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ASSESSMENT OF BIOLOGICAL EFFECTS UNDER THE CONDITIONS OF COMBINED EXPOSURE TO HARMFUL PRODUCTION FACTORS

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Introduction. The current state of the industrial environment is represented by various combinations of production factors that can negatively affect a person in the production process. The problem with this issue is that currently, there are no algorithms for determining which of the factors makes a greater contribution to the formation of the corresponding reaction of the organism to the combined influence of factors and which patterns of the corresponding reaction of the organism may occur. At the same time, the knowledge of such regularities is an important task for occupational medicine in terms of the analysis of occupational conditions and the state of health of the working staff.

The aim of the study was to establish the changes of an animal organism under the simultaneous exposure to electromagnetic radiation (EMR) and moderately low temperature (MLT) and to determine the contribution of each factor to the overall biological effect using mathematical analysis.

Materials and methods of research. The biological effects of the combined influence of EMR and MLT in an experiment on laboratory animals (male rats) were investigated. Biochemical parameters were determined, in particular, the state of peroxidation, antioxidant protection, lipid and carbohydrate metabolism, the status of trace elements and immunological reactivity in terms of cellular and humoral links, reproductive function. The proposed mathematical approach based on artificial intelligence was used (fuzzy membership level) to determine the intensity of EMR and MLT under the combined influence of these factors and to establish the most informative indices using factor analysis (principal components analysis).

Results. The results showed that, under the combined influence of factors, the biological effects were manifested by the following phenomena: morphological changes in the organs of internal secretion (EMR and MLT with the equal degree of contribution = 0.50), increased peroxidation with simultaneous moderate inhibition of antioxidant protection, increased lipid metabolism with dyslipoprotein phenomena (degree of contribution for MLT was 0.67, degree of contribution for EMR was 0.33). As for the immune system, phase changes in the work of both cellular and humoral units (degree of contribution for EMR was 0.53, degree of contribution for MLT was 0.47), disorders in the functioning of sperm (degree of contribution for EMR was 0.55, degree of contribution for MLT was = 0.45) were revealed.

Conclusions. The method of determining the intensity of the impact allowed to reveal that each of the factors had its share of contribution to the studied systems. According to the general calculations, the largest contribution was made by MLT, which added 54 % of its contribution to the overall biological effect against EMR, for which the share of the contribution was only 46 %. Such an analysis made it possible to predict the likely response of the whole organism and to develop reasonably appropriate preventive measures.

Key words: electromagnetic radiation, moderately low temperature, combined effect, experiment, clustering, principal component analysis

Introduction

The current ecological situation is characterized by a possible combination of some factors. Numerous studies show that the combined effect of environmental factors differs from their isolated effects. There is a urgent need to determine the nature of the body's reactions, reveal subtle biological mechanisms that form the corresponding reaction and to reveal the degree of membership level of each factor to the total effect [1].

The electromagnetic radiation (EMR) and moderately low temperature (MLT) are rather significant factors, which often accompany the effect of the mentioned anthropogenic factor.

With technological progress, there is a growing interest in studying such a phenomenon as «electromagnetic pollution», which is stipulated by the widespread use of EMR [2, 3]. The wide use of industrial technologies and household appliances, which is accompanied by the generation of EMR, creates a

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potential possibility for its influence on the human body in everyday life, as well as at the workplace [3]. EMR has a various biological activity influencing different organs and systems. The greatest biological activity is manifested in the central nervous system, cardiovascular, endocrine, and immune systems [2, 4].

In addition, the air temperature is an important environmental factor, which can provide comfortable or uncomfortable living conditions. The adverse effect of air temperature on the body is possible under various circumstances, especially in winter, when the air temperature decreases [5]. Cold stress induces the activation of the most important regulatory systems in order to maintain a constant body temperature [6, 7].

In this connection, the problem of analyzing the body's reactions to the combined effect of mentioned factors is extremely relevant and requires a comprehensive study. However, the analysis of multifactorial effects also requires adequate mathematical and statistical methods of analysis, which, in turn, will determine the share of the contribution of each active factor to the overall biological effect, as well as define the relationship between the indices in order to reveal the mechanisms of formation of the effects obtained [8].

The aim of the study was to reveal the changes of an animal organism under the simultaneous exposure to EMR and MLT and to determine the contribution of each factor to the overall biological effect using mathematical analysis.

Materials and methods of research

The study of the combination of EMR with MLT in a subchronic experiment lasted for 30 days. The research was performed on WAG rats (n = 172). The animals were divided into four experimental groups: 1st – a control group, where the animals were in comfortable temperature conditions; 2nd – a group of isolated action of MLT equal to + 4 °C; 3rd – a group exposed to EMR only with the operating frequency in the flat-parallel capacitor – 70 kHz and the intensity of the electric component of electromagnetic field in the working volume of the capacitor of 600 V/m; 4th – a group exposed to a combined effect of these two factors. The experiment was modeled in a special exposure chamber [29]. The animals were under the influence of environmental factors for 4 hours daily.

For immunological, biochemical studies and identification of functional status of spermatozoa, the rats were removed from each group at each stage (5, 15, 30 days) of the experiment.

Changes in the body were assessed according to blood parameters (lipid peroxidation (LPO), antioxidant system (AOS), lipoprotein metabolism, general metabolic processes, phagocytosis, compliment system C3, C4, C5, immunoglobulins IgA, IgM, IgG) – 36 indices in total, functional status of spermatozoa (total concentration of spermatozoa, concentration of morphologically normal spermatozoa forms, concentration of pathological forms of spermatozoa in sperm, number of motile, non-motile and dead cells, time of motility, acid and osmotic resistance) – 9 indices in total.

The morphometric analysis of the spleen, testes, thyroid gland, adrenal glands and liver were performed at the end of experiment (30 days) – 18 indices.

The next stage was the use of mathematical and statistical methods for analyzing the obtained data to determine the contribution of EMR and moderately low temperature effects to the overall biological effect.

$$dist_q\,(c_{il},c_{il}) = \sum_{i=1}^{n} |c_{iq} - c_{iq}|, \qquad (1)$$

where c_{iq} – center of each group.

After calculating the distance, it was necessary to determine the degree of contribution of each of the experimental groups to the group of combined influence of factors.

$$ml_q = \frac{dist_q^{-1}}{\sum(dist_q^{-1})}\ . \qquad (2)$$

The last stage was to determine the informativeness of indices to reveal the nature of biological effects in the combined action of factors.

$$d(x_{il}(z), \hat{y}_{il}^{(1)}) = \sum_{i=1}^{N}\sum_{l=1}^{q} |x_{il}(k) - \hat{y}_{il}^{(1)}|, \qquad (3)$$

where $x_{il}(k)$ – vector that contains all features and $\hat{y}_{il}^{(1)}(X)$ – first principal component of multidimensional matrix of features.

Results and their discussion

According to the study conducted on the 5th day of the experiment, the biological effects were characterized

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by the following phenomena: activation of LPS processes with simultaneous suppression of AOC; imbalance in the lipid spectrum, with prevalence of atherogenicity; the decrease in phagocytic activity and humoral unit of the immune response (according to IgM and the component of the complement system C4), which was compensated by an increase in the formation of extracellular neutrophils; disorders of spermatozoa function in terms of: an increase in pathological forms of spermatozoa, the number of non-motile spermatozoa and a simultaneous decrease in morphologically normal forms of spermatozoa and time of their motility.

On the 15th day, qualitative changes were similar to the initial ones, but more pronounced in biochemical parameters and functional status of spermatozoa. However, the opposite effect (stimulation) in the immune system was noted.

On the 30th day, the mentioned effects only intensified.

The calculation of the degree of contribution of the group of combined effect and groups of isolated effect to the overall impact was made. The latter data enabled to calculate the contribution of each factor to the overall biological effect.

We obtain a matrix of distances, the first row corresponds to the 5-th day of experiment ($q = 1$), the second row – 15-th day of experiment ($q = 2$) and the third row – 30-th day of experiment ($q = 3$). These distances allow us to find membership levels.

The smaller the distance between the cluster centers, the greater the contribution.

This mathematic model reflects changes in biochemical and immunological indices, parameters of functional status of spermatozoa and morphometric indices of internal organs under the combined effect of MLT and EMR.

Therefore, the greatest negative impact on the 5-th day of the experiment was due to MLT, with a degree of contribution of $\mu_1 = 0.53$, against EMR which was equal to $\mu_2 = 0.47$ (Figure 1).

When calculating the distance between the centers of the clusters, the total membership level at the stage of the 15th day for the group was MLT $\mu_1 = 0.55$, and for the group of isolated exposure to EMR $\mu_2 = 0.45$ (Figure 2).

According to the results of the mathematical analysis, the membership level for 30 days for the group of isolated exposure to MLT was also higher and amounted to $\mu_1 = 0.52$ against the group of isolated exposure to EMR $\mu_2 = 0.48$ (Figure 3).

Thus, the total degree of MLT membership level in the total biological effect was 0.54 against EMR, which was equal to 0.46.

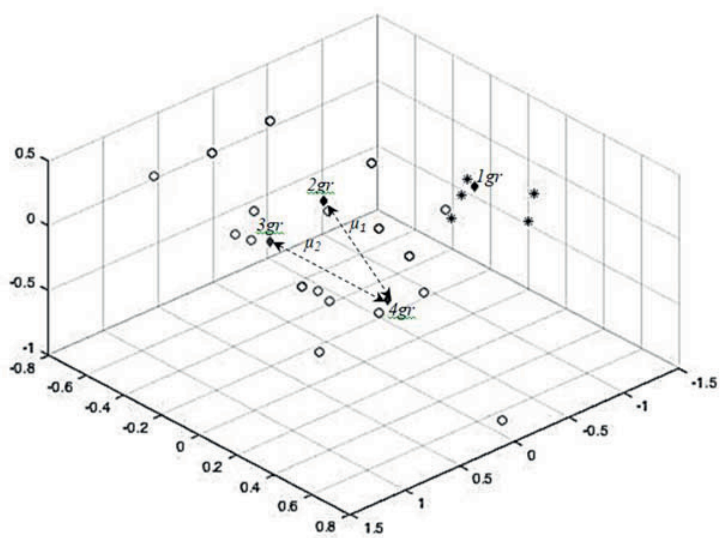

Figure 1. Spatial visualization of changes in general indices under the combined effect of EMR and MLT. The 5th day of the experiment

***Note.** Here and in Figure 2, 3: *o – groups exposed to EMR only, exposed to MLT only, exposed to combined EMR and MLT, *control group; 1 gr ♦ – cluster center of control group, 2 gr ♦ – cluster center of group exposed to MLT only, 3 gr ♦ – cluster center of group exposed to EMR only, 4 gr ♦ – cluster center of group exposed to combined EMR and MLT; μ_1 – degree of contribution between groups exposed to MLT only and exposed to combined EMR and MLT, μ_2 – degree of contribution between groups exposed to EMR only and exposed to combined EMR and MLT.*

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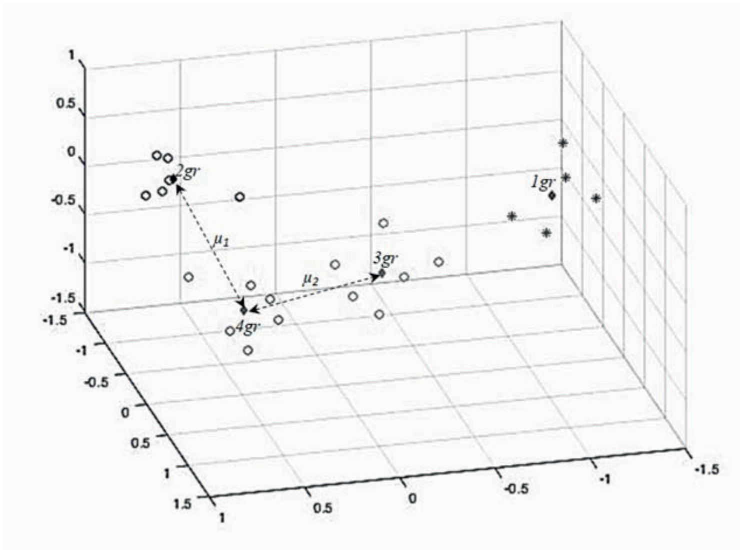

Figure 2. Spatial visualization of changes in general indices under the combined effect of EMR and MLT. The 15th day of the experiment

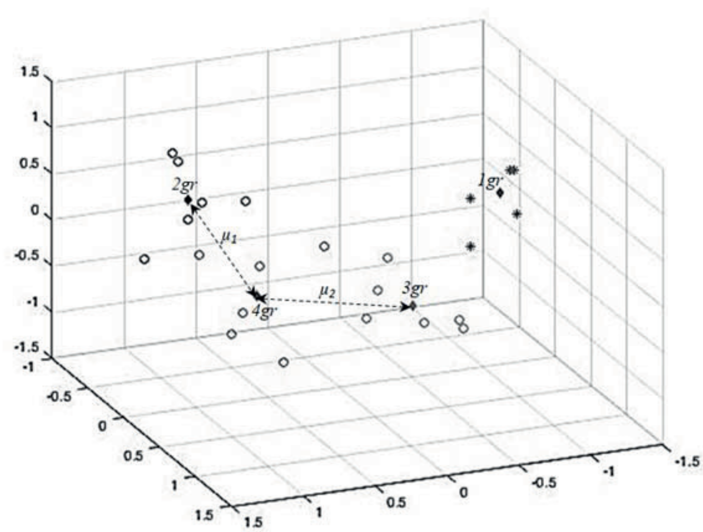

Figure 3. Spatial visualization of changes in general indices under the combined effect of EMR and MLT. The 30th day of the experiment

However, this mathematical approach allowed us to determine not only the contribution of each factor to the overall biological effect, but also the share of the contribution of each factor to individual systems as well.

The data analysis revealed the following results. EMR had the greatest negative impact on the reproductive and immune systems; degree of contribution was 0.55 and 0.53, respectively. MLT had a degree of contribution 0.45 (reproductive system) and 0.47 (immune system). The general metabolic processes were mostly affected by MLT, with a degree of contribution 0.67, EMR had the value 0.33. The morphological changes of the internal organs were caused by mentioned factors with the equal degree of contribution (0.50).

The method of the Principal Component Analysis enabled to determine the criterion-significant indices in the experiment, allowing to obtain a description of the development of biological effects.

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Criteria-significant indices were determined: the concentration of malondialdehyde (MDA), low-density lipoproteins, the activity of superoxide dismutase and catalase, the nitroblue tetrazolium test (NBT-test), the width of the fasciculate zone and glomerular zone of the adrenal glands, the number of motile and non-motile spermatozoa and the total concentration of spermatozoa.

The method of determining the intensity of factors, developed on the basis of factor and cluster analyzes, allowed to find out the degree of contribution of groups of isolated influence of factors to the group of combined influence both on separate functional systems and on the total sum of all studied indices at all stages. The data, criterion-significant indices in the dynamics of the experiment, which allow to characterize the development of biological effects, determine the leading role of each factor in the development of biological effects and predict the possibility of changes in the body's responses to changes in intensity and duration.

Using this approach, it was found that the temperature factor is a leading factor in the formation of biological effects, as low temperature is a strong stressor that causes significant physiological changes in the body to maintain thermogenesis, which can complicate the response to other factors [9]. The essence of stress is to enhance the work of all organs and systems of the body in a critical situation, which helps to adapt to the environment.

The corresponding response of the organism to the combined influence of factors was manifested primarily by the regulation of the neuroendocrine system, which may affect the acceleration of oxidation processes [10], the strengthening of which was proved in our study by determining the informative rate of malonic dialdehyde. It is obvious that the formation of free radicals is one of the universal pathogenetic mechanisms due to the oxidation of LDL and VLDL [11, 12]. As a result of intensive lipolysis in the blood and cell membranes, the levels of triglycerides, fatty acids and cholesterol increase. This nonspecific reaction leads to an imbalance in the system of prooxidants/antioxidants [13].

Support for thermomeostasis is provided by the integrative interaction of various body systems, including the immune and reproductive systems. The observed decrease in the functional activity of neutrophils and the functional state of sperm can be explained from the standpoint of the development of a stress response as a result of influence of low temperatures [18, 19].

The analysis of the combined influence of factors shows that in response to the influence of stress factors, a nonspecific response of the organism unfolds, the mechanism of which is inherent in the influence of low temperatures. The action of cold enhances the effect of EMR on the body in terms of biochemical parameters and inhibits the stimulating effect of EMR in terms of immunology.

Conclusion

The used mathematical approach allowed us to analyze the data obtained in the course of the animal experiment and to identify the key part of each factor studied in conditions of their combined effects on different systems and organs.

The analysis of the data revealed the following phenomenon: EMR had the greatest negative impact on the reproductive and immune systems. The general metabolic processes were mostly affected by MLT.

The total degree of MLT membership level in the total biological effect was 0.46 against EMR, which was equal to 0.54.

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ОЦІНКА БІОЛОГІЧНИХ ЕФЕКТІВ В УМОВАХ СПОЛУЧЕНОГО ВПЛИВУ ШКІДЛИВИХ ФАКТОРІВ ВИРОБНИЦТВА

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Вступ. Сучасний стан виробничого середовища представлений різноманітними комбінаціями виробничих факторів, які можуть негативно впливати на людину в трудовому процесі. Проблематика цього питання полягає в тому, що наразі відсутні алгоритми визначення того, який з факторів робить більший внесок у формування відповідної реакції організму на сполучений вплив чинників та які виникають закономірності відповідної реакції організму, що є важливим завданням для медицини праці задля ефективного аналізу виробничих умов та здоров'я працюючого контингенту.

Мета дослідження. Встановити зміни в організмі тварин при одночасному впливі електромагнітного випромінювання (ЕМВ) і помірно низької температури (ПНТ) та за допомогою математичного аналізу визначити внесок кожного з факторів у загальний біологічний ефект.

Матеріали та методи дослідження. Досліджено біологічні ефекти сполученого впливу ЕМВ і ПНТ в експерименті на лабораторних тваринах (щурах-самцях). Визначали біохімічні показники, зокрема, стан перекисного окиснення, антиоксидантного захисту, ліпідного та вуглеводного обмінів, мікроелементний статус та імунологічну реактивність клітинної та гуморальної ланок, репродуктивну функцію. Запропоновано математичний підхід на основі штучного інтелекту (розрахунок рівня нечіткої належності) для визначення інтенсивності ЕМВ і ПНТ за умов сполученого впливу цих факторів і встановлення найінформативніших показників за допомогою факторного аналізу (метод головних компонент).

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Результати. Встановлено, що за умов сполученого впливу факторів біологічними ефектами є морфологічні зміни в органах внутрішньої секреції (ЕМВ і ПНТ з рівною часткою внеску 0,50), посилення перекисного окиснення з одночасним помірним пригніченням антиоксидантного захисту, посилення ліпідного обміну з дисліпопротеїновими явищами (частка внеску для ПНТ – 0,67, частка внеску для ЕМВ – 0,33). Із боку імунної системи фазові зміни в роботі як клітинних, так і гуморальних одиниць (частка внеску для ЕМВ – 0,53, частка внеску для ПНТ – 0,47), порушення функціонування сперматозоїдів (частка внеску для ЕМВ – 0,55, частка внеску для ПНТ – 0,45).
Висновки. Методика визначення інтенсивності впливу дозволила встановити, що кожен з факторів мав свою частку внеску в досліджувані системи. Найбільший внесок зробила ПНТ, яка додала 54 % своєї частки в загальний біологічний ефект проти ЕМВ, для якого частка внеску становила 46 %. Такий аналіз дозволив передбачити ймовірну реакцію всього організму й обґрунтовано розробити відповідні профілактичні заходи.

Ключові слова: електромагнітне випромінювання, помірно низька температура, комбінована дія, експеримент, кластеризація, аналіз головних компонент

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ОЦЕНКА БИОЛОГИЧЕСКИХ ЭФФЕКТОВ В УСЛОВИЯХ СОЧЕТАННОГО ВОЗДЕЙСТВИЯ ВРЕДНЫХ ФАКТОРОВ ПРОИЗВОДСТВА

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Введение. Современное состояние производственной среды представлено разнообразными комбинациями производственных факторов, которые могут негативно влиять на человека в трудовом процессе. Проблематика этого вопроса заключается в том, что пока отсутствуют алгоритмы определения того, какой из факторов вносит больший вклад в формирование ответной реакции организма на сочетанное влияние факторов и какие возникают закономерности ответной реакции организма, что является важной задачей для медицины труда с целью эффективного анализа производственных условий и состояния здоровья работающего контингента.
Цель исследования. Установить изменения в организме животных при одновременном воздействии электромагнитного излучения (ЭМИ) и умеренно низкой температуры (УНТ), а также с помощью математического анализа определить вклад каждого из факторов в общий биологический эффект.
Материалы и методы исследования. Исследованы биологические эффекты сочетанного влияния ЭМИ и УНТ в эксперименте на лабораторных животных (крысах-самцах). Определяли биохимические показатели, в частности, состояние перекисного окисления, антиоксидантной защиты, липидного и углеводного обменов, микроэлементный статус и иммунологическую реактивность клеточного и гуморального звеньев, репродуктивную функцию. Предложен математический подход на основе искусственного интеллекта (расчет уровня нечеткой принадлежности) для определения интенсивности ЭМИ и УНТ при условии сочетанного воздействия этих факторов и установления наиболее информативных показателей с помощью факторного анализа (метод главных компонент).
Результаты. Установлено, что в условиях сочетанного влияния факторов биологическими эффектами были: морфологические изменения в органах внутренней секреции (ЭМИ и УНТ с равной долей вклада 0,50), усиление перекисного окисления с одновременным умеренным угнетением антиоксидантной защиты, усиление липидного обмена с дислипопротеиновыми явлениями (доля вклада для УНТ – 0,67, доля вклада для ЭМИ – 0,33). Со стороны иммунной системы фазные изменения в работе как клеточных, так и гуморальных единиц (доля вклада для ЭМИ – 0,53, доля вклада для УНТ – 0,47), нарушение функционирования сперматозоидов (доля вклада для ЭМИ – 0,55, доля вклада для УНТ – 0,45).
Выводы. Методика определения интенсивности влияния позволила установить, что каждый из факторов имел свою долю вклада в исследуемые системы. Наибольший вклад внесла УНТ, которая прибавила 54 % своей доли вклада в общий биологический эффект против ЭМИ, для которого доля вклада составляла 46 %. Такой анализ позволил предположить вероятную реакцию всего организма и обоснованно разработать соответствующие профилактические меры.

Ключевые слова: электромагнитное излучение, умеренно низкая температура, комбинированное действие, эксперимент, кластеризация, анализ главных компонент

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