subgroup B. Thus, rats in this subgroup differed from rats in subgroup B in significantly higher (2–4 times) activity of lymphoproliferative process in thymus (Fig. 8). Animals from subgroup B had an evident atrophy of some thymus lobules, whereas in other lobules the hypoplasia in the organ lymphoid parenchyma has been significantly less evident as compared with the reference group. At the same time the weighting factor of thymus has been significantly reduced in comparison with the rats in subgroups A and C (Fig. 8). The presence of evident contacts between tissue basophils and thymocytes (not less than 3–4 in majority of visual fields with magnification 400) has been observed in all the animals in subgroup A, whereas in subgroup B in some cases under partial tumor regression such contacts were single.

Differences between the animals in subgroup A and B were also noted when studying blood leukocytes functional activity (Table 1 and 2). The lowest activity of enzyme-dehydrogenases in lymphocytes (Table 1) and the highest phagocytic activity of neutrophils and monocytes (Table 2) have been noted in male rats in subgroup A. In Subgroup B the values of neutrophils phagocytic activity indicators have been the lowest, the decrease in monocytes phagocytic activity and leukocyte content in peripheral blood as compared with subgroup A (Table 1 and 2) have been observed. At the same time in both subgroups of animals with effective EMR action the indicator of the α-GPDH enzyme activity has been reduced by 22–29 % in comparison with the values in the reference group (Table 1).

In our opinion, the data on differences in the considered indicators in subgroup A and B animals, demonstrated in Table 1 and 2 and in Fig.8, may denote the phase changes in the blood thymocytes and leukocytes state, which accompany the anti-tumor effect of the modulated EHF EMR.

It is known that the accidental involution of thymus may be caused by not only lymphoproliferative processes inhibition but also tEMRorary migration of activated thymocytes from thymus due to the immune processes stimulation [16]. We can assume that on certain stages of the EHF EMR anti-tumor effect realization the migration of thymocytes occurred in tissue owing to their necessity to carry out systemic and/or local reactions.

Such an activation of the lymphocytes regulatory functions in tissues could evidently be the reason for decrease in weighting coefficient and thymus cortical substance width, reduction of total blood leukocytes content in animals of subgroup B, as well as a decrease in the α-GPDH enzyme activity in lymphocytes of the male rats in both considered subgroups, which is known for its close relation to realization of the regulatory functions [17]. After the tissue processes with participation of lymphocytes finished, there probably
occurred the restoration of thymocytes pool, thymus mass, and leukocytes level in blood (subgroup A). Herewith the restoration of the blood lymphocytes functional state could last longer than replacement of the lymphocytes pool in blood and thymus, that resulted in the lowest SDH activity in lymphocytes of the subgroup A animals, which experienced the complete realization of the considered potential.

A relatively high phagocytic activity of monocytes and neutrophils in blood of the subgroup A animals (Table 2) could be a reflection of mobilization of other, not lymphocytic, parts of the effector anti-tumor mechanisms influenced by EHF EMR. Probably, the activation of monocytes and neutrophils in subgroup A was close to its maximum, that could be proved by relatively low values of their activity indices in NBT-test.

Large size of most of regressing tumors, synchronism, delay in approach and dynamics of regression, as well as absence of intoxication signs in animals suggested activation of tumor-specific immunity with the help of modulated EHF EMR and elimination of tumor cells by apoptosis. This result corresponded to the most successful variants of immunotherapy of different malignant tumors [18]. At the same time complete regression of relatively small tumors (up to 1.7 cm³) under the effect of EHF EMR with poly-frequency modulation was, on the contrary, caused by the processes not related to the tumor antigen presentation. Thus, the results of test on cytotoxic activity of natural killer blood cells with the use of K562 tumor cells culture indicated the highest value of the given indice exactly in these cases (cytotoxic index 5.8–6.0 when maximum value of the given indicator in other examined animals 4.6, p < 0.05).

Probably, just due to the fact that the exposure with the modulated EHF EMR started before the Pliss lymphosarcoma inoculation, in the organism of at least some of the animals it was possible to activate the mechanisms inhibiting the development processes of tumor avoidance from immune recognition and cytotoxic factor effect.

Conclusions

The obtained results allow speaking about evident changes in different segments of the immune system, accompanying or anticipating the anti-tumor effects of microwave range EMR, used as factors of the activation therapy. Besides, the effect of the studied low intensity EMR could cause the immunological processes development in 3 main directions related to the following:

- increase in differentiation of malignant cells leading to partial restoration of their structural-functional properties typical for normal cells, and a sharp decrease in their life cycle,
- development of the antigen presentation processes and further effective destruction of tumor cells by apoptosis induction,
- cytotoxic action of natural killer cells, macrophages, other immune system cells and their factors (Fig. 9).

Thus, the anti-tumor influence of the microwave range EMR as an activation therapy factor may probably be caused by different immune mechanisms. The obtained results allow defining the direction of further improvement of the activation therapy methods and development of effective anti-tumor exposures. The fact that the activation of anti-tumor resistance mechanisms is provided by the studied EMR anti-stressor effect allows speaking about the absence of restrictions when using EMR for patients with cardiovascular pathology.

Statement on ethical issues

Research involving people and/or animals is in full compliance with current national and international ethical standards.

Conflict of interest

None declared.

Author contributions

The authors read the ICMJE criteria for authorship and approved the final manuscript.

References

The Nobel Congress, the XI International meeting-conference of Nobel Prize Laureates and Nobelists "Science, technologies, society and the International Nobel Movement", devoted to the jubilees of the Nobel family famous inventions and the 30th anniversary of the International Information Nobel Centre (IINC), took place in Tambov from 24 to 28 October 2017.

The aim of the Congress is to expand and strengthen the mutual cooperation of Nobelists of Russia and foreign countries, summarize and determine the ways for the further development of Nobelistics and the International Nobel Movement, their integration into the world scientific and educational sphere.

The Congress organizers are the following: International Informational Nobel Center (IINC) (Russia, Tambov); Tambov Regional Branch for Nobelistics of the Russian Academy of Natural Sciences (Russia, Tambov); Russian New University (Russia, Moscow); Tambov State Technical University (Russia, Tambov); the non-public joint-stock company SALUS (Russia, Moscow) is the general sponsor of the Congress.

An active part in all the Congress activities was taken by the honorary guests, great-grandchildren of Ludvig Nobel and co-founders of the Nobel Sustainability Trust (NST) Professor Sven Michael Nobel (Sweden, Stockholm) and lawyer Sven Peter Emanuel Nobel (Sweden, Stockholm), as well as the Executive Assistant of the NST Chair, Miss Sweden 2015 Stina Nordlander (Sweden, Stockholm), Chairman of the Russian Union of Non-State Higher Educational Institutions Professor V.A.Zernov, leaders of Tambov and the Tambov Region, scientists from Austria, Germany, Israel, Italy, USA, etc. The total number of the Congress participants is 87 people from 11 countries of the world, 56 reports were submitted.

At the press conference on the first day of the Congress, the co-founders of the Nobel Sustainability Trust introduced the director-representative of NST in Russia V.M.Tyutyunnik, and for the first time officially announced the establishment of a new Nobel’s Prize on technologies of sustainable development and alternative energy. The NST management together with the NAO SALUS General Director E.A.Ivankov (Moscow) and the NST Director-Representative in Russia decided that the first award will be held in May 2019 in St.-Petersburg (Russia). The award committee (country and university are not determined so far) will function similarly to the Nobel Prize Committees on Physics, Chemistry, Physiology or Medicine, Literature and Peace. Nominations will be accepted from basic universities of the world, including three Russian universities.

The Congress also established the International Union of Nobelists, head-quartered in Tambov.

The XII Nobel Congress will be held in Tambov in August 2019.

Professor V.M.Tyutyunnik,
Chairman of the Organizing Committee of the Nobel Congress in Tambov, Russia
Morphologic correlates of the multimodal electromagnetic exposures efficiency (as exemplified by the experiment tumor growth)

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Aims
The aim of the present paper is to study the morphofunctional changes in the tumor tissue and peripheral zone in experimental animals under the influence of extremely high-frequency electromagnetic radiation (EHF EMR). Combined effect of EHF EMR and ultra-low-frequency magnetic field (ULF MF) without the use of special anti-tumor agents is to be studied as well.

Materials and methods
The experiments have been performed on 56 outbred male rats with inoculated sarcoma 45. Low intensity EHF EMR and ULF MF exposures have been carried out only on animals’ heads. Histological and morphometric study of the sarcoma 45 tumor tissue and its peripheral area has been performed.

Results
In case of the combined electromagnetic exposure (in 65% of cases) against the background of the sT able anti-stressor adaptation reactions an evident anti-tumor effect has been observed. Under effective combined electromagnetic action, lymphocyte infiltration of the tumor tissue and its peripheral zone was enhanced not only in comparison with the reference group, but also compared to the EHF EMR monoexposure.

Conclusions
Thus, significant enhancement of the tumor tissue infiltration by leukocyte cells under the combined effect of EHF EMR and ULF MF may indicate mobilization of the local immune mechanisms due to the activation of the neuroendocrine and immune system organs.

Keywords
Electromagnetic exposure, Anti-tumor effect, Tumor infiltration,
initiated using the Yav-1 device (42.2 GHz, 10 mW/cm²), the induction coil and the purpose designed modulator based on a special-shaped signal generator. Under the combined action the modulated millimeter radiation (EHF EMR) and the ultra-low frequency magnetic field (ULF MF) radiation of small induction (1 mT) has been simultaneously used. The frequency of the ULF MF varied in the range of 0.8–7.8 Hz in accordance with the EHF EMR modulation mode. The animal’s head has been exposed to the actions [1, 2]. The sessions have been conducted 4–5 times a week during 4 weeks.

The tumor volume has been determined 2 times a week. The tumor linear dimensions have been measured in three mutually perpendicular planes. Then its volume has been determined by the formula $V = (a \cdot b \cdot c) / 6$, where a, b, c are the linear dimensions of the tumor in cm, V is the tumor volume.

At the end of the experiments, the animals were sacrificed by means of an etheric over-narcotization. The 10% solution of neutral formalin and the Carnoy’s fixative have been used to fix the material. The tumor sections have been stained with hematoxylin-eosin and according to the Brache method. Morphometry of the tumor tissue and its peripheral zone has been conducted with the help of the automated measuring system “SAGA” on the basis of the PC. The study of the cellular composition of the tumor has been performed with an increase of x by 1000. The number of lymphocytes and plasmocytes in each 10th field of vision per 1000 cells has been determined in the tumor tissue (innc. 1000). In the peripheral zone of the tumor counted has been the number of macrophages, lymphocytes, plasmocytes, tissue basophils in each 5th field of vision, and then the average number has been determined.

As a signal indicator of the AR type and intensity, the blood leukocytes formula counted for 200 cells has been used [14, 17]. The AR condition has been determined before the beginning of the course of actions, during the experiment (1 or 2 times a week) and on the day of the rats slaughter.

The statistical analysis of the study results has been carried out with the help of t-criterion by Student and the Wilcoxon-Mann-Whitney criterion.

Result and discussions

During the experiment an active growth of C-45 has been observed in the reference group of rats. Under the action of the polyfrequently modulated EHF EMR as a monofactor, a slow (by 36 %, $p < 0.05$) inhibition of sarcoma 45 growth has been observed 2 weeks after the beginning of the exposures. This inhibition turned out to be Table and a week later the differences ceased to be reliable in comparison with the tumor size in rats of the reference group (Fig. 1).

In the group of animals receiving the combined electromagnetic effect (n = 20), a significant anti-tumor effect has been observed in a part of the rats. It has allowed distinguishing in the group two subgroups of animals that had significant differences in the dynamics of tumor size. In the first subgroup (EHF EMR + ULF MF1, Figure 1), consisting of 13 animals (65 % of animals in the group), a pronounced inhibition of sarcoma 45 growth and even tumor regression in 4 cases have been observed. In the 2nd subgroup (EHF EMR + ULF MF2, Fig. 1), consisting of 7 rats (35 % of animals in the group), no anti-tumor effect has been observed.

The histological examination of the tumor tissue in the reference group animals has showed marked signs of its intensive growth with violation of the capsule integrity and sprouting into the neighboring tissues. Only small groups of dystrophically altered tumor cells have been noted. The infiltration of the tumor tissue by the immune system cells has also been poorly pronounced (Fig. 2, Table 1).
Micropicture of sarcoma 45 under the EHF EMR action has indicated the development of degenerative-dystrophic changes in some cells and a tendency to decrease in their density that led to only an unstable inhibition of the tumor.

In cases of the most pronounced anti-tumor effect of the combined action, i.e. regression of sarcoma 45, significant growth of connective tissue infiltrated by immune competent cells has been observed at the site of the regressed tumor.

The data on the immune system cells, located in sarcoma 45 tissue in the studied group rats (Table 1), were of great interest. Both effects contributed to intensification in lymphocyte infiltration of the tumor tissue. The number of tissue-infiltrating immune cells has increased in both the EHF EMR and the combined electromagnetic effect groups.

Thus, in animals of the EHF EMR monoaction group the number of lymphocytes increased by 2.3 times, and the number of plasmocytes increased by 4.6 times as compared with the reference group (Table 1). Under the combined electromagnetic exposure in the 1st subgroup in cases of the tumor growth inhibition the number of lymphocytes in tumor has been 3.8 times higher compared with the reference, and 1.6 times higher than in the group where only the EHF EMR has been applied (Table 1). The plasmatic cell number has increased 4.7 times on the average under the EHF EMR monoaction. Under the combined exposure it has increased 6.7 times in the 1st subgroup in case of inhibition of the tumor growth and by 4.0 times in the 2nd subgroup of animals as compared with the reference (Table 1).

In the peripheral tumor area the largest number of lymphocytes, plasmocytes and tissue basophils has been observed under the combined electromagnetic exposure (EHF EMR + ULF MF) in the 1st subgroup of animals. In the reference group rats a minimal content of the immune system cells has been observed in the tumor periphery: single lymphocytes and low frequency tissue basophils; no plasmocytes and macrophages observed.

In the EHF EMR group the number of lymphocytes in the peripheral tumor area increased by 7.8 times as compared with the reference group. In the 2nd subgroup of animals receiving the combined exposure, the content of lymphocytes (and the tumor micropicture as a whole) has not differed significantly from the EHF EMR monoaction group (Table 2).

Under the combined exposure in the 1st subgroup, where the lymphocytes number has been maximal (Fig. 2), the given marker has 8.3 times exceeded the reference values and differed (reliably or on a tendency level) from the markers in the 2nd subgroup and the EHF EMR monoaction group (Table 2). In cases of the sarcoma 45 growth inhibition under the effect of EHF EMR and ULF MF in peripheral part of the tumor a tendency to an increase in the lymphocytes mean number in the field of vision has been observed.

Thus, the higher is the exposure efficiency the more significant is the content of lymphocytes in the tumor periphery (Fig. 3).

Besides, under the studied electromagnetic exposures observed has been appearance of macrophages and plasmocytes in the peripheral tumor area (Fig. 3), that has not been noted in the reference group.
Table 1. Markers of sarcoma 45 tissue infiltration in albino outbred rats by the immune system cells under the EHF EMR action and the evident tumor growth inhibition under the influence of combined electromagnetic exposure (EHF EMR + ULF MF)

<table>
<thead>
<tr>
<th>Marker</th>
<th>Reference (n=16)</th>
<th>EHF EMR (n=20)</th>
<th>EHF EMR + ULF MF</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Weight of tumor, g</td>
<td>16,5±1,5</td>
<td>13,6±1,0</td>
</tr>
<tr>
<td>Tissue of tumor</td>
<td>Number of lymphocytes (%)</td>
<td>34,54±2,63</td>
<td>80,82±10,47 ●</td>
</tr>
<tr>
<td></td>
<td>of tumor</td>
<td>0,74±0,42</td>
<td>3,45±1,08 ●</td>
</tr>
</tbody>
</table>

Note: Differences in relation to: reference ● – p<0,05–0,001; from the EHF EMR group: ▼ – p<0,05; from subgroup 2 EHF EMR + ULF MF: ▼ – p<0,05, Field of the vision area – 6787,12 μm². EHF + ULF MF is a subgroup of animals with an evident anti-tumor effect.

Table 2. The content of immune cells in sarcoma 45 peripheral area in albino outbred rats under the EHF EMR action and the evident tumor growth inhibition under the influence of combined electromagnetic exposure (EHF EMR + ULF MF)

<table>
<thead>
<tr>
<th>Number of immune cells in the field of vision</th>
<th>Reference (n=16)</th>
<th>EHF EMR (n=20)</th>
<th>EHF EMR + ULF MF</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Weight of tumor, g</td>
<td>16,5±1,5</td>
<td>13,6±1,0</td>
</tr>
<tr>
<td></td>
<td>Macrophages</td>
<td>0</td>
<td>4,5±0,9 ●</td>
</tr>
<tr>
<td></td>
<td>Lymphocytes</td>
<td>3,2±0,3</td>
<td>23,5±1,1 ●</td>
</tr>
<tr>
<td></td>
<td>Plasmocytes</td>
<td>0</td>
<td>4,6±0,6 ●</td>
</tr>
<tr>
<td></td>
<td>Tissue basophils</td>
<td>0,16±0,06</td>
<td>0,21±0,04</td>
</tr>
</tbody>
</table>

Note: Differences in relation to: reference ● – p<0,05–0,001; from the EHF EMR group: ▼ – p<0,05; from subgroup 2 EHF EMR + ULF MF: ▼ – p<0,05. Field of vision area – 6787.12 μm². EHF EMR+ULF MF – combined action of poly-frequency modulated EHF EMR and ultra low frequency magnetic field (ULF MF).

The plasmocytes content in the rats of the 1st subgroup with the tumor growth inhibition under the effect of the combined exposure has exceeded the number of these cells in the field of vision by 27 % and 41 % in the animals of subgroup 2 and the EHF EMR monoaction group, respectively (Table 2). The macrophage content in the tumor peripheral part has not reliably differed in the observed groups. At the same time it should be noted that this marker has reached its maximum value (up to 10 cells in the field of vision) in rats with tumor regression under combined action of EHF EMR and ULF MF.

The tissue basophil content in the tumor peripheral area has reliably increased as compared with this marker value in the reference group only in cases of effective combined electromagnetic exposure (subgroup 1). The mean number of tissue basophiles (in the field of vision) in animals with tumor growth inhibition under the influence of combined exposure has been almost 3 times higher than in the reference group rats, and more than 2 times exceeded this marker in the 2nd subgroup rats and the animals exposed to only EHF EMR (Table 2).

Conclusions

Thus, the obtained results indicate high anti-tumor efficiency of electromagnetic exposures under their use in the activation therapy modes. Low intensity and localization of exposures (to animal’s head) precluded the possibility of their direct damaging effect on tumor cells. At the same time significant enhancement in the tumor tissue infiltration by leukocyte cells may denote mobilization of the local immune mechanisms due to activation of neuroendocrine and immune system organs.

The tumor tissue histological and morphometric analysis under the studied exposures indicate their mediated damaging effect on sarcoma 45 cells. Moreover, the most significant effect has been noted under combined use of...
EHF EMR and ultra low frequency magnetic field. At the same time the qualitative and quantitative composition of the immune system cells infiltrating the tumor tissue has corresponded to the anti-tumor effect intensity. The appearance under effective exposures of plasmocytes and macrophages, which have been practically absent in the reference group animal tumors, as well as a reliable increase in tissue basophil content, may indicate immediate participation of the noted immune system cells in the mechanisms leading to tumor cells damage.

Thus, combined electromagnetic exposure (in case of appearance of s. Table increased activation AR), as opposed to the EHF EMR monoaction, has provided effective regulatory influences of the neuroendocrine system structures on the immune system ones and led to evident tumor tissue damage, mediated by the immune system effector elements.

Statement on ethical issues
Research involving people and/or animals is in full compliance with current national and international ethical standards.

Conflict of interest
None declared.

Author contributions
The authors read the ICMJE criteria for authorship and approved the final manuscript.

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Pathogenetic substantiation for anti-tumor effectiveness of experimental electromagnetic therapy

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Aim
The objectives to be pursued by our studies are to assess some effects made by experimental actions of ultra-low frequencies electromagnetic field (ULF EMF) in combination with the SCENAR therapy on morphological changes in lung tissues and the regulation structures considering tumor growth dynamics.

Materials and methods
We investigated sex hormone and prolactin concentrations in 156 male rates of outbred strains in the tumor growth dynamics in tissues of the lungs and the hypophysis, and we also studied morphological changes in the lung tissue. All animals were divided into two groups: the reference group, covering the rodents not affected by any exposures, and the experimental group of the rats, who were subjected to ULF EMF exposure in combination with the SCENAR therapy. Both therapeutic exposures were applied upon expiration of 1 week after tumor cell inoculation in a lung.

Results
The applied combination of the experimental therapy, involving the central and local electromagnetic actions, has resulted in the reliably established prolongation of life spans in tumor-bearing animals as well as in the negative dynamics in tumor growing; it has produced effects both on the hormonal homeostasis subsystems: the hypophysis and the lung tissue affected by the cancer tumor. Histological examination of the prepared tissue samples taken from the regressing tumor in the lungs has demonstrated that there has been a certain amount of the tumor cells, which have been excluded from proliferation with taking an opportunity to differentiate.

Conclusions
The central action produced by ULF EMF is a trigger responsible for the neuro-hormonal regulation that promotes a temporal manifestation of an anti-tumor effect and that in combination with the SCENAR therapy provides for the compensatory-adaptational re-setting of the respiratory system and prolonged life maintenance in the animals.

Keywords
ULF EMF, SCENAR, Experimental tumor in lungs, Anti-tumor effect, Morphology, Sex hormones, Prolactin, Hypophysis

Imprint

Introduction
The pathogenetic mechanisms of progression of a tumor disease are determined by the possibility of coordination and integration of re-setting of homeostasis in a tumor-bearing organism which is governed by the central regulation of the processes at the organ-, cell- and molecule-related levels [1, 2]. Therefore, designing therapy strategies based on these assumptions is most promising in applied oncology.

A biological groundwork for the phenomenon of high sensitivity of an organism to low-intensity factors of electromagnetic and electrical nature is that their deficient influence intensity is compensated by gaining a signal character by them; in doing so, a re-coding of the information carrier takes place at the level of the neuro-humoral mechanisms [3, 4]. Much evidence in reference literature shows that there is an anti-tumor effect detectable that is made by electromagnetic influences applied in experimental studies, along with their use for complex therapy in oncology treatment practice [5–8]. So, some research work has demonstrated that the effects produced by ULF EMF on the central structures in the brain in the post-surgery period in lung cancer patients contribute to a reduction in the complication occurrence and to a re-
liably established prolongation of the 3-year survival span [9]. In this case, we observe normalization of the regulatory function of the hypophysis and the epiphysis that is supported by indicators of the hormonal activity of the thyroid glands and the androgenic areas in the adrenal glands and gonads.

Applications of ULF EMF in complex treatment of colorectal cancer patients have made it possible to considerably improve the two-year survival rates after radical surgery and prolong an average life span in cases of palliative surgical interventions for localized tumor processes [10].

Use of the adjuvant chemotherapy supported by ULF EMF focusing on the brain for treatment of malignant glial tumors in our practice allowed us to obtain an immediate clinical effect for 93,3 % of the patients, reliably establish the two year recurrence-free survival rate and minimize symptoms of neurological toxicity in them [11].

Previously we mentioned that we had revealed the role of a change in the hormonal homeostasis indicators of the lungs and the hypophysis in the dynamics of progression of inoculated malignant tumor in the lungs, along with the morphological correlates of development of the experimental tumor in the lung tissue [12, 13]. The pathogenetic significance of the said factors may be confirmed by a pronounced efficacy due to their anti-tumor effect potentiality.

In this context, it should be stated that the aim of our offered research work was to assess effects produced by experimental ultra-low frequency magnetic fields in combination with the SCENAR therapy, considering morphofunctional changes in the lung tissue and in the regulatory structures in the tumor growth dynamics.

Materials and methods

In sets of our experiments used were 156 male albino rats of outbred strains. Malignant tumors developed in the animals after they had been given a single intravenous injection of sarcoma 45 cells into their subclavian veins. Taking into account the tumor growth dynamics (upon expiration of 1, 3, 5 and 45 weeks from inoculation), we examined the morphofunctional changes in the lung tissue. With the use of the method of an immunoferment analysis we quantified progesterone, testosterone, estradiol and prolactin in the lung tissue as well as gonadotropins: luteinizing and follicle stimulating hormones (LSH and FSH, respectively) plus prolactin in the hypophysis tissue. All animals were divided into two groups as follows:

Group 1 (the reference group, n = 54) showing tumor growth without any influences or exposures applied;

Group 2 (the test group, n = 102) to cover the rodents exposed to ULF EMF in combination with SCENAR.

The experimental therapy was carried out by ULF EMF acting on the brain (with the use of device Gradient-2 manufactured by R&D Company PULSE at All-Russia Research Institution GRADIENT, Rostov-on-Don, Russia), in a discrete sequence of frequencies 0,03 Hz – 0,3 Hz – 9 Hz with the a signal-related exposure of 5 minutes – 1 minute – 1 minute, respectively, and at an intensity of 5 mT – 3,5 mT – 2,8 mT, respectively, the values of which were decreased exponentially by a factor of 0,7. Upon expiration of 15-20 minutes after completion of each magnetotherapy session, the skin areas in the projection of the lungs and the sternum were treated with a pulse electric current supplied by the SCENAR equipment (manufactured by Closed-End Joint Stock Company “R&D Department RHYTHM”, Taganrog, Russia) producing varying frequencies from 15,3 Hz to 33,6 Hz. Both treatment actions were started in 1 week after the S 45 tumor inoculation into the lungs, and all treatment sessions, performed usually at 9 or 10 am., covered 5 weeks (4 times a week).

All manipulations in animals in experimentation, including sacrifice procedures, were in accordance with applicable rules, guidelines and regulations prescribed by European Convention for the Protection of Vertebrate Animals used for Experimental and Other Scientific Purposes, Strasbourg, 18.III.1986. Statistics data were processed with the use of our original Software Statistica 10.0 with performing Student’s t-test (p < 0.05).

Results

The results of our analysis show that the applied influence has led to a reliably established prolongation of a life span in the animals affected by the growing malignant tumor in the lung (see Table 1 herein). The animals within the applied therapy test group, belonging to the cohort with a favorable effect, demonstrated a life span 9 times longer than that recorded in the reference group and 3 times longer than that found in a part of the reference group showing no therapy effect. Differences between the reference group and the individual subgroups within the experimental test group were also detected in the sizes, the number of
the regional nodes involved and timing of the node detectability. We found that the detectable involvement of the regional nodes in 33.3% of the animals in the test group took place 2–3 weeks later as compared to the time in question in the rodents in the reference group. It should be mentioned that the tumor-affected nodes in the majority of the test group animals could not be visually identified over the course of the entire experiment.

When carrying out morphological examinations of the lung tissue upon expiration of 1 week from the time of inoculation with Sarcoma 45, i.e. prior to the therapy action (the ULF EMF and SCENAR therapy), it was found that the primary alterations in the tissue in the upper and bottom lobes in the right and left lungs are the same as it was the case with the micro-picture identified in the animals in the reference group. A first triggering mechanism of a pathologic influence exerted by tumor cells on the lung tissue might be a reduced blood flow rate and ample large-focal hemorphages. Actually, we observed that the entire tissue of the lung was completely soaked with blood, and the interalveolar spaces were filled with a large body of erythrocytes and lymphocyte clusters (see Figure 1 herein). As a result, due to such “an invasion attack” some large-focal dystelectasis areas, interstitial pneumonia and microabscess areas in alveolar spaces were made evident.

In week 2 upon inoculation with the tumor cells (the beginning of the ULF EMF and SCENAR treatment), in the upper and bottom lobes observed was a degradation of the bronchiol and alveolar structures not only due to large-scale blood cell aggregations, but mainly due to an egress of tumor cells, leaving the blood vessels, and their growing as thin prolonged filamentous extensions (see Figure 1b herein). Formation of large tumor cell clusters took place, which demonstrated a curved boundary line separating them from the blood-soaked lung tissue (see Figure 1c herein). Formation of a tumor node was characterized by a compression and destruction of the lung tissue structures, closely packed tumor cells, which retained the typical S-45 spindle shape and maintained their high proliferative potential that was evidenced by abundant pathological mitotic figures (see Figure 1d given herein).

The morphological evidence for the fact that the true tumor process was in progress in the lungs in the rats came from the successive short-time going of the tumor cells spread by way of the blood hematogenous route to formation of tumor cell populations in the form of thin extensions and, finally, in the form of a completely formed tumor node.

When continuing the experimental treatment, as designed by us, we observed some destructive alterations in the tumor node structures in the lungs. Starting with week 3 upon the inoculation with S-45, we observed the mobilization of the immune system at the cellular level and formation of a condensed circumferential structure made by matured lymphocytes along the periphery of the tumor node (see Figure 2a herein). Lymphoid cells in concert with histiocytes, macrophages and fibroblasts penetrated into spacing between some individual clusters of the tumor cells, which had their common boundary line. In other

<table>
<thead>
<tr>
<th>Parameter, indicator</th>
<th>Reference group (exposure-free) (n=22)</th>
<th>Test group (upon EMF exposure)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Life span, days (weeks)</td>
<td>38.5±3.5 (5–6 weeks)</td>
<td>108.5±3.5 ¹ (15–16 weeks)</td>
</tr>
<tr>
<td>Time of tumor node detectable involvement after S-45 inoculation, weeks</td>
<td>3–4</td>
<td>5–6</td>
</tr>
<tr>
<td>The number of the regional detectable tumor nodes involved, pcs.</td>
<td>5–7</td>
<td>Up to 6</td>
</tr>
<tr>
<td>Sizes of timing-related detectable involved tumor nodes, cm³</td>
<td>0.5±0.05</td>
<td>0.4±0.05</td>
</tr>
</tbody>
</table>

Notes: ¹ – statistically significantly reliable in relation to data on the exposure-free group (p≤0.05); ² – statistically significantly reliable in relation to the no-effect group upon completed exposure (p≤0.05).
Fig. 1 а–d. Lung tissue in a rat inoculated with Sarcoma 45 upon the ULF EMF and SCENAR treatment: а) a large-focal hemorrhage, interstitial pneumonia in progress, microabscesses and dystelectasis, upon expiration of 1–2 weeks from the date of inoculation; b) degradation of bronchiol-alveolar structures, egress of tumor cells leaving the blood vessels and formation by them of thin prolonged filamentous extensions; c) formation of large-sized tumor cell clusters with a curved boundary line; d) formation of a tumor node with compressed lung structures. Hematoxylin and eosin staining. Magnification x40.

words, one of the primary markers of the control influence made on the malignant tumor localized within an organ should be a disruption of the territorial integrity of the tumor accompanied by formation of some micro-populations making clusters as a sort of syncytial tissues (see Figure 2b herein).

In the lung tumor tissue, there were sizable areas of individual or small groups of tumor cells identified, which were separated by some activated cells of connective tissue with a predominance of lymphocytes and fibroblasts, which attained various configurations: from a prolonged to a round one (see Figure 2c herein). We noted nuclear chromatin clumps in some individual areas within the clusters of bare hyper-chromated nuclei (see Figure 2d). The cells were turned to ghost-type structures with retaining their unclear-cut profiles of the melt cytoplasm (see Figure 2e herein). In weeks 5–6 we were able to identify some amorphous, optically homogenous, areas produced from materials due to lysing of the tumor malignant cells as well as due to growing of the new connective tissue within the lung tissue in the rats, who demonstrated a pronounced anti-tumor effect.

Under microscopic examination, identified were some individual groups of the tumor cells with signs of loss of the malignant character as follows: loss of predominance of the typical spindle-shaping and attaining a more round contour profile, with ordering the cells to form single-layer structures of linear- or follicular-type design (please, refer to Figure 3a, b herein).

An intensification of the processes of cell regeneration was noted in the lung tissue with a marked anti-tumor effect. The cellular sources of these processes were fibroblasts, alveolocytes of type II, macrophages, and endotheliocytes of blood capillaries. In our field of
vision we detected sizable areas filled with fibroblasts (see Figure 3c herein). In addition, we identified accumulations of alveolocytes of type II (pneumocytes of the IIInd order) representing an intensively secrete releasing, epithelial, and proliferating surface cell, the main component of which is dipalmitoylphosphatidylcholine (see Figure 3d herein).

The condition of some connective-tissue related elements like macrophages, mast cells, elastic, collagen and reticular fibers, suggested that some protection and compensation processes were being formed in the rat lungs affected by the tumor, under the effective treatment with ULF EMF and SCENAR.

When examining our specimens, we detected sometimes elastic structures of peri- and interlobular, intervascular, perivasal and peribronchial localization. The fibers demonstrated a layered order and exhibited their clear-cut excessive curvature pattern (see Figure 4a herein). An increase in the content of the reticular fibers indicated that metabolism processes in the lungs were intensified. Under a relative stabilization of the lung condition, the relationship among the fiber types was maintained due to an increase in the

![Image](image_url)

Fig. 2 a-e. Lung tissue inoculated with Sarcoma 45 in a rat, upon the ULF EMF and SCENAR treatment, status in week 3-4 from the date of inoculation: a) mobilization of immunocytes along the periphery of the tumor node; b) separation of tumor conglomerates by activated cells of connective tissue; c) disruption of intercellular contacts of tumor cells under penetrating lymphocytes and fibroblasts; d) clumping and fragmentation of nuclear chromatin in tumor cells; e) cytoplasm melting in tumor cells with their turning to “ghosts”. Hematoxylin and eosin staining. Magnification x40.
number of the collagen fibers referred to the elastic and reticular ones.

The stroma tissue was formed by fibers grouped into bundles due to the lateral aggregation at the bronchiolar structures (see Figure 4b). Following the run of the fibers, we observed some isthmi and flask-shaped protrusions: they were the false bronchi surrounded by accumulations of the bronchi-associated lymphoid elements (see Figure 4c). In the bronchi of the rat lung, there ciliated epithelium lined with the smooth muscle fibers forming the typical sublayer, basal cells layer and cylindrical epithelium (see Figure 4d) was retained.

An examination of the level of steroid hormones and prolactin concentrations demonstrated pronounced dependence of these parameters on the effectiveness of the experimental therapy as well as on the tumor growth time (see Table 2 herein).

Upon expiration of 3 weeks from the time of tumor inoculation, the prolactin concentration in the lungs in the rats without effect upon completed combined electromagnetic treatment showed no statistically significant difference from the same parameter, recorded for the reference group, and exceeded the normal values by 40 %. At the same time, while the prolactin level in the tissues of the lungs of the animals demonstrating an effect upon the experimental anti-tumor therapy was higher than the normal values by 28 %, it remained by 28,2 % lower than that determined in the lung tissues in the reference group rats. During the said test period, there was no significant difference in the levels of testosterone and estradiol concentrations in the lung tissue in the rats of the “no-effect” group from those in the reference group identified, and the said parameters were reduced by 57 % and 32.6 %, respectively, as compared to the normal values. We
had a completely different situation with the parameters recorded in the lung tissue in the rats responding effectively to the applied treatment: the levels of testosterone and estradiol concentrations remained within the range of the values identified in the intact animals.

Let us analyze the concentrations of progesterone in the two groups upon expiration of 3 weeks from the date of inoculation. In the lung tissue in rats, demonstrating the experimental therapy effect, the said parameter increased practically by 33 % referred to the values recorded in the intact animals and exceeded the same value identified in the reference group by a factor of 4,2. The progesterone concentration in the lung tissue in the “no-effect” group rats decreased by a factor of 1,9 referred to the normal values, but it remained by 62,9 % higher than that recorded in the reference group rodents.

The concentrations of prolactin and estradiol in the animals, demonstrated a favorable response to the anti-tumor treatment, within 5 weeks upon the inoculation, remained unchanged as compared to the previous test period, and the concentrations of testosterone and progesterone were reduced by a factor of 1,9 and by a factor of 1,8, respectively. As to the lung tissue in the “no-effect” group rats, it was the concentration of progesterone only, which changed as compared to the period examined before: it decreased by a factor of 1,9. It should be noted that in the period 5 weeks upon the inoculation with the tumor, the differences in the parameters under examination between the “effect-demonstrating” group rats and those in the “no-effect” group referred to the concentration levels of prolactin and progesterone only, whereas the concentrations of testosterone and estradiol identified in the lung tissue in either group were at the same levels.

Fig. 4a-d. Lung tissue inoculated with Sarcoma 45 in a rat, upon the ULF EMF and SCENAR effective treatment, status in week 5-6 from the time of inoculation: a) growing of elastic, collagen and reticular fibers with a excessive curvature pattern; b) lateral peribronchial aggregation of stromal elements like bundles; c) accumulations of bronchus associated lymphoid elements – BALT structure; d) preservation of ciliated epithelium in bronchi, lined by smooth muscle fibers, basal cells and cylindrical epithelium. Magnification x100. Hematoxylin and eosin staining.
Table 2. Concentrations of sex hormones in the lung tissue in male rats: the normal values vs. actual values in dynamics in rats upon inoculation with S-45

<table>
<thead>
<tr>
<th>Test period</th>
<th>Prolactin (ng/g tissue)</th>
<th>Progesteron (ng/g tissue)</th>
<th>Testosterone (ng/g tissue)</th>
<th>Estradiol (ng/g tissue)</th>
</tr>
</thead>
<tbody>
<tr>
<td>The background data</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(intact animals)</td>
<td>206.8±14.3</td>
<td>165.2±11.5</td>
<td>170.7±13.2</td>
<td>114.9±10.5</td>
</tr>
<tr>
<td>Reference group</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 3 upon inoculation</td>
<td>315.8±18.8</td>
<td>52.3±3.0</td>
<td>68.9±4.7</td>
<td>71.9±5.8</td>
</tr>
<tr>
<td>Week 5 upon inoculation</td>
<td>391.7±23.6</td>
<td>46.3±2.8</td>
<td>46.4±4.3</td>
<td>46.1±3.7</td>
</tr>
<tr>
<td>“Effect-demonstrating” group</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 3 upon inoculation</td>
<td>265.2±11.7</td>
<td>219.5±17.2</td>
<td>152.5±13.2</td>
<td>93.0±8.8</td>
</tr>
<tr>
<td>Week 5 upon inoculation</td>
<td>267.3±18.1</td>
<td>120.1±9.7</td>
<td>81.6±7.3</td>
<td>83.4±7.3</td>
</tr>
<tr>
<td>“No-effect” group</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 3 upon inoculation</td>
<td>289.4±9.3</td>
<td>85.2±4.5</td>
<td>73.4±1.1</td>
<td>77.4±5.4</td>
</tr>
<tr>
<td>Week 5 upon inoculation</td>
<td>328.3±26.1</td>
<td>45.6±1.9</td>
<td>74.1±6.3</td>
<td>77.1±3.2</td>
</tr>
</tbody>
</table>

Notes: 1 – statistically significant in relation to the background data (p<0.01); 2 – statistically significant in relation to the reference group data (p<0.01); 3 – statistically significant in relation to the data of the previous period of experiment (p<0.01); 4 – statistically significant in relation to the data of the “effect-demonstrating” group (p<0.01).

Table 3. Concentrations of hormones in hypophysis tissue in male and female rats within the norms and those identified for dynamics of tumor growth upon S-45 inoculation

<table>
<thead>
<tr>
<th>Test period</th>
<th>Prolactin (%)</th>
<th>LH (%)</th>
<th>FSH (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Background data</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(intact animals)</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Reference group</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 3 upon inoculation</td>
<td>217.4±11.9</td>
<td>173.7±12.2</td>
<td>35.4±2.1</td>
</tr>
<tr>
<td>Week 5 upon inoculation</td>
<td>63.6±7.7</td>
<td>233.3±17.5</td>
<td>16.8±1.2</td>
</tr>
<tr>
<td>“Effect-demonstrating” group</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 3 upon inoculation</td>
<td>132.4±9.2</td>
<td>90.8±8.7</td>
<td>80.7±5.6</td>
</tr>
<tr>
<td>Week 5 upon inoculation</td>
<td>135.1±4.5</td>
<td>85.4±1.2</td>
<td>43.6±1.2</td>
</tr>
<tr>
<td>“No-effect” group</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 3 upon inoculation</td>
<td>179.4±15.8</td>
<td>142.9±10.3</td>
<td>59.6±2.2</td>
</tr>
<tr>
<td>Week 5 upon inoculation</td>
<td>79.2±5.6</td>
<td>173.3±13.4</td>
<td>30.1±2.7</td>
</tr>
</tbody>
</table>

Notes: 1 – statistically significant in relation to the background data (p<0.01); 2 – statistically significant in relation to the reference group data (p<0.01); 3 – statistically significant in relation to the data of the previous period of experiment (p<0.01); 4 – statistically significant in relation to the data of the “effect-demonstrating” group (p<0.01).
Another intriguing fact was that we observed some alterations in the hypophysis in rats upon inoculation of a malignant tumor into the lung under the experimental therapy (see Table 3 herein).

So, upon expiration of 3 weeks after the tumor inoculation, the concentration level of prolactin in the hypophysis in rats, demonstrated a favorable response to the anti-tumor treatment, increased by 32.4% as compared to the same parameter in the intact animals, and in the reference group it was recorded to be more than doubled. A statistically significant change in the LH concentration within the indicated period of time was not detected, whereas it exceeded the normal values by 73.7% in the reference group. The concentration level of FSH in the hypophysis in the "effect-demonstrating" group rats, recorded 3 weeks after the inoculation, lowered by 19.3% as against 64.6% in the reference group animals. Upon expiration of 5 weeks after the inoculation, it was established that it was the concentration of FSH in the hypophysis in the "effect-demonstrating" group rats upon completion of the anti-tumor experimental therapy that changed: it was by 46% lower than that found in the previous period; but at the same time, the value remained higher by a factor of 2.6 as compared to that in the reference group.

The animals in the "no-effect" group demonstrated the concentrations of prolactin, LH and FSH intermediate between the values recorded in the reference group and those in the group with a pronounced anti-tumor effect.

Conclusions
The proposed combined application of the experimental therapy based on the central and local electromagnetic action has exerted an effect on both of the constituents of homeostasis: the hypophysis and the tumor-affected lung tissue. It is likely that this has created such conditions of the hormonal "environment", when and where the tumor has not been able to grow and has experienced a structural involution. Under the histological examinations of the prepared samples, taken from the regressing tumor in the lung upon completion of the treatment, the fact has been noticed that a number of the tumor cells terminate proliferation and acquire a differentiated state. The mechanism of this transformation remains unclear, but it is known that in order to induce differentiation it is required to provide a pre-specified normal hormonal status. When compared the prolactin concentration level in the tumor-affected lungs in the rats of the "effect-demonstrating" group to that in the "no-effect" group, it is easy to note the normalization thereof in the first case considered and an essential increase in the level in the second case.

It is probably that the action adequate in its intensity and frequency, applied to the projection of the hypophysis, is capable of providing the stable character of its command issuing system responsible for the regulation of the level of prolactin and sex-related hormone concentrations, and the local treatment with SCENAR facilitates retarding of pathological stimulation of hormones in the tissue-related homeostasis.

The central action produced by ULF EMF is a trigger of the neuro-humoral regulation with increasing of the general and local anti-stress potential and the associated level of the organism resistance that promotes the temporal manifestation of the anti-cancer effect, and in combination with the SCENAR therapy it provides the compensatory-adaptational re-setting of the respiratory system and prolongation of a life span in general.

Statement on ethical issues
Research involving people and/or animals is in full compliance with current national and international ethical standards.

Conflict of interest
None declared.

Author contributions
The authors read the ICMJE criteria for authorship and approved the final manuscript.

References.
Use of physical factors of electromagnetic nature for decreasing complications in respiratory and cardiovascular systems in patients after surgical treatment of lung cancer

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Abstract
Surgical treatment of lung cancer is associated with a high risk of postoperative complications in cardiovascular and respiratory systems. Studied is the ability to prevent the postoperative complications in respiratory and cardiovascular system when exposed to ultra low frequency magnetic field in the given category of patients. To achieve this aim 126 patients, operated for lung cancer, have been exposed to ultra low frequency magnetotherapy on the brain occipital area during the early postoperative period. The Spectr-2 device for broad range magnetotherapy has served as an electromagnetic field source. Demonstrated is almost a two times decrease in a total number of postoperative complications as compared with the patients not subjected to the treatment. It has been established that using the electromagnetic nature factors leads to normalization in respiratory and cardiovascular activity and prevents development of many severe complications such as pulmonary embolism, acute myocardial infarction, disturbed cerebral circulation.

Keywords
Lung cancer, Magnetotherapy, Postoperative complications

Introduction
Lung cancer remains one of the most important medical and socio-economic problems in Russia and major developed countries. Nowadays surgical treatment is the main method giving hope for prolongation and improvement of life quality for lung cancer patients. Total number of complications and postoperative lethality in surgical treatment of lung cancer has always been considered high, and nowadays the frequency and probability of their development remain quite variable and depend on many factors. It is stated that in some cases an operative ablation of malignant tumor may cause acute stimulation of metastatic process due to traumatism of such operations [1–2]. This phenomenon is based on a number of mechanisms. The most important of them are stressor reactions which determine the nonspecific basis of pathology and aggressive treatment methods [2–3]. The correction of the mechanisms underlying pathological stress reaction may prevent the stimulation of metastasis, as well as reduce the probability of complications development in cardiovascular and respiratory systems and improve the results of tumor patients surgical treatment.

The phenomenon of anti-stressor effect of a weak ultra low frequency magnetic field (ULF MF) is established and becomes more and more applied in experimental oncology clinical practice. [4, 5, 11, 12]. When the specially developed ULF MF modes influence the brain hypothalamic area (CNS), the anti-stressor reactions are formed. These reactions elevate the general resistance of the organism including antitumor resistance [6–10].

The aim of the present paper is to study the ability to prevent the postoperative complications in respiratory and cardiovascular system when exposed to ULF MF during early postoperative period in lung cancer patients.

Materials and methods
At the RRIO thoracic surgery department analyzed are the data on 702 lung cancer patients after surgical treatment over a period of 7 years (1996–2003). 126 patients have been exposed to the ULF MF mag-
netotherapy on the brain occipital area during the postoperative period. The Spectr-2 device with microprocessor control of induction, frequency and exposure for broad range magnetotherapy has served as a source for electromagnetic fields in order to protect the organism from the operational stress action. The central ULF MF exposure (on a head) in accordance with the presence of locus of a high magneto receptivity of the signal in occipital area [6] has been carried out. ULF MF has been applied every day starting from the second day after surgery according to the developed program of a sequential one session long increase in frequency of magnetic field in the range of endogenous brain frequency rhythms within the framework of the following mode: 0.03 Hz – 5 min; 0.3 Hz – 3 min; 9 Hz – 1 min, with a fixed magnetic field (MF) intensity. The initial MF intensity has been 5 mT, and then it has been gradually reduced with intervals of 2–3 days to 0.8 mT taking into account the exponential law with transition coefficient equal to 0.7 for maintaining the stable adaptation anti-stressor reactions. Patients not exposed to the magnetic field (574 individuals) have been a reference group. Comparing these groups of patients we have studied the immediate results (complications, postoperative lethality) of the surgical treatment. In both groups male subjects prevail, the mean age is practically the same: 54.8 and 56.5 years. The patients have been distributed similarly according to the tumor process stage in both the 1st and the reference group. The volume of surgical interventions, including extended, combined, economic and test operations, has been the same in these groups. The quantity of performed pneumonectomies has prevailed (Table 1).

In the compared groups the patients with an evident concomitant pathology, mostly chronic lung and car-

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**Table 1. Volume and frequency of operations, performed in the surgical treatment group and at postoperative MT**

<table>
<thead>
<tr>
<th>Operation volume</th>
<th>Operation (n=574)</th>
<th>Operation + MT (n=126)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>absolute unit</td>
<td>%</td>
</tr>
<tr>
<td>Pneumonectomies</td>
<td>293</td>
<td>51.0±2.9</td>
</tr>
<tr>
<td>extended</td>
<td>188</td>
<td>66.2±2.0</td>
</tr>
<tr>
<td>combined</td>
<td>73</td>
<td>24.7±2.5</td>
</tr>
<tr>
<td>Lobectomy and bilobectomy</td>
<td>185</td>
<td>32.1±1.8</td>
</tr>
<tr>
<td>Economic resections</td>
<td>41</td>
<td>7.1±1.1</td>
</tr>
<tr>
<td>Trial thoracotomy</td>
<td>55</td>
<td>9.5±1.2</td>
</tr>
</tbody>
</table>

**Table 2. Distribution of patients depending on the concomitant diseases in the surgical treatment group and at postoperative MT**

<table>
<thead>
<tr>
<th>Pathology</th>
<th>Operation (n=574)</th>
<th>Operation + MT (n=126)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>absolute unit</td>
<td>%</td>
</tr>
<tr>
<td>Cardiac ischemic disease</td>
<td>174</td>
<td>30.3±1.9</td>
</tr>
<tr>
<td>Hypertensive disease</td>
<td>129</td>
<td>22.5±1.7</td>
</tr>
<tr>
<td>Chronic bronchitis</td>
<td>396</td>
<td>69.0±1.9</td>
</tr>
<tr>
<td>Pneumosclerosis</td>
<td>137</td>
<td>23.9±1.8</td>
</tr>
<tr>
<td>Silicosis</td>
<td>29</td>
<td>5.1±0.9</td>
</tr>
<tr>
<td>Gastritis, peptic ulcer disease</td>
<td>74</td>
<td>12.9±1.4</td>
</tr>
<tr>
<td>Diabetes</td>
<td>56</td>
<td>9.8±1.5</td>
</tr>
<tr>
<td>Obliterative disease vessels</td>
<td>42</td>
<td>7.3±1.1</td>
</tr>
<tr>
<td>Varicose vein disease</td>
<td>29</td>
<td>5.1±0.9</td>
</tr>
<tr>
<td>Other</td>
<td>149</td>
<td>25.9±1.8</td>
</tr>
<tr>
<td>Total</td>
<td>1215</td>
<td>211.8</td>
</tr>
</tbody>
</table>
The adaptation reactions have been identified according to the blood formula signal indices. The leucocytes cell composition has been defined by calculating 200 cells in Romanovsky-Giemsa-stained blood smears. The level of lymphocytes taking into account the content of eosinophils, monocytes and a total number of leucocytes as a reaction tension criterion has served as a signal criterion for the adaptation reaction.

**Results and discussions**

The analysis has demonstrated that in the group of postoperative exposure to alternating magnetic field observed is almost a two times decrease in a total number of postoperative complications as compared with the patients not subjected to such a treatment after operation (19.9 ± 3.5 % and 34.3 ± 2.0 %, respectively). Differences in the structure of complications in these compared groups have also been observed (Table 3). Noted has been the absence of complications related to the coagulability increase in the ULF MF postoperative exposure patients, i.e. no cases of pulmonary embolism (PE), myocardial infarction, cerebral circulation violation have been observed, whereas in the reference group after surgical treatment the total number of these complications has been 3.5 % (20 patients). Besides, in 7 patients (1.2 %) other thrombogenic complications such as superior vena cava and aortic abdominal thrombosis have been observed.

<table>
<thead>
<tr>
<th>Complications</th>
<th>Form of treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Surgical treatment (n=574)</td>
</tr>
<tr>
<td></td>
<td>Surgical treatment + MT (n=126)</td>
</tr>
<tr>
<td>Wound abscess</td>
<td>oсложнения</td>
</tr>
<tr>
<td>Bronchus suture failure</td>
<td>33 (5,7)</td>
</tr>
<tr>
<td>Emphysema</td>
<td>23 (4,0)</td>
</tr>
<tr>
<td>after pneumonectomy</td>
<td>19 (3,3)</td>
</tr>
<tr>
<td>after lobectomy</td>
<td>15 (2,6)</td>
</tr>
<tr>
<td>Residual cavity after</td>
<td>4 (0,7)</td>
</tr>
<tr>
<td>Pericarditis</td>
<td>13 (2,3)</td>
</tr>
<tr>
<td>Mediastinitis</td>
<td>3 (0,5)</td>
</tr>
<tr>
<td>Pneumonia</td>
<td>15 (2,6)</td>
</tr>
<tr>
<td>Cardiovascular insufficiency</td>
<td>31 (5,4%)</td>
</tr>
<tr>
<td>PATE</td>
<td>12 (2,1)</td>
</tr>
<tr>
<td>Myocardial infarction</td>
<td>5 (0,9)</td>
</tr>
<tr>
<td>Disorder of cerebral circulation</td>
<td>3 (0,5)</td>
</tr>
<tr>
<td>Other thrombogenic complications</td>
<td>7 (1,2)</td>
</tr>
<tr>
<td>Postoperative hemorrhage</td>
<td>5 (0,9)</td>
</tr>
<tr>
<td>Stress surgical complications</td>
<td>7 (1,2)</td>
</tr>
<tr>
<td>ulcerative hemorrhage</td>
<td>5 (0,9)</td>
</tr>
<tr>
<td>perforated ulcer</td>
<td>2 (0,35)</td>
</tr>
<tr>
<td>Acute retention of urine</td>
<td>1 (0,2)</td>
</tr>
<tr>
<td>Intestinal distention</td>
<td>17 (3,0)</td>
</tr>
<tr>
<td>Total</td>
<td>197 (34,3±2,0)</td>
</tr>
</tbody>
</table>
In lung cancer patients after surgical interventions quite often observed are the complications immediately related to the stressor action of the surgical operation itself. These are gastric and intestinal bleeding, perforated stomach and duodenal ulcers. The accompanying ULF MF-therapy preventive effect has been manifested in the following form. Firstly, in the group of postoperative ULF MF exposure patients such complications have not appeared, though 23 patients in anamnesis had stomach and intestinal diseases, namely gastritis, stomach and duodenal ulcer.

Secondly, in the group of postoperative magnetotherapy observed has been a decrease in the number of postoperative pneumonia and cardiovascular complications from 2.6% to 1.6% and 5.4% to 1.6%, respectively.

Thirdly, the most significant result has been a more than 2 times decrease in the postoperative lethality in patients exposed to magnetotherapy as compared to the patients not exposed to it (3.2 and 7.3%). It is logical to assume that decrease in lethality has been caused by the absence or reduction of a number of many above mentioned complications as well as improvement in their treatment results.

To control the mechanisms of lung cancer accompanying therapy the functional activity indicators of central nervous system (CNS), endocrine and immune systems has been studied. These indicators serve as physiological correlates for the ULF MF therapeutic action. In brain cortex stabilization of indicators of biopotentials spatial synchronization for alpha-and beta-rhythm ranges has been noted. Typical for oncology patients negative dynamics of slow-wave processes capacity has been 3.6 times reduced after conducting the poly-frequency therapy, that denotes direct correlation with a decrease in the oncology patients vegetative lability (Fig. 1).

Actually, according to the hormonal indicators, as opposed to the reference, the increased level of cortisol and adrenalin has normalized. The reduced melatonin-forming function of epiphysis and activity of thyroid glands and gonads have been restored (Fig. 2).

The cellular immunity indicators have normalized at the same time. The T-lymphocytes percentage has grown due to the helper-inductor cells which express marker CD4+ and the mitogen response of T- and B-cells to phytohemagglutinin (PHA) and lipopolysaccharide (LPS).

The adaptation reaction structure has been studied in lung cancer patients before the operation, as well as on day 1, 7 and 14 after the operation in the group of patients subjected to surgical treatment only (26 individuals) and exposed to the ULF MF after the operation (25 individuals) (Fig. 3).

Comparison of the initial adaptation reaction structure in the reference group patients has demonstrated that on day 1 after the operation the frequency of the acute stress development has 10 times increased, and the chronic stress has 6.5 times increased. The stress reaction part has been 88.5%, that is adequate to high traumatism of surgical intervention inducing the stressor effect. Despite the increased term of post-

Fig. 1. Some poly-system factors identifying possibility for increasing the non-specific and anti-tumor resistance

<table>
<thead>
<tr>
<th>α - rhythm</th>
<th>β - rhythm</th>
<th>θ - rhythm</th>
<th>Δ - rhythm</th>
</tr>
</thead>
<tbody>
<tr>
<td>before treatment</td>
<td>after treatment</td>
<td>before treatment</td>
<td>after treatment</td>
</tr>
<tr>
<td>1900</td>
<td>2180</td>
<td>60</td>
<td>90</td>
</tr>
<tr>
<td>before treatment</td>
<td>after treatment</td>
<td>before treatment</td>
<td>after treatment</td>
</tr>
<tr>
<td>307</td>
<td>92</td>
<td>1000</td>
<td>996</td>
</tr>
<tr>
<td>before treatment</td>
<td>after treatment</td>
<td>before treatment</td>
<td>after treatment</td>
</tr>
<tr>
<td>1700</td>
<td>2200</td>
<td>360</td>
<td>1000</td>
</tr>
</tbody>
</table>

Accurate intensification of spatial synchronization of cortex bio-potentials in α - band (p < 0.03), β - band (p < 0.05)
Fig. 2. Some indicators of the neuroendocrine and immune systems activity in the compared groups of lung cancer patients (reference, initial state, treatment with and without the ULF MF)
operative period, on day 7 after the operation in this group of patients the state of protective-compensatory mechanisms has not altered, they have remained suppressed due to dominance of chronic stress. Two weeks after the operation the frequency of the chronic stress occurrence has reduced by 19.3 % but still remained quite high (69.2 %). It can be explained by a prolonged transition from the acute form of this reaction into the chronic dominant one, as well as a rather low level of anti-stressor reactions.

The identification of the adaptation reactions by leukogram in the main group of the lung cancer patients has detected that a day after the surgical intervention the adaptive transition dynamics has been similar to the reference. This similarity consisted in total development of stress, which occupied 96 % of all the reaction structure. However, on day 7 after the operation, as opposed to the reference group where the above stressor dynamics of adaptive rearrangements remained, in the main group the acute stress development frequency reduced by 9 times, and the chronic stress development frequency reduced by 3.8 times as a result of the stressor reactions transition to the anti-stressor. On day 14 of the postoperative period in the group of patients exposed to the ULF MF formation of stable anti-stressor reactions has already been observed. The frequency of stress identification in the general structure of adaptive response has reduced by 17.3 times. The coefficient of correlation between the anti-stressor reaction and stress, as a reflection of the integral bioadaptive potential of the organism as a whole, has increased. Thus, the reliable intergroup differences in the frequency of the anti-stressor reaction development and the coefficient of correlation between the anti-stressor reactions and stress in identical terms of investigation indicate the evident regulatory type of the ULF MF exposures on the organism integral systems. These systems form the physiological archetype of the adaptive response aimed at increasing the active resistance, thus contributing to functional rehabilitation of lung cancer patients subjected to a traumatic surgical intervention.

**Conclusions**

The analysis of immediate surgical treatment results has demonstrated that the ULF MF application during the postoperative period allows reducing the total number of postoperative complications and lethality, eliminating the development of various severe postoperative complications in respiratory and cardiovascular system such as pulmonary embolism, acute myocardial infarction, cerebral circulation violation. The ULF MF application enables improving the course of postoperative period accelerating the process of patients’ rehabilitation due to correction of the adaptive reaction mechanisms and nonspecific resistance increase.

**Statement on ethical issues**

Research involving people and/or animals is in full compliance with current national and international ethical standards.

**Conflict of interest**

None declared.

**Author contributions**

The authors read the ICMJE criteria for authorship and approved the final manuscript.
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Theory of health: successful translation into the real
Influence of postoperative magnetotherapy on homeostasis central regulation efficiency

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Abstract
The world scientific literature is rich in sources considering pathological changes in the central nervous system (CNS) in case of tumor growth. The topical issue is the study of mechanisms providing a stable performance of the brain as the highest section of nervous system which largely predetermine the oncology patients life quality.

Aim
The aim of the present research is to study the dynamics of bioelectric markers of the CNS functional state in lung cancer patients during an early postoperative period for the purpose of evaluation of the conducted magnetotherapy adaptive and correcting effects.

Materials and methods
25 lung cancer male patients have been examined. The reference group patients received a standard postoperative therapy, while in the main group it has been accompanied by the complex modulated ultra low frequency magnetic field exposure applied to the occipital area. Before the operation and 10 days after it in all the patients EEGs have been recorded unipolarly, according to system “10/20” as well as the galvanic skin resistance measured in reference points using the R. Voll electro-acupuncture method.

Results
It is demonstrated that the postoperative magnetotherapy application in lung cancer patients is accompanied with a significant exceedance of the spatial synchronization values over the corresponding markers only in case of surgical treatment: in alpha band between the central and temporal area in the left hemisphere, and between the similar areas in the right hemisphere; in beta-band between the symmetrical areas in the temporal and central cortex. The observed compensatory-reduction processes in the lung cancer patients brain bioelectric activity correlate to a 1.8 times decrease in the frequency of asymmetries in the galvanic skin resistance values in 2 pairs of hypothalamus reference points, that may indicate a certain improvement in the organism higher vegetative regulation.

Conclusions
Thus, we may assert that the trend to the symmetrization of the spatial synchronization markers and the electro-acupuncture measurement results indicate a better preservation of the CNS functional state in the main group patients, that is the result of the weak low frequency magnetic field anti-stressor and protective effect on the brain.

Keywords
Magnetotherapy, Ultra low frequency magnetic field, Brain bioelectric activity, Cortical biopotential spatial synchronization, Electro-acupuncture, Hypothalamus, Homeostasis, Adaptation, Lung cancer

Introduction
Nowadays the effect of low frequency varying magnetic fields of weak intensity (ULF MF) is attributed to information-related exposures, which are effective remedies for the regulatory system state correction and the organism non-specific resistance increase [1, 2]. It is demonstrated that electromagnetic fields of weak intensity with the multi-frequency signal, which includes the frequencies of the brain endogenous rhythmic activity, have an anti-stressor effect and contribute to a rise in the organism non-specific resistance, including the anti-tumor one [3, 4]. The role of the ULF MF in the anti-tumor effect initiation
is related to a possibility of response rearrangement of the regulation central mechanisms, which forms the basis for directed correction of homeostasis.

According to A.A. Ukhtomsky, a biological rhythm, or a physiological interval of time, is a system-forming factor. Equalization of these physiological intervals, or lability, provides interaction between different structures, their incorporation in a certain working ensemble. Just the idea of A.A. Ukhtomsky about the isolability role in establishing the relations between separate nervous centers has allowed us to consider that the distribution over the cortical surface of the segments with similar bioelectric rhythmic activity on certain stages of the conditioned reflex system formation is a direct evidence of the identical course of processes in time, and reflects the mutual adjustability of neural structures permitting the possibility of their working connections. In relation to the above, the study of the cortical biopotential spatial synchronization processes under the effect of the complex modulated ULF MF is reasonable and urgent.

Besides, evaluation of the functional state of hypothalamus as a central controlling segment of the developing adaptation state and the most important center of vegetative regulation, as well as the organs and systems under its control, has provoked our great interest. It is well-known that examination of the reference points (RP) in the paired meridians of general measuring profile by the method of electro-acupuncture according to Voll (EAV) allows judging disbalances in the organism subsystems under malignant tumors of different localizations [6, 7]. Informativity of the method in evaluation of adequacy, tolerance and efficiency of the anti-tumor treatment [8, 9] has been demonstrated. The results of the EAV electrical parameters measurement in lung cancer have been obtained [10].

Thus, the aim of the present research is to study the dynamics of bioelectric markers of the CNS functional state in lung cancer patients during an early postoperative period for the purpose of evaluation of the conducted magnetotherapy adaptive and correcting effects.

Materials and methods
25 male lung cancer patients have been examined. The reference group patients (n=12) received a standard postoperative therapy, whereas in the main test group (n=13) it has been accompanied with the complex modulated ultra low frequency magnetic field exposures to the occipital area. Before the operation and 10 days after it in all the patients the EEG has been recorded and the galvanic skin resistance measured in reference points using the EAV method.

To record the EEG the brain electrical activity analyzer Encephalan-131-01, Medikom MTD, Taganrog, has been applied. The electrodes on the patient’s head have been placed according to international system 10/20, the biopotentials recorded unipolarly. The degree of interrelation between the neocortex areas (levels of spatial synchronization) has been defined according to the cross-correlation coefficient (CC), calculated for 16 intra- and interhemispheric pairs of leads: front (F), central (C), temporal (T) and occipital (O) in alpha- and beta-range frequencies. Studied have been the global and local SSCB. For the global SSCB the sum of correlation coefficients (CC) between all the cortical areas has been determined, and for the local SSCB the CC for separate segments of neocortex have been calculated. Relations between the global and the local SSCB are rather complicated and still not clarified. Therefore, it is recommended to perform a parallel control using both markers to better evaluate the human brain functional state [11].

Reflex-diagnostic complex Rista-EPD, OKB RITM, Taganrog, has been used to carry out the EAV measurements. Using this complex the evoked electrical conductivity (resistance) in reference points (RP) of skin surface has been measured. The loading galvanic skin test with constant current up to 12.25 µA and voltage up to 2.07 V has been applied for measurements. The result of interaction “stimulus-receptor” has been recorded in conventional units (c.u.). The obtained data are interpreted as deviations in the organ and system energetic and functional state appeared at the given moment and enrich available laboratory, instrumental and clinical results [6].

Results and discussions
When studying the total level of synchronization in alpha range it has been demonstrated that it has not changed in relation to the preoperative values, and in the end of the treatment course it has been identically evident in both main test group and reference group of patients. The analysis of this marker in beta-range has demonstrated absence of changes in the main test group and a tendency to reduction in the reference group by the end of the treatment course (p<0,1) (Table. 1).
Table 1. Dynamics of values of total level of synchronism of cortical bio-potentials of brain in lung cancer patients at different stages of treatment

<table>
<thead>
<tr>
<th>EEG rhythms</th>
<th>Stages of investigation</th>
<th>ULF MF, n=13</th>
<th>Reference group, n=9</th>
<th>ULF MF, n=13</th>
<th>Reference group, n=9</th>
</tr>
</thead>
<tbody>
<tr>
<td>alpha-rhythm</td>
<td>Preoperatively</td>
<td>6.80±0.35</td>
<td>6.63±0.42</td>
<td>6.60±0.43</td>
<td>6.32±0.44</td>
</tr>
<tr>
<td>beta-rhythm</td>
<td>Postoperatively</td>
<td>6.70±0.30</td>
<td>5.87±0.43</td>
<td>6.74±0.50</td>
<td>4.83±0.47*</td>
</tr>
</tbody>
</table>

Note: * – the difference on the level of tendency (p<0.1) as compared with the preoperative values in reference group for beta-rhythm.

The data obtained by M.N. Livanov [5] show that the global SSCB marker reflects readiness of a subject to act. However, it is not always possible to differentiate various functional states using the level of global SSCB. In these cases the local SSCB marker is used. Intensification of the local SSCB in the cortical areas, which are mostly specifically involved depending on execution of certain functions, is considered as an optimal functional state marker necessary for their effective realization. Besides, determinative may be not the change in total synchronism but the formation of foci of bioelectric process maximum synchronism.

In our investigation the analysis of the spatial synchronization marker in separate pairs of leads has allowed revealing reliably significant differences between the groups at postoperative stage of examination both in alpha- and beta- range of frequencies. It has been established that the values of synchronization between the leads in both main and reference groups have undergone slight changes in the end of the treatment. However, the direction of these changes in the compared groups has been exactly opposite, as a rule. As a result, on day 14 after the operation the differences in average group values of synchronization between some neocortex area biopotentials have reached a reliable level, the main test group having greater values at that.

Thus, in alpha range of frequencies a reliable difference in the marker values has been detected for symmetrical central temporal pairs of leads to right (C_3T_4) and left (C_4T_3) hemisphere. For pair C_3T_4 these values in the main test group have made 0.91 ± 0.03 as against 0.72 ± 0.08 in the reference group (p < 0.05), for pair C_3T_3 – 0.88 ± 0.02 and 0.78 ± 0.05, respectively (p < 0.05). Between the symmetrical temporal leads T_3T_4 observed has been similar correlation in values of the synchronization marker in the compared groups (0.33 ± 0.09 and 0.51 ± 0.07, respectively), however, only on the level of tendency (p < 0.1) (Fig. 1a).

The analysis of the spatial synchronization markers in beta-band has demonstrated presence of reliable differences between the groups in pairs of symmetrical central (C_3C_4) and temporal (T_3T_4) leads, as well as in the occipital temporal pair of the right hemisphere (O_2T_4). Thus, for pair of C_3C_4 leads the average group synchronization marker after the ULF MF course has value 0.54 ± 0.05 as against 0.41 ± 0.04 in the reference group (p < 0.05), for pair T_3T_4 the value is 0.25 ± 0.05 and 0.06 ± 0.04 (p < 0.05), for O_2T_4 – 0.38 ± 0.06 and 0.15 ± 0.09, respectively (p < 0.05) (Fig. 1b).

The detected peculiarities of the SSCB marker dynamics in pair of leads T_3T_4 for beta-rhythm have allowed us to assume that the tendency to increase in synchronization between the temporal areas of cortex, which has been outlined in alpha range of frequencies after the magnetotherapy course, has not been accidental. In our opinion, the tendency to symmetrization of the CC values, as well as to the increase in the spatial synchronization of cortical processes between symmetrical segments in the large hemisphere cortex, is a natural reflection of optimization in medullary processes caused by the poly-frequency electromagnetic exposure.

E.A. Soroka and coauthors accentuate the necessity for combined activity of temporal areas in both hemispheres for adequate implementation of complex organized functions [12]. According to D.M. Tsarapina and coauthors [13], intensification of interhemispheric relations of EEG, especially temporal and lower frontal segments, accompanies a coordinated cooperative activity of the left and right hemispheres. It is known that an extremely significant biological meaning is attached to pair work of large hemispheres, which consists in providing integral performance of the organism [14], determining a successful result of the self-regulation processes [15].

In the present study of the EAV markers special attention has been paid to the state of paired RP of hy-
pothalamus as a center, which carries out an important role in the development of adaptation reactions [16] and determines individual resistance to stressor actions [17].

The preoperative examination of patients in the studied groups using the EAV method has shown that the state of sympathetic and parasympathetic divisions of vegetative nervous system has been sufficiently balanced and expressed in normoergic response at a “hand-to-hand” lead. After the operation the normoergic state has changed to hypoergic, being evident to a greater or lesser extent in both groups of patients (Table. 2). As known, the hypoergic state corresponds to prevalence of parasympathetic part in regulation of vegetative functions of the organism [18].

The percentage of acute asymmetries in reference points of general measuring profile, which reflects a degree of deregulation in the organism bioenergetic state, under the stress action of surgical intervention has reliably increased as compared with the initial values only in the reference group of patients (р < 0,05). In the first group due to the protecting ULF MF action this growth has been insignificant (by only 2 % as against 75 % in the reference group) (Table. 2).

We have paid special attention to the asymmetries in pairs of hypothalamic points, which reflect the state of regulatory center of the organism adaptive capabilities. The frequency of occurrence of acute asymmetries in one or two pairs of measuring points in hypothalamus has changed (in relation to the preoperative values) depending on the treatment method. Thus, after the course of postoperative magnetotherapy the similar marker has decreased by 43 %, On the contrary, in the reference group after surgical interven-__tion the frequency of the paired asymmetry detection has significantly increased, exceeding the preoperative values by 34 % (Table. 2, Fig. 3).

The analysis of difference in absolute values of galvanic skin resistance between paired points of hypothalamus has demonstrated the similar direction of changes. In other words, when applying the ULF MF a reliable decrease in the marker almost by 45 % (р < 0,05) occurred, while in the reference group the growth of this marker has been 72 % as compared with the preoperative value (р = 0,05) (Table. 2, Fig. 4).

The data on energetic and functional state of the patients’ measuring profile organs and systems obtained using the EAV, allows concluding that, first of all, the positive results of the applied exposures are manifested on both the level of controlling part of adaptation, a hypothalamus, and the level of general measuring profile. The first ones are especially evident, probably because just the hypothalamic area of brain is a central part of reacting to electromagnetic fields. Secondly, application of relative (asymmetry in measuring points), not absolute (hand-to-hand) markers of oncology patient organs and systems energetic and functional state in EAV is the most informative.

Such methodological approach has already justified itself when trying to mediatly define a degree of prevalence of the tumor process in colorectal cancer, being under control or having lost control of regulation on the higher part of vegetative nervous system [19]. Taking into account the general pathogenetic basics of the tumor progression we may consider that in lung cancer the unfavorable predictive marker of

Fig. 1. Dynamics of the spatial synchronization markers after the ULF MF exposure for alpha- (a) and beta- (b) rhythm:
- reliably higher in the main test group than in the reference one (p<0,05);
- – higher in the main test group than in the reference one, on the level of tendency (p<0,1)

Fig. 2. Dynamics of percentage of acute asymmetries in the general measuring profile reference points
Table 2. Dynamics of the organism functional state markers according to EAV on the stages of surgical treatment of lung cancer patients

<table>
<thead>
<tr>
<th>Groups of patients</th>
<th>Investigated markers</th>
<th>Stages of investigation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Preoperatively</td>
</tr>
<tr>
<td>Reference</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(n=14)</td>
<td>Hand-to-hand (c.u.)</td>
<td>84,5±1,2</td>
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<tr>
<td></td>
<td>As. total (%)(^1)</td>
<td>17,7±3,5</td>
</tr>
<tr>
<td></td>
<td>As. Hypothalamus RP (%)(^2)</td>
<td>50</td>
</tr>
<tr>
<td></td>
<td>Δ Hypothalamus RP(^3)</td>
<td>3,1±0,5</td>
</tr>
<tr>
<td>ULF MF</td>
<td>Hand-to-hand (c.u.)</td>
<td>82,8±1,4</td>
</tr>
<tr>
<td>(n=14)</td>
<td>As. total (%)(^1)</td>
<td>34,4±6,4</td>
</tr>
<tr>
<td></td>
<td>As. Hypothalamus RP (%)(^2)</td>
<td>78</td>
</tr>
<tr>
<td></td>
<td>Δ Hypothalamus RP (c.u.)(^3)</td>
<td>6,9±1,2</td>
</tr>
</tbody>
</table>

Note: \(^1\) – percentage of acute asymmetries in general measuring profile; \(^2\) – percentage of patients with asymmetries in paired RP of hypothalamus; \(^3\) – difference in values of markers in paired RP of hypothalamus

the vegetative function disregulation is a high specific weight of acute asymmetries in both pairs of the hypothalamus measuring points. In its turn, we consider a decrease in their quantity as a sign of the regulatory process optimization under the effect of weak low frequency electromagnetic field.

Thus, we may assert that the spatial synchronization marker dynamics in alpha- and beta-range of frequencies, as well as the EAV results in the main test group patients indicate the better preservation of the CNS functional state resulting from anti-stressor and protective effect of weak low frequency magnetic field on brain.

Conclusions
The postoperative magnetotherapy application in lung cancer patients is accompanied with a reliable (p < 0,05) exceedance of the spatial synchronization values over the corresponding markers only in case of surgical treatment: in the alpha range of frequencies between the central and temporal area in left hemisphere, between the similar areas in the right hemi-
sphere, in the beta range between the symmetrical areas in the temporal and central cortex. The observed compensatory-reduction processes in the lung cancer patients brain bioelectric activity correlate to a 1.8 times decrease in the frequency of asymmetries in the galvanic skin resistance values in 2 pairs of hypothalamus reference points (according to EAV), that may indicate a certain improvement in the organism higher vegetative regulation under the effect of weak low frequency magnetic field.

Statement on ethical issues
Research involving people and/or animals is in full compliance with current national and international ethical standards.

Conflict of interest
None declared.

Author contributions
The authors read the ICMJE criteria for authorship and approved the final manuscript.

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Neurophysiological status and adaptation responses upon application of electromagnetic fields in complex treatment of patients with malignant gliomas of the brain

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Aim
The aim of this paper is to discover some advantages and benefits of the complex treatment of patients with malignant gliomas of the brain with the use of electromagnetic field therapy.

Materials and methods
Our examination focused on patients suffering from malignant gliomas of the brain. One cohort of them (the reference group covering 30 patients) received surgery and chemoradiotherapy (CHRT) without electromagnetic field exposure of the brain, while the other group (the main test group which included 30 individuals) was subjected to the same therapy, but accompanied by the electromagnetic field treatment of the brain. Varying and permanent magnetic field exposure sessions were carried out from day 1 till day 20 in the context of the complex therapy. All in all, every patient received 4 courses of the systemic chemotherapy, with an interruption of 4 weeks, in combination with exposures to electromagnetic field applied to the brain area. An assessment of efficacy of the adjuvant chemoradiotherapy was performed upon expiration of 12–14 days from the date of the last injection of chemotherapy drugs according to applicable WHO recommendations for standardizing in assessment of treatment outcomes.

Results
An application of the combined magnetotherapy, which includes both the central and local electromagnetic actions, in the treatment of malignant gliomas of the brain results in a statistically significant increase in the general and recurrence-free two-year survival, improves the scale grade level of the ECOG/WHO performance status grading and the Karnofsky index according to the Karnofsky performance status scale, provides for reduction of neurological toxicity symptoms and makes a pronounced anti-stressor effect, namely, leads to predominance of the reactions of training, calm and elevated activation in the framework of adaptation reactions of the physiological types.

Conclusions
The central and local actions by ultra-low frequency magnetic field (ULF MF) used in the therapy of malignant gliomas of the brain should be considered as a trigger of the neuro-hormonal regulation that is favorable for reducing toxicity influence by chemoradiotherapy on the brain and that is beneficial for improving life quality and prolonging a life span for the above patients.

Keywords
Neurooncology, Malignant gliomas of the brain, ULF MF, ECOG/WHO performance status, The Karnofsky index, Neurophysiological status, Adaptation reactions

Imprint

Introduction
Brain tumors range from 2 to 8 % of all neoplasms in total. It is known that recent international reports represent a picture of a considerable growth of the incidence rate of the brain tumors in populations throughout the world. As to Russia, we can state the incidence rate of the brain tumors is 10 cases per 100 000 population, with a half of the tumors located immediately inside the brain. By estimating the primary tumors of the brain in the adult population in Russia, statistics data show that gliomas of the brain accounts for 40–46 % of the cases, with 55 % of malignant tumors among them. Considering the glial tumors, the largest proportion is made by glioblastomas (16,7 %) as well as by astrocytic tumors (17,9 %), and in this case the malignant types of astrocytomas tend to occur 1,3 times more frequent.
ly in male individuals and 2 times more frequently in females as compared to the benign neoplasms [1, 2].

Within the last ten years, owing to drastic advancements in neuroanesthesia and significant improvements in microsurgery techniques, a great success has been made in the field of surgery of external brain tumors (ectopic pituitary adenoma, meningioma etc.), but the problem of therapy of patients suffering from malignant gliomas remains unresolved, and the recent results associated with the topical problem cannot be considered quite satisfactory [3–5]. An average life span of patients with malignant gliomas covers 12 months only, and the 1-year survival does not exceed 68%; the 2-year survival is not higher than 27%; and the 3-year survival is under 4.3% [6].

In the last few years a trend is evident to a wider use of magnetic fields as a component in the complex therapy of tumors, the mechanism of influence of which is directed both immediately on the malignant tumor and the adaptation and protection systems of the organism of the patient to be treated [7–9]. The action on the brain by magnetic field as the component of the complex therapy of the brain malignant tumors is viewed as reasonable, since it meets the most advanced requirements, which are placed for eligible therapy of malignant tumors of the brain, as indicated below: firstly, magnetic field acts on the perifocal glia producing cytostatic, anti-edema and anti-inflammatory effects, elevating efficacy and reducing toxicity of chemoradiotherapy; secondly, magnetic field exerts an influence on hypothalamus realizing the systemic effect to enhance non-specific and anti-tumor resistance of the organism of an affected individual [10–16].

Materials and methods

Our investigation was concerned with patients who suffered from malignant gliomas of the brain. One cohort of the patients (the reference group covering 30 individuals) received surgery and chemoradiotherapy without exposure of the brain to electromagnetic field, while the other cohort of the affected persons (the main test group consisting of 30 patients) received the same medical treatment, but accompanied by exposures to electromagnetic fields, targeted at the brain. The therapy procedure for the patients in the main test group was conducted as follows: upon the relevant morphological verification as the basis of the diagnosis, every patient with malignant glioma of the brain was exposed to magnetic field targeted at the brain, namely, the hypothalamus area: applied was ULF MF at frequencies of 0.03, 3 and 9 Hz, with an exponentially changing induction from 5 to 0.8 mT, with an exposure time from 7 to 1 minute; upon expiration of 20 minutes applied was permanent magnetic field with an induction of 20 mT, under an exposure time of 15 min, to cover the surgery-affected area of the patient. Later on, the treated patient was subjected to tele-gammatheapy with Cobalt isotope (Co 60) sources using equipment Agate-R or Rocus to cover the tumor bed in the regimen of the classic fractionation: a single focus dose of 2–2.6 Gy on the field, to the total focus dose of 60 Gy plus the systemic chemotherapy based on the use of Carmustine (BiCNU) in physiological sodium chloride solution – 150 mg/m² on day 1 & day 3, and Etoposide – 60 mg/m² on days 1–3.

Exposure sessions with applied varying and permanent magnetic field were carried out from day 1 to day 20 of the treatment in the same regimens and sequences. The patient received a total of 4 courses of the systemic chemotherapy with an interval of 4 weeks, in combination with sessions of exposure to electromagnetic field.

For the patients in the reference group, the chemoradiotherapy courses were completed by giving the same dose of gammatheapy and providing the identical doses of chemodrugs as it was the case with the main test group.

An evaluation of efficacy of the adjuvant chemotherapry was performed upon expiration of 12–14 days after the last injection of chemotherapy drugs in accordance with WHO Recommendations for standardized interpretation of the treatment outcomes in grading the evidence.

An objective treatment effect was assessed upon completion of course 4 of the therapy by studying the dynamics of tumor sizes according to data obtained by roentgen ray computed tomography with the use of Toshiba-Asteion VR 2002 equipped with Workstation Vitrea-2. The immediate results of the treatment were analyzed by investigations of the observed general two-year survival and the recurrence-free survival of the patients. The survival data were calculated according to the classical non-parametric statistic method, namely, the Kaplan–Meier estimator.

In doing so, diagnosed were the following most informative parameters:

1. The immediate and nearest outcomes of the new treatment method for patients suffering from ma-
lignant gliomas of the brain, as offered by us, versus the conventional chemoradiotherapy, using the Kaplan-Meier estimator for an eligible interpretation of the results.

2. Dynamics of the general performance and quality of life for the patients at stages of their specific treatment in the groups under study by tracing changes in their ECOG Performance Status and their Karnofsky Performance Status according to the ECOG/WHO numbering scale and the Karnofsky index between 100 and 0, respectively.

3. The pattern of the adaptation reactions was assessed before and after surgery in the early post-operative period as well as after chemoradiotherapy accompanied by magnetotherapy. Blood count was performed according to Giemsa-Romanowsky blood film staining technique for a 200 cell count. In addition to the statistics data required for an identification of the actual type of the adaptation reaction, we determined a common group-related pattern of the adaptation response with calculating the synthetic parameter: it was the coefficient of relation between the anti-stressor reactions and stress ($K = AC / C$) that made possible to properly estimate the efficacy of the action of the physical factor in question.

**Results and discussion**

An immediate objective clinical effect (remaining in remission) upon chemoradiotherapy, accompanied by the magnetic field treatment, was achieved in 28 of the 30 patients (93,3 ± 4,6 %) who received chemoradiotherapy and magnetotherapy that was by 2,3 times more often than it was the case with that group, where the patients were subjected to the conventional chemotherapy only (12 individuals – 40 ± 9,1 %). Progression of the disease was observed in the main test group in 2 patients only (6,7 ± 4,6 %), while in the group of patients, who received the conventional chemoradiotherapy only, we recorded 14 individuals with the progressing tumor (46,7 ± 8,5 %), i.e. it was 7 times more frequently as compared to the main test group. In the main test group, all individuals suffering from gliomas survived till the end of course 4 of the medication therapy, while in the reference group 4 persons died before the completion of the same therapy course (13,3 ± 6,1 %) (see Figure 1 herein).

Our analysis has shown that the recorded general 2-year survival in the patients of the main test group has reached 40,0 ± 8,9 % that is 2,4 times higher than that indicator recorded for the group subjected to the conventional chemoradiotherapy, which amounts to 16,7 ± 7,2 %.

Our investigation of the observed 2-year recurrence-free survival data for the patients with malignant gliomas of the brain, depending on the medication therapy, has demonstrated that the above mentioned indicator for those patients, who received chemoradiotherapy accompanied by magnetotherapy, has reached 20,0 ± 7,1%, i.e. it is 6 times less than that found in the main test group.

The median survival in the patients of the groups in question was reported to be as follows: in the reference group it was 9 months, and in the main test group it reached 16 months, i.e. it was established to be 1,7 times higher for the cohort received chemoradiotherapy and magnetotherapy (see Figure 2 herein).

The recurrence-free median in the patients of the considered groups was recorded to be as follows: in the reference group it was 6 months, and in the main test group we observed 12 months, i.e. the indicator was 2 times higher as compared to the reference (see Figure 3 herein). When analyzing the survival data, we obtained statistically reliable significant differences between the main test group and the reference cohort ($p < 0,05$).
Fig. 2. General 2-year survival data in patients suffering from malignant gliomas of the brain upon completion of chemoradiotherapy with magnetic field treatment versus chemoradiotherapy without magnetotherapy, based on the Kaplan–Meier estimator.

Fig. 3. Data on two-year recurrence-free survival of patients with malignant gliomas of the brain upon completion of chemoradiotherapy accompanied by magnetic field treatment vs. chemoradioterapy alone, based on the Kaplan–Meier estimator.
Our enquiries into the topics of the dynamics of the ECOG performance status grading and the Karnofsky indices in the patients under studies, which should be considered as parameters reflecting the subjective general response by an organism to chemoradiotherapy exposures, demonstrated that the same data, which showed before their therapy practically no differences in the patients from the groups under studies, upon completion of the 4 therapy courses were found to be statistically significantly better for those patients, who additionally received magnetotherapy.

The differences in the Karnofsky index values, established between the groups, increased from 1 % to 7,7 % (р < 0,05) (see Figure 4 herein). Whereas an average grade according to the ECOG Performance Status Scale for patients with malignant gliomas of the brain was recorded to be 2,20 ± 0,08 in the main test group and 2,25 ± 0,09 in the reference group, the respective indicators identified in both groups upon completion of the 4 therapy courses were reported to be 2,06 ± 0,09 and 2,43 ± 0,014, respectively, that allowed us to establish the presence of a statistically significantly better dynamics of the indicator in those patients, who received magnetotherapy in addition to the conventional therapy (р < 0,05). The patients with the better dynamics were capable of all selfcare and even able to carry out everyday work of a light nature.

By this means we succeeded to achieve the better dynamics in the ECOG Performance Status grading and the Karnofsky index values in patients of the main test group subjected to CHRT accompanied by MT. Our analysis of such a subjective criterion like neurologi-
Fig. 7. Intergroup differences in the coefficient of the relation between the anti-stressor reactions and stress upon completion of the complex therapy comprising magnetotherapy in patients with malignant gliomas of the brain.

The quintessence of the inter-group differences was calculation of a synthetic parameter: it was the CAS/S coefficient to represent the relation between an integral of the anti-stressor reactions and stress that is capable of quantifying the final result of the application of magnetotherapy by testing the types of reactions and identifying the frequency of the occurrence of development of anti-stressor reactions and stress in patients with malignant gliomas of the brain. The dynamics of adaptive resetting patterns, as illustrated in Figure 7 herein, exhibited essential intergroup differences in CAS/S both at the initial and final stages of therapy.

At the post-operative observation stage, before the beginning of the adjuvant therapy, we noted a tendency of bringing the CAS/S values in the groups under studies closer together, i.e. equalization of the CAS/S values. But upon completion of the basic therapy accompanied by the electromagnetic field treatment we detected the predominance of the anti-stressor reactions that was favorable for an increase in resistance and beneficial for the functional rehabilitation; that promoted an improvement in the quality of life of the patients.

**Conclusions**

1. The application of the adjuvant chemoradiotherapy accompanied by the magnetic field treatment, targeted at the brain in the context of therapy of malignant gliomas of the brain, results in a statistically significant increase in the general 2-year survival by 2.4 times (40.0 % against 16.7 %) as well as an increase in the 2-year recurrence-free survival by 6 times (from 3.3 % to 20.00 %), (p < 0.05).

2. The use of adjuvant chemoradiotherapy in combination with magnetotherapy in treatment of malignant glial tumors of the brain improves the ECOG Performance Status grades and provides for the better Karnofsky index values. The established intergroup differences in the Karnofsky indices have shown a rise from 1 % to 7.7 % (p < 0.05). Before the therapy, an average ECOG Performance Status grade in patients suffering from malignant gliomas of the brain has been found to be 2.20 ± 0.08 in the main test group and 2.25 ± 0.09 in the reference group, respectively, whereas upon completion of the 4 therapy courses the relevant status has been assessed as 2.06 ± 0.09 and 2.43 ± 0.014, respectively, that allows identifying a statistically significant better dynamics of the data in patients, who received MT (p < 0.05).

3. The utilization of the adjuvant chemoradiotherapy combined with magnetotherapy in treatment of malignant glial tumors of the brain reduces symptoms of neurological toxicity, as compared to the reference group, as follows: general brain symptoms by 24 %; motor disorders by 26.9 %, sensory processing disorders by 16.7 %, speech disorders by 10.7 % and higher nervous activity disorders by 13.9 % recorded within the time close to surgery (6 months after the surgery operation). It contributes to a better dynamics of the ECOG Performance Status grading (0.32 against 0.18) and in the Karnofsky index values (9.3 against 2.6 %).
fect: it provides for predominance of training, calm and elevated reactions in the pattern of the physiological reactions in 73.4% of the cases; it decreases the stress share by 2.9 times that promotes increasing of the coefficient of the relation between the anti-stress or reactions and stress by 10.8 times, as against the level identified in chemoradiotherapy treatment without magnetotherapy (p < 0.05).

Statement on ethical issues
Research involving people and/or animals is in full compliance with current national and international ethical standards.

Conflict of interest
None declared.

Author contributions
The authors read the ICMJE criteria for authorship and approved the final manuscript.

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Xenon effect on electro-physiological markers in oncology patients with postcastration syndrome during early post-operative period: pilot studies

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Abstract
Breast cancer (BC) takes a leading position among all malignant neoplasms in women [1]. One of the critical aspects of the radical treatment of BC is the development of post-castration syndrome (PCS) in patients of reproductive age, who as a rule reach physiological, social and professional maturity, competence and status in the period up to 45 years of age. Traditionally used hormone replacement therapy, despite its high efficiency, has a number of contraindications and side effects. Therefore, it is urgent to develop and implement in clinical practice the effective methods of rehabilitation of patients with PCS. One of the main areas of research is the study of the prospects for the use of therapeutic xenon-oxygen mixtures in restorative medicine, as it has a pronounced anti-stress, hormone-modulating, antidepressant and sedative effect. Accordingly, the aim of the present paper is to study the effect of xenon on cardiovascular system markers, brain bioelectric activity and psycho-emotional state in oncology patients with hormone-positive breast cancer in the stage of surgical castration. 8 female oncology patients have been examined: the main group consists of women who received inhalations with a xenon-oxygen mixture on the fifth day after the surgery, and the reference group are the patients with the main course of treatment only. The electrocardiogram and electroencephalogram recording and the 8-color Lüscher test have been performed before, after surgery (ovariectomy) and after the completion of therapy. It has been established that the use of xenon-oxygen mixture in the main group of patients allows reducing the manifestation of somatic and psycho-emotional changes peculiar to PCS. After the course completion no stress of regulatory systems has been observed in patients, a tendency to normalization of myocardial metabolism, positive changes in bioelectrical activity and an improvement in the psychoemotional state have been revealed.

Keywords
Postcastration syndrome, Xenon, EEG, ECG, Psycho-emotional state

Introduction
Breast cancer (BC) takes a leading position among all malignant neoplasms in women [1]. One of the critical aspects of the radical treatment of BC is the reproductive age of patients, who as a rule reach the physiological, social and professional maturity, competence and status in the period up to 45 years of age. The carried out hormone-reducing operations necessarily deprive young patients of their priorities for quality of life, health, development and self-realization.

Total ovariectomy is performed at the stage of surgical castration in case of hormone-dependent breast cancer after radical mastectomy. Already during the first hours and days after the moment of castration, significant violations of the hypothalamic, endocrine, neurovegetative, hemodynamic regulatory systems are revealed, as well as deep neuropsychic disorders [2, 3]. The consequences of postcastration syndrome (PCS) development in patients of reproductive age after performing radical surgery are unpredictable and complex for correction [3]. Nowadays drugs for hormone replacement therapy are used for PCS therapy. Despite their high effectiveness, they have a number of contraindications and side effects, which allows using them only as a short-term therapy [4].

Imprint
Therefore, it is urgent to develop and implement in clinical practice the effective methods of rehabilitation of patients with PCS. One of the main areas of research is the study of the prospects for the use of therapeutic xenon-oxygen mixtures in restorative medicine [5, 6]. The therapeutic effect of xenon is determined by high sensitivity of cardiovascular [6-8] and nervous system [6] to it. Xenon improves the processes of metabolism and the cerebral tissues oxygenation [9]. Xenon has a pronounced anti-stress, hormone-modulating, anti-depressant and sedative effect and is promising in the treatment of PCS [5].

Accordingly, the aim of the present paper is to study the effect of xenon on cardiovascular system and brain bioelectric activity indices and psychological state in oncology patients with hormone-positive breast cancer during the stage of surgical castration.

Materials and methods

8 female patients of reproductive age (under 45 years of age, mean age 36.4 ± 0.5) with the diagnosis of hormone-positive breast cancer, being at the stage of surgical castration (ovariectomy) after mastectomy and chemotherapy have been examined. The patients received comprehensive treatment at the FGBU RRIO over the 2016 to 2017 period. All examination protocols have been prepared in accordance with the Helsinki Declaration (1964) ethical standards and approved by the university committee on ethics.

Five procedures of xenon-oxygen inhalations every other day in the morning have been prescribed to the main group of patients on the fourth day after the surgical intervention (ovarioectomy) [5]. The supply of xenon-oxygen mixture has been carried out through the face mask creating a hermetic closed circuit. A comfortable for the patient percentage of xenon in the gas mixture, usually 18–20 vol. %, has been established. The gas flow rate under conditions of hermeticity of the circuit is on average 1.8–2 liters. Duration of the inhalation is 12–17 minutes (saturation time, 5 minutes before the appearance of gas action signs, then breathing in a closed circuit, with additional portion oxygenation, for leveling the possible signs of hypercapnia, namely nausea, vomiting, redness of the skin, etc.). After inhalation the patients stayed in a state of superficial sleep, which resulted in the increase in duration of the recovery period, a comfortable state without external factors. During the procedure the parameters of hemodynamics and gas exchange have been monitored interactively. The reference group of patients received only the main therapy.

Prior to the surgical intervention (1st stage of the study), on the third day after the operation (stage 2) and after the therapy completion (stage 3), the electrocardiogram (ECG), the electroencephalogram (EEG) of calm wakefulness and the 8-color test by M. Lüscher [10] have been recorded.

ECG has been recorded using a single lead placed on a bone in the aortic area during 30 seconds in the lying position with the help of Cardiocode device (Taganrog, Russia). Heart hemodynamic parameters, the tension index of the regulatory systems by Baevsky, metabolism of myocardium (oxygen, lactate and phosphocreatine quantity) have been calculated using the Cardiocode software. Heart hemodynamic indicators have been evaluated using the data on blood phase volumes calculated noninvasively by means of substitution of cardiac cycle phase durations into the G. Poyedintsev – O. Voronova equation of hemodynamics [11]:

- SV – stroke volume of blood, ml;
- PV1 – blood volume entering the ventricle during early diastole, ml;
- PV2 – blood volume entering the ventricle during atrial systole, ml;
- PV3 – blood volume ejected from the ventricle during rapid ejection, ml;
- PV4 – blood volume ejected from the ventricle during slow ejection, ml;
- PV5 – blood volume pumped by the ascending aorta during systole, ml.

According to the variation pulsometer results the regulatory systems tension index (TI) by Baevsky has been calculated: 100–500 conventional units is a norm, more than 500 c.u. is overtension, less than 100 c.u. is a weak state of the regulatory systems [11]. The stability of state has been evaluated by the RR-intervals and blood stroke volumes (SV) scatterograms.

The changes in myocardium metabolism has been evaluated by the oxygen, lactate and phosphocreatine quantity, calculated indirectly with the help of Cardiocode software. A parameter value within the range of 0.7…0.85, 0.6…0.65, 0.5…0.55 has been considered as a norm for aerobic process; 3.0…7.0 for anaerobic-glycolitic process and 2.0…4.0 for anaerobic-alcoholic one [11].

The EEG has been recorded in 19 monopolar leads (Fp1, Fp2, F7, F3, Fz, F4, F8, T3, C3, Cz, C4, T4, T5, P3, Pz, P4, T6, O1, O2), placed according to the 10-20 sys-
tem, using the electroencephalograph-recorder “Encephalan-EEGR-19/26” (Medikom MTD, Taganrog). Calculated has been the spectral capacity of EEG in a state of calm wakefulness with closed eyes in frequency range 0.5–18.0 Hz using the Fourier transformation (FFT). Artifacts have been excluded from the analysis. The data have been processed with the use of software package Statistica10. A statistical analysis of changes in the sum of EEG spectral capacity during different treatment stages has been carried out with the help of the Kolmogorov-Smirnov nonparametric test (p < 0.05).

The following parameters have been evaluated by the Lüscher 8-color test [10]: the level of anxiety, the prevailing emotional background and the state of the organism energetic balance. The level of anxiety has been interpreted depending on location of four main colors of the test according to the method [10]. The stable emotional background of an individual has been determined using the rate of the total deviation from the autogenous norm calculated according to A.I. Yuriev’s formula [10]. The organism energetic balance has been characterized by the vegetative coefficient for the Lüscher test proposed by K. Shiposh, and the vegetative balance coefficient [10]. The coefficient values differ from 0.2 to 5 points and are interpreted in the following way: chronic overstrain, compensated state of fatigue, optimal efficiency and superexcitation.

The study results have been processed with the use of Statistica10 software package.

**Results**

The basic heart hemodynamic parameters calculation in patients of the main and reference group has allowed detecting absence of their disorders at different stages of the study. Each parameter value is presented in Table 1.

The calculation of the regulatory systems TI and the analysis of the RR intervals and stroke blood volumes scatterograms in the reference group patients before ovariectomy and after the therapy completion has showed the absence of regulatory systems tension (TI = 472 (SD = 23.2) conventional units and TI = 397 (SD = 46.2) conventional units, respectively). But this state has been unstable (Fig. 1А and 1С). After the ovariectomy (Fig. 1В) in this group patients a mean-stable state of the regulatory system tension (TI = 919 (SD = 33.8) conventional units) has been detected.

Evaluation of the parameters of myocardial metabolism in patients of the reference group has demonstrated low oxygen values (0.37 (SD = 0.02) conventional units and 0.14 (SD = 0.02) conventional units, respectively), high lactate indices (0.8 (SD = 0.04) conventional units (SD = 1.4) conventional units, respectively) with a normal content of phosphocreatine (3.3 (SD = 1.2) conventional units and 2.6 (SD = 0.6) conventional units, respectively) at the first and second stages of the study. At the third stage of the study after the therapy completion with a reduced quantity of oxygen (0.11 (SD = 0.04) conventional units), the quantity of phosphocreatine has decreased below normal (0.48 (SD = 0.03) conventional units), the quantity of lactate has normalized (4.2 (SD = 0.4) conventional units).

In the main group patients at the first (Fig. 2А) and second (Fig. 2B) stages of the study noted has been the unstable state of the regulatory systems tension (TI = 677 (SD = 35.2) conventional units and 533 (SD = 21.4) conventional units, respectively). At the third stage (Fig. 2С) no regulatory system tension

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**Table 1. The basic heart hemodynamic parameters in patients at three stages of the study**

<table>
<thead>
<tr>
<th>Hemodynamic parameters</th>
<th>Reference group</th>
<th>Main group</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before operation</td>
<td>After operation</td>
</tr>
<tr>
<td>SV</td>
<td>59.2 (0.8)</td>
<td>57.2 (7.2)</td>
</tr>
<tr>
<td>PV1</td>
<td>38.8 (2.5)</td>
<td>32.7 (3.4)</td>
</tr>
<tr>
<td>PV2</td>
<td>20.5 (2.6)</td>
<td>24.6 (10.4)</td>
</tr>
<tr>
<td>PV3</td>
<td>35.2 (0.5)</td>
<td>33.9 (4.3)</td>
</tr>
<tr>
<td>PV4</td>
<td>24.1 (0.4)</td>
<td>23.4 (2.9)</td>
</tr>
<tr>
<td>PV5</td>
<td>8.8 (0.7)</td>
<td>8.48 (0.8)</td>
</tr>
</tbody>
</table>

Note: In the table the parameters' mean values and their standard deviation are presented.
Fig. 1. Scatterograms of RR-intervals and blood stroke volumes (SV) in the reference group patients at the first (A), second (B) and third (C) stage of the study.

(TI – 176 (15.2) conventional units) has been observed and this state has been moderate-stable.

Evaluation of the myocardium energetic state in the main group patients at the first and second stage of the study, before and after the ovariectomy, has demonstrated a low content of oxygen (0.25 (0.02) conventional units, 0.5 (SD = 0.02) and 0.42 (SD = 0.07 conventional units), respectively) and phosphocreatine (0.36 (SD = 0.01) conventional units and 0.4 (SD = 0.01) conventional units, respectively) with increased content of lactate (22.6 (SD = 2.5) conventional units and 18.3 (SD = 1.6) conventional units, respectively). At the third stage of the study after the therapy completion increased content of oxygen in myocardium has been revealed (0.51 (SD = 0.02) conventional units), when approaching the rates of lactate (8.5 (SD = 1.4) conventional units) and phosphocreatine (0.96 (SD = 0.3) conventional units) to the norm.

At all the experiment stages the patients in the reference group of EEG in the state of calm wakefulness with closed eyes have been characterized by irregular disorganized alpha-activity in a form of wave groups and low index (30–40 %), not modulated by the amplitude keeping the zone differences. Against the background of the polymorphic activity the separate groups of slow activity, low amplitude and low index (up to 25 %) are presented. Before the surgery and after the therapy completion beta-activity in the form of mean and high index (40–51 %) wave groups has also been noted. Figure 3 shows the localization of foci of the most pronounced activity of the EEG: at the first stage (3B) the focus of the alpha activity has been localized in the right parietal area, the beta activity focus has been localized in the right temporal area, the delta activity focus – in the left frontal-anteroposterior area. At the second stage (Figure 3C) the focus of the alpha activity shifted to the left temporal area, the delta activity focus shifted to the right frontal-anteroposterior area. At the third stage (Figure 3D) the focus of alpha activity has been localized in the central-parietal area, the beta activity focus has been localized in the right temporal area, and the delta activity focus – in the left temporal area.

The evaluation of the spectral capacity change (the Kolmogorov-Smirnov test, p < 0.05) as compared with the first stage has demonstrated reduction in the spectral capacity in the delta-rhythm range and retardation of alpha-rhythm at the second stage of the study. Decrease in the alpha-activity capacity is noted at the third stage of the study (Fig. 3A).
In the main group patients at the first stage of the study, irregular, insufficiently organized alpha-activity in the form of high amplitude and index rhythm (70–77 %) with smoothed area differences has dominated on the EEG before the ovariectomy, the localization focus is in the right parietal area (Fig. 4B). Also pronounced is the beta-activity in the form of high index (39–43 %), mean amplitude and low frequency rhythm, not localized. Demonstrated is slow activity of the mean index (up to 30) with amplitude up to 38 µV in the form of separate waves, localized in the left temporal area (Fig. 4B).

At the second stage of the study, after the ovariectomy, irregular, insufficiently organized alpha-activity in the form of a high amplitude rhythm, mean index (up to 64 %), localized in the right occipital area, has been observed on the EEG (Fig. 4C). Delta-activity in the form of rhythm of 1.0 Hz frequency, mean amplitude, with the localization focus in the right anteriofrontal area has dominated. As compared with the first stage detected is decrease in capacity of alpha-activity (the Kolmogorov-Smirnov test, p < 0.05, Fig. 4A).

At the third stage of the study, after the therapy completion an irregular, insufficiently organized alpha activity in the form of a high amplitude rhythm, a high index (69–73 %), dominated on the EEG, with smoothed zonal differences, the focus of localization being in the left parietal area (Fig. 4D). The delta-activity of mean index (up to 40 %) in the form of separate waves, localized in the right anteriofrontal area (Fig. 2D) is also evident. As compared with the second stage identified is rise in the delta- and alpha-activity capacity. The delta-activity capacity is also higher compared with the first stage. Noted is decrease in the capacity of the low frequency alpha-activity and increase in the high frequency one (the Kolmogorov-Smirnov test, p < 0.05, Fig. 2A).

As a result of the Lüscher 8-color test the general features which describe psychological state of the reference and main group patients on each stage of the study have been separated. In the reference group patients at the first stage of the study the psychoemotional state has been characterized by the presence of anxiety, psychological adaptation and optimal working efficiency. Beginning with the second stage of the study, in some patients at the second and third stages of the study an actively-dependent position focused on the opinion of significant individuals, without manifestations of anxiety and stress, was pronounced. In another part of the patients at the second stage of the study a state of anxiety expressed in dissatisfaction.
with the situation, vulnerability, and compensated by sthenic type of reaction, active position, pronounced emotivity, has been detected. On the third stage the stress state manifestations, caused by the feeling of frustration and overtension of efforts aimed at obstacles overcoming are typical for the above mentioned patients. The stress state is compensated by means of a passively defensive position. Negative emotions, bad mood, unpleasant feelings and fatigue prevail.

In the main group patients at the first stage of the study moderately anxious state expressed in frustration, uncertainty, dependence, suspiciousness and fears for health, has been marked. In major cases the given state has been compensated by a passively-meditative position, control of the state and concern for health. Psychological adaptiveness, stable emotional state and optimal working efficiency have also been marked. At the second stage no manifestations of anxiety and stress have been detected in the patients. A need for bright feelings, self-confidence, positive emotions, optimism and optimal working efficiency have been marked. The vegetative coefficient index indicates the compensated state of fatigue. After the therapy completion, improvement in general condition and mood, reduction of pain in the area of the postoperative wound, normalization of night sleep, increase in attention concentration and activity, optimism and positive attitude toward continuing the treatment have been noted.

Discussion

On the basis of the obtained data we established that the post-castration syndrome in oncology patients during the early postoperative period in most cases is characterized by the presence of anxiety with prevalence of negative emotions, disorder in adaptation process, sleeping and general state, state of overstrain, that corresponds to literature data on cognitive function aggravation in patients with breast cancer after surgical treatment [12, 13]. Disorder in the neurohumoral background in the patients caused moderate change of bioelectric activity in brain mainly of a regulatory type due to regulating systems dysfunction [14], tension in the regulatory systems and disorders in the myocardium energetic state, indicating the system overtension and exhaustion.

In patients exposed to the course of xenon oxygen mixture inhalation additionally to the main therapy, also detected is absence of tension in the regulatory systems and a tendency to normalization of the myocardium metabolism. Thus, confirmed is the data on cardioprotective effect of xenon which consists in maintaining the cardiovascular stability [15]. At the
same time absence of the xenon effect on general haemodynamic parameters has been observed [16]. Detected are the changes in EEG consisting in increase in capacity of alpha-activity in high frequencies. This fact proves the data on the activating effect of xenon [17]. Absence of the beta-rhythm, the rhythm of tension is characteristic of the above mentioned EEG changes as well. At the same time in the reference group patients the EEG is characterized by slowdown in the alpha-rhythm and presence of beta-activity. Detected is improvement of psychological state. Thus, the positive effect of xenon in oncology patients in early postoperative period involved neurohumoral stabilization and functional state optimization of the patients’ state [15, 18]. The ability to prevent disorders associated with the post-castration syndrome will allow the patients to maintain their functional and social status, which is a prerequisite for a full life [19].

Conclusions
1. Application of xenon-oxygen inhalation in the early postoperative period in oncology patients with PCS has demonstrated decrease in the regulatory system tension (according to the TI data) and normalization of myocardium metabolism indicators with the absence of its effect on general hemodynamic parameters.
2. Against the background of taking the xenon-oxygen inhalations revealed have been the shifts in the amplitude-frequency markers of the EEG towards its activation and improvement in general psycho-emotional state of the patients.

Statement on ethical issues
Research involving people and/or animals is in full compliance with current national and international ethical standards.

Conflict of interest
None declared.

Author contributions
The authors read the ICMJE criteria for authorship and approved the final manuscript.

References


Pathogenetic peculiarities of sclerosing of blood vessels in hemangioma in minor aged children upon exposure by electromagnetic optical spectrum radiation

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Aim
Our aim was to study pathogenetically significant markers of sclerosing of hemangiomas in infants upon their exposure to optical radiation in the infrared region of the spectrum by analyzing thermography and morphology examination results.

Materials and methods
The basis for the thermographic study was our observation for 200 patients (birth to age 1), who received the hemangioma treatment in 2014–2016. We investigated thermograms of each vascular tumor in infants both before and after the treatment conducted with the use of the infrared thermography (IRT) method. An IRT session was carried out upon a calm, quiet staying of an undressed infant in a room at an ambient temperature of 22°C and humidity of 0.009–0.012 kg/m³. To record heat fields in correspondence with the actual heat state of the tumor areas under studies, we used our thermo-vision device RADUGA-6 manufactured by AOMZ. The IRT data were compared with those obtained in ultrasound examinations. Diagnostic indicators of sclerosing, upon testing blood serum specimens, taken from 5 practically healthy and 25 hemangioma-affected children, were analyzed with the use of the cuneiform dehydration technique. Microscopic examination of the sampled specimen structures was performed with the LEICA DM SL2 microscope supported with original software MOR-PHOTEST.

Results
It has been established that an alteration of the heat field structure in a vascular malformation depends on a treatment stage. When sclerosing the blood vessels in the malformation, the thermo-graphic indicators show a sharp decrease (from 39.90°C at a temperature gradient of +3.060°C to 36.00°C at a temperature gradient of +0.10°C) and approach the normal values. Structural changes in serum facies analyzed at each stage of the treatment reflected the respective alterations both in the infant organism and the malformation. Some criteria have been identified by us to indicate sclerosing of the malformation, which are as follows: wide radial cracks, incomplete concretions, Sierpinski-carpet and leaf-type structural patterns. The relevant ultrasound examination data have confirmed our suggestions that there is a reduction in hemangioma sizes, the absence of blood flow and a blood supplying blood vessel as well as regression of hemangiomas against the background of electromagnetic radiation in the optical infrared region of the spectrum.

Conclusion
It has been found that evidence data on temperature distribution in hemangiomas obtained with remote infrared thermography are in good correlation with the criteria of biological fluid morphology as well as the relevant ultrasound scanning data. The factual evidence makes possible to provide an objective assessment of the states and stages of development of the vascular malformations in order to select the most adequate therapy.

Keywords
Electromagnetic radiation of optical spectrum, Remote infrared thermography, Morphology of blood serum, Hemangioma

Imprint

Introduction
Among all benign tumors, quite often found can be in infants hemangiomas with locations on the skin, in soft and mucous tissues: their percentage reaches from 50 to 80 %. They may occur in 1,1–2,6 % of the newborns, and by the age of 1 the occurrence rate of the malformations may achieve 10,1 %; they are characterized by a fast, locally destructing growth. We observe an increase in the percentage of infants show-
ing the proliferating skin or underskin hemangiomas complicated by ulcer, inflammation and bleeding [4]. Considering this factual material, the problem of effective treating hemangiomas in infants remains topical at the present time.

An application of the Photo-Chromotherapy Method (PCM) is based on the capability of the monochrome red light of inducing photo-bio-adaptive processes of repair and regenerative tissues in an infantile organism, with an increase in synthetic, phagocytic activity of neutrophils as well as an increase of indicators of the lymphoid subsystem of immunity that favors the stabilization of growth, so that regression of vascular malformations on the skin is promoted. Using PCM, we can induce vasodilation of the micro-blood vessels, activate the processes responsible for sclerosing of main blood supplying vessels within the tumor, and intensify humoral factors of the regulation of the local blood flow. The phototherapy with the monochrome red light results in an improvement of peroxidative and energetic metabolism, harmonization of the internal processes in the organism and elevation of non-specific resistance of the infant [4].

An adequate modern understanding of changes occurring in hemangioma during the treatment, including that with PCT, is not possible without assessments of prognostic criteria, which reflect the dynamics of the sclerosing processes in the mentioned vascular malformation type.

In this connection, undeniably, there should be fundamental research to be conducted in order to discover the essence of the light emitting diode emission effect [10, 12]. One of the key acceptors of such effect might be biological fluids, among them blood serum. The cuneiform dehydration technique implies some specific conditions onbio-fluid the self-organization that results in obtaining a solid film (a facies) showing specific patterns which reflect certain individual biological parameters of homeostasis [3, 7, 8].

The specific structural patterns composed by the contained molecules form the respective system-and subsystem-related levels as well as the local structures at the macro-level which may be visually examined. At the present time, in blood serum in adults revealed have been a number of local mor-phological structured markers, indicating certain pathological processes, including that of sclerosing of blood vessels. So, when blood vessels are affected by sclerosing, the blood facies in adults demonstrate leaf-type structural patterns; and, as this takes place, it should be mentioned that the greater is the relation between the cross-sectional line and the longitudinal line size of the leaf pattern, the more pronounced are sclerotic alterations therein [3, 7]. As to infants with hemangiomas, regression of this sort of vascular tumors is determined by sclerosing of the major blood-supplying vessels in the hemangioma under treatment. In doing so, we should note that identifying markers of this process may bear witness to the efficacy of the applied therapy measures and detection of the reserves of the infant’s health in order to take the proper treatment measures aimed at strengthening of the reserves and prevention of the reserve depletion.

Among the existing methods of diagnostics, thermography of blood vessels occupies a highly important place because of due considerations of a correlation between the intensity and evidence of clinical manifestations of a disease and an actual temperature on the skin surface [9, 11]. Recently the infrared thermography (IRT) has been widely used as one of the methods of non-invasive diagnostics that is very topical for infantile diagnostics [5, 6, 8]. The temperature measured on the skin of an infant under the rest conditions, considering microclimate, excludes any changes in the body temperature and is said to be an integral indicator of the degree of functional activity of the vascular tumor or the tumor-projected-area organ.

The IRT method is based on recording and visualizing of temperature fields with the use of specific imaging equipment, namely, thermovision devices. The main element of the device is a sensitive receiver of the infra-red radiation; the receiver is responsible for conversion of the thermal radiation emitted by the human body in the infra-red wavelength range into electrical signals which are automatically processed and transformed into images of an area under examinations, which are called thermograms [9]. Thermograms obtained with the thermovision device exhibit temperature field distributions which correspond to the thermal state of the specified areas. The data delivered with the equipment represent both anatomical & topographical and functional alterations in the affected zone [1, 2, 9, 11]. However, it should be mentioned that the infrared thermography applied for the purpose of tumor growth diagnostics has limited applications both in Russia and abroad [9, 11].

Considering the above, we should state that searching for the proper criteria of sclerosing blood vessels
according to the morphological picture of dehydrated serum films in children aged under 1 with hemangiomas becomes topical at every stage of photo-chromotherapy (PCT).

Materials and methods

Our clinical studies are based on observing patients covering the cohort from newborns to infants aged under 1, who have been subjected to PCT of hemangiomas in the red spectral region in the period 2014–2016. When conducting the studies, we specified some applicable criteria for an evaluation of the efficacy of the carried-out therapy as follows: a reduction in sizes of malformations, the appearance of sharp tumor contours, a decrease in intensity of blood flow, changes in gradients of temperatures between the tumor and the enclosing tissue, since such type of dynamic monitoring of the hemangioma condition makes possible to obtain objective evidence data on therapy outcomes and facilitate individualization of the patient treatment tactics. The tumor tissue due to its intense metabolism demonstrate in more frequent cases a higher temperature as against the enclosing tissue, that is recorded in the relevant projection as an enhanced infrared radiation, and it is just this phenomenon that is used as the basic principle for the IRT diagnostics in clinical oncology. In pathogenesis of hemangiomas in children aged under 1, it is possible to detect alterations of the vascular bed within the pathological focus region, which may have manifestations in one form or another to a different degree. The IRT examinations in each child undressed were carried out upon expiration of 15 minutes of rest staying of an infant in a room at an ambient air temperature of 22°C and with an ambient air humidity of 0.009–0.012 kg/m³. To record the relevant thermal fields, which reflect the respective thermal condition of the tumor regions to be examined, utilized was thermovision device RADUGA-6 manufactured by AOMZ (Russia). The same patients were subjected to ultrasound scanning with the use of sonographic equipment as listed below: LOGIQ 400 MD, HDI-1500, HDI-400, ATL and USA with linear-type piezo-sensors at frequencies 7,0–12,0 and 9–15 MHz. Utilizing the Brightness-Mode, we assessed sizes, echogenicity, the presence or the absence of tumor encapsulation, contours of the malformation; in the color Doppler sonographic mapping (CDM) regime and in the color Doppler energy (CDE) mode we examined the character of blood flow and the presence or the absence of a blood-supplying vessel. The revealed alterations therein were recorded in the two mutually perpendicular projections that offered a possibility to produce 3D-images of the pathological focus regions.

Our experimental studies implied tests of blood samples taken from 25 infants, aged under 1, diagnosed with skin hemangiomas, at every stage of photo-chromotherapy, as against blood specimens taken from the practically healthy infants. Blood sampling was performed before the experimental PCT, subsequently upon completion of PCT course 1, 2 and 3 as well as 1 year after health restoration; at the same time blood tests were carried out in the cohort of the healthy children; in each case a blood sample volume was 0.5 cm³. All collected blood specimens were centrifugically processed at a centrifuge speed of 1000 RPM, so that serum samples were separated and tested with the use of cuneiform dehydration [3]. A drop of the biological fluid amounting to 15–20 µL (5–7 mm in diameter) was placed onto a degreased microscope glass slide with a lecithine substrate, using a micro-dropper with a dosing device. Thereupon, the drop was dried at an ambient air temperature 20–25°C, avoiding air draft, under an ambient air relative humidity 65–70 %. The drop to be dried should be completely immovable, and the microscope glass should be located in a strictly horizontal position. The time required for drying (till the time of the structural pattern analysis) should be 18–24 hours. The microscopic examination in our case was performed with microscope instrumentation LEICA DM SL2, with an application of original Software Morphotest. Morpho-logical analysis was carried out with utilization of optical light, dark field and polarization micro-copy with a magnification of x5, x10, x40 and x100, respectively [3].

The facies type reflecting the pattern of symmetry, the number and shapes of major system- and subsystem-related criteria, comprising cracks, isolated areas and concretions, might be identified to be either of physiology nature being represented by the radial and (or) partially radial types, or of pathology character being composed by the irradial, circular, amorphic, double-facies types, as well as to be of stable or non-stable types. In order to determine the facies and their structural type, namely, stable or not stable type, pathologically stable or nor stable type, we also conducted an additional drop testing of the serum sample taken from the same child.
Results and discussion

Upon thermometry of hemangiomas with different locations, we have obtained evidence as indicated below. The obtained thermal profile differed in colors of the elements on mapping images that made possible to identify separating lines of individual temperature regions in different parts in the child body and the vascular malformation. It was established that on thermograms abnormally cold zones were blue colored; hot areas were black, red or orange; isothermal regions were represented in yellow and green. According to such color map of the tumor, we were capable of visually identifying its extension and sizes, location and activity of the processes occurring therein. The smallest size of the tumor focus for hemangiomas detected with RIT was 1 mm in diameter. The conducted thermometry allowed reliably revealing an increase in temperature in the tumor region under examination. A local increase in a temperature in hemangiomas was contoured as a hyper-thermic focus area with a gradient of temperatures from + 3,0°C to + 0,2°C. The maximum temperature in the tumor was recorded to be 39,0°C. Upon assessing the patterns of the thermal map we classified the following types of the tumors: hyper-

Fig. 1 a, b, c and d. Variety in thermal maps representing cavernous hemangiomas in chest wall skin area
thermal, hypothermal and isothermal malformations. During the growth and the stabilization of the process, the contours for the majority of the tumors were found to be clear-cut on the thermograms, while during the tumor regression we noted diffuse contour lines. As to the children upon successful treatment of hemangiomas, the indicators of the thermal field between the symmetrical areas showed practically no difference from the normal ones, and the temperature distributions were recorded to be from 35.9 °C to 36.0 °C, with a gradient under 0.1 °C. The thermograms of such children were isothermal. Heat fields were represented in yellow and green. Figure 1 exhibits an example of various thermographic maps of cavernous hemangiomas in the chest in children aged under 1.

A hemangioma in Figure 1a demonstrates sharply cut boundaries with the distinct maximum of light effect that is dark-color-marked as an extensive area of hyperthermy. The structure of the thermal field is homogenous. It should be noted there is a disorder in the thermal symmetry. Determined can be a hyperthermal focus with a temperature gradient of +2.25 °C. Disorders in symmetry with a high gradient of temperatures can be used as one of the criteria for thermo-vision diagnostics to indicate that there is an active growth process of the vascular malformation [1, 2, 6]. Clinically, in our case, the hemangioma is characterized by an active proliferative growth.

Figure 1b gives a thermogram of a cavernous hemangioma which is represented by some regions, differing in degrees of the expressiveness of the light effect, where the pattern of the thermal field is inhomogeneous. The central zone of hyperthermy is clear-cut (dark-colored); the border line of the hyperthermal field is well-marked and is bounded by a less hot (red color) zone with a sharp transition into a zone of iso- and hypthermy (yellow and green colored on the thermogram). There is a disorder in the thermal symmetry available. Identified can be a hyperthermal focus with a temperature gradient of +0.8 °C in the centre and +0.5 °C along the periphery of the vascular malformation. Such alterations in the thermogram can bear witness to the fact that there is a stabilization of the tumor processes. The thermographic mapping pattern of the cavernous hemangioma in Figure 1c is not sharp; the contours are not symmetrical; structurally, the thermal field shows non-uniformity. The foci of the hyperthermal fields are bounded by some areas of hypo-and isothermy. The contour-related asymmetry is enhanced, and the hyperthermal focus shows a temperature gradient from +0.3 ° to +0.2 °C. Clinically, the given hemangioma can be classified as regressing. In Figure 1, examining the hemangioma area, we can find some insignificant regions with a pronounced minimum of brightness as a hyperthermy zone; the malformation depicted herein is mainly of isothermal pattern. The boundary lines are not clear, and the contours are indistinctly seen. The structure of the thermal field is homogenous; no temperature gradients are found; the malformation is isothermical with reference to the enclosing tissues. It might be viewed as an indicator of involution of the vascular malformation, i.e. hemangioma.

Informativity and reliability of the data delivered by remote IRT in PCT of hemangiomas in infants are about 100 %. But taking into account the fact that remote IRT is a non-invasive, safe and rather simple technique, which may be applied as many times as it is required for the same infantile individual, this new technology can be successfully recommended for individual administration of power exposure dosing under the specified PCT parameters depending on the IRT initial data.

By this means, based on the IRT initial data on actual temperature distributions in hemangiomas and enclosing tissues, we are capable of objectively assessing the condition and the stage of progression of the vascular malformation in order to select the most adequate therapy.

Our analysis of the serum facies in children has demonstrated distinct differences in the groups under studies. When identifying the facies of serum in infants with complicated hemangiomas before PHT, it
was established that in the common group the facies with extremely low level of structuring were dominating: the texture of the facies were found to be either of amorphical or irra-dial type; it was identified as pathologically stable (see Figure 2 herein).

Upon microscopic examination of the solid films of serum in infants after completion of PCT course 1, there was another morphotype of the facies identified (see Figure 3a, b). Most of children under examination demonstrated a partially radial type of the systemic organization of the facies of blood serum, and only in one case the radial type of the facies was found. Some local pathological formations as markers of hypoxic and inflammatory processes (wide radial cracks, the Sierpinski carpets, leaf-and bundle-type structures and incomelated concretions) appeared. The mentioned alterations in the bio-fluid indicated that there were changes in the homeostatic processes in the child organism targeted at sclerosing the vessels in the hemangioma that was evidenced by the signal markers of sclerosing appeared in most children: they were leaf-type structures appeared therein. But it should be noted that at that stage of PCT the alterations were of physiologically unstable nature.

The visualized representation of hemangiomas in infants demonstrated that there were small-scaled bright spots on its surface that was an indication of the beginning of the sclerosing processes.

The structure of the facies of blood serum in children with hemangiomas upon course two of PCT at-
tested that there was a restoration of the normal types of the morphostructure as a reflection of the appearing anti-tumor and bio-adaptive effects of PCT (see Figure 4a, b herein).

The architectural structure of the facies changed: the type of symmetry became radial; formation of isolated regions and concretions took place that was an indication of the normalization of the first and second levels of the self-organization. In some infants we revealed some blood serum facies showing some cracks atypical in sizes: the cracks were either shortened to one-half of the facies radius, or they were longer in their length intersecting the facies circumference.

Visual imaging of hemangiomas detected that there were some largesized bright sclerotic fields appeared throughout its entire surface, and the tumor demonstrated shrinkage; in palpation it was found that the tissue was soft.

Upon completion of course three of PCT, the structure of the blood serum facies approached the normal type recorded for conventionally healthy infants without hemangiomas. The sclerosing markers were absent (see Figures 5–7 herein).

Examinations of the facies of blood serum in infants carried out 1 year after the recovery confirmed that there were normal types of the facies with clearcut radial structure of the cracks identical to those found in healthy children. No markers of pathological processes like inflammation, hypoxia or sclerosing were detected. According to the visual images, at the location of hemangioma a soft connective tissue scar was identified.

By this means upon performing the analysis of the criteria of crystallization, an identification of system-, sub-system-related and local signs of self-organization of the bio-fluids as well as markers of pathological processes in blood serum in the infants at every stage of PCT, it has been established that a restoration of level I and II of self-organization takes place (the system-related concentrated waves, radial symmetry of cracks, appearance of isolated regions and concretions). Already upon completion of course two of PCT, we have been able to clearly identify that the criteria of sclerosing of blood vessels in hemangioma in the morphotype of blood serum have been met; the occurrence rate of pathological process markers like inflammations, hypoxia and intoxication has decreased. Upon completion of the PCT courses, the morphotype of the facies do not show any differences from that found in healthy children.
When analyzing the data obtained with ultrasound scanning, the following factual evidence was produced: before therapy, in 57.8% of the cases recorded were hemangiomas with a higher echogenicity; in 77.7% of the cases the contours thereof were not clear, and in 100% of the cases encapsulation was not found. The type of blood flow was detected to be either central in 38.8% of the cases or mixed in 27.8% of the cases; in 70.2% of the cases detected was an arterial blood supplying vessel. In dopplerography recorded were high values of blood flow velocity ($V_{\text{max}} \geq 16s/s$). That should be treated as factual evidence for the presence of proliferation of the angiomatis process. Upon reaching the required clinical effect, at the first stages, there were clear-cut boundaries of the malformation identified, a decrease in echo-density and a significant reduction in volumes and intensity of blood flow. During the hemangioma regression, the malformation was not detectable. The clinical picture for the children was characterized by the following: after PCT course 1, regression of hemangioma was recorded nearly for a half of the group under studies; in case of cavernous hemangiomas, regression was recorded only for 15% of the infants. According to the ultrasound scanning data, we detected a reduction in sizes of the tumor by 46–71%, a decrease in blood flow velocity by 11–22%, and encapsulation was recorded in 40% of the cases. Upon PCT course 3 and 4, the percentage of the children showing regression of the tumor was increasing. We recorded in those infants a reduction in sizes of the malformation by 98%; we identified a weak periphery blood flow or the absence thereof in 96% of the studied cases; clear boundaries or encapsulation were found in 98%, and the disappearance of the blood supplying vessel was recorded in 80% of the cases. Upon PCT course 4, the rest of the children with simple hemangiomas demonstrated regression of the hemangiomas, and for the other children showing other forms of hemangiomas recorded were percent-ages of stabilization and regression. In cases complicated by hemangioma ulcers, in PCT obtained were data on some local effects of monochromic low intensity red-light emission on pyoinflammato-ry processes within the affected wound area: sclerosing of the main blood supplying vessel took place, activation of connective tissue elements (fibroblasts, histiocytes, macrophages, lymphocytes, building-up of bactericidal action of neutrophiles) resulted in an accelerated cleansing of the wound with removal of necrotic mass, a reduction by more than two times in duration of excudative phase of the inflammation process with synchronous developing of granulation tissue, so that epitheliali-za-tion of the wound, on the average, was completed
on day 5 from the date of the beginning of the light therapy that was considerably shorter than the respective reference parameters: it became shorter by three or five times (see Figure 8a and b herein).

**Conclusion**

Electromagnetic influence provided by the optical range in the spectral red light region initiates regression of the vascular malformations in children aged under one. The clinical effect is provided due to inducing the sclerosing process in blood vessels in hemangiomas. The data obtained with the use of remote IRT on actual temperature distributions in hemangiomas, the criteria for morphology of bio-fluids and the data produced with ultrasound scanning are in good correlation; they make it possible to objectively assess the condition of the vascular bed in the tumor. Some markers of sclerosing blood vessels identified according to the blood serum morphology data and IRT evidence can be useful for an assessment of efficacy of the treatment carried out.

**Statement on ethical issues**

Research involving people and/or animals is in full compliance with current national and international ethical standards.

**Conflict of interest**

None declared.

**Author contributions**

The authors read the ICMJE criteria for authorship and approved the final manuscript.

**References**


Socio-cultural factors of human surviving under the conditions of the Leningrad blockade

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Aim
The aim of the article is to show the significance of the axiological approach in explaining the place of socio-cultural factors, such as social status, ethical values and art, in survival of human population under the conditions of the Leningrad blockade, which reveal in a new methodological viewpoint the significance of the fundamental discoveries, made by Soviet scientists, still underestimated, in biology and medicine (high viability of a part of the population under extremely severe conditions, possibility of activating the hidden reserves of the organism on the verge of life and death, dysregulation pathology, high life potential of the human body, dependence of severity and outcomes of alimentary dystrophy on the psychoemotional status), which have an enduring value and are very topical in our days of significant growth rate of occurrence of extreme situations like environmental and anthropogenic disasters, local armed conflicts and criminal events.

Materials and methods
In this article the following theoretical tools are applied: a widely used in science neurobiological approach; a new approach to the nature of art as a means of human survival; a cosmological model of the «world of many worlds»; a concept of space ethics and the corrected «hierarchy of needs» by A. Maslow. The axiological approach is chosen as the methodology and logic of the scientific research, which is, in its turn, a component of the modern scientific cognition that makes possible to show the significance of socio-cultural factors of human survival in extreme situations.

Results and discussion
For the first time in the philosophical literature, the role of socio-cultural factors as a methodological and logical explanation of scientific discoveries in biology and medicine during the Leningrad blockade is shown, which makes it possible to reveal the significance of these factors under the conditions of the growth of occurrence rate of contemporary extreme situations. For the first time a hypothesis has been put forward, according to which the diversity of the systems of ethical values of human society is rooted in the depths of the Multiverse.

Conclusion
It is suggested to use the results of discoveries in biology and medicine dated back to the Leningrad blockade together with the revealed mechanisms of such socio-cultural factors as social status, ethical and artistic values, which represent the concentrated aesthetics, for human survival under extreme situations of the modern world.

Keywords
Methodology, Logics, Extreme situation, Social status, Ethical values, Aesthetic values, Multiverse, Medicine, Biology, Discovery, Alimentary dystrophy, Pathology, Blockade

Imprint

Introduction
At present, the dynamic development of the world is characterized by such a significant increase in the number of extreme situations that they become a daily feature of modern life. Now, a sharp increase in the occurrence rate of natural and anthropogenic environmental disasters, local armed conflicts and terrorist acts, leading to mass casualties, is observed. This means that inevitability of extreme situations at the beginning of the 21st century must be taken as a quite certain reality, which makes it necessary to take measures to minimize their destructive consequences. The extreme nature of the vital activity of the modern complex non-linear information society with its latest scientific achievements and high technologies is caused by all the previous development of the society during nearly the whole of the last century. It is quite natural that in the scientific literature considered are the problems of genesis of extreme situations generated by nature itself and the dynamics of war in world
The paradox of the modern world lies in the fact that the growth of the extreme situations number is accompanied by modern achievements in the field of science, especially neurobiology, the latest technologies such as information and communication, nano-, bio-, cognitive and social sciences. A great importance in this regard, in opinion of Russian researchers V.B. Simonenko and S.V. Magayeva, have still not fully appreciated achievements in biology and medicine made under the conditions of the Leningrad blockade, which itself is a monstrous experiment that placed the human body «on the verge of the existence” [2]. Fundamental discoveries in biology and medicine, made under the conditions of the Leningrad blockade, prove to be very promising for researching the natural mechanisms of survival and recovery of the human body in pathologies emerging in extreme situations.

**Materials and methods**

Scientific research shows that the Leningrad blockade was the most difficult, massive and lasting extreme situation in the history of mankind. The specific aggravation of the situation consists in the action of three pathogenic factors, such as the psychological pressure of the 900-day siege of the city with bombing strikes and artillery shelling; almost full hunger and fierce cold of the first blockade winter. “Success in studying the blockade and postblockade pathology is still little known and unrealized. Meanwhile, an analysis of scientific works by clinicians, pathophysiologists, pathoanatomists and histologists allows us to consider that during the severe time of the blockade, major discoveries were made in the field of biology and medicine, which until now have not been properly evaluated by experts” [2–5].

Soviet biologists and physicians under the extreme conditions of the Leningrad blockade managed to collect valuable material on the basics of survival and recovery of the organism subjected to the shock in its vital activity foundations, using the pathogenesis of alimentary dystrophy as the main cause of human death events. The studies of the clinicians of the blockade time revealed the hidden reserves of the organism and the importance of the psychosomatic factor for the emergency situations outcomes, what at the beginning of the 21st century determined their need for studying the regularities of the organism’s vital activity in everyday extreme situations. In their fundamental research «The Leningrad blockade: discoveries in biology and medicine» V.B. Simonenko and S.V. Magayeva showed that for the theory and practice of modern medicine and biology of great significance are the following discoveries of the Leningrad blockade: the appearance of a high life potential of the human body; activation of natural mechanisms of viability in a significant part of the human population in the long-term extreme situation; diseases of nervous regulation of functions and their significance for the outcomes of an extreme situation; dependence of severity and outcomes of alimentary dystrophy on the psycho-emotional status; dependence of severity and outcomes of the atrophic process on the preservation of apoptosis regulation at the organism level; possibility of natural reduction of atherosclerotic lesions in the vascular wall; a key role of nervous mechanisms in the pathogenesis of hypertensive disease [2].

The significance of the blockade discoveries in biology and medicine is that they determine the ways to develop the activation of natural sanogenetic mechanisms and increase the vitality of the organism in extreme situations. Besides, they contain empirical facts confirming the importance of such socio-cultural factors as social status, ethical and artistic values for human survival in extreme situations. First of all, the survival statistics shows that those who had the social status of workers of defensive and rehabilitation institutions, as well as representatives of public military authorities, who were engaged in securing the life of besieged Leningrad, survived [2] that received its scientific justification in modern research. Thus, D. Rock in his monograph «Brain. Instructions for use: how to use your capabilities to the maximum and without overloads” emphasizes the importance of the social status in human life:»Status is one of the main engines of social behavior, in addition to community and justice”. In order to maintain or improve their status, people are ready to go very far. The feelings obtained when the status increases may be more positive than money, and if it decreases, it is felt almost as a danger to life «[6]. The status represents one more primary reinforcement or threat. In case of determining the attitude of associates to the status of a person,
his brain uses the same schemes and mechanisms as when determining the attitude to other basic needs that provide survival. Moreover, the neurobiological studies show that the human brain is a unity of the left and right hemispheres, each of which performs its specific functions, including the left hemisphere is focused on the rationalization of human behavior, which «apparently has a considerable value in terms of survival” [7].

This fact fits perfectly into the latest neurobiological studies, according to which the human brain has a neural social network responsible for the relationship of the individual with society; it is similar to neural networks that control vision, movement or hearing: «We are born with a social network» [6]. The famous hierarchy of needs by A. Maslow needs some correction considering the latest neurobiological research. «Many modern studies show that the brain, when interacting with social needs, uses the same neural networks as it is the case with survival. Hunger and ostracism activate the same reactions to threat and pain, involve the same neural networks «[6]. This neurobiological approach perfectly fits the research of the neurophysiological foundations of ethics, which have now become widespread. The dynamic development of science and the latest technologies (information, computer, gene, etc.) posed the problem of the neurophysiological foundations of ethics, which have strategic importance of nano-, bio-, info-, cogno-, and social technologies (NBICS-technologies) for the beginning of the new, fourth industrial revolution (Industry 4.0), literally turning the whole way of life both of the society and an individual therein. The ethics literature states that nowadays people express concern for the environment, as its destruction is able to put an end to our civilization. However, only a few try to comprehend the fundamental, strategic significance of the moral, or ethical, environment in the life activity of a human individual and the society [8]. The ethical environment is a kind of mirror, showing a person’s reflection in the eyes of the world, although he often does not feel the impact of this environment. This ethical environment is generated and supported by human individuals, who, in this sense, are ethical animals raised in a certain socio-cultural environment that facilitates the implementation of a life-affirming or life-defying worldview.

In his monograph «Culture and Ethics», outstanding humanist thinker A. Schweitzer justifies the ethics of reverence for life, which is the interpenetration of the ethics of self-improvement and the ethics of self-denial. The core of this interpenetration is the elementary thinking that generates reverence for life, as it proceeds from the fundamental problems of man's relationship to the world, the meaning of life and the essence of good. Here we are speaking about the relationship of a man as a finite being to the infinite being of the surrounding world, which has an ethical character that was emphasized by A. Schweitzer [12]. He shows that a man bears a subjective responsibility that goes to infinity and represents a responsibility for the whole life that is in the sphere of his influence. Herein, a man becomes internally free from the outside world and at the same time strives to fulfill his responsibility, and that is the ethics [12]. In other words, the source of ethics is the man's deep awareness of the world-affirmation, which, together with the life-affirmation, is naturally inherent in his will to live and which he strives to realize.

The subjective responsibility of a human individual is the ethics that is of cosmic character, which is the connection of man's ultimate being with the infinite being of the surrounding world, a link constantly generated by culture that represents the unlikely potency of an infinite nature. It is clear that the subjectivity of a human individual lies not only in culture, but also in the biological evolutionary process appeared due to the mutual influence of the necessity and a chance. Recent
scientific achievements in the field of an understanding of the nature of life show that the understanding of biological evolution, developed by the science of the 20th century, is obsolete and incomplete [13]. This thesis is argued in the monograph by the foreign researcher E.V. Kunin «The logic of the case. On the nature and origin of biological evolution", which uses a new perspective on the probability of a spontaneous appearance of life, generated by modern cosmology. It should be borne in mind that «any time-stable replicator» is meant under the form of life therein [13]. It has been just any replicating system that is capable of evolution due to the joint action of drift and natural selection that allows us to approach the problem of the origin of life within the framework of modern cosmology. Unlike the old cosmological concepts, treating the finite Universe, the recently introduced model of the «world of many worlds» (WMW) is an infinite multiverse with an infinite number of island universes, which cardinaly changes the very definitions of the possible, probable and accidental. Basing on the WMW model, E.V. Kunin draws the following fundamental conclusion: «Thus, the spontaneous emergence of complex systems that could be considered practically impossible in the final universe becomes not only possible, but also inevitable in the WMW, although the a priori a probability of the vast majority of stories occurring in a given universe is vanishing small. This new force of a chance, reinforced by the anthropic reasoning, has profound consequences for our understanding of any phenomenon in the universe, including life on the Earth” [13]. Consequently, the hypothetical assumption that the human society, which appeared as a result of cosmo-bio-co-sociological evolution, evolved a variety of systems of ethical values, ultimately conditioned by the Multiverse, has the right to exist. Thus, everything is built in accordance with a chain as follows: life arises in an infinite multiverse (infinite being), then society appears with its culture and a human being (ultimate being), which is an ethical animal living in the ethical environment created by himself.

The scientific studies establish a fundamental empirical fact, according to which any system of ethical values is conditioned by a set of cosmophysical fluctuations of space-time, which also influence the ethical environment permeating the aggregate of social, political, economic, informational and other environments of society. These cosmophysical fluctuations of space-time are also manifested in fluctuations in the activity of the Sun, which lead to changes in the psyche of human individuals, to «hot» and «colored» revolutions, insurrections, rebellions, as shown by A.L. Chizhevsky [14]. It is clear that in such emergency situations a radical change in the ethical environment occurs, when the old system of ethical values is disintegrated, accompanied by negative, destructive social processes.

It should be noted that during the Leningrad blockade the ethical environment of the Soviet society was preserved with its high spirituality and patriotism, which strengthened the motivation to survive and facilitated the experience of the burden of blockade life [2]. As the blockade of Leningrad is 900 days of inhuman suffering, courage and heroism of its inhabitants. The «Blockade book» by A. Adamovich, based on original materials, documents, letters, memoirs of the survivor of the siege, tells about the defenders of the city, the heroic dramatic days of the defense of Leningrad during the Great Patriotic War [15]. No less remarkable is the book by S. Yarovoy “The blockade ethics", which describes the moral values of the Leningraders died in 1941-1942. The book's narrative is the description how people found their death in broken streets and frozen houses, in endless queues for bread, others managed to survive in inhuman conditions, but they all brought to us a story about their experience of torment, their perseverance, their pity and humanity, about how people gave their helping hands to each other in the blockade nightmare [16]. It is shown that works of art served as one of the powerful means of maintaining ethical standards, contributing to the survival of a human under the terrible conditions. «The thirst for art was simultaneously suppressed and consolidated under the blockade conditions» [16].

Art contributed to a strong motivation to survive on the brink of death, forced to exert strong-willed efforts to combat death, included creative brain activity in an extreme situation, as evidenced by the activities of legendary poet and publicist Olga Fedorovna Berggolts, scientist Mikhail Vasilievich Chernorutsky, who developed the foundations of the of alimentary dystrophy pathogenesis, the survivor of the siege and painter Elena Oskarovna Martilla and thousands of other famous and unknown devotees, martyrs and heroes. It is quite logical that in the speech dedicated to his 90th anniversary D.S. Likhachev cited his vexed during the Blockade thought that “a book saves from...
death”. He believed that during frenzied hunger just the distraction from thoughts about food alienated the irreversible stage of alimentary dystrophy: “Books, namely stories and tales, and especially poetry, helped us a lot: they diverted our attention and created the conditions, under which a person continued to live, not “ate himself”” [2]. On the whole, art contributed to Leningraders’ survival, because it gave them a sense of high dignity, a sense of a person recognition that comes from the nature of art.

The Atlas of World Art proposes a new view on art as one of the oldest, most widespread and important types of human activity, and that is one of the reasons for the unchanged significance of art in human life to provide an elementary survival through an aesthetic form [17]. Therefore, it becomes clear why the work of art so strongly affects the emotional and intellectual world of a man, why many Leningraders under the extreme conditions of the blockade could survive through communication with works of art (it is not accident that not only Leningrad, but also all the front soldiers were thrilled by the Dmitry Shostakovich’s concert by the Leningrad Philharmonic in November 1942).

Results and discussions

In the present paper the axiological approach has been used for the first time as a methodological and logical basis for studying the materials and results obtained by Soviet scientists during the Leningrad blockade, and the most important discovery of them in human biology is the confirmation of the high vitality of a part of the population under the extreme conditions. The importance of the axiological approach in the field of biology and medicine to explain the mechanisms of the influence of such socio-cultural factors as social status, ethical and artistic values for human survival in life-threatening situations has been stated for the first time. A hypothesis that any system of ethical values has its roots in the depths of the Multiverse, which explains its significance for human survival under the extreme conditions, has been put forward. It is shown that art plays an important role in preserving the vitality of man, and the very feeling of beauty (the quintessence of which is art), in accordance with the ideas of such great physicists as S. Drell, A. Einstein, W. Heisenberg, F. Capra, J. Wheeler et al., affects the psychophysical processes of the human body due to the importance of the «aesthetic principle» in intranuclear and cosmic processes. The axiological and neuro-physiological approaches make it possible to develop mechanisms of human survival in extreme situations based on the use of such new high technologies as nano-, info-, bio-, cognitive and social ones.

Conclusion

For the first time identified is the importance of the axiological approach combined with neurobiology as a methodology and logic of scientific cognition, in which an important role is assigned to the perfect discoveries made by Soviet scientists under the conditions of the Leningrad blockade in the field of biology and medicine, which have an enduring significance for human survival in extreme situations that have become everyday’s phenomena in the modern world. Basing on the axiological and neurobiological methodology, the possibilities of using such socio-cultural factors as social status, ethical and aesthetic values, allowing minimizing the consequences of extreme situations for a human individual, are shown for the first time. A hypothesis has been put forward that the effectiveness of ethical values in human survival in extreme situations is due to the fact that these values are rooted in the depths of the Multiverse. The importance of the aesthetic values (whose concentrated representation is art), which is based on intranuclear and cosmic processes, is shown. Prospects for further minimizing some consequences of extreme situations for humans are associated with the use of a number of scientific methodologies, such as axiology, neurobiology, artificial intelligence, robotics, nanotechnology, biomedical technology, and others.

Statement on ethical issues

Research involving people and/or animals is in full compliance with current national and international ethical standards.

Conflict of interest
None declared.

Author contributions
The authors read the ICMJE criteria for authorship and approved the final manuscript.

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Bio-liquid morphological analysis

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Aim

Information is presented on the new scientific line in medicine and biology: bio-liquid morphology. The interdisciplinary character of the given research area is emphasized. The problems and prospects of bio-liquid morphological analysis development both in applied and fundamental aspects are discussed.

Keywords

Optical microscopy, Bio-liquid morphological analysis, New diagnostic technologies, Structure quantitative processing, Protein self-organization

Imprint

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Structure, being the most important quality of any material object, on one hand mirrors the conditions of its formation and on the other hand determines its physical and operational properties. Therefore, considerable attention is paid to structure in various areas of science, including biology and medicine.

Until recently in medical practice, the biological tissue structure examination was applied only to cellular and higher forms of its organization. Genetically determinate cellular forms of living systems are well known. They are studied within frameworks of such disciplines as anatomy, histology, cytology, etc. Structures of bio-liquids for medicine, practical medicine in particular, are still not adequately investigated. The concept of structure has been considered to be inapplicable to biological liquids because of the extremely high variability of their molecular composition and the character of the components interaction. Along with this, at the molecular and permolecular levels, the bio-liquids possess determinate, rather stable orderliness.

In the 1990s in Russia, a new scientific line in medicine and biology was formed: bio-liquid morphology [1]. It is methodically based on the biological liquid being brought into solid phase with a following examination with an optical microscope. The obtained data contain information not only of the concentration of elements – constituents of such complex system – but of the character of their interrelation as well. Morphological parameters of the biological liquids have distinct peculiarities and are used as the diagnostic criteria for determining diverse pathological conditions in different organs and systems, homeostasis stability degree, biological age, and the applied therapy efficiency. At present, the given scientific line is widely introduced in medicine and is already applied in therapy, surgery, pediatrics, urology, etc.[2, 3, 4, 5]. Research in the area of biological liquid morphology is not only of practical importance, but also discloses principally novel approaches in the living matter organization study, and is of interest not for clinicians, biologists, and biophysicists only, but for investigators in the area of complex system behavior and self-organization as well.

Bio-liquids participate in all processes of intra- and extracellular metabolism. Among the bio-liquids are blood serum, lymph, cerebrospinal fluid, secretions of endocrine and exocrine glands, urine, etc. Biochemical, physical-chemical, immunological, and other methods of bio-liquid examination give fragmentary data on the body’s condition, while the structural analysis allows information of the system as a whole to be obtained. To extract such information, a special method has been developed of bio-liquid dehydration with preset angular profile of the mass being dried –
cuneiform dehydration method. At cuneiform dehydration, the molecules and permolecular complexes are distributed in strictly defined places of the drying drop surface in the form of concentration zones. As a result, a facies is formed, the structure-specific character of which is determined by the entire set of qualitative and quantitative parameters of the substances present in the bio-liquid (Fig. 1). That is, the bio-liquid dehydrated drop is a standard thin slice of highly mobile tissue, the structures of which are being ordered and fixed during the dehydration process.

The facies appearance is used now to diagnose a wide circle of diseases. Pathological changes that take place in the body lead to violation of the chemical activity of its structures, which is reflected in morphology of the bio-liquid solid phase.

For the blood serum facies of a healthy man, symmetry of the cracks allocation is typical. The cracks are radially oriented, positioned throughout the entire facies area, and divided into radial sectors approximately equal in size, in which the concretions are situated (Fig. 1A).

Pathological states are characterized by an asymmetrical disposition of cracks, which, in their turn, form irregularly located sectors and singularities (Fig. 1B). The structure of facies of biological liquids with small protein content and increased salt content (saliva, urine, etc) differs considerably from the blood serum structure. Facies of such bio-liquids have two areas: a peripheral area shaped as a protein ring and a central area filled with salt dendrites (Fig. 1C).

Today, markers of sclerotic changes, inflammatory processes, and other pathological body conditions have been established. The obtained results of the morphologic analysis show good agreement with the clinical data of the examined persons’ health, and the biochemical and immunological tests.

In studying a material structure, the exploration of structural peculiarities of its separate elements is of great importance. In terms of spatial microstructure geometry, any material could be conceived as a conglomerate, consisting of a set of microscopic bodies filling a certain space and connected with each other by the contact surfaces (microparticles). They are structural units of a microscopic structure, much like the elementary cell is a structural unit of crystalline structure. Among the structural elements of bioliquid facies are cracks (Fig. 2A), concretions (Fig. 2A), dendrites (Fig. 2B), foreign inclusions (Fig. 2C), etc.
At present, to analyze bio-liquid structure, the descriptive approach is used. Quantitative estimation of the dehydrated bio-liquid structure makes it possible to involve the effective mathematic device as well as the computer technique to find analytical dependences, which will enable us to reveal the nature of the physical-chemical processes that take place in the material. In addition, quantitative description of the bio-liquid morphological parameters will allow new diagnostic tests to be developed.

When studying the bio-structural effects, we noticed some analogies in the character of aging and cracking processes in polymers and bio-liquids. This made it possible to use the mathematic device developed for solid-object, multiple-fracture processes describing for structures of blood serum and protein preparations objectivization[6].

In the course of investigations, the hardware-software system “Morphotest” was developed[7]. The computer-aided system of biological liquid structure analysis allows facies images to be obtained, digitized, recognized, and analyzed. The possibility of photographs, files, and assays evolving into analysis essentially widens the system capabilities. Special software methods of image improvement, original techniques to reveal diagnostically valuable structural elements, allow facies qualitative characteristics to be obtained.

A “Morphology” database has been created. It is constantly populated by different types of pathologies and more precise quantitative criteria are elaborated.

The method enables the effects to be visualized and studied, which earlier could not be observed [8].

Now high sensibility of living organisms to weak physical fields is of interest (small radiation doses, low-intensity electro-magnetic fields, laser radiation). That is why the task of detecting these reactions and studying the mechanisms of their transformation into biological functions now appears. The bio-liquid could be considered as an indicator of weak physical signals. The bio-liquid provides a simple model, useful and convenient for qualitatively and quantitatively describing certain cooperative processes that take place at a molecular and permolecular level under the influence of low intensity factors.

The blood serum compound composition, along with the diversity of the body’s physical-chemical processes, makes it difficult to decide what specific factors result in formation of those particular structures observed in the facies. In order to reveal the causes leading to some particular pathological structures formation, we are investigating protein artificial aging using simple models (albumin, in particular)[9]. The use of model liquids of simple composition allows us to reduce to a minimum the number of factors affect-
ing the structure formation, as well as to provide the varying liquid parameters controlled. The present researches, in the authors’ opinion, are to stimulate investigations of the self-organization processes in protein – basic component of the living matter.

At present, a large amount of experimental data on the bio-liquid structures has been accumulated. The data classification, explanation in unified terms, and search for new directions for further research are the main tasks of this novel scientific line development.

In conclusion, we would like to note that the body’s bio-liquids, as an object to be studied, are of interest for specialists of several scientific lines, because this is a problem of great scientific interest and practical potentialities. The task of establishing the mechanisms of bio-liquid structure formation is one of the most complicated problems of physical-chemical hydrodynamics, theory of heat and mass transfer, and phase changes. It is a part of the general scientific problem of isotropy, order, and chaos relations (self-organization).

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