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Complex-spectrum magnetic environment enhances and/or modifies bioeffects of hypokinetic stress condition: An animal study

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Abstract

Monitoring of cardio-vascular function in astronauts on Russian space station Soyuz revealed a decrease of heart rate variability during periods of increased geomagnetic activity, which is related to increased risk of cardio-vascular disorders. Spaceflight electric and magnetic environments are characterized by complex combination of static and time-varying components in ULF–ELF (ULF: 0–10 Hz; ELF: 10–1000 Hz) range and by high variability. The objective of this study was to investigate the possible influence of these magnetic fields on rats to understand the pathway regarding functional state of cardio-vascular system. Magnetic field-pattern with variable complex spectra in 0–50 Hz frequency range was simulated using three-axial Helmholtz coils and special computer-based equipment. The effect of the magnetic field-exposure on rats was also tested in combination with hypokinetic stress condition, which is typical for manned space missions. It was revealed that variable complex-spectrum magnetic field acts as a weak or moderate stress-like factor and can increase loading for regulatory mechanisms of cardio-vascular system. Various functional shifts can be amplified and modified, when the magnetic field-exposure is combined with hypokinesia. Our results support the idea that variable complex-spectrum MF action involves sympathetic activation, overload in cholesterol transport in blood and also secretor activation of tissue basophyls (mast cells) which can influence the haemodynamics. These functional shifts might lead to increased risk of cardio-vascular diseases.

Keywords: Electromagnetic environment; Biological response; Spaceflight conditions; Stress factor; Hypokinesia; Cardio-vascular system

1. Introduction

Natural and artificial electromagnetic fields (EMF) are currently viewed as important ecological factors. Spaceflight electric and magnetic environments are characterized by complex combination of static and time-varying components in ultra-low and extremely low frequency range (ULF: 0–10 Hz, ULF: 10–1000 Hz). During last decades it was shown by many authors that ULF–ELF electric and magnetic fields (MF) may produce biological effects and consequently may be a possible source for health problems. Most studies targeted industrial frequencies of 50–60 Hz. However, it has been reported that natural and technological MF in wide ULF–ELF frequency range also can be linked to different health problems, as increase in incidence of brain cancer, of leukemia, and cardiac diseases as well as to increase in work and traffic accidents (see e.g., Ptitsyna et al., 1995, 1998; Roederer, 1995; Mitsutake et al., 2005 and Refs. therein).

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Monitoring of cardio-vascular function in astronauts on Russian space station Soyuz revealed a decrease of heart rate variability during periods of increased geomagnetic activity (Baevsky et al., 1998), which is related to increased risk of cardio-vascular disorders. However, the physiological and metabolic mechanisms of possible biological activity of variable complex-spectra MF typical for spaceflight environments remain unknown.

The objective of this study was to investigate the influence of variable MF with complex spectra in 0–50 Hz frequency range on several metabolic patterns that can reflect negative shifts for animal cardio-vascular system under normal and stress conditions. Hypokinetic conditions that are typical for space manned missions were chosen as a stress factor.

2. Experimental design and methods

To investigate possible biological effects of the complex spectra MF-pattern, we used not-bread male rats six months of age. Animals were tested in ‘‘open field’’ to estimate the constitutional characteristics of organisms. The ‘‘open field’’ test characterizes the integral response of animals to the new surrounding which represents a weak stress-factor.

Four animal groups were used in the experiment: (i) control ($n = 15$); (ii) exposed to complex spectra MF ($n = 15$); (iii) stressed by hypokinesia ($n = 15$); (iv) simultaneously stressed and exposed to MF ($n = 15$). The control group (i) was located in the laboratory near the stressed animals (iii). Each group includes the same number of animals with low ($n = 5$), middle ($n = 5$) and high ($n = 5$) activity in ‘‘open field’’. Such a procedure allowed us to form identical experimental groups regarding individual psycho-physiological features of the animals and also to analyze the dependence of MF- and stress-induced response on individual-typological peculiarities of animal organism.

The experiments were carried out from June 8 to June 10, 1999 for simple MF-exposure and from June 15 to June 23, 1999 for multiple MF-exposure. Hypokinetic stress was developed by placing the animals in narrow boxes during 23 h a day. The rest and feeding of animals, the cleaning of boxes have been done during 1 h. Animals were kept under the natural changes of diurnal luminosity. It is known that the effect of a load (‘‘stress’’) depends on the stage of circadian rhythm. However, it relates to a short-time load. The design of our experiment (constant limitation of motive activity for 23 h per day) allowed us not to take into account the phases of diurnal biorhythms.

The time of MF-exposure of animals was 3 h, from 9.00 to 12.00. In the experiments with simple MF-exposure the hypokinetic stress started 3 days before the experiment. In experiments with multiple MF influence the hypokinetic stress started and finished simultaneously with MF-exposures (the duration of the experiment was 9 days, daily 3-h MF-exposure). Such experimental design was used because the development of stress-reaction in white rats is a phase process (Temuryants et al., 1989), which includes the alarm phase, adaptation and disadaptation phases. Since hypokinesia is a moderate stress-factor, the third phase does not develop. The first phase of the stress-reaction (alarm phase) is most pronounced on the third day. During this phase animals are highly sensitive to various factors, supposedly also to simple MF-exposure. The phase of adaptation is clearly manifested only after 8th–10th days of hypokinesia. Therefore, it was interesting to estimate the influence of multiple MF-exposures on the development of adaptation in animal organism to hypokinetic stress.

One of the aims of this study was to design, construct and validate a three-axis MF-exposure facility in which appropriately controlled conditions could be maintained (see Fig. 1). We designed and developed an exposure facility which allows creating varying and/or static MF in each axis separately or simultaneously. MF can be generated both in a manual mode and automatically by means of a host computer. The exposure facility includes a reference magnetometric device MVC-2 for simultaneous monitoring of static and variable MF during experiments. MVC-2 is a waveform capture system with sampling rate up to 200 Hz. The computer-based waveform capture system MVC-2 was used for testing characteristics of the constructed exposure facility: shielding factor of the shield room, Helmholtz coil constant, and the spatial homogeneity of generated MF in the exposure volume. The shielding factor in the center of device for static fields ranged from 3.85 for Z direction to 19.1 for Y direction. It was defined by the work exposure volume, where non-linearity of generated MF was less than 10%. This exposure facility was used for studying response of rats to MF. The MF-pattern with complex spectra in ULF–ELF range used in our experiment is shown in

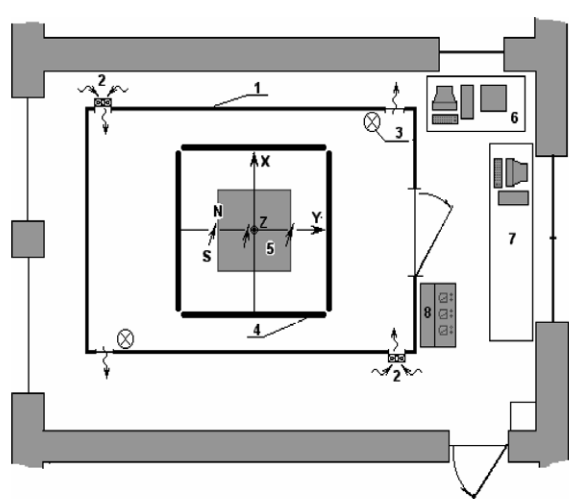

Fig. 1. The shield chamber and devices for MF simulation. 1, shield walls; 2, ventilation outlets; 3, bulbs of direct current (100 W); 4, three pairs of orthogonal Helmholtz coils; 5, work area; 6, the computer-based device for MF measurement; 7, computer for MF simulation; 8, code-analogue counter and three-channel amplifier.

Fig. 2. In the biological experiments this MF-pattern was cyclically repeated during the exposure time.

The following metabolic indices in blood were estimated in this experiment: total non-polar lipids, total cholesterol, cholesterol in α- and β-lipoproteines, total phospholipids, fatty acids, secondary lipid peroxidation products, total tiols. Also the degranulation rate of mast cells was used as one of the test of biological activity of complex spectra MF.

We used the following statistical parameters: mean, standard deviation, standard error. Student's *t*-test was used to clarify whether the difference between two means was statistically significant. Revealed differences were considered statistically significant when the *p*-value (probability for the found results to be accidental) was below 0.05 ($p < 0.05$), and as a tendency, when the *p*-value was below 0.10 ($p < 0.10$). Bonferroni's correction were not used, attention was concentrated on pair comparison.

3. Results

First, it is necessary to note that it is seen (Figs. 3 and 7) rather big differences between the two control groups (simple and multiple exposure groups). It is commonly known that oscillations of physiological and metabolic indices within the limits of physiological norm, with periods from a few hours to a few days, are usual for the organism of man and animals. This circumstance is the cause of the indicated differences between control groups in our experiments, since the experiments with simple and multiple MF-exposure were carried out in different days. Therefore, the estimation of effects of MF-exposures was conducted exceptionally in relation to the correspondent controls.

We found statistically significant MF-associated changes for various chosen indices. These changes depend on a functional state of the organism and its individual-typological features. The most interesting results are presented in Figs. 3–8 as examples of specific biological effects of MF influence.

Mast cells are located in all tissues and synthesize a great variety of biologically active substances, such as histamine, serotonine, prostaglandines, heparine, proteases, etc. They play an important role in the regulation of several links in immune system, and also in regional haemodynamics, trophic processes and tissue transformation. A statistically significant increase of degranulation rate was revealed

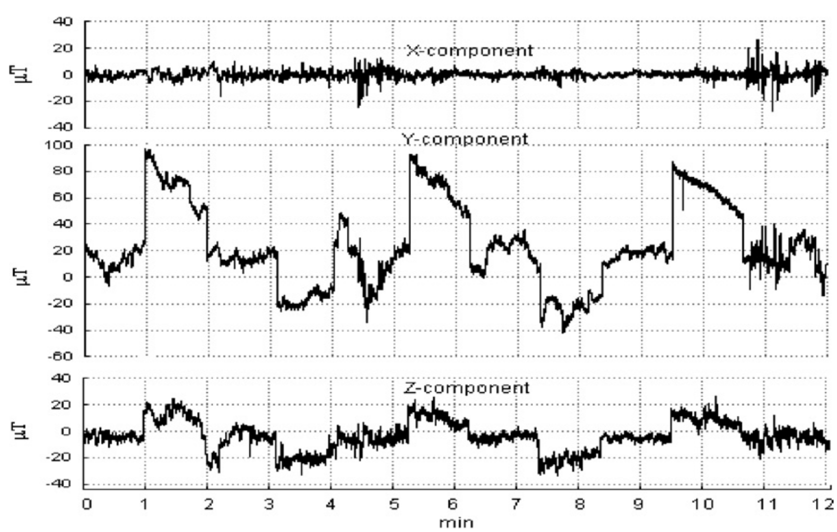

Fig. 2. The MF-pattern generated by the computer-based facility.

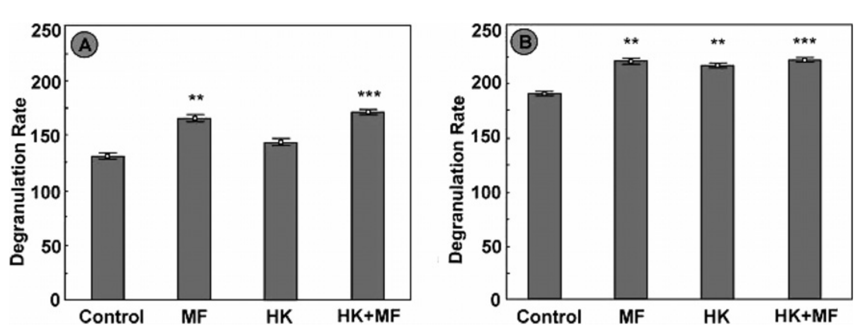

Fig. 3. Degranulation rate of mast cells in animals under simple (A) and multiple (B) MF-exposure (without considering the individual-typological features of animals). Here and in the next Figures: control, biological control; MF, MF-exposure; HK, hypokinetic stress; HK + MF, hypokinetic stress and MF-exposure simultaneously. Statistical significance of differences relatively to control, $^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.001$, t, $p < 0.10$ (tendency).

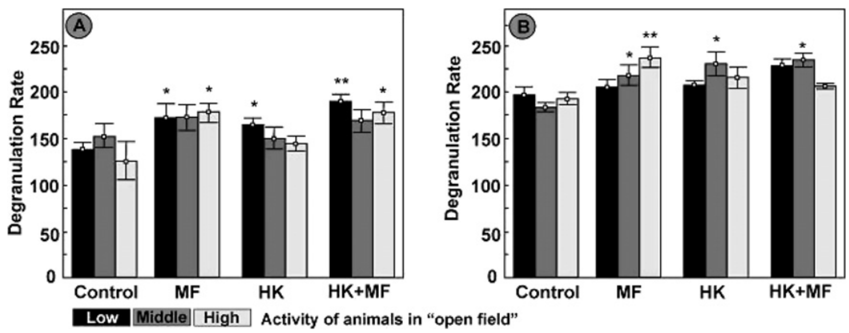

Fig. 4. Degranulation rate of mast cells in animals with different individual-typological features under simple (A) and multiple (B) MF-exposure.

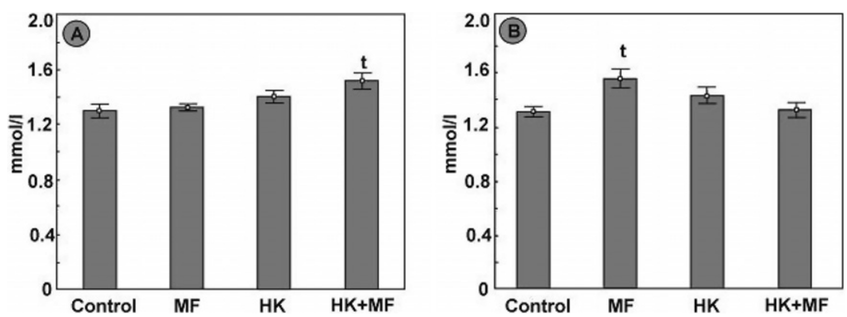

Fig. 5. Levels of total cholesterol in blood plasma of animals under simple (A) and multiple (B) MF-exposure (without considering the individual-typological features of animals).

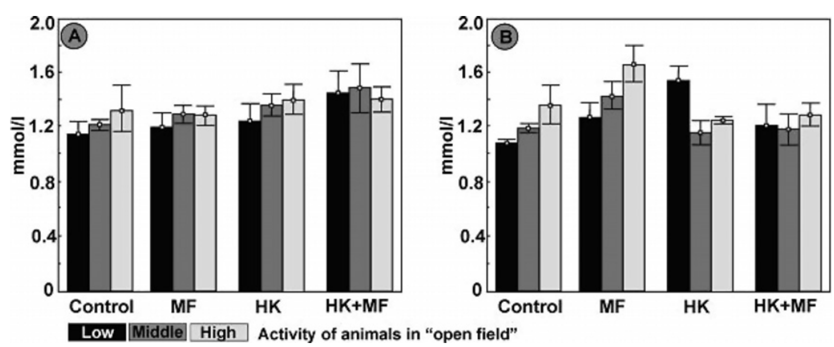

Fig. 6. Levels of total cholesterol in blood plasma of animals with different individual-typological features under simple (A) and multiple (B) MF-exposure.

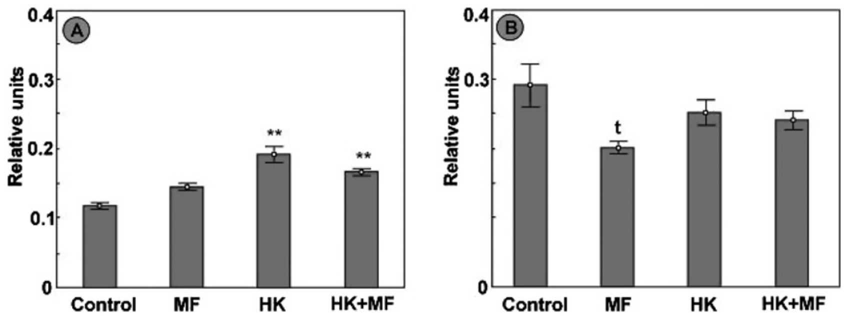

Fig. 7. The ratio between the level of total cholesterol and phospholipids in blood plasma of animals under simple (A) and multiple (B) MF-exposure (without considering the individual-typological features of animals).

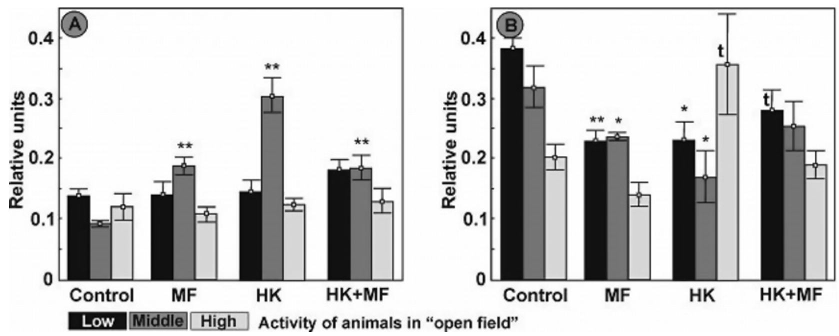

Fig. 8. The ratio between the level of total cholesterol and phospholipids in blood plasma of animals with different individual-typological features under simple (A) and multiple (B) MF-exposure.

under both simple and multiple MF-exposure (Figs. 3 and 4). Our results provide evidence that the response of mast cells to complex-spectrum MF is similar to the response to stress factors of different nature. The functional activity of mast cell can be increased owing to raised concentration of stress-hormones in experiments *in vivo* (Baldwin, 2006). Besides, the direct response of mast cells to MF was shown in experiments *in vitro* (Abu Khada and Martynyuk, 2001).

The different response of mast cell in animals with different individual-typological (constitutional) features (Fig. 4) indicates that MF-induced cell response depends on hormonal and immune status of organism. For example, in animals which showed high activity in "open field" and low anxiety, the MF-induced response of mast cells exceeds the response to hypokinetic stress (Fig. 4). Therefore the mast cells, as well as pinealocytes in pineal gland in CNS, seem to be cellular elements sensitive to MF. Possibly, the MF-induced increase of degranulation rate and the subsequent change of capillary blood-flux in different tissues can be one of the reasons for a raised functional loading of cardio-vascular system in men and animals. On the other hand, the activity of mast cells play a key role in immune processes which are responsible for the development of allergy. Therefore, we can suppose that MF is an environmental factor which can increase the sensitivity of the organism to different allergic agents.

The changes of the level of total lipids, fatty acids, phospholipids in blood were revealed in experiments with simple and multiple MF-exposure. The shifts in the concentration of cholesterol and cholesterol/phospholipids ratio are shown in Figs. 5 and 6, Figs. 7 and 8, respectively. It is known that cholesterol is an important component of cell membranes, but high level of cholesterol in blood is one of major risks for coronary heart disease, which can lead to heart attacks. Therefore, the indices of cholesterol level, as well as cholesterol/phospholipids ratio, are widely used in medicine for prognosis of atherosclerosis development.

Our results show a weak increase of total cholesterol in blood plasma after simple and multiple MF-exposures (see Fig. 5). On the contrary, a detailed analysis showed a steady tendency to increase blood cholesterol in animals with low and middle activity in "open field" (see Fig. 6). It should be noted that the found change in terms of cholesterol is on the borderline of statistical significance level. However, these steady tendencies of changes in biological indices deserve attention in view of future planning of more detailed researches.

The simultaneous MF-induced changes of phospholipid level in blood plasma result in changes of cholesterol/phospholipids ratio (see Figs. 7 and 8). Thus, also in this case (as for degranulation rate in mast cells) complex-spectrum MF acts as a stress-like factor. Therefore, the development of deeper and long-term functional shifts in cardio-vascular system in population, which is chronically exposed to complex-spectrum MF, could be connected to shifts in lipid metabolism, first of all in cholesterol link.

4. Summary and conclusion

- Statistically significant shifts of functional activity have been found in different metabolic and physiological processes under simple and/or multiple influence of MF with complex spectra in low frequency range. The value and direction of functional shifts caused by MF influence depend on individual-typological (constitutional) features and also on physiological state (norm, stress) of organism.
- Among the studied biological parameters, the degranulation rate of mast cells and also the indexes of lipid metabolism and cholesterol transportation in the blood system seem to be the most sensitive to MF-exposure.
- The exposure of organism to complex spectra MF modifies the development of moderate stress-reaction in the organism. In many cases complex spectra MF acts as a weak or moderate stress-like factor, which amplifies or modifies the functional shifts caused by other stress-factors, such as the hypokinetic stress.

Our results indicate that MF with highly variable complex spectra, typical for spaceflight environments, can be one of the factors increasing the loading for regulatory mechanisms of cardio-vascular system. This increased loading could lead to development of classical pathogenesis states such as myocardial ischemia, arrhythmia and others, which lead to increased risk of cardio-vascular catastrophes. Moreover, these MF could increase the sensitivity of organism to different allergic agents.

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