JPRS 63321 30 October 1974

# BIOLOGICAL EFFECTS OF RADIOFREQUENCY ELECTROMAGNETIC FIELDS

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#### NOTE

Title and Subtitle BIOLOGICAL EFFECTS OF RADIOFREQUENCY	
BIOLOGICAL EFFECTS OF RADIOFREQUENCY	5. Report Date
AND	30 October 1974
ELECTROMAGNETIC FIELDS	6.
Author(s) Z. V. Gordon, Editor	8. Performing Organization Rept. No.
Performing Organization Name and Address	10. Project/Task/Work Unit No.
Joint Publications Research Service	11 Contract /Great No
Arlington, Virginia 22201	, in contract, Grant No.
, Sponsoring Organization Name and Address	Covered
As above	
	14.
. Supplementary Notes	·
O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY	RADIOCHASTOT,
NO 4, 1973, FIOSCOW	
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The Report contains results of studies of hygieni	c standards at industrial
sites. Data is also presented on in-depth studie	s of the mechanism of action
of electromagnetic fields	
	· · ·
. Key Words and Document Analysis. 170. Descriptors	
USSR	
Biology and Medical Sciences	
Environmental Biology Electromagnetism	
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b. Identifiers Open-Ended Terms : COSATI Field/Group 06F Availability Statement Unlimited Availability	19. Security Class (This Report) 266
b. Identifiers Open-Ended Terms : COSATI Field/Group 06F Availability Statement Unlimited Availability Sold by NTIS	19. Security Class (This Report)       21. No. of Pages         UNCLASSIFIED       266         20. Security Class (This 22. Price

### JPRS 63321

30 October 1974

Page

# BIOLOGICAL EFFECTS OF RADIOFREQUENCY ELECTROMAGNETIC FIELDS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian No 4, 1973

[Book edited by Z. V. Gordon, signed to press 14 December 1973, 400 copies]

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#### FOREWORD

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 p 5

[Article by Academician A.I. Berg]

/Text/ The problem pertaining to the influence exerted on the human organism by radiofrequency electromagnetic fields, which are a factor of the industrial environment and of every-day living conditions, continues not only to be important but also acquires special significance in view of developments in science and technology. This applies, first of all, to very low intensity radiofrequencies, particularly the superhigh frequencies.

During the last 20 years numerous studies have been published in the Soviet Union and abroad dealing with this problem. Among these studies a significant place is held by the work conducted at the Laboratory of Radiofrequency Electromagnetic Waves of the Order of the Red Banner of Labor Institute of Labor Hygiene and Occupational Diseases of the Academy of Medical Sciences USSR.

This, the fourth, collection describing the work of this laboratory covers results obtained during the 1968-1972 period and undoubtedly represents a fundamentally new outlook. Contained are results of studies and investigations on hygienic standards during a period of extensive application of safety measures developed by Soviet scientists for improving the sanitary level of industrial sites. Results are also presented on in-depth studies on the mechanism of action of electromagnetic fields, including those at the level of fine physiological regulation, which are of interest not only in biology and medicine, but also in the sense of bionics.

It cannot be doubted that the publication of the present compendium will have a profound effect on a wide circle of scientific and practical workers in the biomedical, engineering and technical fields.

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K.V. Nikonova and I.P. Sokolova have shown convincingly in animal experimentation that at high intensities of combined radiation synergism prevails, while in the case of long-term experiments with low intensities of the combined factors, the changes that resulted were no greater than when the factors were employed alone. It should be noted that at low levels of radiation the importance of the SHF factor is more pronounced in the combination. These findings guided our approach in setting the hygienic standards for combined effects of SHF and soft x-rays.

A combination of SHF and elevated air temperature is frequently encountered in the industrial situation, or it is related to climatic conditions, and appears as an important hygienic factor which required special studies for its resolution.

At the present time, on the basis of the experimental studies of K.V. Nikonova, as well as on the basis of the data of V.A. Zhuravleva /1972/, there are reasons for believing that synergism prevails when the effects of SHF and heat are combined, and that furthermore, only heat alone induces lesser changes in terms of a number of indices than the combination of heat and SHF of athermal intensity. This emphasizes the significant changes that occur within the organism, and which apparently are due to the important role of microwave irradiation. These investigations require further expansion particularly when dealing with low intensity levels of irradiation, since they will be highly significant in setting hygienic standards for cases in which combination of SHF and heat exist.

The fact that due consideration was given to sanitary regulations in planning and scientific organization of work, resulted in effective hygienic protection of the personnel of radio and television stations from irradiation, maintained their work capability, created favorable conditions for work at the industrial sites, and so forth. This entire set of measures significantly improved working conditions, and was largely due to the efforts of a group of scientific workers under the leadership of P.P. Fukalova.

A new trend in our investigations deals with determining the significance of conditions of irradiation to which personnel working with emitters of electromagnetic waves (EMW), including microwaves, are exposed. It is generally recognized that until recent years hygienic studies did not take into account the dynamics of irradiation of workers employed in handling microwave emitters. The second group consisted of subjects who commenced work at the installation after 1960, when the hygienic standards which we had formulated (1957) were already largely in force.

Finally, the third group consisted of individuals who first started work at that enterprise during the last five or more years, and were not subjected to irradiation exceeding 10 mW/ sq cm.

The clinical picture of the first two groups has been described by specialists at our clinic and at other clinics (E.A. Drogochina, M.N. Sadchikova, et al., N.V. Tyagin, N.V. Uspenskaya); they are characterized by the degree of clinical manifestations and the presence or absence of restorative processes.

With respect to the third group, it can only be pointed out that some of these individuals showed limited changes which became evident during the clinical observation, but that they differed little from changes in the control group, and consequently, they cannot be related to the effects of SHF radiation.

On the basis of this we feel that our closest attention must be accorded to the second group. The clinical picture of this group continues to remain unfavorable despite the relatively favorable working conditions. This may either be due to inadequacies in the hygienic standards, or to intermittent radiation.

In order to understand the mechanism of action of radiofrequency electromagnetic fields (EMF) on the entire organism, it is necessary to investigate every level of its vital activities. In connection with this it is necessary to conduct in-depth experimental and theoretical studies on the mechanism of the athermal action of microwave radiation on the molecules, elucidate the direct effects of microwaves on the excitability of isolated cells, and in connection with this, study its effects on ion transport across cell membranes, and so forth.

Already at the present time, in some cases, we may speak of a correlation of results of biophysical investigations with clinical findings. Thus, the data of V.M. Shtemler on the direct effects of microwave irradiation on the membranes of isolated erythrocytes, which results in changes in the transmembrane transport of Na+ and K+, and consequently a change in the transmembrane gradient, are in agreement with the results of clinical investigations (G.G. Lysina, et al.) is involved in the genesis of the primary syndromes in subjects exposed to irradiation under commercial conditions have been confirmed in both acute and chronic experiments on animals.

In the view of M.S. Bychkov, the data which have been obtained on cortical and subcortical relationships in the mechanism of action of the microwaves, point to the rationale of attempting to strike a regulatory balance between stimulants and tranquilizers as part of the therapeutic measures taken to combat the pathogenetic effects. At the present time this aspect still remains a point of discussion. However, in view of its practical significance, it obviously deserves special experimental studies, as well as further clinical observations.

V.N. Dumkin and S.P. Korenevskaya have observed alterations in glucocorticoid function of the adrenal glands in patients with manifested forms of radiowave disease, and interpreted this as resulting from primary lesions in the deep structures of the brain which regulate the metabolism and synthesis of corticosteroids; dysfunction in the hypothalamus-hypophysisadrenal cortex system is obviously responsible for the clinical syndromes of the disease.

According to experimental data, changes which arise in organisms under the influence of intermittent and constant low intensity radiation also clearly point to the susceptibility of the hypothalamus-hypophysis-adrenal cortex system to microwaves. This is supported by data on pathomorphological investigations of the hypothalamus which indicate changes in secretory activity of the supraoptic and paraventricular nuclei (M.S. Tolgskaya, et al.).

This is also indicated by eosinopenia in irradiated animals in response to a strong sound stimulus, and the absence of such a reaction in control animals (I.A. Kitsovskaya, E.I. Polukhina). Changes in the weight of endocrine glands (hypophysis, adrenal glands), also may indicate their altered functions (N.K. Demokidova). In addition, changes were also noted in the degree of diuresis and the excretion of Na and K in the urine (N.K. Demokidova). A tendency -- which frequently bordered on significance -- was seen in the increase of epinephrine in the adrenal glands in animals during exposure to low intensity irradiation (I.A. Kitsovskaya).

The problem of the effects of radiofrequency EMW on the reproductive function is quite timely. Experimental studies conducted on animals by A.N. Bereznitskaya and her collaborators, have shown quite convincingly that nonthermogenic Both clinical and experimental studies support the contention which we expressed earlier that to a large extent the biological effect of radiofrequencies is due to disturbances in regulatory processes.

Particularly pertinent data on the biological effects of microwaves have been obtained in recent years on the effects of very low intensity irradiation (up to 150  $\mu$ W/sq cm ; Table 1).

Studies on the problems of biological effects of radiofrequency EMW were designed to provide hygienic standards for this factor. However, the problem of standardization still requires further detailed and complex investigations.

The criteria which are utilized by institutes of work hygiene and occupational diseases for setting standards represent complex investigations consisting of three major components;

 Hygienic evaluation of the working conditions of persons working with sources of radiofrequency EMW, i.e. determination of the actual intensities of radiation.

2. Dynamic clinical observations on the state of health of people working with sources of radiofrequency EMW radiation over a period of several years (5 and more).

Comparison of clinical and hygienic data permit more accurate determination of the work conditions and state of health of the individual.

3. Experimental studies on animals designed to yield threshold values for functional changes which appear largely during chronic exposure (from 4 - 10 months for the different biological indices).

It is of advantage to discuss this problem in somewhat more detail without, however, any pretention at complete coverage.

The problem lies in the fact that, unfortunately, the concept of "threshold reaction" from the point of view of medical evaluation of a biological effect and, all the more so, the setting of standards up to the present time remains indefinite due to the fact that there are as yet no clear-cut and widely applicable pathophysiological criteria for distinguishing between, for example, protective adaptive reactions and compensatory reactions, as well as between regulatory reactions in an emergency situation, and pathological reactions at the level of various systems, including the central nervous system, as well as the regulatory processes at other levels. Table 2. Soviet Hygienic Standards for Radiofrequency Electromagnetic Fields

#### Таблица 2

#### Отечественные гигиеннческие нормы для электромагнитных полей радиочастот

		(2)	(2)
	(1) Лианален радлоролн	ПЛУ дия про- фессиональных групи	(Э) ПЛУ для населения
4) 5) 67 8) 9)	Средние волиы (СВ) 100 кгц—3 МГц Короткие волиы 3—30 МГц Ультракороткие 30-50 МГц 50-300 МГи Микроволиы 300300 000 МГи	50 В/м 5 А/м 20 Е/м 10 В/м 5 В/м 10 мкВт/см <sup>2</sup>	10 В/м 4 В/м 1 В/м 1 мкВт/см <sup>2</sup>

Key:

# 1. Radiowave band

- Maximum permissible level for occupational groups
   50 V/m, 5 A/m, 20 V/m, 10 V/m, 5 V/m, 10 µW/sq cm
   Maximum permissible level for the population
  - 10 V/m, 4 V/m, 1 V/m, 1 µW/sq cm
- 4. Medium waves (MW)
- 5. 100 KHz-3 MHz
- 6. Short waves 3-30 MHz 👘
- 7. Ultrashort 30-50 MHz
- 8. 50-300 MHz
- 9. Microwaves 300-300,000 MHz

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Table 1. Some Results of Experimental Studies on the Biological Effects of Very Low Intensity Microwaves (Up to 150 µW/sq cm)

#### Таблица 1

	(1) Исследованные функции	(2) Характер изменения	DILM, MEBT/ CN <sup>4</sup>	По данним исследова- телей	
(5)	Вес тела	(13) О(ффвание в весе (хролический опыт)	150	В. В., Мар- ков	(25)
(6)	Артериальнос давле- нис	2-фазное течение с выряженным гипотен- зивным эффектом (хронический очыт)	150	В. В. Мар- ков	(25)
(7)	Воспрововодительная функция	Спрасние способности к онлодотверснию, уманывение численности потомства, узеля- чение количества пенолнонениях вотомков, увелячение змбриодальной смертности и и т. я. (хоонический опыт)	150	А. Н Берез- икцкая и со- авторы	(26) 1 1
(8)	Центрольная нервиая система	1. Изменение ЭЭГ с преимущественной (16) синхропизанией (острый опыт)	1020 ii 60.1ee	З. В. Гвоз- дикова ч др., Ч. Асабаев	(27) F
		2. Бавариантиме сдвиги с преобладанием (17) яктиваций (хроянческый опыт)	150	М. С. Быч- коэ В. В. Марков В. М. Рычко-	(28 <b>)</b>
		(18) 3. Биляриантиме сдвиги в подкорково-ство-		ва	J
(9)	Электромнография	зовых структурах (хронический опыт) (Эр)изение электрической активиости ДЕ	150	Е. В. Мар- ков	(29)
(10)	Система гипотала- мусуннофизкора	1. Изменение веса же из внутренной сек- (19)реции (гинофиза, падночечников)	150	Н. К. Демо- кидоър	(30)
	валноченников	2. Изменсине нейросократорной функции (20) сопотелебсуба	150	М. С. Толг- скал и со	(31)
		3. Тепленция к узоличению ссдержения (21 корадренящия в надаллечниках	150	авторы И. А. Клиов- ская Э. И. Полу-	(32) T X
(11)	Обмен веществ	Сленти в волносолевом обмене (внаседение (22) воды. Na и K, общего азога)	150	хива Н. К. Демо- кидова	(33)
(12)	Иммунология	Угнетение фагоцитарной реакции нейтро- (23) филов	150	А. П. Вол- кова В. В. Мар- ков	(34) л л

Сводка некоторых результатов экспериментальных исследований биоэффектов микроволновых воздействий весьма малых интенсириостей (до 150 мкВт/см<sup>2</sup>)

Кеу:

1. Function

2. Change

- 3. Power flux density , ww/sq cm
- 4. Investigator
- 5. Body weight
- 6. Arterial pressure
- 7. Reproductive function
- 8. Central nervous system
- 9. Electromyography

## UDC 621.397.618:658.387

MAJOR TRENDS IN THE SCIENTIFIC ORGANIZATION OF WORK AT RADIO AND TELEVISION STATIONS

Moscow O BIOLOGICHESTKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 15-20

Article by P.P. Fukalova/

/Text/ Studies have been conducted which have demonstrated that the creation of an effective protection and rational organization of labor at radio and television stations can be achieved by adherence to the fundamental principles and standards dealing with radiation protection in combination with the regulations of NOT /scientific organization of labor/, as well as organizational, technical, esthetic, ergonomic, and hygienic measures.

Hygienic investigations designed to test the effectiveness of the protective methods which we have proposed have shown that the fundamental principles and the standard decisions which we have recommended (1964, 1966, 1968), could not be immediately be put into effect at those enterprises where, from the hygienic point of view, defective equipment is still utilized, as well as out-of-date forms of labor organization.

Significant levels of electromagnetic radiation prevail in situations where use is made of multistage unshielded cascades and circuits, exposed high-frequency energy transmission lines, antennae commutators and switches, as well as cable networks connected to powerful high-frequency cascades.

Cumbersome and irrationally constructed long wave (LW) and medium wave (MW) transmitters, as well as short wave (SW) and ultra-short wave (UW) transmitters with manual commutation of energy to the antenna from each transmitter, are employed without adequate protection against radiation, not to mention the ergonomic shortcomings of this equipment with respect to working conditions. Thus, when old systems of LW and MW transmitters are used, the most radical method which can be instituted for protecting the personnel from radiation consists of installing new transmitters, and locating them in a manner which is in accordance with the principles of the scientific organization of labor.

The results of our measurements of field intensities at the work locations involved in servicing the old and the new transmitters are presented in Table 1.

Table 1 indicates that as a result of the replacement of the old transmitters with the new transmitters, field intensities in the work rooms decreased on the order of 10 to 100-fold.

In situations in which old systems of SW transmitters are employed, the personnel may be protected from irradiation either by replacement of these transmitters, or by improved shielding of the old transmitters, as well as by replacement of manual commutation by automatic commutation, and a more rational layout of the instrumentation.

It should be pointed out that the rational disposition of the transmitters in the presence of manual commutation can be achieved primarily through its reconstruction and replacement by automatic commutation.

A scheme for replacing multi-row disposition of the transmitters by single- and double-row dispositions are given in Figures 2 and 3 (a, b). In distinction to multi-row disposition of the transmitters (Fig. 2) with a manual method of commutation of the energy, and a large number of feeder lines which in the working regions create tens and more electric fields with intensities of 100 V/m, single- and double-row disposition of the transmitters (Fig. 3, a and b) with replacement of manual commutation by automatic commutation, and displacement of the commutators into a different room, decreases or completely eliminates the passage of feeders to the working rooms.

Furthermore, disposition of the control panel in front of the transmitters, located in a single row, assures convenient working conditions for the personnel on duty who are responsible for the proper function of these transmitters.

Implementation of the measures which included shielding and rational layout for the equipment decreased the radiation to the maximum permissible level, and even lower. The light tones of the color schemes in the interior of the installation fit in well with the gray color of the transmitters, as well as with the rational utilization of natural light and illumination provided by daylight lamps which are located in the panels of the white ceiling.

Comfortable surroundings have been created for the service personnel by the complete isolation of the control and transmitter operating room, air-conditioning, and the decorative layout.

Physiological studies conducted under the conditions in which the new organization of labor prevails, have shown that the workers retain their work capacity as indicated by the stability of the parameters which were under study -- without significant differences -- during the course of the day (static force of the muscles of the right hand, correctional test, visual motor reaction; Fig. 1).

Therefore, the extensive technical reorganization at the radio transmission centers, which implemented protective measures based on the principles of NOT, have created favorable sanitary and hygienic conditions, as well as rational forms of labor organization which assure a high level of work capacity on the part of the personnel.



Figure 1. Physiological Investigations on Individuals Employed Under Conditions of New and Old Forms of Labor Drganization

Key:

- 1. Static force, right hand
- 2. Correctional test
- 3. Selection
- 4. Time , sec
- 5. Time, min
- 6. Time, msec
- 7. Number of errprs
- 8. Number of determinations
- 9. Conditions:
- 10. --- out-of-date labor organization
- 11. - new labor organization



Figure 3. Disposition cof Transmitters With Automatic Commutator in a Separate Room. a) Single Row. b) Double Row

Key:

- 1. Commutator
- 2. Transmitter
- 3. Control panel

### UDC 614.87:537.868.029.64

HYGIENIC EVALUATION OF WORKING CONDITIONS INVOLVING RADIOWAVE EMITTERS ON THE BASIS OF DYNAMIC STUDIES ON THE NATURE OF RADIATION DURING A WORK SHIFT

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 21-25

[Article by V. V. Markov]

[Text]

The present article deals with material on hygienic evaluation of the working conditions of operators of SHF equipment which was obtained on the basis of dynamic investigations of the nature of irradiation during the period of a work shift . The results showed that this occupational group is usually exposed to intermittent radiation which is characterized by alternating periods of radiation and intervals without radiation, and a random variation in the forces and temporal parameters. The resultant data were treated by the mathematical theory for random processes.

Until recently, studies dealing with the working conditions of the various industrial enterprises of individuals handling emitters of microwave radiation regarded the process of radiation as uniform and continuous. Such assumptions were used in the formulation of hygienic standards for this industrial factor. However, this approach does not correspond to actual industrial conditions; it has come to be appreciated that people working with emitters of microwaves in many instances are exposed to intermittent and non-uniform radiation. Consequently, it is obvious that evaluations of working conditions which are based on the assumption that these occupational groups are exposed to continuous radiation do not correspond to the actual conditions of microwave radiation. instrument PO-1, model "Medik". PFD gave us the time an operator performed his work which was accompanied by emission of EMF energy. During every measurement the data were copied on several occasions from the instrument and as a final result we took the maximum intensity of EMF. Time studies were performed with an ordinary stopwatch during the course of an entire shift. During that time we noted whether the operator was or was not exposed to irradiation with EMF energy.

On the basis of the instrumental and the time course studies we constructed individual curves for irradiation of each operator alone, and then on the basis of these curves we constructed generalized curves for the entire group of workers who were involved in maintaining the same piece of equipment.

The working place of the operators was a transceiver booth of the radar station. Depending on the type of equipment that was being produced, several generators were located in the booth with different modes of microwave generation. The process of adjusting, regulating and testing the SHF radar equipment required frequent turning on and off of high voltage (the correction of malfunctions required that the high voltage be turned off).

During work the operator moves in the booth and adjusts successively one after another of the generators; during that time he may be subjected to intermittent microwave irradiation with different intensities and of different durations and different periods without radiation. PFD may vary significantly during the shift when several generators are functioning.

Several locations may be designated in the booth in which the operator does most of his work on adjusting the transmitting radar equipment. Preliminary measurements show that all types of equipment have in common a narrowly directed type of radiation which is directed at the place of work through slits in the casings, slits between the doors and the body of the cabinet as well as slits in the flanges and so forth. This results primarily in the irradiation of the upper part of the body.

Instrumental measurements of the EMF intensities involved in adjusting type A equipment at six points in the working zone of 13 operators showed that there was significant differences in the PFD values depending on the piece of equipment that was being serviced. In this situation maximum intensity did not exceed 100  $\mu$ W/sq cm and the minimum intensity did not fall below 6  $\mu$ W/sq cm. minutes and not more than 83 minutes with an average value of 57 minutes or 16% of the time of the entire shift.

The results of the instrumental and the time studies showed that the working conditions of the operators handling type V radar equipment are completely satisfactory.

Obviously, individual irradiation data may not give an adequately satisfactory representation of the possible variations in radiation under actual industrial conditions and for that reason we shall discuss general graphical analysis of the material. This analysis revealed that the maintenance, operation and testing of the full line of radar equipment, irregardless of the type of equipment that was involved, is accompanied by irradiation with microwave EMF which has a pronounced intermittent character. The fundamental parameters of intermittent radiation are as follows: EMF intensity, the duration of exposure of a biological object to EMF, and the interval (absence of radiation). On the basis of the nature of the intermittence of the individual parameters we found it possible to isolate two fundamental types of intermittent radiation within which we may place every type of radiation encountered by us, with due allowances. In the first type (Fig. 1) intermittency is evident in every major parameter; in the second type only the duration of radiation and the interval between exposures showed intermittency. In addition the intensity of radiation remains constant during the entire period of study or varies within narrow limits (from 10-15%, Fig. 2).

Different operators, including the same operator during different shifts, are exposed to radiation which differs in its characteristics.

All of the above considerations point to the fact that irradiation of the radar equipment operators is a chance process: this process describes the different PFD values which vary with time and which cannot be foreseen. Identification of intermittent radiation under industrial conditions as a random process requires adequate mathematical means for treating the data. For that reason it was necessary to use the mathematical theory applicable to random process according to which the fundamental characteristics of the random process are represented by its mathematical expectancy  $(m_y)$  dispersion  $(D_i)$ and the correlation function of the random process  $(K_y)$ .

Results of individual mathematical treatment of each type of product demonstrated that, although different in their details, the characteristics of a random process have common features.



Figure 1. First Type of Irradiation Characterized by Intermittence of Each Parameter -- Intensity, Period of Irradiation, and Intervals Between Irradiation



Figure 2. Second Type of Intermittent Irradiation Characterized by Intermittence of Two Parameters -- Period of Irradiation and Intervals Between Irradiation

At the same time the territory of the airports and the adjacent rayons are continuously being developed by the construction of technical, administrative and residential buildings of various heights.

The significant increase in RS power and increased directional effect of the antennae points to the obvious need for the proper type of mutual distribution of RS in various industrial and civil buildings.

An important problem in the planning, installation and utilization of powerful SHF emitting objects at CA airports is the problem of sanitary and protective zones between RS and other objects at the airport (buildings, waiting platforms, terminals and so on).

The correct solution of these problems requires accurate knowledge of the PFD of the SHF which is created by RS antennas at different distances from the objects being irradiated. In view of this we conducted a series of experiments on the determination of the levels of irradiation of the territory at a number of large airports in the USSR that are equipped with scanning RS.

The topography of the locality, the height at which the radio antenna is located as well as the working angle of inclination of the antenna dish differ in each airport. This makes it very difficult to compare and evaluate the results of measurements conducted at the individual airports.

As a result of this we developed a method for conducting the measurements and for the treatment of the results of the data which would permit standardization of the conditions and generalization.

By representing the results in this form it would make it possible to utilize them in planning new airports and in determining the proper place for locating the scanning RS on their territory.

Studies on the existing PFD levels were conducted over a period of 3 years--from 1968-1970.

Our course of studies consisted of 3 stages: preparation, measurements, and treatment of the results and their classification.

to the source of radiation. Correction in the angle of measurement can easily be achieved by simple geometrical means.

4. Communications were maintained by means of radio. To that end we employed short wave and ultrashort wave portable radio transmitters which made it possible to maintain radio communication over the entire distance which we covered in our studies.

**PFD** measurements:

1. The measurements were conducted with a PO-1 instrument. The antenna of the measuring instrument was oriented toward the center of the lower RS antenna dish. When measurements were conducted above ground the instrument was mounted on a tripod which was fixed into a receptacle of the automatic lift. The radio transmitter was also located there.

2. The RS antenna system was directed toward the selected route of measurement. The upper and lower antenna dishes of the radar station were set in a position which corresponded to the most negative angle of inclination.

3. Once the antennas of the measuring instrument and the RS were oriented in the proper direction, the radar was turned on and put into operation. Once the RS antenna and the measuring instrument antenna were properly aligned, adjustments were made for maximum emission. Then the RS antenna was fixed and a series of measurements were conducted at one selected distance and at one height with the angle of inclination of the lower antenna dish being varied from its most negative value to its most positive value at intervals of 0.5°.

4. After a series of measurements had been made the emitter was turned off, but the position of the radar antenna was not changed since the measurements were repeated at other heights at the same distance.

5. PFD measurements were conducted in a similar manner at the different distance markers. Every result of the measurements was recorded in a table. The following data were recorded in tabular form: distance between the point of emission and measurement, the height of the point of measurement, the angle of measurement, the angle of incline of the antenna, and the measured PFD values.

In view of the fact that at each airport the topography of the locality, the height of the RS antenna, the working angle of inclination of the antenna dish are different, it is extremely difficult to compare the results of measurements conducted at

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In order to apply the resulting PFD values in these coordinates to all airports, we employed the greatest and the smallest PFD values of all the measurements, since they reflected the possible range of irradiation of the territory of the airports.

The results of our measurements make it possible to determine possible PFD levels for SHF fields at various distances from the source of emission and at different heights, taking into consideration the topography of the region.

On the basis of these measurements it will be possible in the future to plan zones of discontinuity between RS and various objects with a satisfactory degree of certainty, for practical purposes, without conducting actual instrumentation studies.

Formulation of concrete methods for this type of planning is presently under study.



Coordinates of the Point of Measurement

### Figure 1.

Key:

- 1. Point of emission
- 2. Point of measurement
- 3. Measurement line (for given measurement point)
- 4. Axis of the first lobe
- 5. Horizon line

The distribution of MRS with respect to other buildings and constructions at the airport as well as residential, community, and industrial buildings is dictated by the technical requirements of MRS equipment for its proper operation and the maximum permissible level of power flux density (PFD) of SHF.

Care, protection and prevention must be utilized in order to prevent adverse effects of SHF irradiation and the SHF equipment must be located in such a manner that its radiation remains within acceptable standards.

We conducted instrumental measurements in a locality to obtain quantitative information on the intensity of SHF radiation emitted by MRS antennae. The measurements were performed with a PO-1 instrument according to a strictly adhered to system:

1. From the point at which MRS was located we selected a direction which was characteristic of that region and designated points of measurement at every 50-100 m.

2. Measurements at every point were conducted at several heights ranging from 1.5-15 m (at 3 m intervals).

3. The MRS antenna is oriented in a chosen direction and the antenna of the measuring instrument from the point of measurement is oriented toward the center of the MRS antenna. When the antennae have been mutually aligned the radar station is adjusted for maximum emission.

4. The MRS antenna is fixed and a series of measurements are conducted at different angles of inclination of the antenna at intervals of 0.25°.

5. Analogous measurements are conducted along several different directions.

Every measurement was performed 3 times and the arithmetic mean for the 3 values was recorded. Communication was maintained by means of portable radio stations; the necessary height was achieved by an automatic lift SPO-19. PFD values were obtained under conditions of visual range between the source of radiation and the point of measurement during different times of the year (winter, spring, summer) under different metereological conditions.

The results of the measurements indicated that during operations of MRS stations great care must be exercised when work is conducted in the zone of radiation as well as in the

Table 1.

Рабочий диапазон	ина ППМ, зон мкВт/см <sup>3</sup> 1) (2)	(3) Раднус зоны, м, для высот над местностью		
(1)		h=1,5 м	<i>h=</i> 4,5 м	h7,5 м
Сантиметровый	1	2850	3800	3700
	10	1550	1500	1200
Миллиметровый	1	3300	3500	3800
(5)	10	1300	1350	1450

.

Key: 1. Working band 2. PFD, µW/cm<sup>2</sup> 3. Zone radium, m, for heights above the locality 4. Centimeter

.

5. Millimeter



Figure 2. The Relationship Between PFD and Distance for the Millimeter Band

1. measurement height h = 1.5 m Key: measurement height h = 4.5 m
 measurement height h = 7.5 m



Figure 3. The Relationship Between PFD and Distance for Measurements at Different Heights at Maximum Emission (Millimeter Band)

Key: 1. Calculated 2. h = 7.5 m 3. h = 1.5 m

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Key: 1. h = 4.5 m 2. h = 7.5 m

UDC 621.371.029(-21)

DISTRIBUTION OF ULTRASHORT WAVE FIELDS IN THE VICINITIES OF URBAN TELEVISION CENTERS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 37-42

[Article by N. D. Khramova, V. A. Miroyedov, and V. V. Yur'yev]

[Text]

Instrumental measurements were made of the electromagnetic field intensities around television centers. Good agreement was obtained between calculated and measured data. At a distance of 250-750 m from a television center the intensity of the electromagnetic field noes not exceed 1 V/m.

The distances which are covered by radio and television have been greatly expanded by the large network of radio stations designed for broadcasting and communication, the increase in the number of television centers and relays, the tendency to use superhigh frequency as well as the increase in the power of the transmitters and improvements in the antenna systems.

At the same time the rapid rate of civilian and industrial construction has resulted in the fact that radio and televi-sion stations find themselves surrounded by residential homes.

Consequently, people are exposed to the potential hazard of irradiation by radio waves.

We've studied the distribution of electromagnetic fields around large television stations in the USSR.

Television communication in the USSR is conducted at frequencies ranging from 48.5-230 MHz with corresponding wavelengths of 6.25-1.3 m. Cmnidirectional multi-storied turnstile antennas are employed with a gain factor of about 4. The antennas are located on supports at a height of 140-160 m and higher, up to 190-230 m.

Generally, 3 characteristic zones are considered in a high frequency field of radiation: the adjacent zone, the intermediate zone, and the distant (wave) zone.

In considering field distribution in populated areas around television centers we are concerned only with the wave zone.

However, the conditions for the dissemination of electromagnetic energy in the city represents a very complex problem since, from the point of view of dissemination of the radio waves, the city can be regarded as a highly rugged terrain. For all practical purposes, it is extremely difficult to determine field intensity by means of calculations in such a situation.

We elected to employ instrumental evaluation of electromagnetic field intensity. Furthermore, in hygienic practice it has come to be accepted to express the intensity of radiation in distant zones in units of power flux density. We chose to evaluate this factor in terms of the intensity of its electrical component, i.e., in volts per meter (V/m). This was advantageous because in determining the levels of radiation in residential areas we had to conduct measurements both outside of buildings and within buildings (on staircases, in apartments, and so forth).

Under these conditions we have to deal with limited spaces and a large number of secondary emitters. Consequently measurements that were conducted within buildings--irregardless of their distance from the antenna of the television center--are related to measurements conducted in the adjacent zone where the intensity of radiation is commonly evaluated in terms of the intensity of the components of the field, in part in terms of the electrical component, i.e., in volts per meter.

During the period 1970-1971 we investigated 9 television centers and relay stations. Studies on the distribution of field intensity were conducted in the greatest detail in the cities of Tartu, Tallin and Riga.

The measurements were conducted with instrument PZ-2. In order to increase the sensitivity of the instrument we employed an additional removable antenna, in addition to the standard antenna which came with the instrument. The instrument was calibrated in a field of plane-parallel capacitor at the Leningrad Tekhnikum of Instrument Construction and Automation. Calibration was conducted with both a standard antenna and the accessory removable antenna.

A detailed plan for carrying out the measurement was devised. To that end, we laid out 3 routes along which we conducted our measurements which were most representative of the residential area; the measurements were conducted in 3 to 4 different directions for a distance of up to 3 km. Along the selected routes measurements were conducted at intervals of 50 m up to a distance of 1.5 km and then at intervals of 100 m. At each point 3 measurements were made and the arithmetical mean was determined. The measurements were largely conducted at the height of a human being--1.5 m above the ground level.

Now, let us consider the intensity distributions of electromagnetic fields in some cities. A composite plot is presented in Fig. 1 which delineates the relationship between the field intensity at a distance from several television stations in the USSR on the basis of the measurement conducted from 1969-1970.

In 1971 we repeated in greater detail our measurements in the residential zones around the television centers in Tartu and Tallin.

The vicinity around the Tartu television center is characterized by a virtually flat region which consists of one and two storied cottages and gardens; the street plan in that area is straightforward and uncomplicated (Fig. 2).

In Fig. 2 we have indicated the routes of measurements which radiate from the television station. In addition, we made measurements along 2 circular routes around the television station (one larger and one smaller).

Measurements of field intensity conducted in areas close to this television center have given us a very clear picture of field distribution (Fig. 3).

Comparison of the field characteristics for the Tartu television center which were conducted in 1971 and 1970 (Fig. 1 curve 3), we can see that up to a distance of 1 km from the television center the field intensity was greater in 1970. This difference can easily be explained on the basis of the fact that in 1970 a shortwave radio center was located at the territory of the television center. In 1971 this radio center was moved outside the center by a distance of approximately 8 km and, consequently, the field intensity around the television center decreased.

A different type of residential construction is found in Tallin. In Tallin the television center is located in a residential area consisting of 5 to 7 storied buildings. This significantly alters the distribution of the ultrashort wave field and decreases the average level of intensity (Fig. 4).

In Tallin, in addition to the measurements conducted at 1.5 m, we also conducted measurements at heights of 4, 8, and 12 m with the assistance of an automatic lift. Measurements at the various heights were conducted on the same streets and at the same points as were the measurements which were conducted without the assistance of an automatic lift. In addition, measurements were also conducted at the same height and at the same distances from the television center on staircases in buildings which were next to windows which were facing the television station.

The results of these measurements are given in Fig. 5(a,b).

Inside the buildings the field intensities were 2.5-7 times lower than they were at the equivalent point out on the street. Furthermore, the closer we came to the television center the more significant the differences became.

Within the conditions prevalent in a city an accurate mathematical calculation of the distribution of the field intensity which was generated by the antennas of the television center could not be made in view of the non-uniformity of the city building constructions, the frequent irregular street plans, the fact that various materials were used in the constructions as well as the presence of a large number of reflecting surfaces.

Therefore, our approach to this problem was largely utilitarian.

On the basis of our experimental investigations we attempted to calculate the expected field intensities by using the interference formula of B. A. Vvedenskiy which characterizes the dependence of the intensity of an electric field on the distance, wavelength, and the height of the antenna.

$$E = \frac{2,18 \sqrt{PD}}{r^{2\lambda}} h_1 h_2,$$

where E is the expected field intensity, P is the power of the transmitter,

- D is the antenna gain
- r is the distance from the antenna to the point at which E was determined
- **1** is the wavelength
- $h_1$  is the height of the phase center of emitting antenna,  $h_2$  is the height of the point of measurement.

However, the applicability of this formula is limited by the following consideration

$$h_1h_2 \leqslant \frac{rh}{18}$$
.

This condition may be met in the region of meter waves when the heights  $h_1$  and  $h_2$  are comparable.

In our case, since the height of the emitting antenna was more than 100 m, as a rule, and the height at the point of measurement was 1.5 m, determination of the effective (actual) value for the field intensity can be determined more readily by using the following formula

$$E_{\mathbf{g}} = \frac{173 \, V \overline{P[\mathbf{kW}] D}}{r \, [\mathbf{km}]} \cdot V[\mathbf{mV/m}].$$

We introduced the coefficient V which represents the attenuation factor which takes into consideration the attenuation of the field due to its distribution over a highly rugged terrain (this term may be used to describe large cities). The value of the attenuation factor may be determined from the following expression

$$V = 1,7 \frac{h_1(\mathbf{M}) \cdot h_2(\mathbf{M})}{r(\mathbf{M})} .$$

In Figs. 3 and 4 the thick line indicates the calculated relationship between field intensity and distance.

The calculated relationship between field intensity and the distance conforms with the rounded curve for the maximum values of intensity of the electric field which were obtained on the basis of experimental measurements in these localities. The agreement between calculated and experimental values is fairly good. On the basis of the comparative results presented in Figs. 3 and 4 we may conclude that the attenuation factor could be decreased somewhat.

The fact, that a certain relationship prevails in the agreement between the calculated and experimental values, is supported by the fact that the agreement in the results was better in Tartu. In Tartu the field is distributed more evenly since in the vicinity of the Tartu television center the residential area consists of buildings of approximately the same height (1-2 stories and the topography of the region is relatively flat).

Analysis of the experimental data obtained in the vicinity of various television centers in the USSR has shown that at a distance of 250-750 m the local population is exposed to radiation which is equal to 1 V/m; these distances are indicated in Figs. 1, 3 and 4 and the value for the level of intensity is in accord with the maximum permissible level for ultrashort wave radiation.

This work requires further study in order to obtain unequivocal agreement between experimental and calculated values, the introduction of correctional coefficients, and the determination of the applicability of the formulas to various environmental conditions (the type of construction, the height of the building, the design, the constructional materials, as well as the distance from the point of emission, the parameters of the emitting antenna, and so on).



Figure 1. Distribution of USW Fields in Cities Around Television Centers According to 1969-1970 Measurements. 1) Saratov. 2) Vinnitsa.
3) Donetsk. 4) Tallin. 5) Tartu. 6) Vilnyus



Figure 2. Build-Up Area in the Vicinity of the Tartu Relevision Center



Figure 3. USW Field Distribution Around the Tartu Television Center


Figure 4. USW Field Distribution Around the Tallin Television Center



Figure 5. The Relationship Between Field Intensity and Height at Different Distances from the Television Center (Tallin)

UDC 613.62: [614.87:537.868.029.64]

THE CLINIC, PATHOGENESIS, TREATMENT, AND OUTCOME OF RADIOWAVE SICKNESS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 43-48

[Article by M.N. Sadchikova and K.V. Glotova]

/Text/ The results of long years of clinical observations have demonstrated that prolonged work under conditions of exposure to SHF electromagnetic waves of significant intensities (up to several mW/sq cm), may result in the development of an occupational disease -- the radiowave sickness. Three essential syndromes of this disease have been identified: the asthenic syndrome, the astheno-vegetative syndrome with vascular dysfunction, and the hypothalamic syndrome. Studies on the mechanism of the neurovascular impairment have revealed the significant role of the deep structures of brain, including the hypothalamic regions. Essential principles have been formulated to cover drug therapy, and late results of treatment.

Many studies are available in the literature which deal with the biological effects of superhigh frequency electromagnetic fields on the human and animal organisms.

The results of clinical studies conducted by a number of authors /N.V. Tyagin, 1960, 1962, 1963, 1965, 1971; A.G. Panov and N.V. Tyagin, 1966; N.V. Uspenskaya, 1961, 1963; Ye. V. Gembitskiy, 1962, 1970; V.N. Gur'yev, 1962, 1963; I.R. Ramzen-Evdokimov and V.A. Sorokin, 1970; E. Klimkova-Deutschova, 1957, 1963; and others/, as well as our own observations, indicate that prolonged work under conditions in which the individual is exposed to SHF electromagnetic fields may lead to the development of a disease entity, the clinical picture of which is characterized by changes in the functional state of the nervous and the cardiovascular systems. However, up to the present time, a number of questions dealing with the clinical aspects, pathogenesis, and treatment of this disease have not been adequately studied.

Studies which have been conducted over a period of many years at the clinic of the Institute of Labor Hygiene and Occupational Diseases of the Academy of Medical Sciences USSR on a large group of people who were employed in the radio industry, have made it possible to elucidate the nature of the effects of SHF electromagnetic fields of different intensities on the states of the nervous, cardiovascular, neurohumoral, endocrinemetabolic, and other systems, and have also made it possible to investigate the sequence of clinical manifestations of this disease, as well as to classify it and isolate an independent nosologic disease entity -- the radiowave sickness.

These investigations have convinced us that a profound professional pathology arises frequently in people who have, for long periods of time, been employed under conditions where they came into contact with sources of SHF electromagnetic energy and where the intensity of irradiation may attain several milliwatts per square centimeter.

A decrease in the incidence of occupational pathology is observed under the effects of SHF electromagnetic fields of low intensity which do not exceed a hundredth of a milliwatt per square centimeter, and during exposure to more intense radiation for short periods of time.

Simultaneously, vegetative vascular dysfunctions continue to be detected which are related primarily to the increased excitability of the sympathetic branch of the vegetative nervous system.

Exposure to SHF electromagnetic fields of even smaller intensity (from fractions to units of microwatt/square centimeter) do not evoke changes in the state of health of the workers, which could be connected to the exposure.

We have determined that in this professional pathology changes in the central nervous system are of primary importance, particularly in its vegetative branches, and in the cardiovascular system, which are characterized by a unique clinical symptomatology with two forms of vegetative reactions which are determined by dysfunction in the interaction between the excitability of the sympathetic and the parasympathetic branches of the central autonomic formations in the brain, and the state of the cortical processes. On the basis of our own observations, and the data in the literature, the clinical picture of radiowave sickness may be represented by the following three syndromes.

The asthenic syndrome is encountered in the initial stages of the disease. It seems to be largely based on "exhaustion" of the central nervous system. The vegetative changes are totally defined with dominance of vagotonia, arterial hypotension, and bradycardia. On the whole, the syndrome has a favorable outcome, and does not decrease the work capacity of the patients for long periods of time.

The astheno-vegetative syndrome with vascular dysfunction is the most frequently encountered entity, and is usually seen in moderate and severe stages of the disease. The clinical picture of this syndrome is marked by more pronounced asthenic phenomena and primary features attributed to vegetative dysfunctions which are related to elevated excitability of the sympathetic branch of the vegetative nervous system, vascular instability with hypertension, and vasospasms. The latter factors frequently determine the severity of the disease.

At a given stage of the increasing pathological phenomena, the hypothalamic syndrome (vegetative vascular form) arises, which is characterized by the development of paroxysmal states in the form of sympathoadrenal crises. The course of the asthenovegetative and the hypothalamic syndromes are protracted, and frequently lead to decreased work capability.

In advance stages of the disease, with increasing asthenic phenomena, emotional and vegetative vascular instability, and particularly in cases characterized by paroxysmal states, a clinical picture of ischemic heart disease develops, along with hypertension, and sometimes changes in the dynamics of brain circulation which sharply decrease the work capability of the patients.

The fact that vasospastic phenomena are encountered in moderate and advanced stages of the radiowave sickness has been supported by instrumental investigations. Thus, the results of rheographic investigations of cerebral hemodynamics have demonstrated a decrease in pulse volume, and increased tone of intra- and extracranial blood vessels which are returned to the normal state by the nitroglycerin test. Mechanocardiographic data have indicated increased tone of muscular vessels, and an increase in peripheral resistance. Electroencephalographic and certain biochemical studies are in agreement with the clinical features of the radiowave disease and its course. During initial stages of the disease the patients may evidence desynchronization of the alpha activity, a stability in the alpha rhythm, or desynchronization of the alpha rhythm; in moderate and advanced stages of the disease, bilateral synchronous discharges of the theta and delta waves are evident, and sometimes diffuse slow waves are detected, particularly under the influence of hyperventilation which indicates that the subcortical structures are involved in the pathological process.

Changes in the carbohydrate metabolism correlate well with the tendencies in the vegetative reactions, particularly in the changes of the sugar curves following glucose loading. Thus, in the vegetative vascular changes resulting in vagotonia, we encounter depressed curves, while in vegetative vascular dysfunctions which result in sympathetic tone along with sympathoadrenal crises, we encounter so-called diabetic or doublepeaked sugar curves. Changes in the sugar curves may be related to impairment of the mechanisms responsible for the regulation of homeostasis, and primarily this would involve deficiencies in the hypothalamus - hypophysis-adrenal cortex system. In order to evaluate the functional state of this change we investigated the levels of certain mediators of the nervous system, such as blood levels of histamine and acetylcholine, urinary levels of epinephrine and norepinephrine, as well as the glucocorticoid function of the adrenal glands.

Blood levels of histamine in patients with radiowave sickness are elevated. Changes in the concentration of acetylcholine are less pronounced; however, they are accompanied by changes in the activity of cholinesterase.

In moderate and pronounced stages of the disease the ratio of epinephrine and norepinephrine is decreased along with normal parameters -- actual variations and mean values --for the levels of catecholamines in 24 hour urine samples. In paroxysmal states a sharp variation is evident in the secretion of adrenalin, and a definite rhythm in the excretion of norepinephrine characterized by increased secretion of norepinephrine in the evening and at night, which points to dysfunction in the mechanisms responsible for the regulation of the activity of the sympathoadrenal system.

Results of investigations on the glucocorticoid function of the adrenal glands -- conducted by S.P. Korenevskaya -- have pointed to definite changes in the levels and relationships of cortisol, cortisone, and their tetrahydro derivatives. Changes in the relationships between the levels of tetrahydro derivatives of cortisol and cortisone, as well as between cortisol and cortisone, indicate dysfunctions in the mechanisms responsible for the transformation of cortisol into cortisone.

Subcutaneous administration of small doses of epinephrine (0.3 ml of 0.1% solution) elicits marked autonomic vascular reactions in the patients, as well as more distinct changes in the daily dynamics of catecholamine excretion, and impairment in the glucocorticoid function of the adrenal glands.

Development of pronounced vegetative vascular reactions in patients with radiowave sickness in response to the administration of small doses of epinephrine suggests significant impairment in the vegetative formations of the brain. These findings make it seem likely that the dysfunctions of the functional state of the sympathoadrenal system and the glucocorticoid function of the adrenal glands in patients with radiowave disease are the sequelae of primary lesions in the deep structures of the brain which are responsible for the central regulation of the neurohumoral and the neurohormonal processes in the organism. The investigations which have been conducted are in agreement with the clinical and electroencephalographic findings on the involvement of the deep structures of the brain, including in part the hypothalamic structures, Furthermore, dysfunction of the hypothalamic-hypophysealadrenal cortical system which arises on the basis of neurasthenia may be highly important in the pathogenic mechanisms responsible for the development of the clinical symptoms of the radiowave sickness.

On the basis of the pathogenic mechanisms of radiowave disease, treatment of the neurovascular dysfunctions should be conducted with consideration of the etiology of the disease, the state of the cortical function, and the predominant type of vegetative reaction which is involved in the development of a given syndrome, as well as the general state of the patient, and individual peculiarities. This has been covered in the publications of Ye. V. Gembitskiy /1970/.

Analysis of the results obtained with hospital treatment of 152 patients (129 men, 23 women, of relatively young age -80% up to 40 years of age) helped elucidate the principles of drug therapy of patients with various clinical syndromes of the radiowave disease. Patients with asthenic syndrome should be treated with analeptic and sedative therapy (bromine preparations, leonorus, valerian, hawthorn, korvalol /transliteration/) in combination with stimulants (sodium arsenate, strychnine nitrate, securenin, tincture of ginsen root, pankrotin /transliteration/, and others). When vagotonia predominates the vegetative vascular dysfunction, treatment should be instituted with cholinolytic drugs (atropine, amizil /transliteration/) as well as preparations with combined effects (belloyd, bellaspon /both transliterations/), and small doses of subcutaneous insulin followed by intravenous glucose infusion.

Therapy should include therapeutic gymnastics, massage, hydrotherapy (cold baths, circulating shower baths) as well as psychotherapeutic talks.

In the case of patients with astheno-vegetative syndrome and hypothalamic syndrome, and which also evidence marked vegetative vascular dysfunction and sympathoadrenal crises, they should, in addition to analeptic and sedative therapy, receive weak tranquilizers (seduksen, elenium, trioksazin /all transliterations/) and antihistamines (dimedrol, pipol'fen, suprastin /all transliterations/) which potentiate their actions as well as vasodilators. Of the large arsenal of modern vasodilators, preference should be given to magnesium sulfate which also possesses sedative qualities in combination with reserpine which is a weak tranquilizer, and hypotensive ganglionic blocking agent. In the presence of persistent hypertension, agents must be included which act directly on the vascular wall (papaverine, eufillin /transliterated/, no-shpa /transliteration/). Patients with coronary insufficiency should receive preparations which improve coronary circulation and myocardial metabolism (validol, nitroglycerin, intensain, panangin, izoptin [all transliterations]) as well as analgesics (injections of anal'gin /transliteration/).

Excellent therapeutic effects are obtained by injections of ATP, vitamins of group B, glutamic acid, oxygen therapy which improve the metabolism of amines and oxidative processes in the organism.

In view of the elevated reactivity of the patients, the selection of the appropriate therapeutic agents and doses requires a highly individualized approach. Particular care must be exercised in assigning drugs with narcotic action.

In cases of sympathoadrenal crises, sympatholytic drugs (small doses of chlorpromazine or propazin /transliteration/)

are recommended in combination with injections of vasodilators and analgesics.

The effectiveness of treatment is determined not only by the use of the proper drugs but also by the duration of the course of treatment, and the appropriate type of job placement.

Superior therapeutic results are obtained in patients with the asthenic syndrome in whom clinical improvement appears in two to three weeks. Following discharge from the hospital, 54 patients (out of 61) with the asthenic syndrome returned to their previous work; 7 were assigned to other types of jobs.

It was more difficult to achieve therapeutic effects in patients with astheno-vegetative and hypothalamic syndromes. Following a three- to six-week period of treatment. 35 of the 80 patients with the astheno-vegetative syndrome returned for their previous work and 7 were assigned to different types of work. In the case of 28 patients with the astheno-vegetative syndrome, and 2 patients with the hypothalamic syndromes, temporary release from work had to be obtained for occupational reasons for periods of 1 to 2 months; simultaneously, these patients underwent further treatment under ambulatory conditions or at sanitoria or health resorts. Eleven patients with the astheno-vegetative syndrome, and eight patients with the hypothalamic syndrome, were classified as Group III occupational invalids.

Further studies were conducted on a group of 100 patients. The periods of observation lasted from one to ten years, but in the majority of cases the observations were conducted for three to six years. At the time of the last examination most of the patients were between from 40 to 45 years of age.

The results of the investigations showed that despite repeated courses of treatment, temporary relief from irradiation, the course of the disease became progressively more severe as the patients returned to their previous conditions of work, and this was particularly evident in patients with astheno-vegetative and hypothalamic syndromes (Table 1, group A). Paroxysmal states are frequently noted in such patients, and in advance cases hypertension and ischemic heart disease appear. The work capacity of such individuals becomes limited, and in consequence, they were either classified as Group III, or even Group II occupational invalids, or else they were assigned to different tasks without an adverse change in their classification rating. At the same time, cessation of work under conditions of irradiation, results essentially in the stabilization of the process (Table 1, group B).

Therefore, the results of our long-term followup indicate that in moderate and severe forms of the disease, which are characterized by the development of the astheno-vegetative and the hypothalamic syndromes, work under conditions of exposure to irradiation should be terminated.

Individuals with initial forms of the disease may return to previous conditions of work under the condition that they be carefully followed and periodically undergo repeated courses of treatment.

Timely job placement of the patients, along with therapeutic and prophylactic measures, create conditions which lead to improvement in the state of health and prolong the work activity of patients with radiowave sickness. Table 1. Results of Dynamic Studies on Radiowave Sickness Patients With Continuous Exposure to Radiation at Work (Group A) and after Termination of Exposure (Group B)

(1)	(2)		( <u>4</u> ) xa	рактер те	н викоре	трудоспо	cofilioers	(янсло ,	1041L)
Клирические		TEAL		ация	÷(7)	Tp(18)	юсобен	Перев	еден на
снидромы	Грулпа	Ко.1-во бо	Висстано	Стабиллэ:	Прогресс	Lia npex- Tien paco-	ания облу-	нинал III груп- (13)	ИДИОСТЬ II Группа (12)
Астенический (14)	А Б	24 5	3	13 2	11	13	8 5	3	=
истено-вегетатив- най с сосуди- стой дисфунк- цией (15)	А́ Б	47 16		. <u> </u>	47		15 7	29 8	3 1
І поталамический (вегетативно-со- судпетая форма)	А Б	2 6		5	2 1			2 5	
(17) Beero:		100							

47

1

5

Key:

- 1. Clinical syndromes
- 2. Group
- 3. Number of patients
- 4. Course and work capacity (number of individuals)
- 5. Recovery
- 6. Stabilization
- 7. Progression
- 8. Work capacity
- 9. Declared as invalids
- 10. At previous work
- 11. Unexposed
- 12. Group III
- 13. Group II
- 14. Asthenic
- 15. Astheno-vegetative with vascular dysfunction
- 16. Hypothalamic (vegetative vascular form)
- 17. Total

### UDC 614.87:537.868.029.64

STATE OF THE BLOOD SYSTEM UNDER THE INFLUENCE OF SHF FIELDS OF VARIOUS INTENSITIES AND IN RADIOWAVE SICKNESS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 49-54

/Article by V.V. Sokolov, I.A. Gribova, N.A. Chulina, M.N. Gorizontova, and M.N. Sadchikova/

[Text] A sequence of changes was noted in the blood system under the influence of high intensity SHF electromagnetic fields which initially consisted of stimulation and subsequent prolonged cytopenia which was directly related to the intensity of radiation. Changes in bone marrow hemopoiesis had the distinct character of intense compensatory regeneration.

At the present time a significant number of experimental and clinical investigations have accumulated which show that the blood system does not remain indifferent to the effects of SHF electromagnetic fields. Studies of professional groups demonstrated a variety of changes in the blood picture: stimulation of hemopoiesis with leukocytosis /L.M. Kapitanenko, 1964; V.N. Gur'yev, 1967, and others/, increased levels of erythrocytes, hemoglobin, and reticulocytes /N.V. Uspenskaya, 1959/, as well as cytopenic reactions which frequently consisted of leukopenia /F.A. Gromov, 1966; M.P. Troyanskiy, et al., 1967; V.D. Maslennikov, 1969; and others/, and less frequently, of throm-bocytopenia /N.V. Tyagin, 1968; S.S. Rogusskiy, O.A. Ulitskiy, and B.N. Bartsevich, 1970; and others Z, and erythrocytopenta [V.V. Sokolov, and N.A. Chulina, 19647. The development of relative lymphocytosis, monocytosis, and eosinophilia, have also been described /F.A. Gromov, 1966; Ye. V. Gembitskiy, 1966; P.N. Fafanov, 1968; Hines and Randall, 1952; Barron, Love, and Barraff, 1956; Haduch, 1960; Jranyi, 1960; and others/. Qualitative changes in the blood cells were evident in metabolic impairment /Ī.V. Zakharov, 1967, 1968; A. Ya. Kholodnyy, et al., 1968; I.S. Dronov, 1968; and others/, and in the phagocytic activity of leukocytes /A.I. Ivanov, and

B.A. Chukhlovin, 1968; Ye. I. Smurova, 1967; and others/, as well as in the decrease of osmotic and acid resistance of erythrocytes /S.V. Balakireva, et al., 1969; and others/. Bone marrow samples obtained by puncture revealed moderate activation of the erythroid /R.D. Zhuk, 1967/, granulocytic /Ye. V. Gembitskiy, et al., 1968, 1969/, or both series /N.V.Uspenskaya, 1963/, along with a more limited response on the part of the reticular plasmacytic series.

The variety of hematologic reactions in persons who are exposed to the same factor, and the tendencies evident in these reactions which in one case may reflect stimulation of hemopoiesis, and in another case lead to the development of a cytopenic reaction, lead to the conclusion that certain phases exist in the changes of the blood, and that the data must be analyzed in terms of the intensity of the radiowaves, as well as the duration os exposure, and the nature of the general clinical manifestations.

Investigations were conducted on 131 persons (115 men, 16 females) with various forms of the radiowave sickness. The asthenic syndrome and manifestations of vegetative vascular dysfunction with predominance of vagotonia, was evident in 47 patients. In 84 cases, the astheno-vegetative syndrome was diagnosed with the sympathetic tone type of vegetative vascular dysfunction. In eight of these patients the vegetative vascular form of the hypothalamic syndrome was evident with sympathoadrenal crises. Most of the patients were 40 years old or less. The duration of employment ranged from five to ten years or more. In past years the intensity of the electromagnetic fields was significant and reached several mW/sq cm. Blood studies on patients with radiowave sickness showed a significant decrease in the concentration of thrombocytes and leukocytes (due to neutropenia with relative lymphocytosis; Table 1). Thus, the number of leukocytes in the patients under investigation was, on the average,  $5599 \pm 89$  and  $6600 \pm 73$  in the controls which consisted of 800 clinically healthy people, and the mean number of thrombocytes was decreased to  $221700 \pm 4120$  (248000  $\pm 1920$  in the controls). However, in the presence of the general tendency for a decrease in the number of leukocytes and thrombocytes, cases with marked leukopenia and thrombocytopenia were not encountered among the patients with the radiowave disease. In only individual cases was the leukocyte count less than 4000 per pliter, while the level of thrombocytes did not fall lower than 150000 per The erythrocyte counts and hemoglobin levels were uliter. normal. In addition, there was a tendency toward reticulocytosis (up to 16 to 17%). Cytometric studies on erythrocytes conducted on 35 patients with radiowave disease, showed a

decrease in the average diameter of the erythrocytes, and an increase in their thickness, while the volume of the cells remained unchanged; this indicated a tendency toward spherocytosis on the part of the erythrocytes. It is known that an increase in the spherical nature of erythrocytes decreases their osmotic stablility. Acid erythrograms were employed to obtain the distribution of the erythrocytes in terms of their age and stability. Investigations conducted on 41 patients showed shifts both toward a decrease, as well as an increase, in the acid stability of erythrocytes. The latter indicated that there was an increased input of young, more stable, erythrocytes into the blood stream, and this was in good agreement with the presence of reticulocytosis.

Cytochemical investigations on leukocytes obtained from 20 patients showed metabolic changes which were reflected in their relationships between neutrophils which contained alkaline and acid phosphatases, toward a predominance of the latter type of neutrophils. There was also an increase in the number of lymphocytes which reacted positively in the acid phosphatase test.

Studies were conducted on bone marrow hemopoiesis in patients with radiowave disease; the changes which were observed were characterized by an increase in the number of erythronormoblasts primarily on account of the polychromatophilic cells. While the normal value for the number of erythroid elements was  $20.53 \pm 0.378$  in the investigated patients, this value approached a mean of  $24.7 \pm 1.59$  (P < 0.05), and in individual subjects, it had even increased to 35.41%.

On the other hand, the number of neutrophilic cells was decreased (52.92  $\pm$  1.96 with 60.82  $\pm$  0.51 for the normal value) due to the number of mature segmentonuclear forms, the number of which, in a number of cases, decreased to 9-11%. Lymphocyte counts were frequently elevated. Determination of cells which were in a state of mitosis showed that in patients with radiowave disease, there was a tendency for cells in a state of division to increase. While in the controls 5.42  $\pm$ 0.40 cells out of a thousand nucleated cells were dividing, in the investigated individuals, the number of mitoses was increased to 7.67  $\pm$  0.59 (P < 0.05). Furthermore, the increase in proliferation occurred largely on account of the more intense division of the cells of the erythroid series (24.5  $\pm$ 2.86 with 21.25  $\pm$  1.34 for the controls), while division of the granulocytes changes to only a small extent. Cytogenetic investigations of the dividing cells in the bone marrow did not show an increase in the number of aberrant mitoses in comparison with the controls. Only an increase in the number of aneuploid cells was detected. They were encountered two to three times as frequently as in the controls and were largely represented by hypoploid forms.

The data just presented support a contention that SHF fields exert an effect on the blood system, and that both quantitative and qualitative changes are encountered in the blood cells. However, even in the presence of clinical symptomatology of the radiowave disease, these changes are not great and do not appear in all cases of the disease. Once they arise they do not show a tendency to progress. The blood pictures were identical (Table 1) both in the light forms of the disease (asthenic syndrome with vegetative vascular changes of the vagotonia type -- subgroup I) as well as in the clinically more severe forms of the radiowave sickness (astheno-vegetative syndrome with sympathetic tone type of vegetative vascular dysfunction, and the hypothalamic syndrome -- subgroup II).

Dynamic studies conducted on 40 patients which were first studied at the time the diagnosis was made, and two to six years after their work was terminated under conditions where they were exposed to electromagnetic irradiation, demonstrated that the hematologic changes in radiowave sickness are reversible. Cessation of exposure to SHF led, in the vast majority of patients, to a progressive normalization of hemopoiesis.

In addition, the structure and chromosomal composition of cultured lymphocytes obtained from the blood was conducted on patients that had sustained radiowave disease, and had not been working in conditions in which they were exposed to electromagnetic fields for two to four years (Table 2). Analysis of the metaphasic plates did not reveal changes either in the number of aberrant cells, nor in the number of chromosomal aberrations per 100 cells. However, there was a difference in the relationship between the number of chromatid aberrations, and chromosomal aberrations. In the group which we investigated, there was a relative increase in the number of aberrations of the chromosomal type, i.e. paired fragments and chromosomal exchanges and the relationship between the chromatoid and chromosomal aberrations was 0.4:1, with 1.1:1 in the control.

As was the case in studies on the metaphasic plates, bone marrow cells showed an increased number of aneuploid cells which was primarily due to hypoploid cells with 44-45 chromosomes and less. The average number of an uploid cells in the experimental group was 10.5%, and in the control group, it was 5.8% (P < 0.05).

Cytogenetic changes which were detected in persons with radiowave sickness after a fairly long period of time after work was terminated, are non-specific in nature, but still indicate qualitative deficiencies in the blood cells at this period of time.

Special attention must be accorded to the sequence of changes which occur in the blood system under the influence of SHF fields, and on the effects of duration and intensity of radiation. It is self-evident that the primary reaction of the blood system to exposure to relatively high intensities of SHF electromagnetic fields, consists of stimulation of hemopoiesis with the development of moderate leukocytosis, erythrocytosis, and reticulocytosis. This type of reaction was encountered in past years when the intensity of radiation at the different locations was fairly high, and the duration of exposure may have lasted for several years /N.V. Uspenskaya, 1959; and others 7. We encountered the stimulatory effects with leukocytosis in workers that had been exposed for short periods of time to electromagnetic fields with intensities of several mW/sq cm /V.V. Sokolova and M.N. Ariyevich, 19507. Ye. V. Gembitskiy /19697 and others, also regard leukocytosis as a primary response to SHF. Subsequently, with the increase in duration of exposure and the developments of symptoms of the radiowave sickness, the stimulatory response is replaced by cytopenic reactions which most frequently consist of leukopenia and thrombocytopenia.

We investigated the effects of the intensity of the electromagnetic fields of the nature of the hematological changes at one of the enterprises of the radio industry where, as a result of the public health measures which were taken, a sharp decrease in the intensity of radiation occurred beginning with 1960. The first group consisted of 60 subjects who worked there until 1960 when the intensity of the electromagnetic fields approached several mW/sq cm. The second group (65 subjects) consisted of individuals who commenced work after 1960 when the intensity of radiation as a rule did not exceed hundredths of mW/sq cm, and short term exposures to more intense radiation occurred only sporadically. The groups differed little in terms of sex, age, and the type of work they performed. The length of service of every individual was approximately ten years. Results of our studies demonstrated significant differences in the hematological indices of

those two groups (Table 3). The subjects that had been exposed to high intensity SHF for long periods of time, evidenced marked cytopenic reaction with a decrease in the levels of leukocytes, thrombocytes, as well as erythrocytes. Prolonged exposure to low intensity radio waves elicited only a limited decrease in the number of thrombocytes and the hemoglobin concentration as well as a tendency toward erythrocytopenia.

In summarizing the results of our investigations, we may state that the blood system responds to exposure to SHF electromagnetic fields. The nature of the changes to a large extent depends on the intensity and duration of the exposure to the radiowaves; however, even in the presence of symptomatology indicative of radiowave sickness, the hematological changes are not extensive, and do not appear in all cases, and furthermore, do not evidence a marked tendency to progress. Termination of contact with SHF leads to gradual normalization of hemopoiesis. Our experience, as well as the data in the literature, indicate that there is no reason to believe that hypoplastic changes and leukoses may arise as a result of exposure to electromagnetic fields of the radiofrequency range which was being considered. These data indicate that electromagnetic fields of intensities which do not elicit a thermal effect, do not directly act on the processes of cellular proliferation.

As a result of the changes induced in the nervous system by these factors, changes occur in the regulation of the neurovascular and humoral systems, including the blood system . As a consequence of this cellular metabolism is altered. Degenerative changes accelerate the death of cells. Evidence for qualitative deficiencies in the blood of the patients is indicated by the tendency toward sporulation of the erythrocytes, indications of metabolic changes in leukocytes, as well as the increase in the number of hypoploid cells. Changes which are evident in the bone marrow may apparently be regarded as a compensatory reaction of intensified regeneration. A direct and specific effect of SHF fields on the blood cells themselves cannot be excluded, however, at the present time, this question remains open, and shall be the subject of further studies.

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Table 1. Peripheral Blood Indices in Radiowave Sickness

	•	• •	•				
Грунпы (1)	Эритро-	Гемогло.	Цветной	Рекулоци-	Тромбоци-	РОЭ,	Лейкони-
	циги, млп.	бан, т/?;	показатель	аы, г.	зы, тыс.	мм/ч	ты, тыс.
	(2)	(3)	(4)	(5)	(0)	-(1)	1 (0)
Контрольная	4,70 <u>-</u>	14.7±	0,95±	7,2 <del>∃:</del>	$245.0\pm$	6,0±	6,49 <del>-1-</del>
(9) (800 чел.)	±0,017	±0,04	土0,002	- <u>1</u> :0,25	$\pm 1.60$	±0,21	±0,058
(10) Основная	4,77 <u>±</u>	14,8 <b>±</b>	0,93 <u>:</u> ±	8,3û <u>-</u>	221,7±	4,8 <u>±</u>	5,60±
(131 чел.)	±0,035	≿0,09	±:0,002	±0,38 *	±:4,12 *	±0,13	±0,082 *
I подгруппа	4,74±	14,7-上	0,91 <u>士</u>	7,94±	219,1±	4,2±	5,39±
(11) (47 чел.)	±0,063	土0,10	士0,003	≓≥0,59	∃:7,93	±0,42	±0,141
II подгруппа	$4,78\pm \pm 0,039$	14,8±	0,92 <b>±</b> -	8,60±	223,2±	5,1±	5,71±
(12) (84 чел.)		.±0,12	-±0,003	±0,51	±4,90	±0,57	±0,115
\$	1						

(14)Продолжение

	(13) Лебкоцитарски формула, и								
Грушпы (1)	базофилы	-ыроппафа ыл	ивлочко- ядерные	сегменто- ядерные	лимфоцаты	моноциата			
	1 (157 -	1.103		(10)	- TTT	(20)			
(9) (800 чел.)	$0.5\pm \pm 0.01$	2.6≟ :±0.08	5.0 .!: ::::0.11	56,9:± +0.31	32,8 <u>-1-</u> -+0.30	$5.7 \pm 0.10$			
Основиля (21)(131 чел.)	0,5± ±0,07	2,8± ±0,27	5,2± -±0,22	52,6± ±0,06 *	34,6± ±0,51 *	5,9± ±0,25			
(11) (47 чсл.)	0,4± ±0,13	3,1± ±0,29	5,3± ±0,39	52,1 <u>-1-</u> ±1,24	34,6± ±1,15	5,7± ±0,33			
11 подгруппа (12) (84 чел.)	0,5± ±0,07	2,7 土 土0,33	5,2 <u>⊣</u> ₌ ⊐:0,29	52,4 ± ±0,71	34,5± ±:0,61	6,0± ±0,33			

(22) рименание Волоск таблицах сполучкой (\*) обозначены различия, доспи ающие статистической достоверности (p<0.05).

Key:

- 1. Group
  - 2. Erythrocytes, millions
  - 3. Hemoglobin, gm%
  - 4. Color index
  - 5. Reticulocytes, %
  - 6. Thrombocytes, thousands
  - 7. ESR, mm/hr
  - 8. Leukocytes, thousands
  - 9. Control (800 persons)
- 10. Reference (131 persons)
- 11. Subgroup I (47 persons)

Table 1. Continued

- Key: 12. Subgroup II (84 persons)
  - 13. Leukocyte formula, %
  - 14. Continued
  - 15. Basophils
  - 16. Eosinophils
  - 17. Stabnuclears
  - 18. Segementonuclears
  - 19. Lymphocytes
  - 20. Monocytes
  - 21. Reference group (131 persons)
  - 22. Annotation. In each table the asterisk (\*) denotes differences approaching statistical significance (P < 0.05)
- Table 2. Cytogenetic Indices in Cultured Lymphocytes of Individuals Had Sustained Radiowave Sickness

	(1) Наименование труппы	ห์กลแพะตรมดา สมัคญกลารานเร และรายห, วิ	ห้อมพาธุราภุณ นด้อยคุณหมี กล 100 และราดแ	Количетво аберрадий хроматрывого така на 160 клеток	Колечество аберракси хремасодно- го тена ис го клеток	Соотнолсяны хромандина и хромосты- цый аберра- цый	Количество алеундиклик клеток, %
(7)	Контроль (30 чел., 6000 метафаз) .	(2) 1,25	(9) 1,35	<b>(3)</b> 0,72	(ц) 0,63	(5) 1,1 : 1,0	(6) 5,8
(8)	Основлая грувпа (12 чел., 1200 метафаз)	1,33	1,42	0,42	1,0	0,4 : 1,0 *	10,5 *

Key:

1. Group designation

- 2. Number of aberrant cells, %
- 3. Number of chromatid--type aberrations/100 cells
- 4. Number of chromosomal-type aberrations/100 cells
- 5. Ratio of chromatid to chromosomal aberrations
- 6. Number of aneuploid cells, %
- 7. Control (30 persons, 6000 metaphases)
- Reference group (12 persons, 1200 metaphases)
  Number of aberrations/100 cells

# Table 3. Peripheral Blood Indices in Groups Exposed to SHF Fields of Various Intensities

-	Грушлы (1)	Эритроци- ты, млн. (2)	Гемогло- бщ, т/•а	Илетной наказа- 7)Л	Ретикуло- цаты, %	Тромбоци- ты, тыс. (6)	РОЭ, мм/ч <b>(7)</b>	Лейкоци- ты, тыс. (8)
(9)	Контрольная	4,70 <u>-</u>	4,7±	0,95±	7,2±	245,0±	6.0±	6,49±
	(800 чел.)	±0,617	±0,04	±0,002	±0,25	±1,60	±0,21	±0,058
<b>(</b> 10)	I групна (60 чел.)	$4,29 \pm \pm 0,041$ *	13.8± ±0,16 *	0,95± ±0,003	8,3± -±0,41 *	196,0 <u>+</u> ±3,57 *	5,0 <u>+</u> ±0,52	5,67± ±0,184 *
<b>(1</b> 1)	11 группа	4,56 <u>+</u>	14,1±	0,94 <u>+</u>	6,8±	225,0 <del>±</del>	4,7±	6,74±
	(65 чел.)	±0,058	±0,11	±0,003	±0,48	±5,5 *	±0,57	-+_0,245

-						(12) <sub>Пµ</sub>	одолжение			
			Лейкоцитариая формула, %							
	<sup>Грунны</sup> (1)	базофилы	аозинофи- лы (14)	палочке- ядерные (15)	сетменто- ядерење -(16)	аямфоциты (17)	моноциты (18)			
(9)	Контрольная (800-чел.)	0,5 <u>⊣</u> - ±0,01	2.6± ±0,03	5,0≟ ±0,11	56,9 <u>+</u> ±0,31	$32.8\pm \pm 0.30$	$5.7 \pm \pm 0,10$			
(10)	I группа (СО чсл.)	$0.5\pm \pm 0.008$	2.7± ±0,33	6,4:± .±0,42	53,4± ± 1,0 *	34,8± ±0,90 *	6,0∄- :±0,36			
(11)	II группа (65 чел.)	0,6± ±0,07	2,0± ±0,18	$5.4 \pm \pm 0.30$	55,0± ±±1,2	32,3± ±1,16	6,1±± ±0,29			

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Key:

- 1. Group
  - Erythrocytes, millions
    Hemoglobin, gm/%
    Color Index

· · ·

- Reticulocytes, %
  Thrombocytes, thousands
- 7. ESR, mm/hr

- 8. Leukocytes, thousands 9. Control, 800 persons 10. Group I, 60 persons 11. Group II, 65 persons
- 12. Continuation
- 13. Basophils
- 14. Eosinophils

- 15. Stabnuclear
- Segmentonuclear
  Segmentonuclear
  Lymphocytes
  Monocytes

UDC 613.62:/614.87:537.868.029.64/

### GLUCOCORTICOID FUNCTION OF THE ADRENALS IN RADIOWAVE SICKNESS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 55-57

[Article by V.N. Dumkin and S.P. Korenevskaya]

Studies are conducted on corticosteroid excretion in patients with moderate and pronounced manifestations of radiowave sickness by means of their fractionation from urinary extracts by silica gel thin-layer chromatography (TLC). Changes in glucocorticoid metabolism in patients with radiowave sickness are related to damage of the deep structures of the brain which are responsible for regulating the activity of the hypophysis-adrenal cortex system.

It has been recognized that prolonged employment under conditions of intense exposure to SHF radiowaves may lead to the development of a disease which is characterized by a complex of neurological, neurocirculatory, vegetative, and endocrine metabolic dysfunctions /E.A. Drogochina and M.N. Sadchikova, 1964; N.V. Tyagin, 1971; and others/.

Detailed biochemical investigations conducted at our clinic on patients with radiowave disease have demonstrated a number of dysfunctions which are obviously the results of changes in the functional state of the mesodiencephalic regions of the brain /I.V. Pavlova, et al., 19707.

These data are in agreement with the results of clinical studies of E.A. Drogochina and M.N. Sadchikova /1964/, as well as with the investigations of M.S. Tolgskaya and Z.V. Gordon /1971/ who demonstrated significant changes in the cells of the hypothalamic-hypophysial region in animals that had been exposed for long periods of time to the effects of SHF radiowaves. The present study was conducted to define more precisely the metabolism of glucocorticoid hormones of the adrenal glands, and also the state of the central mechanisms of homeostatic regulation, and the formations which activate the hypophysisadrenal cortex system in patients with radiowave sickness.

Clinical observations were conducted of 20 patients who at work, especially during the first few years, were subjected to significant intensities of **S**HF electromagnetic fields in the centimeter range; the patients were from 33-46 years old, and their duration of service ranged from 10-20 years.

The clinical picture of the subjects under investigation was characterized by a complex variety of vegetative vascular dysfunctions and presence of crises, cerebral or coronary vascular insufficiencies, pronounced emotional ability, and asthenic manifestations. Depending on the severity and persistence of these manifestations, the forms of the radiowave disease were classified as moderate or pronounced (M.N. Sadchikova).

In the present study, we separated hydrocortisone (F), cortisone (E), their tetrahydro derivatives (THF, THE), and tetrahydro-17-hydroxy-11-deoxycorticosterone (THS) from urinary extracts by means of silica gel TLC /0. Adamec, et al., 1962/. Control data were obtained on 10 clinically healthy individuals between the ages of 30 and 45 years. The control data obtained in our investigations did not differ significantly from the data in the literature /K.V. Kruzhinina, 1968; and others/.

The average results for the background excretion of corticosteroids (mg/day) in the controls and in the patients (I), as well as the excretion in patients on the day of the adrenaline test (II), are presented in Table 1.

It is evident from Table 1 that the levels of these hormones have undergone significant changes including a decrease in the total hormone excretion  $\Sigma(P < 0.01)$  as well as a decrease in tetrahydrocortisone (P < 0.01), and cortisone (P < 0.02) with an insignificant increase in the THS fraction (P < 0.01). Changes in the average value for the F/E ratio indicate disturbances in the mechanism responsible for the transformation of cortisol into cortisone in the patients of this group.

Administration of small doses of adrenalin to healthy people does not result in a significant clinical reaction or activation of the hypophysis-adrenal cortex system which is due to the patency of the central mechanisms responsible for regulating homeostasis. Administration of small doses of adrenalin to patients with hypothalamic lesions leads to the development of vegetative vascular crises and changes in the excretion of corticosteroids  $/\overline{G}$ .L. Shreyberg, 1962/. In the majority of our patients, the subcutaneous administration of 0.3 mls of 0.1% solution of adrenaline hydrochloride resulted in the development of marked vegetative vascular reactions or crises, usually of the mixed type.

As is evident in Table 1, administration of adrenalin to the patients resulted in statistically significant (P < 0.01)increase in  $\Sigma$  and to some extent, an increase in THF, which indicates activation of the hypophysis adrenal-cortex system. However, changes in the levels of the THS, F and E fractions, as well as the F/E ratio, became even more pronounced. This obviously represents persistence of abnormalities in patients with the radiowave sickness, as well as alteration in the transformation of cortisol into cortisone.

It should be mentioned that similar changes in the glucocorticoid function of the adrenal glands have been detected by us in patients with toxic diencephalopathies in chronic intoxication with neurotropic poisons. These considerations led us to the conclusion that the changes in the glucocorticoid function of adrenal glands which we detected in patients with pronounced form of radiowave disease are sequelae of original lesions in the deep structures of the brain which are responsible for the central regulation of corticosteroid synthesis and metabolism.

The resultant data also suggest that the dysfunction in the hypothalamus hypophysis-adrenal cortex system, which appear at a certain stage of development of the pathologic process, may have a pathogenic significance and play a definite role in the formation of the clinical syndrome of radiowave sickness. This provides a basis for designing certain rational approaches in therapy.

Table 1.

	THP	THE	THS	P	E	ם	<u> </u>
Control	0,53±0,7	2,0 <u>±0,14</u>	0,01 <b>±0</b> ,005	0.21±0,02	0,31±0,05,	3,42±0,18	0,67
I	0,61±0,08	1,96±0,26	0,07±0,01	0,25±0,06	0,16±0,03	3,07.±0, <b>33</b>	1,6
11	0,75±0,09	2,00±0,23	0,08±0,02	0,37±0,08	0,14±0,02	3,33±0,0 <b>37</b>	2,6

#### UDC 614.87:537.868.029.64

ELECTROGRAPHIC DATA ON THE EFFECTS OF VERY WEAK MICROWAVES AT THE LEVEL OF THE MIDBRAIN RETICULAR FORMATION-HYPOTHALAMUS-CEREBRAL CORTEX LEVEL

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 58-65

[Article by M.S. Bychkov and I.S. Dronov]

Acute experiments on rabbits exposed to low intensities (up to 100  $\mu$ W/sq cm) microwaves yielded data on parallel changes in the background electrical activity of the cerebral cortex, the reticular formation (RF) of the brain stem, and in the posterior hypothalamus, which were characteristic of certain intersystemic changes in the brain, and indicated the important role of the subcortical-brain stem structures in the mechanism by which the microwaves elicit their biological effects in the CNS. Separate long-term experiments were conducted on the interaction between the anterior and the posterior regions of the hypothalamus, which is characterized by a phasic development of the response to microwaves.

Until recent times, the problem of the importance of the subcortical-brain stem structures, and particularly of the brain stem RF, and the hypothalamus, and the mechanism of action of microwaves on CNS at low (on the order of one mW/ sq cm) and, of even greater significance, very low intensities (up to 100  $\mu$ W/sq cm) -- that is to say, when non-thermal conditions of irradiation were employed -- have been virtually unstudied in electrographic experiments.

In the present article, two types of data are presented on the effects of very weak microwaves: data on parallel recording of the background activity of the cortex of the cerebral hemispheres, brain stem RF, and of the posterior hypothalamus obtained in acute experiments on rabbits, and electrographic data on changes in the anterior and posterior hypothalamus, which were obtained on rabbits in chronic experiments. Experiments on parallel recordings of the biopotentials of the cortex, the mesencephalic RF, and the posterior hypothalamus were conducted on 24 animals with a powerflux density (PFD) of 100  $\mu$ W/sq cm, and a 30 minute duration of exposure. A general summary of the results obtained at the end of the exposure, with respect to each of the investigated structures of the brain, is presented in a comparable manner with respect to the frequency of the given reaction in the diagrams of Figure 1.

Of primary interest here is the predominance of the reaction of deactivation, which is distributed among the investigated structures of the brain in the following manner: the deactivation reaction clearly predominates over other effects (in the present case, activation) in the posterior hypothalamus (in 18 rabbits against 6), was found to be the predominant form of reaction in the cortex (in 14 rabbits out of the 24 rabbits), and finally, was encountered very frequently, but did not predominate over the other effects in the brain stem RF (deactivation was found to occur in 12 out of the 24 rabbits).

If we relate deactivation only to activation, then the dominance of deactivation in the cortex and the posterior hypothalamus becomes even more significant. However, in the brain stem RF the predominance of the deactivation reaction over the activation reaction is insignificant.

Consequently, in distinction to the data obtained with respect to the cortex of the cerebral hemispheres and the hypothalamus, one cannot speak about a predominance of the deactivation reaction in the brain stem RF -- in the latter case, the incidence of activation and deactivation was approximately equal.

The maximum number of noticeable reactions (irregardless of the trend) was detected in the hypothalamus -- 100%. The absence of significant changes ("null" effect) is encountered in the cortex and the RF in approximately equal number of cases (in respectively 4 and 3 rabbits out of the total number of 24 rabbits).

Equal number of rabbits showed activation in the cortex and the hypothalamus; this number was significantly smaller than the number of rabbits showing deactivation in these structures. At the same time, activation in the mid-brain RF was encountered much less frequently than in the cortex and the hypothalamus. Above represented data which characterized the relationship in the various alterations in the structures of the brain from the point of view of the frequency of the appearance of a given effect in that formation. In addition -- and perhaps of even greater importance -- it is of interest to determine the distribution of these changes in the structures under consideration in every individual rabbit.

Among the 24 rabbits we found 5 variants (I-V) of combinations of these changes which arose in the cortex of the cerebral hemispheres, the posterior hypothalamus, and the brain stem RF. These variants (syndromes) are presented in the diagrams in Figure 2, where comparisons are made of the frequencies with which each variant was observed.

By neglecting syndromes IV and V, which occurred very infrequently (variant IV occurred in two cases, and variant V in one case out of a total of 24 rabbits) we can turn our attention to discussing the first three variants, and the conclusions which can be based on them. [This portion of the Russian text contains typographical errors -- translator].

First, variant I, which consists of a generalized deactivation reaction, shows clear dominance in all three of the investigated structures, while variant II (generalized activation), and variant III (absence of an effect in the cortex, deactivation in the hypothalamus, and activation in the RF, are encountered in an approximately equal number of cases. [This portion of the Russian text has typographic errors -- translator].

Second, generalized reactions show great predominance, i.e. unequivocal changes are present in the functional state of the brain structures under discussion (variants I and II together encompass 17 of the animals, that is, the majority of the animals that were studied).

Three, an effect in the cortex is always observed when the changes in the functional states of the hypothalamus and the RF (variants I and II) are identical; it is absent in those cases in which the responses in the hypothalamus and the RF are of contrasting nature (variant III), and, in addition, in each of those four cases, the interrelationships were identical: activation in the RF, and deactivation in the hypothalamus.

Variant III does not occur very often, but in our view it is of great interest -- particularly in relation to variants I and II -- since here we deal with four experiments which yield uniform results, thereby indicating that a SHF field may elicit not only identical, but also contrasting changes in the hypothalamus and the brain stem RF, and in this situation it appears as if though a mutual neutralization takes place of the effects of these subcortical structures exert on the cortex.

It is characteristic that not a single one of the other variants which is presented in Figure 2, contradicts variant III; variant III accounts for all those cases in which a null effect is evident in the cortex, and for all those cases in which the hypothalamus and the RF evidence contrasting changes.

Taking into consideration all of the five variants of the overall effect of the SHF field, it becomes evident that there are distinct, direct functional correlations between the changes in the cortex and the hypothalamus: inhibition in the cortex is correlated with inhibition in the hypothalamus and excitation in the cortex is in direct correlation with excitation in the hypothalamus. Naturally this suggests that the hypothalamus may have an important role to play in the development of the response of the cortex or the cerebral hemispheres to the field.

We may add another consideration to this. The inhibitory correlation between the cortex and the hypothalamus, is encountered only in the inhibitory (variant I) or "null" effect in the RF (variant IV), while an "excitational" type of correlation between the cortex and the hypothalamus occurs only in cases of stimulation (variant II), or, again, in the case of a "null" effect in the RF (variant V); furthermore, both type of correlations in the overwhelming majority of cases were encountered in situations in which there was a definite response and not a "null" effect in the RF. This suggests that under the influence of a SHF field, the effects of the hypothalamus on the cortex are either mediated by the reticular formation of the mid-brain (all the more so since the hypothalamus contains reticular structures), or that a parallel influence is exerted on the cortex by the RF; it is most likely, however, that both of these mechanisms interact.

If, however, antagonistic interactions come into play between the hypothalamus and the RF, then, as noted earlier (variant III), the effect of the SHF field on the cortex is blocked.

In this manner the significance of the RF in the mechanism of action of the SHF field on the CNS became quite evident.

In comparing the data which referred to the hypothalamus and the RF, we may come to the conclusion that binding correlations between changes in these two structures are absent, although there is a tendency for identical changes, particularly when they are inhibitory. In connection with this, we would like to point out that we did not encounter a single case in which stimulation of the posterior hypothalamus was combined with inhibition in the RF.

Finally, another consideration which is worthy of note, consists of the fact that in excitation of RF (and this occurred rather frequently -- in nine out of twenty four rabbits), the cortex showed other excitation or a null effect (variant I and II), and never an inhibitory effect, i.e. an effect which was usually typical and strongly expressed in this formation.

Analysis of the electrograms during different stages of exposure showed that the responses in many cases were characterized by a definite evolution of the response. The first changes are noted in the cortex; they occur very early, by the fifth to the tenth minute of irradiation. As a rule they consist of deactivation from the very beginning, even in those rabbits in which the final effect in the cortex is represented either by activation or appears as a null effect. In subsequent stages, these changes in the cortex are usually expressed either by progressive aggravation of the initial deactivation effect, or, on the contrary, by activation up to (variant III) or above (variant II), the starting level.

Changes in the hypothalamus and the mid-brain RF occur simultaneously (or almost simultaneously) during the tenth to the fifteenth minute of exposure to the SHF field, or even later. Usually those rabbits who in the final analysis show a generalized deactivation reaction (variant I) the initial changes in the RF consist of activation, and only with the duration of further radiation, this state is replaced by deactivation. The initial effects which are noted on the hypothalogram may either appear as activation or as deactivation, and later these initial features are either retained or are replaced by the opposite process.

Characteristically, development of activation in either one of the subcortical structures, or in both of these structures simultaneously, was correlated with attenuation of the primary deactivation reaction in the cortex of the cerebral hemispheres, and as deactivation in these subcortical structures developed, particularly in the hypothalamus, deactivation in the cortex was enhanced. Figure 3 contains an electrogram from an experiment in which the most frequent effect in response to the SHF field was obtained -- a generalized reaction of deactivation (variant I).

The resultant show, on the one hand, the primary deactivation of the cortex in response to SHF fields, and on the other hand, the subsequent effects on this response from the hypothalamus and the mid-brain RF, whose interaction in influencing the mechanism of response of the cortex to the SHF may take various forms, and may lead in the final analysis, either to enhancement of the primary effect of SHF field on the cortex, to a restitution of the initial level of the functional state of the cortex, or finally may lead to generalized activation of the cortex.

Now let us consider data which describe changes in the different regions of the hypothalamus in chronic experiments.

Studies on rabbits show the distinct changes in the hypothalamus on exposure to a PFD of 50  $\mu$ W/sq cm, which were particularly pronounced during the initial periods of the daily exposure.

Figure 4 represents the most characteristic response obtained which consisted of a generalized deactivation in the hypothalamus following a week of irradiation. However, contrasting alterations may be noted in the anterior and the posterior hypothalamus (Figure 5 demonstrates deactivation in the posterior regions of the hypothalamus in the presence of activation in the anterior regions).

The relationship of the temporary spatial changes in the anterior and the posterior hypothalamus indicate that the latter region is much more susceptible to the effects of radiation. This is evident by the earlier appearance of changes, or the greater degree of the response in the posterior hypothalamus (Figure 4, and especially Figure 6, where distinct deactivation is apparent in the posterior hypothalamus with a very insignificant analogous change in the anterior hypothalamus after the second week of irradiation; the generalized response here is relative, and the primary unit responsible for the systemic reaction appears to be the posterior hypothalamus).

These effects suggest that the posterior hypothalamus is of primary importance in the integrated effect of microwaves at the diencephalic level, and this includes the possible effects of the posterior hypothalamus on the anterior hypothalamus. Figure 7 illustrates the dynamics of change in the hypothalamus during the period of irradiation. In addition to a more pronounced response in the posterior hypothalamus at a given stage of chronic irradiation, Figure 7 also illustrates the phasic evolution of the response to the microwaves. During the initial stages (two weeks), the response is expressed very strongly (in the form of a generalized deactivation); subsequently the response becomes highly attenuated, and in the present case, the response takes the form of generalized activation at the end.

Although changes in the background activity of the hypothalamus are rather distinct, after one to two weeks of irradiation, they either weaken or are maintained at the previous level. Cumulation was not noted, at least not at the levels of radiation employed here.

The latter consideration obviously mitigates to a certain degree the hygienic evaluation of this factor at the intensities at which it was employed. However, we obviously cannot state that this indicates complete well-being. The fact that the persistent effects which we noted can at least be treated as an adaptive and a compensatory reaction, cannot be regarded entirely as an acceptable situation from the hygienic point of view. First of all, it has not been proven that this is, in fact, an adaptive-compensatory reaction. Secondly, one cannot regard as completely harmless the prolonged changes in the functional state of the diencephalic system, all the more so since we obviously deal with tension responses which border on stress.

On the basis of the above data presented here, as well as on the known discordancies which may arise in the interaction of the specific and nonspecific afferent systems in the thalamocortico-thalamic cycle which was described previously by one of us [M.S. Bychkov, 1967, 1970 -- PFD 20-30  $\mu$ W/sq cm] we may consider that the concept of the diencephalic genesis of the primary clinical syndromes in people subjected to low intensity SHF fields, has been entirely supported by direct experimentation, including the chronic experiments.

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- Figure 1. Relationships of Various Responses to Microwaves in the Cortex of the Cerebral Hemispheres, the Posterior Hypothalamus, and the Brain Stem RF. Numbers Indicate Rabbits With a Given Response.
- Key: 1. Cortex
  - 2. Posterior hypothalamus
  - 3. Brain Stem RF
  - 4. Absence of SHF effect
  - 5. Activation
  - 6. Deactivation



- Figure 2. Syndromes of Reticular-Hypothalamic-Cortical Changes in Rabbits Induced by Exposure to Microwaves. Arabic Numbers Indicate the Number of Rabbits With a Given Syndrome; Roman Numeral Indicate Syndromes.
- Key:
- 1. Absence of SHF effect
  - 2. Activation
  - 3. Brain stem RF
  - 4. a) cerebral hemisphere cortex
  - 5. b) posterior hypothalamus
  - 6. c) brain stem RF

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Figure 3. Dynamics of the Microwave Effect on the Rabbit Brain. Syndrome No. I. 1) Backround. 2, 3, 4, 5) Respectively 10, 15, 20, and 25 Minutes After Initiation of Irradiation. Top to Bottom: Visual Cortex of the Cerebral Hemispheres, Posterior Hypothalamus, Midbrain RF. Here and in Fig. 4 the Time Scale is One Second; 100 µV Calibration Potential.



Figure 4. Changes in the Rabbit Hypothalamogram Induced by One Week of Radiation. Top to Bottom: One Second Time Interval; Anterior Hypothalamus to the Left; Posterior Hypothalamus to the Right; Posterior Hypothalamus to the Left. I) Backround. II) After a Week of Irradiation.



Figure 5. Same as Fig. 4 in Another Rabbit. Top to Bottom: One Second Time Interval; Anterior Hypothalamus to the Right and Left; Posterior Hypothalamus to the Right and Left.







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Figure 7. Index of Rhythm Assimilation in the Hypothalamus At Different Periods During Long-Term Exposure to Microwaves

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ELECTROENCEPHALOGRAPHIC CHANGES UNDER THE INFLUENCE OF LOW INTENSITY CHRONIC MICROWAVE IRRADIATIONS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 66-70

/Article by M.S. Bychkov, V.V. Markov, and V.M. Rychkov/

Chronic experiments conducted on rabbits have demonstrated that intermittent and continuous irradiation of the animals with low intensity microwaves elicit qualitatively and quantitatively, different changes in the electroencephalograms (EEG), under otherwise equivalent energetic characteristics and duration of exposure. Intermittent radiation was seen to be somewhat more effective particularly in reference to the recovery times.

Until recent times, EEG investigations on the biological effects of microwaves have been conducted almost exclusively with continuous regimes of irradiation as well as constant intensity of the latter. However, under actual conditions people are exposed to alternating radiations under industrial conditions which are characterized by alternations in the periods of irradiation, and intervals of various durations in combination with variability in the intensity of radiation, i.e. the radiation is intermittent in terms of a number of parameters.

The present investigation consisted of a chronic (two month) experiment on rabbits with removable electrodes for recording EEGs in order to elucidate the nature of the effect of intermittent microwave radiation ( $\lambda = 10$  cm, pulse generator), employing an experimental model of irradiation which, in principle, adequately simulated industrial conditions (regime No. 1 of V.V. Markov /1972/): 150  $\mu$ W/sq cm for 8 minutes; 10 minute rest period; 60  $\mu$ W/sq cm for 8 minutes; 240  $\mu$ W/sq cm for 6 minutes; 34 minute rest period; 320  $\mu$ W/sq cm for 12 minutes; 60  $\mu$ W/sq cm for 8 minutes; 14 minute rest period; 60  $\mu$ W/sq cm for 8 minutes; 150  $\mu$ W/sq cm for 8 minutes. The total time of actual irradiation was one hour. This was the first series of experiments (series I). A parallel series of experiments, series C, was conducted with continuous radiation -- 153  $\mu$ W/sq cm for one hour. Each experimental series encompassed 12 rabbits. In both series of experiments, the incident energy, as well as the total period of exposure were equal. A control series of experiments (series K /kontrol'/) were conducted on eight rabbits, also for a period of two months.

As the primary indicator, we selected the activation and deactivation changes which are represented by the corresponding alterations in the relationship for the relative values for slow and rapid fluctuations (on the basis of automated evaluation of EEG).

Figure 1 illustrates the dynamics of the primary indicator (the relative value expressed as the percent of the slow activity of the visual cortex) in control rabbits over a period of nine weeks, which was taken as the physiological norm. We can appreciate that there were no significant fluctuations in this index. As a result, the following fluctuations within the period of one week were selected as the physiological standards; an increase in the relative value of slow activity (synchronization)up to 10% of the overall background electrical activity, and a decrease (desynchronization) of up to 12% of the overall activity.

In the experimental series, as can be appreciated from Figure 1, there is a clear tendency toward a decrease in the relative values for the slow activity during the initial period of chronic irradiation.

In Tables 1 and 2 data are presented for the experimental series which represent changes in synchronization or desynchronization which exceeded the fluctuations in the control animals. Since changes were detected only during the first half of the period of irradiation, the data which are presented refer only to this stage of the experiments.

Let us consider Figure 2 in which the extent of the changes in activation and deactivation are presented. The plot shows that shifts toward activation dominate in both regimes of microwave radiation, and are relatively similar in both series; normalization was encountered in series C after 15 sessions of radiation, while in series I, marked changes in both parameters were retained for yet another week of irradiation.
By means of the U criterion of Wilcoxon-Mann-Whitney, we determined that the differences between the activation effects in both series after the first five sessions of radiation, were insignificant; after ten sessions the differences became significant (at  $\Delta_{\rm C}$  = 13.7,  $\Delta_{\rm I}$  = 16) which indicate a somewhat greater biological effectiveness of intermittent radiation.

The data which have been presented, therefore, show distinct, although limited, changes in the spontaneous electrical activity in the case of both series of animals, with somewhat greater changes evident in series I during the period of time which corresponded to ten radiation exposures, and retention of this effect in this series of animals for an additional week of continued irradiation, after which, even in this series, a return to the normal level was evident.

Are ogous relationships in effects with respect to time -- but which were more pronounced than those discussed above -- were seen in studies on the reactivity to sub-stress functional stimuli in the form of prolonged (two minutes) combined stimulation with light flashes and intermittent sound, with the intensity and the frequency of both stimuli being altered in contrasting directions.

Biopotential integrator studies, which had been conducted earlier by one of us (M.S. Bychkov) on a large group of intact animals (110 rabbits), revealed two types of responses to the present type of stimulation which, from the point of view of the theory of automatic regulation, may be regarded as a transient process: type A -- desynchronization -- occurred in 44% of the rabbits, and type B -- synchronization -- occurred in 43% of the rabbits. Thirteen percent of the rabbits were nonreactive. In addition, within type A and B responses, we detected a number of subtypes of the transient process. These subtypes are indicated in Figures 3 and 4 where point 1 indicates the deviation from the background during the two minute period of stimulation, and points 2, 3 and 4 indicate t. recovery dynamics during the next three-two minute intervals of the aftereffects. On the left of the curves are indicated the frequency of the appropriate subtype responses which are expressed as the percent of the total number of animals under investigation.

Subtypes of the transient process -- which represent deviations from optimal and suboptimal regimes of regulation -- were encountered in isolated situations in a relatively large number of animals. This applies first of all to subtypes A'3 (significant deviation) and A<sup>3</sup> -- an extreme case of system modulation (the so-called over-regulation), and secondly, to subtypes B'l' and B"" -- extreme states of modulation which reflected functional deficiency could be characterized as profound neurasthenia.

Returning to our discussion of the effects of continuous and intermittent radiation during chronic experiments, we should like to point out that we did not observe any deviations in the control rabbits from the "standards" noted earlier throughout the course of the study. However, in the irradiated animals of both series during the first few weeks (i.e. during the preadaptational period) we encountered a significant increase in the number of responses which constituted deviations from optimal and suboptimal regulatory regimes. Thus, for example, if prior to irradiation in series I rabbits we did not detect any deviations at all, and in series C deviations amounted to only 8% (subtype A"), then after a week of radiation the frequency of deviation in series C (subtype A' and B") and in series I (subtype A" and A"") deviations were noted in one-third of the animals, and after two weeks, in half of the animals (in series I -- A', A", and B'"; in series C --A', A", B'", and B"").

Furthermore, as was the case in studies on spontaneous electrical activity, recovery (adaptation) in series I was prolonged for one week in comparison with series C (after three weeks deviations in series I were encountered four times more frequently than in series C).

In addition, there were also certain fine differences in the effects elicited by both types of radiation regimes, namely:

1. Among the reactions which took place in the regimes which were close to being optimal, in series I more persistent transient processes predominated (time constant data) in comparison with series C.

2. In series I, in distinction to series C, there was a fairly distinct tendency toward amplification of non-linearity which was in part apparent in the increase in divergence between theoretical and experimental models, and amplification of the disproportion between adjacent amplitudes of deviation on the curves of the transient process.

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The mathematical models which were obtained with the assistance of the theory of automatic regulation led to the conclusion that the factor of intermittence adds a very definite and a specific dimension to the effects of the microwaves.

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Nevertheless, the major result was, without doubt, the difference in the recovery times. In interpreting this in terms of adaptation, we may speak of the well known deadaptational effect of intermittence.

The material which we have just presented indicates that intermittent microwave radiation has somewhat greater biological consequences, at least under the conditions employed in our study.



Figure 1. Changes in the Average (Median as % of Backround) of the Arbitrary Value of Slow Activity in the General EEG Rhythm System of the Rabbit Taken as 100%. Confidence Intervals for the Median Taken From the Table of A.P. Ashmarin and A.V. Vorob'yev Based on Van-Der-Varden Data. Table 1. Bivariant Shifts in Series C (in Relation to the Backround)

(1) Недели после облу- чения	% слу- чаео (2)	L Δ, ţ <sub>ΰ</sub>	% слу- чаев (2)	۵, %	% слу- чаев (2)	Δ, 5	% C.IV- 436B	4 Δ. %
Δ+ Δ 3) Отсутствие изменений	33 58 9	4 16,6 —	17 66 16	2 13,7 	100	1 1	100	

(Ц) Примечания. 1. В таблице приведены значения отклонений ( $\Delta$ ) за пределы принятых эталонов нормы (контроль):  $\Delta$ +дерактивация,  $\Delta$  — активация. 2. В графе «Отсутствие изменения» показаны случан, когда изменения не превышают эталопных границ.

Key:

- 1. Week after irradiation
- 2. % of cases
- 3. Absence of change
- 4. Annotations. 1) Table presents deviation values ( ) exceeding accepted normal standards (controls): + deactivation, deactivation. 2) "Absence of change" indicates cases in which changes do not exceed standard limits.

Table 2. Bivariant Shifts in Series I (in Relation to the Backround)

(1)	1		2		3		4	
Недели после облу- чения	% слу- чаец	Δ, %	% слу- чаев	Δ, 🤧	% слу- чаев	A, 96	75 слу- Чаев	Δ. %
	(2)		(2)		(2)		(2)	1
	17 58	6 14	33 58	4 16	25 66	8 12	8	1
<ol> <li>Отсутствие</li> <li>изменений</li> </ol>	25		9	_	9		92	
	I							

Key:

- 1. Week after irradiation
  - 2. % of cases
  - 3. Absence of change



- Figure 2. Bivariant Changes in Experimental Series Indicating Deviations in Arbitrary Values for Slow Activation (in the General Rhythm System) as % of the Control. Fluctuations in the Control Taken as Zero Level.
- Key:
- 1. Weeks 2. Irradiation regime:
- 3. Intermittent
- 4. Continuous
- 5. Control



Figure 3. Type A Transitional Process in Intact Rabbits (Explanations in the Text)

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Figure 4. Type Transitional Process in Intact Rabbits (Explanation in the Text) 94

## UDC 614.87:537.868.029.64

THE EFFECTS OF CONTINUOUS AND INTERMITTENT MICROWAVE RADIATION ON WEIGHT AND ARTERIAL PRESSURE DYNAMICS OF ANIMALS IN CHRONIC EXPERIMENTS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 71-75

[Article by V.V. Markov]

Data are presented on the comparative biological evaluation of two types of microwave radiation (continuous and intermittent) of non-thermal intensities which were equal in terms of the total incident energy and the duration of exposure. Intermittent radiation had a more pronounced biological effect as evidenced by the significant lag in weight of and more pronounced hypotension.

Extensive experimental data which have been described in the literature on the biological effects of microwaves deal with the effects of continuous radiation /A. Denier, 1933; H. Kuttig, 1955; Z.V. Gordon, Ye. A. Lobanova, and M.S. Tolgskaya, 1955; Z.V. Gordon, 1960, 1964, 1966; Yu. A. Osipov, 1962; I.R. Petrov, and A.G. Subbota, 1966; N.V. Tyagin, 1968; and others/. The effects of other regimes of microwave irradiation have remained virtually unstudied.

We have conducted comparative studies on the effects of intermittent and continuous irradiation with microwave electromagnetic fields (EMF) on weight dynamics and arterial pressure (AP) of animals in chronic experiments. Details of the irradiation regimes have been described in our joint articles with M.S. Bychkov and V.M. Rychkova which are included in the present volume.

The studies were conducted on 36 white rats divided into three groups, each consisting of 12 animals. For studies on weight dynamics, the groups were selected in such a manner that the

initial mean weights for each group were equivalent. Animals in the control and the experimental groups were maintained under identical conditions.

Three series of experiments were conducted. In the first series, series I, the animals were exposed to intermittent microwave irradiation, and in the second series, series C, they were exposed to continuous radiation; the third series, series K [kontrol'] served as the control group.

During the first three months there were no significant differences in the dynamics of the mean weights for the animals of the three groups (Figure 1); the observations were conducted for a period of seven months. A significant lag in weight gain became apparent only by the fourth month of irradiation in animals exposed to intermittent EMF in comparison with animals subjected to continuous irradiation. Subsequently, this lag was retained not only during the period of irradiation, but also during the period of recovery. Statistical analysis of the weight differences of the animals in series I and C by the method of Student, showed that the differences in the mean weights at the end of the fourth month were statistically significant (P < 0.05). At subsequent periods of time, in many cases, the reliability approached the threshold of probability (0.95). The latter factor supports the contention that animals exposed to intermittent radiation showed a tendency to lag in weight throughout the course of the entire experiment. Weight dynamics of the animals exposed to continuous EMF did not differ from the weight dynamics of the control series.

Comparative evaluation of the regression series, which reflect the dependence of the change in weight on the number of exposures, was conducted according to the cirterion of series differences, and the transcendence of one series over the other [I.A. Plokhinskiy, 1970; algorithm 31]. Furthermore, each experimental series was comparative with the control series. The results are presented in Table 1. Also included are standard values for a transformed Fisher's criterion which corresponds to the thresholds of probability (0.95--0.99--0.999).

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The data presented in Table 1 show a distinct effect -- which, incidently, was highly significant in terms of the criterion according to which one series exceeds the other -- for intermittent radiation in the absence of significant differences between the effects achieved with continuous radiation and the control data, as determined by the given algorithm.

Consequently, the following conclusions may be reached on the basis of the comparison of the effects of intermittent and continuous regimes of radiation:

1. Continuous irradiation with low (non-thermal) intensities does not influence the dynamics of the experimental animals.

2. Intermittent exposure to radiation with the parameters in question leads to a significant ( $\beta \ge 0.95$ ) inhibition of weight gain by the animals in prolonged experiments.

A plethysmometric method was employed to measure AP (modified instrument of A. Kh. Kogan, 1959).

Changes in the AP under both regimes of irradiation are phasic in nature (Figure 2). The initial phase which consists of elevated AP at the end of the fourth week, enters the second phase which consists of a persistent decrease. After irradiation is terminated (recovery phase), a gradual increase in AP takes place during a period of a month and a half until values are reached which are close to the initial values.

Comparison of the nature of the changes in AP in terms of each phase makes it possible to elucidate certain qualitative differences in the effects elicited by intermittent and continuous radiation. While during the first few days (the first and the third) a more distinct and significant (P < 0.05) increase in AP was encountered in animals belonging to series I, while in series C, only a tendency toward an increase in AP was noted, then already by the end of the second week of irradiation, the greatest increase in AP was encountered in series C animals, while in series I, AP remained at dead level, which it had reached by the third day. The significance of the differences between the values for AP of the experiments in this series, as well as in comparison with the controls, was significant according to Student's t test where the probability exceeded 0.95.

Equalization of the empirical regression series in the second phase -- the major phase in terms of its duration -- by means

of a weighted sliding mean, resulted in a very clear picture of the differences in the effects elicited by the regimes of irradiation under comparison. Figure 3 represents theoretical regression lines from the moment of initiation of hypotension, to the end of the recovery phase. No advantage could be gained by equalization of the empirical series in the first phase in view of the fact that it is short.

We found it of particular interest to compare the effects of each radiation regime on the decrease in AP. To that end, we performed dispersional analysis of a unifactorial complex. We selected five gradations of the factor which corresponded to 30, 60, 70, 90, and 110 irradiation sessions. Analysis of the unifactorial complexes conducted on small samples with randomization by the use of a table for random numbers. The results of the analysis is presented in Table 2.

On the basis of the data in Table 2 it is evident that in the case of both regimes, the more probable reaction is one of a fall in AP (for intermittent regime  $\beta > 0.999$ , for continuous regime  $\beta > 0.99$ ). Furthermore, the results of dispersional analysis have shown a significant difference in the extent of the response elicited by the different regimes of irradiation:

1. In terms of the intermittent series of irradiation, the observed degree of influence was 66% ( $n_x^2 = 0.66$ ), and in terms of the continuous series it was 54% ( $n_x^2 = 0.54$ ).

2. For all objects in a given category, the influence of intermittent radiation on AP may account for ( $\beta = 0.95$ ), no less than 51% and no more than 81% ( $n_X^2 = 0.5 \div 0.81$ ) of the overall effect of the sum of factors which are considered in the dispersion analysis. At the same time, the confidence limits for the overall influence of intermittent irradiation ( $n_X^2 = 0.34 \div 0.74$ ) indicate that intermittent radiation may account for ( $\beta = 0.95$ ) only 34-74% of the overall effect of the totality of factors.

Therefore, the greater effectiveness of intermittent radiation vis-a-vis continuous radiation becomes self-evident.

In summarizing the results in respect to blood pressure, we may make the following conclusions:

1. In principle, the response of the vascular system to the effects of intermittent and continuous microwave radiation is essentially identical.

2. During the initial (short) phase of changes in arterial pressure, the hypertensive effect is more significant in animals subjected to continuous EMF radiation in comparison with intermittent radiation; however, its appearance is delayed. During the second phase -- the major phase in terms of duration -- the hypotensive effect is more pronounced in animals exposed to intermittent radiation.

3. The dynamics of change in arterial pressure during the recovery phase do not differ in the two experimental series.



Figure 1. Changes in the Body Weight of Animals in the Comparison Series During the Experiment.

Key:

- 1.⊿, gm
  - 2. Irradiation regime:
- 3. Intermittent
- 4. Continuous
- 5. Control
- 6. Period of irradiation
- 7. Recovery period
- 8. months

		(2) Сравниваемые серин					
	(1) Критерий 	(3) <u>M</u> = K	<u>(</u> <u></u>				
<b>(</b> 5)	Различия рядов (F1)	$F_1 = 2,3$ $F_{5t} = (2,0-2,6-3,5)$	$F_1 = 0, I$ $F_{SI} = (2, 0, -2, 6, -3, 5)$				
(6)	Превышения одного ряда над другим (F <sub>2</sub> )	$F_2 = 10.8$ $F_{SI} = (3.9 - 6.8 - 11.3)$	$F_2 = 0, 1$ $F_{S} = (3.9 - 6, 8 - 11, 3)$				

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Key:

- 1. Criterium
- 2. Compariosn series

• •

- 3. "I"\_"K"
- 4. "C"-"K"
- 5. Series difference (F) 6. Excess of one series over the other (F<sub>2</sub>)



Figure 2. Dynamics of Arterial Pressure Change in Animals of Comparison Series

- Key:
- 1. Regime of irradiation:
  - 2. Intermittent
  - 3. Continuous
  - 4. Control
  - 5. Weeks
  - 6. Days
  - 7. Period of irradiation
  - 8. Recovery period



		(1) <sup>II</sup>	нтермитти	рующий реж	CILM .	1
	(2) Разнообразие	Дисперси (суммы ко ратов)	ии Чис вад- степе свобо	ло Вариа ней (среді хи квада	исы нае Показатель Л <sup>ан)</sup> η <sup>2</sup> =	силы влияния: (6) 0,663±0,053
(8)	Факторнальное (м групповое)	еж- 1145,4		4 286	Его до 6,6 Ф=	$\frac{0,663}{0.053}$ =:12,5
(9)	Случайное (вну групповое)	три- 583,6	2	5 23	Достоверно ,3 F = -	рость по Фишеру: (7) $\frac{286,6}{97.2} = 12,3$
(10)	Общее	1730	2	9 59	$F_{\bullet t} = \{2$	2,8-4,2-6,5 }
		(11)	Непреры	вный режим		
	(2) Разнообразне	Дисперся (суммы ка ратсв)	ин Чис рад- степе своб	ло Вариа ней (сред оды кваар	псы пие Показатель аты) р <sup>2</sup> =	силы влияния: 66 :0.542±0.673
(8)	Факториальное (м групповое).	еж- 878,2	. (4)	) (5) 1   219,5	Бо 50 Ф=-	$\frac{0.542}{0.073} = 7,4$
(9)	Случайное (вну групповое).	три- 741,8	25	5 29,67	2 Достоверно Г = 2	ость по Фишеру: 7 19,550 29,672 = 7,4
(10 <b>)</b>	Общее	1620	29	9 55,8	$F_{\mu} = \{$	2,8-4,2-6,5
	•	(1)	3) Контра	ЛЬ		
	(2) Разнообразие	Дисперсии (суммы квад- ратов)	Число степекся свободы	Вариансы (средние звадраты)	Показатель си п <sup>2</sup> =0.16	лы влияния: (6) 8±0.133
(8) Dan	сторнальное (меж- рупповое)	(3) 204,7	(4) 4	(5)	Ero достов $\Phi = \frac{0,168}{2}$	ерность: (12) -=1.2
(9)Слу	чайное (внутри- групповое)	1012,5	25	40,5	0,133 Достоверность F = 51,2	по Фишеру: (7) —=1,2
(10) 060	цее	1217,2	29	41,8	$F_{**} = \{2, 8-$	4,2-6,5 }

Key:

- 1. Intermittent regime
  - 2. Extent
  - 3. Dispersion (sum of squares)
  - 4. Degrees of freedom
  - 5. Variances (mean-square)
  - 6. Extent of influence
  - 7. Fisher significance
  - 8. Factorial (intergroup)
  - 9. Random (intragroup)
- 10. Total
- 11. Continuous regime

12. Its significance
 13. Control



Figure 3. Change Dynamics in Arterial Pressure in mm Hg in Animals in the Hypotensive Phase (Weighted Slinding Means)

Key:

- 1. Arterial pressure
- 2. Irradiation regime:
- 3. Intermittent
- 4. Continuous
- 5. Control
- 6. Period of irradiation
- 7. Recovery period
- 8. weeks

# UDC 614.87:537.868.029.64

THE EFFECTS OF CONTINUOUS AND INTERMITTENT RADIATION ON THE FUNCTIONAL STATE OF THE HYPOTHALAMIC-HYPOPHYSIS-ADRENAL CORTEX SYSTEM

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 76-79

Article by I.A. Kitsovskaya and E.I. Polukhina

The present article deals with results obtained in investigations conducted on the functional state of the hypothalamus-hypophysis-adrenal cortex system. Intermittent and continuous exposure to microwaves elicited shifts in certain indices which reflect the state of the individual components of the system, as well as of the entire system.

The literature contains data dealing with changes in the system of neurohumoral regulation which accompany various pathological states and functional changes in the organism. As a result of this, a great deal of attention is accorded to investigations on the effects of microwaves on the functional state of the hypothalamus-hypophysis-adrenal cortex system.

The present investigation deals with studies on the effects of low intensity intermittent and continuous irradiation -equivalent in their energetic value -- on the hypothalamushypophysis-adrenal cortex system.

In order to study this system, we conducted two series of experiments. In the first series we employed the Thorn test in its classical form as well as in two modifications: eosinophil counts were obtained prior to and after the administration of 10 units of ACTH, eosinophil counts were obtained prior to and after injection of 0.5 mg/Kg of 0.1% epinephrine, and eosinophil counts were also obtained before and after exposure of the animals to a strong sound stimulus (70 db). In addition, in these experiments lymphocyte counts were also obtained under the same conditions.

In the second series of experiments, studies were conducted on the levels of ascorbic acid in the adrenal glands and their weight coefficients. These experiments were conducted on the animals under the influence of irradiation, and under the influence of stress stimuli in the form of heat and cold.

Table 1 contains data on the differences in eosinophil levels prior to and after the administration of ACTH in control and experimental animals.

As is evident from the data in Table 1, the glucocorticoid function of the adrenals in animals that were exposed to irradiation was completely retained. This is indicated by the decrease in eosinophil numbers by more than 50% in all cases following ACTH administration.

Table 2 contains data on eosinophil levels in the blood prior to and after the administration of adrenalin to control and experimental animals.

The data in Table 2 also indicates a significant decrease in eosinophils (more than 50%) four hours after the administration of adrenalin in animals in each of the investigated groups which also indicates that the normal function of the hypothalamus-hypophysis-adrenal cortex system was retained.

These changes in the eosinophil levels are evoked, in all likelihood, by increased secretion into the blood of the glucocorticoid hormone from the adrenal glands under the indirect influence of the hypothalamus. It has been established that epinephrine, which influences the hypothalamus, is known to elicit the formation of certain mediators in that structure which, on entering the hypophysis, stimulate the excretion of trophic hormones which influence the endocrine glands.

In Table 3 data are presented on blood eosinophil counts expressed as the difference between their numbers prior to and after the animals had been subjected to a strong sound stimulus.

The data in Table 3 indicate that animals in both irradiated groups showed a decrease in the number of eosinophils by more than 50% under the influence of the sound stimulus, while in the animals in the control group, the change was less pronounced. This is probably due to the fact that animals exposed to radiation became more susceptible to various stimuli, in the present case the bell, and in their case it appeared as a very strong stimulus, whereas in the control animals, it did not appear as such.

It is also possible that the difference in the reactions of the irradiated and the control animals to the sound stimulus may be due to certain changes in the central nervous system, since it is known that the participation of the adrenal cortex in the response of an organism to extreme stimuli, may be mediated by the central nervous pathway, where the first participant in the response process appears to be the cerebral cortex.

Confirmation of the fact that in the irradiated animals the hypothalamus-hypophysis-adrenal cortex system remains virtually unchanged, is also suggested by the fact that the level of lymphocytes in the blood decreases following the administration of ACTH, epinephrine, and after exposure to a strong sound stimulus.

As indicated above, in the second series of experiments, the functional state of the adrenal cortex was evaluated on the basis of its level of ascorbic acid and its weight coefficients.

These indices were studied in animals subjected only to radiation, as well as in animals subjected to heat and cold stresses.

If the state of the adrenal cortex is evaluated in terms of its levels of ascorbic acid, then it may be stated that under the influence of radiowave irradiation, their function does not change. However, comparison of the weight coefficients of the adrenals in irradiated and control animals, shows a tendency toward increased weight of the adrenals at all times of the investigation, only in those animals that had been subjected to continuous irradiation. In animals subjected to intermittent irradiation, this type of change was noted only after a week of radiation; at other periods of observations, such changes were not evident.

Animals that had been subjected to continuous radiation showed a tendency toward an increase in the weight of the adrenal glands if, on the day of the investigation, they were subjected to a cold stress (this was evident throughout the entire period of irradiation); however, animals that had been subjected to intermittent radiation showed similar changes only during the first one to two weeks of irradiation.

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Similar changes were noted in irradiated animals that were subjected to heat stress on the day of the determination. These data may perhaps indicate some type of a threshold for changes in the functional state of the adrenal glands since, according to the data of some investigators, one of the initial changes in the adrenals appears to be an increase in their weight coefficients.

Therefore, on the basis of our data, it may be said that continuous and intermittent irradiation with very low intensities does not alter the functional state of the hypothalamushypophysis-adrenal cortex system.

However, the animals are not indifferent to low intensities of irradiation, as indicated by the differences in the reactivity of the irradiated and the control animals to the sound stimulus. Thus, for example, a 70 db sound stimulus appears as a powerful stress factor for the former, while the latter do not respond to it in a similar manner.

Conclusions

1. Exposure to intermittent and continuous radiation of microwaves is virtually without effect on the functional state of the hypothalamus-hypophysis-adrenal cortex system in animals. (However, there is a tendency for the adrenals to gain in weight.)

2. Under the influence of irradiation, regardless of its nature (intermittent or continuous) the sensitivity of animals to the effects of a sound stimulus is altered, i.e. the hypothalamus-hypophysis-adrenal cortex system responds differently to the sound stimulus in irradiated and control animals.

# Table 1. Difference Between Essinophils Numbers Before and After ACTH Administration, %

	(1) Режим облучения		(2) Непрерывный,		Интерынт (3)ющий			Контроль		
(5)	Сроки исследования, не- дели	1	2	4	. 1	2	4	1	2	4
(6)	Содержание эозинофн- лов. %	73,4	6 <b>0,0</b>	50,8	60,5	53,7	65,4	53,4	81,6	54,8

Key: 1. Irradiation conditions

5. Study time, weeks

2. Continuous

- 3. Intermittent
- 4. Control

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6. Eosinophils level, %

Table 2. Difference in Blood Ecsinophil Levels Before and After Epinephrine Administration, %

(1) Режим обдучения	Не	(2) :прерыя:	સમદે	Иптермиттир ющий				(Ц) Контроль		
Сроки исследования, не-	· 1	2	3	1	2	3	1	2	3	
Содержание зозинофи- лов, % . ( <b>б</b> )	91,5	92,6	93,0	93,0	92,2	84,0	90,3	93,3	89,4	

Key: Same as for Fig. 1.

## Table 3. Difference in Blood Ecsinophil Levels Before and After Exposure to a Powerful Sound Stimulus, %

(1) Режим облучения	Но	(2) прерывк	សេអី	Клтермиттирующий			Контроль		
Сроки исследования, не- дели (.5)	1	2	4	1	2	4.	1	2	4
Содержание зозинофи- лов. % . (.С.)	59,2	65, <b>2</b>	51,8	59,4	55,4	52,9	18,8	0,6	31,2

# Key: Same as for Fig. 1.

# UDC 614.87.537.868.029.64

THE EFFECTS OF INTERMITTENT AND CONTINUOUS RADIATION ON THE FUNCTIONAL STATE OF THE ADRENAL MEDULLA

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 80-82

[Article by I.A. Kitsovskaya and E.I. Polukhina]

Studies conducted on the levels of epinephrine and norepinephrine in the adrenal glands showed relative stability of these factors to the effects of intermittent and continuous irradiation with microwaves. Certain changes were evident only in the presence of a cold stress.

A number of authors have demonstrated that the influence of external stimuli is frequently accompanied by changes in the tissue levels of catecholamines [A.M. Baru, 1969; E. Sh. Matlina, and T.B. Rakhmanova, 1967; N.M. Vyaz'mina, and Z. Ye. Pustovoytova, 1969]. In addition, the importance of the catecholamines in the stress reaction is well known.

The object of the present investigation was to study the functional state of the adrenal medulla under the influence of intermittent and continuous radiation. In these investigations we employed two regimes of intermittent and continuous irradiation.

The functional state of the adrenal medulla was evaluated on the basis of its concentrations of epinephrine and norepinephrine. Investigations on the catecholamines were conducted not only during irradiation, but also during the application of heat and cold stresses during irradiation.

The scheme of the first intermittent regime of irradiation which we used consisted of two alternating intensities (250 and 100  $\mu$ W/sq cm) and correspondingly, two durations of exposure (one hour and fifteen minutes) with a continuous regime of 2 mW/sq cm for 30 minutes.

The second regime of intermittent and continuous radiation has been described by V.V. Markov. That regime approaches the regimes which prevail under industrial conditions. In the latter case, irradiation is intertwined with intervals without radiation, and the intensities of irradiation are characterized by greater variability than is the case in the first regime.

It should be pointed out that the maximum intensity of irradiation in this regime amounts to 320 uW/sq cm, and the minimum is 60  $\mu$ W/sq cm. The intensity of continuous radiation is 153  $\mu$ W/sq cm.

Since our primary interest was to investigate the compare the effects of intermittent and continuous radiation, the regimes were adjusted in such a manner that the total energy value of each intermittent radiation was equal to the corresponding value of continuous radiation.

In the first regime of irradiation, studies on the catecholamines were conducted after two, six, and twelve months of irradiation. In the second regime, studies on catecholamines were conducted after one, two, three and four weeks of irradiation.

Results of investigations of catecholamines in the adrenals in the first regime of radiation showed insignificant changes in the levels of adrenalin and noradrenalin (the difference was not statistically different). Thus, after two months of irradiation with a continuous regime of radiation, the levels of norepinephrine were equal to  $211 \pm 43.7$  ng, and in the case of intermittent radiation,  $222.5 \pm 44.2$  ng; at the same time, in the control group, the level was  $139.6 \pm 23$  ng. After six months of irradiation, the corresponding values were  $680 \pm$ 71.1 ng,  $505 \pm 84.9$  ng, and  $590 \pm 62$  ng, respectively, and after 12 months, the corresponding values were  $205 \pm 2$  ng,  $212 \pm 27$  ng, and  $149 \pm 13$  ng.

Thus, an impression is created that there is a tendency for the norepinephrine levels of the adrenal glands to increase throughout the entire period of irradiation of the animals.

In view of the fact that a physiological stress may reveal latent functional changes in the state of the adrenal medulla in the present investigation, we subjected the animals to marked cold and heat influences (these stressful situations were applied immediately before investigations on the levels of the catecholamines ). However, even with the use of physiological stresses, we did not notice significant deviations from the normal levels in the concentrations of epinephrine and norepinephrine. In animals that had been subjected to irradiation for a relatively long period of time (six months) we noted an increase in the levels of epinephrine under both regimes of irradiation, which approached statistical significance and, likewise, a decrease in the level of norepinephrine which approached statistical significance in animals that had been subjected to a continuous regime of irradiation in the presence of a cold stress. Thus, in the case of continuous radiation after a heat stress, the concentration of epinephrine was 1,055 ± 102 ng, and in the case of intermittent radiation, it

was 1,188  $\pm$  117 ng, while in the control animals, it was 990  $\pm$  62 ng. After a cold stress, the concentrations of epinephrine were correspondingly 1,996  $\pm$  56 ng, 1,656  $\pm$  154 ng, and 1,300  $\pm$  100 ng, respectively, and the levels of norepinephrine in this case were 910  $\pm$  52 ng, 245  $\pm$  10.8 ng, and 280  $\pm$  25 ng.

In view of the fact that changes in the functional states of various systems and organs under the influence of external factors frequently can be noted only during the initial periods of influence, we decided to conduct investigations with a second regime of irradiation at earlier periods of time.

As has been noted above, investigations on the effects of the second regime of irradiation on the functional state of the adrenal medulla, were conducted after one, two, three and four weeks of irradiation. In these investigations, as in the previous studies, we employed heat and cold stresses.

The results of these investigations demonstrated that intermittent and continuous regimes of irradiation with microwaves, did not alter the function of the adrenal medulla in the irradiated animals during a period of one month. Only in individual cases did we note a tendency toward an increased norepinephrine content in the adrenal glands. After three weeks of the continuous regime of irradiation, the concentration of norepinephrine was  $307 \pm 42.7$  ng, while in the control animals, the concentration was  $246 \pm 48.2$  ng; in animals subjected to the intermittent regime, the value was  $320 \pm 57.2$  ng, and in the corresponding control group, it was  $246 \pm 48.2$  ng. Under the influence of a heat stress, in the case of the continuous regime of irradiation, the corresponding values were  $420 \pm 33.7$  ng, and  $132 \pm 56$  ng, respectively, while in the intermittent regime of irradiation, the respective values were  $406 \pm 71.9$  ng, and  $132 \pm 56.0$  ng. Conclusions

Low intensities of intermittent and continuous radiation exert virtually no effect on the levels of epinephrine in the adrenal glands.

Norepinephrine shows a tendency to increase, and in a number of cases, this increase comes close to being significant.

No differences were noted in the effects of intermittent and continuous irradiation in terms of the indices which we studied.

# UDC 614.87:537.868.029.64

CERTAIN DATA ON THE BIOLOGICAL EFFECTS OF CONTINUOUS AND INTERMITTENT MICROWAVE RADIATION

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 83-86

Article by N.K. Demokidova

Long-term experiments were conducted on the comparative effects of intermittent and continuous microwave irradiation on certain metabolic indices and the weight of the more important endocrine glands in rats.

It is known that nonthermal intensities of SHF influence protein metabolism in people [V.A. Syngayevskaya, 1962] and animals [S.V. Nikogosyan, 1962, 1968], and that enhancement of dissimilatory processes occurs which, as has been demonstrated by V.A. Syngayevskaya, is accompanied by stimulation of the hypophysis-adrenal cortex system (increased urinary potassium, decreased urinary sodium, increased urinary level of total nitrogen, and of 17-ketosteroids, and so forth).

In the present study we compared the effects of intermittent and continuous microwave ( $\lambda = 10$  cm) irradiation on certain aspects of water, electrolyte, and protein metabolism. In addition, we determined body weight changes and weight coefficients for such endocrine organs as the hypophysis, the adrenals, and the thyroid gland, since all of these factors are interdependent.

White rats, about 200 gm in weight (36 males and 72 females) were divided into groups, each consisting of 12 animals; one group served as the control, a second group was irradiated continuously for one hour with 153  $\mu$ W/sq cm microwaves, and the third group was subjected to the same total amount of incident energy in intermittent exposure to microwaves for

two hours (intensities of 60, 160, 240, and 320  $\mu$ W/sq cm, with the periods of irradiation interrupted by three intervals, lasting from 10 to 34 minutes).

Determinations were conducted on the levels of total nitrogen in the dynamic state (after dilution of the mineralized urine in Conway dishes), sodium and potassium in the urine (by means of flame photometry), and on the extent of daily diuresis expressed in milliliters. After the animals had been sacrificed, the endocrine glands were weighed, and the weight was expressed in mg%.

Table 1 contains data on certain biochemical changes in male rats that had been subjected to the effects of continuous and intermittent radiation. As can be seen from the data in Table 1, intermittent radiation evoked a tendency toward increased excretion of water and sodium (after two weeks, one month, and two months), as well as a statistically significant decrease in the urinary levels of potassium (after two weeks, and at the end of the experiment). After the animals had been sacrificed, the weight of the hypophysis was found to be significantly decreased (Table 2).

Under the influence of continuous radiation, a statistically significant increase in urinary concentration of sodium occurred (after two weeks, one month, and two months). At approximately the same time, there was a tendency toward increased excretion of water. The urinary levels of potassium remained within normal limits. The weight of the hypophysis showed a tendency to decrease. No changes were evident in the levels of total urinary nitrogen, the weight of the thyroid gland, and of the adrenals, nor were there significant changes in the body weight of both groups of animals.

In females, the following changes were noted (Table 3): a) exposure to intermittent radiation evoked the decrease in the excretion of water (during the first month) and sodium (during the third month), the increased excretion of potassium (during the fifth month), and a tendency toward a decrease in the total nitrogen levels in the urine during the entire course of the experiment with a return to normal levels at the end of the investigations; and b) continuous radiation evoked a decrease in the excretion of water throughout the course of the experiment, with increase toward normal levels after five months, decreased excretion of sodium (after two weeks and three months), potassium (during the third month), and total nitrogen (after one, two, and three months). Determinations of the weight coefficients of the endocrine glands in the females two weeks after irradiation yielded the following findings. Intermittent radiation resulted in a decrease in the weight of the hypophysis and the adrenal glands, and evoked a tendency toward an increase in weight of the thyroid gland. Exposure to continuous radiation also elicited a decrease in the weight of the hypophysis and the adrenal glands, and resulted in a significant increase in the weight of the thyroid gland. Five months after initiation of irradiation no changes were noted in the weights of the endocrine glands irrespective of the type of radiation regime that was employed, with the exception of a significant increase of the weight of the hypophysis in rats that were exposed to intermittent microwave radiation. In addition, significant changes in body weights were not noted.

It is generally recognized that changes in the weight of endocrine glands indicate, as a rule, changes in their function.

A number of authors have encountered changes in the function of the endocrine glands under the influence of radiowaves. Thus, F.A. Kolesnik, et al. [1967] found dysfunctions in the endocrine system of people under the influence of SHF, up to 10  $\mu$ W/sq cm in intensities, which in part included depressed function of the hypophysis-adrenal cortex system. Yu. D. Dumanskiy, et al. [1972] in experiments on rats and rabbits, showed that under the influence of pulses (1, 5, and 20  $\mu$ W/ sq cm), and continuous (0.5, 1.5, and 10  $\mu$ W/sq cm) regimes of irradiation with SHF energy ( $\lambda = 3 \div 12$  cm) over a period of four months, resulted in addition to changes in the CNS, an increase in the quantity of 17-ketosteroids in the urine, and a decrease in the weight of the adrenal glands, and their concentrations of ascorbic acid.

M.I. Smirnova and M.N. Sadchikova [1960] observed enhancement of thyroid function in people working with SHF generators. O.I. Shutenko and I.I. Shvayko [1972] used radioactive iodine to demonstrate increased function of the thyroid gland in white rats, that were subjected to decimeter SHF with a power flux density of 1.5 and 10  $\mu$ W/sq cm.

The hypophysis, the adrenal glands, and the thyroid gland are intimate components in the regulation of metabolism by the central nervous system.

Our experiments demonstrate that under the influence of relatively weak SHF fields, significant changes in the weight of these important endocrine glands take place, which are accompanied by changes in water, electrolyte, and in part, protein metabolism. These facts suggest that the effects of radiowaves on metabolism are mediated via the endocrine system.

Conclusions

1. Exposing rats to intermittent and continuous irradiation with microwaves results in the change in weight of such endocrine glands as the hypophysis, the thyroid, and the adrenals. These phenomena are accompanied by biochemical changes in metabolism.

2. Under continuous radiation, metabolic indices and the weight of the endocrine glands at the end of the experiment (four to five months) did not differ from the respective control values.

3. Under intermittent irradiation after four to five months, significant changes were apparent in the urinary levels of potassium, and in the weight of the hypophysis (in females these two factors were increased, and in males they were decreased).

# Table 1. The Effects of Intermittent and Continuous Irradiation on Certain Aspects of Metabolism in Male Rats

Влияние интермиттирующего и непрерывного режимов облучения

Табляца ј

Плн	тельность	(2)	(3) Сут	очное содержан	не, мг	· ·
001	<b>учения</b>	Режим облучения	калий (к.)	натрий	общий азот	Днурез, мл (7)
Две	(8) е недели	(9) Контроль Интермиттирующий	$74.1 \pm 4.9$ $48.8 \pm 7.1$	$9,6\pm1,75$ 11,4 $\pm1,75$	$219,1\pm19,3$ $157,2\pm30,0$	6,3±1,17 5,3±1,01
	Непрерыеный	$90,2\pm6,4$	18,8±3,47 p<0,05	237,2±23,9	6,7±1,38	
Оді	(10) ни месяц	(; •) Контроль Интермиттирующий Непрерывный	32,2±4,7 34,3±4,4 26,9±3,4	$\begin{array}{c c} 17,8\pm2,21\\ 21,3\pm2,16\\ 33,5\pm2,97\\ p<0,001 \end{array}$	92,3±11,7 114,7±27,7 87,6±8,90	5,3±0,96 8,7±1,46 7,8±1,93
Два	(11) амесяца	(9)Контроль Интермиттирующий Непрерывный	39,6±4,3 32,2±4,1 42,6±2,8	$6,7\pm1,83$ 10,0±3,28 13,4±1,75 p<0,05	148.9±12.3 147.5±2.5 182,8±21,6	$4.4 \pm 0.67$ 5.3 $\pm 1.20$ 6.5 $\pm 21.6$
Чет	(12) ыре ме- сяца	(9) <sub>Қонтроль</sub> Интермиттирующий Непьерывный	$47,2\pm5,2$ $32,9\pm3,9$ p<0,05 $50.0\pm6.1$	$28,8\pm8,90$ $29,2\pm4,30$ $24,3\pm3.04$	$242,2\pm19,1$ $249,9\pm23,7$ $278,5\pm31,6$	$7,6 \pm 1,11$ 9,0 $\pm 1,17$ 8,4 $\pm 1,17$

Key:

1. Length of irradiation

- 2. Irradiation conditions
- 3. Daily levels, mg
- 4. Potassium
- 5. Sodium
- 6. Total nitrogen
- 7. Diuresis
- 8. Two weeks
- 9. Control
- Intermittent Continuous
- 10. One month
- 11. Two months
- 12. Four months

Table 2. Body Weigth and Weigth Coefficients of Endocrine Glands of Rats Exposed to Intermittent and Continuous Microwave Irradiation

### Таблица 2

	(1)	(2) O6луч	енне	(3) Bec T	ела, г	Вефрек	оэффициенты ных желеэ	эндокрин-
	во живот- ных и пол	Режин (с)	Длитель- ность	исходный (7)	перед за- боем (9)	гипофиз	надпочеч- янки	щитовит- ная железа (1.1)
(12) (12) (12)	12 самок 12 самок 12 самок	Контроль (13) Интермит- тирующий Непрерыв- ный	Две не- дели (16)	$199 \pm 13,2$ $201 \pm 14,1$ $200 \pm 9,5$	$218 \pm 11,0$ $217 \pm 12,3$ $210 \pm 9,9$	5,7 $\pm$ 0,24 5,07 $\pm$ 0,19 p = 0.05 5,1 $\pm$ 0,12 p < 0,05	$29,1\pm0,63$ $25,8\pm0,64$ p<0,002 $26,6\pm0,92$ p<0,05	8,9±0,50 9,5±0,45 10,5±0,51 p<0.05
(12) (12) (12)	12 самок 12 самок 12 самок	Контроль (13) Интермит- тирующий Непрерыв- ный	Пять месяцев (17)	180±3,9 180±3,5 180±4,4	300 <u>+</u> 3,1 289 <u>-</u> ±3,5 291 ±4,4	$5,95 \pm 0.27$ $6,74 \pm 0.80$ p < 0.05 $6,0 \pm 0.23$	29,2±0,97 30,2±1,51 27,6±0,95	9,5±0,55 9,1±0,49 9,4±0,39
(14) (15) (15)	12 самцов 11 самцов 11 самцов	Контроль (13) Интермит- тирую ций Непрерыв- ный	Четыре месяца (18)	$231 \pm 6,2$ $231 \pm 6,6$ $230 \pm 6,1$	423±13,0 442±16.6 421±9,31	$3,23 \pm 0,08$ $2,53 \pm 0,09$ p < 0,05 $2,98 \pm 0,10$ p > 0,05	12,9±0,58 13,2±0,81 13,7±0,47	10,2 <b>±0,54</b> 9,98 <b>±0,41</b> 9,81 <b>±0,3</b> 4

# Вес тела и весовые коэффициенты эндокринных желез крыс, облучавшихся микроволнами в интермиттирующем и непрерывном режиме

Key:	1.	Animal	number	and	sex
------	----	--------	--------	-----	-----

- 2. Irradiation
- 3. Body weight, gm 4. Weight coefficients of endocrine glands
- 5. Conditions
- 6. Duration
- 7. Initial
- 8. Before sacrifice
- 9. Hypophysis
- 10. Adrenals
- 11. Thyroid
- 12. 12 females
- 13. Control Intermittent Continuous
- 14. 12 males

- 15. 11 males
- 16. Two weeks
- 17. Five months
- 18. Four months

# Table 3. The Effects of Intermittent and Continuous Irradiation on Certain Aspects of Metabolism in Female Rats

#### Таблица З

Лительность	(2) .	(3) Сут	очное содержан	не, кг		
облучення	Режим облучения	калий	натрий	общий азот	Диурез, мл (т)	
(8) Две недели	(9) <sub>Қонтроль</sub> Интермиттирующий Непрерывный	( <b>4</b> ) 62,2±6,80 56,6±6,75 54,9±7,07	(5) 25,7±3,10 20,1±2,30 16,8±2,30 p < 0,05	(6) 187,0±22,7 .158,4±15,4 148,1±16,9	$6,9\pm0,99$ $4,9\pm0,68$ $4,0\pm0,41$ p<0,02	
(10) Один месяц	(9) <sub>Контроль</sub> Интермиттирующий Непрерывный	49,9±5,95 38,6±1,73 43,7±5,18	5,4±1,32 4,6±0,73 9,5±1,58	$184,7 \pm 23,8 \\ 141,9 \pm 12,30 \\ 123,0 \pm 17,6 \\ p = 0.05$	$5,9 \pm 0.99$ $3,2 \pm 0.61$ p < 0.05 $2,7 \pm 0.59$ p < 0.02	
(11) Два месяца	(9) <sub>Контроль</sub> Интермиттирующий Непрерывный	14,5±2,49 16,7±2,50 9,9±1,95	10,8±2,16 9,2±1,70 12,2±2,66	$190,3\pm 39,2 \\116,6\pm 17,8 \\93,7\pm 13,1 \\p < 0,05$	$7,1\pm 1.714,4\pm 0.822,0\pm 0.41p=0.01$	
(12) Три месяца	(9) Контроль Интермиттирующий Непрерывный	$22,8\pm4.9121,3\pm3,708,0\pm1,62p<0,05$	$22,9 \pm 4,08$ $12,2 \pm 1,87$ p < 0,05 $10,1 \pm 1,88$ p < 0,01	$217.6 \pm 42.87 \\ 157.5 \pm 27.9 \\ 90.3 < 12.6 \\ p = 0.01$	7,6±1,45 5,6±0,99 2,7±0,79 $\rho < 0,01$	
(13) Пять меся- цев	(9) Контроль Интермиттирующий Непрерывный	$19,6 \pm 2.01 \\ 30,2 \pm 2.68 \\ p < 0,05 \\ 27,9 \pm 6,98$	$11,0\pm2,21$ 9,1 ± 1,34 12,2 ± 1,42	157,7±21,0 161,5±21,1 148,3±23,50	4,2±0,97 4,5±0,78 3,0±0,61	

Влияние интермиттирующего и непрерывного режимов облучения на некоторые показатели обмена самок крыс

Key:

- 1. Length of irradiation
- 2. Irradiation conditions
- 3. Daily Levels, mg
- 4. Potassium
- 5. Sodium
- 6. Total nitrogen
- 7. Diuresis
- 8. Two weeks
- 9. Control Intermittent Continuous
- 10. One Month
- 11. Two months
- 12. Three months
- 13. Five months

# UDC 614.87:537.868.029.64

THE EFFECTS OF INTERMITTENT AND CONTINUOUS IRRADIATION ON CHANGES IN THE SECRETORY FUNCTION OF THE HYPOTHALAMUS AND CERTAIN ENDOCRINE GLANDS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 87-90

[Article by M.S. Tolgskaya, Z.V. Gordon, V.V. Markov, and R.S. Vorontsov]

Comparative morphological studies were conducted on the biological effects of intermittent and continuous irradiation with microwaves of nonthermal intensities. Studies conducted at the level of the different structures of the hypothalamushypophysis-adrenal cortex system revealed reversible changes. The differences were observed in the restoration of the secretory activity of the hypothalamic nuclei (intermittent radiation resulted in a more prolonged course than did continuous radiation).

At the present time, there is no doubt as to the importance of the hypothalamus in adaptive reactions which maintain the internal milieu of an organism under the influence of a variety of adverse external factors.

The studies of V.I. Zubkova-Mikhaylova, V.F. Mayorova, S.V. Vladimirov, A.A. Voytkevich, A.A. Galoyan, V.V. Aleshin, M.S. Tolgskaya, Z.V. Gordon, and others, demonstrated changes in the amount of neurosecretion in the hypothalamus and the neurohypophysis under the influence of various physical stimuli (sound, light, electrical) chemical substances (x-ray irradiation), and irradiation with electromagnetic waves.

The present investigation is a portion of a complex of studies on the biological effects of intermittent and continuous microwave radiation. A unique feature of the present experiment was that the laboratory conditions were approximated as much as possible to actual industrial conditions, in other words, the experimental regime of irradiation adequately represents the irradiation of workers in the industry.

Of great theoretical and practical interest was the study on the process of neurosecretion in the hypothalamus and its correlation with changes which arise in certain endocrine glands.

Morphological studies on the neurosecretory function of the hypothalamus, neurohypophysis, and the endocrine glands was conducted in parallel with studies on the dynamics in the change in vascular tone.

These investigations were conducted in a chronic experiment of seven months duration. The experimental animals (white rats) were divided into three groups. The first group of animals were subjected to intermittent radiation, the second group was exposed to continuous radiation, and the third group served as controls. Details of the irradiation parameters are given in the article by M.S. Bychkov, et al. Four series of experiments were conducted on each group.

The nuclei of the anterior hypothalamuswere studied in serial sections following staining with aldehydefuchsin by the method of Gomori and of Bargman and Nissl, and for RNA. In all the groups and series, the animals were sacrificed at the same time.

In the first series of both groups of animals, sacrificed at the height of increase in arterial pressure (2-3 weeks after initiation of irradiation), an increase in the accumulation and secretion of the neurosecretory product was evident in the hypothalamic nuclei. There was an increase in the number of neurons whose protoplasm was completely filled up with neurosecretory granules. In the surrounding interstitium, along the course of axons, we frequently encountered drops of neurosecretory material (Figure 1). In addition, many neurons had small vacules in the protoplasm. Furthermore, there were accumulations of prolate drops and beading of the secretion along the axons of the nerve cells, and along the nerve fibers of the hypothalamohypophysial tract.

All of the morphological findings can be regarded as proof for the activity of the anterior nuclei of the hypothalamus, and active secretion into the blood stream. The neurons of the anterior hypothalamus were found to be in various stages of accumulation and secretion of the neurosecretory material in the second series of both groups of animals that were sacrificed 4-5 weeks after irradiation was commenced, when there was some stabilization of the arterial pressure (the arterial pressure was close to the initial background levels). Some neurons were in a state of rest after secreting their material, and Gomori-positive secretory granules were present only in the perinuclear area of the protoplasm. In other cells, the beginnings of resynthesis of the neurosecretory material were evident in the form of dark drops in the protoplasm (Figure 2).

In the interstitium, here and there small drops of neurosecretory material were detected, which were frequently distributed along the course of the fibers of the hypothalamohypophysial tract (Figure 2).

The histological picture showed a certain degree of stabilization or normalization of the neurosecretory activity of the hypothalamus and the neurohypophysis, which in many aspects approximated that found in the control animals.

In the third series of both groups of animals that were sacrificed after the low arterial pressure set in (22 weeks after irradiation began) the nuclei of the anterior hypothalamus were marked by the appearance of neurons that were smaller in size, as well as atrophied neurons. Only isolated neurons contained small secretory droplets in their protoplasm.

Rather frequently, nerve cells were encountered which contained almost no secretory material, and only a thin layer of protoplasm, i.e. exhausted nerve cells. In addition, neurons could be encountered which were in the initial stages of dystrophy with uneven and seemingly "corroded" outlines, and indistinct nuclei (Figure 3).

The descriptions just presented indicate exhaustion of the secretory activity of the anterior nuclei of the hypothalamus and of the neurohypophysis.

In the fourth series of both groups of animals that were sacrificed during the period of recovery (six weeks after irradiation was terminated) at a time when the arterial blood pressure was close to the background level, we noted a picture which was close to that seen in the control group of animals. The neurons of the hypothalamus were in various stages of accumulation and secretion of the neurosecretory material. Some cells were in a state of rest, other cells were in the beginning stages of resynthesis of neurosecretory material, and still other cells were secreting the neurosecretory material into the surrounding interstitium along the course of axons. Droplets of secretion were found in the hypothalamohypophysial tract along the course of nerve fibers, and in the posterior neurohypophysis.

However, in the hypothalamic nuclei were encountered neurons which were in the state of exhaustion and almost completely without protoplasm (the nucleus was surrounded by a thin layer of protoplasm).

Consequently, normalization was evident of the neurosecretory activity in the anterior hypothalamic nuclei. However, it must be noted that in animals that had been subjected to intermittent radiation, the process of recovery was proceeding somewhat more slowly and was less complete than in animals subjected to continuous irradiation, and in addition, a greater number of exhausted neurons were found.

Phasic changes in the functional activity were also encountered by us in morphological studies of the adrenal glands. Morphological studies of the thyroid gland also revealed certain phasic changes in the functional activity of this gland, however, the phasic changes did not always coincide with the changes in the functional activity of the hypothalamus.

The above considerations lead us to conclude that the hypothalamus and the neurohypophysis, along with the adenohypophysis and the adrenal glands, participate actively in the adaptive mechanisms of the organism.

To a large extent our data are in agreement with the results of P.D. Gorizontov [1970] who demonstrated that under the influence of various stress factors, the reactivity of an organism changes during the different periods of the extreme state.

During the first stage -- which consists of the mobilization of the adaptive mechanisms of an organism in response to a pathogenic factor -- there is an increase in the secretion of the hypothalamic nuclei, and increased functional activity of the adrenal glands. During this period, blood pressure of the animals increased. During the second stage -- increased nonspecific resistance -- we found a leveling off of the blood pressure, and at the same time, stabilization (normalization) of the hypothalamic and adrenal functions. On continued irradiation, a third stage of exhaustion sets in. During this period a persistent decrease in blood pressure appeared, and the number of exhausted cells in the hypothalamus increased; in the adrenal cortex the concentration of RNA, DNA and lipids decreased in comparison with the controls.

After irradiation was terminated, there was a gradual increase in the blood pressure, i.e. the state of recovery, and this was accompanied by gradual normalization of the functional activity of the anterior nuclei of the hypothalamus and of the adrenal glands. The phase of recovery was less complete and commenced later in the group of animals that had been subjected to intermittent radiation.

Our data point to the advantages which may be obtained by means of functional and morphological investigations of the neuroendocrine factors which are involved in the mechanisms of homeostasis and adaptation during the course of chronic irradiation with microwaves of low intensities involving different regimes of exposure (intermittent and continuous).


Figure 1. Accumulation of Neurosecretory Material in the Protoplasm of Hypothalamic Neurons. Large Amounts are Evident as Masses and Droplets Distributed Along the Course of Nerve Fibers. Gomori Stain. X 600



Figure 2. Hypothalamic Neurons in Various Stages of Accumulation and Secretion of the Material. X 600



Figure 3. Exhausted Neurons ("Naked Nuclei"). Bargman Stain. X 900

UDC 614.87.537.868.029.64

#### ON SETTING HYGIENIC STANDARDS FOR THE COMBINATION OF SHF AND X-RAYS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 91-94

Article by K.V. Nikonova and I.P. Sokolova

 $\sqrt{T}$ ext $\sqrt{T}$  Results are presented of experimental studies on the nature of the biological effects elicited by combined exposure to 10 cm microwaves and soft-rays with E = 13.5 kev for the purpose of obtaining fundamental data that could be utilized in establishing hygienic standards.

One of the current problems in labor hygiene , including that branch which deals with investigations on the effects of electromagnetic waves in the radiofrequency range, consists of elucidating the effects of all the industrial factors on an organism.

Presently, investigators are devoting considerable attention to the combined effects of SHF electromagnetic waves and x-ray irradiation.

The literature is replete with individual studies indicating that combination of these two factors has very serious consequences for the health of people working under such circumstances /T.K. Butkina, A.S. Vorontsova, Ye.A. Girskaya, et al., 1959; K.N. Klyachina, B.M. Stolbun, I.Ye. Okonishnikova, et al., 1961; A.P. Chesnokova, 1969/.

Few experimental studies are available on the biological effects of exposure to a combination of SHF and x-rays; they pertain to radiations of different energies, and their conclusions are not quite convincing and at times contradictory /K.N. Klyachina, B.M. Stolbun, and I.Ye. Okonishnikova, 1963; K.N. Klyachina, I.Ye. Okonishnikova, and E.A. Kalinina:,1966; K.V. Nikonova and N.D. Khramova, 1968; K.V. Nikonova, N.D. Khramova, and I.P. Sokolova, 1969; K.V. Nikonova, I.P. Sokolova, N.D. Khramova, and M.S. Tolgskaya, 1972; M.S. Sakovskaya, and P.R. Vaynshteyn, 1969; A.N. Liberman, M.S. Sakovskaya, and I. E. Bronshteyn, 1972/. The present study presents the results of experimental investigations on the biological effects following exposure to a combination of 10 cm microwaves and soft  $E_{eff} = 13.5$  kev x-rays which were designed to provide foundations for setting hygienic standards.

The experiments which were conducted with different SHF power flux densities and doses of soft x-rays, demonstrated that the nature of the biological effects in response to combined irradiation depends on the intensities of the factors involved.

Synergism prevails when the combination consists of high intensity SHF and x-rays. In such a situation the primary factor which is responsible for the clinical picture are the soft x-rays.

Thus, exposure to a combination of x-rays in a dose of 2,500 r (once) and SHF with a power flux density of 40 mW/sq cm (for 15 minutes per day for 6 weeks) produce a clinical picture which is characterized by the development of radiation sickness which is expressed by changes in the weight of the animals, decreased number of leukocytes in the peripheral blood, development of skin changes, and decreased testicular weight.

Animals that had been exposed to combinations of radiation evidence a greater percentage of deaths than do the remaining groups (subjected only to x-rays or SHF alone), more pronounced loss in body weight, earlier appearance of and more extensive cutaneous lesions, a more pronounced decrease in peripheral blood leukocytes  $\sqrt{K}$ . V. Nikonova, and N. D. Khramova, 19687. The resultant data make it possible to conclude that under the given conditions, soft x-rays and SHF conduct themselves as synergists -- by synergism we understand an effect which simulates summation, or exceeds it somewhat.

The conclusion regarding the synergism of the effects of SHF and soft x-rays was also found applicable when studies were conducted in which the intensity of SHF was 10 mW/sq cm and that of x-rays 250 r on single exposure, and 50 r/week on multiple exposures. In these experiments, the combinations of irradiation elicited a more pronounced decrease in the number of leukocytes in the peripheral blood.

However, in terms of other indices -- changes in body weight, motor activity, functional state of the central nervous system, reproductive capacity of the animals, and morphological changes in the organs and tissues -- the changes observed on combined exposure did not exceed those seen when either factor was employed alone. It should be noted that as transition is made to lower intensities, the role of UHF in combined irradiation, becomes more prominent. The changes which arise in the number of factors, are largely determined by the radiofrequency irradiation ( changes in body weight, leukocyte formula, hyperplasia of the RES cells in the liver, lymphoid elements in the lungs, and the lymphoid follicles in the intestines).

In setting hygienic standards for these factors, the biological effects elicited by low intensities of irradiation are of the greatest interest.

At the Institute of Labor Hygiene and Occupational Diseases of the Academy of Medical Sciences USSR from 1970 until 1972, extensive studies were conducted on the biological effects of SHF and soft x-rays under conditions of chronic experiments with corresponding levels of intensities of 1 mW/sq cm and 25 r/week. Studies were conducted on weight dynamics, peripheral blood (leukocyte and leukocyte formula, erythrocytes, hemoglobin, reticulocytes, thrombocytes), immunobiological reactivity, functional state of the central nervous system, reproductive capacity, weights of the individual organs, as well as the histological picture of organs and tissues of the irradiated animals.

In order to exclude any chance elements, the basic studies were repeated two to three times.

Comparison of the resultant data, along with due consideration to their reproducibility in repeated experiments, made it possible to determine the most characteristic effects which are due to the combined irradiation.

The resultant data demonstrated that combination of low intensity SHF and soft x-rays elicited different biological effects than those evoked by high intensity radiation. Under conditions of low intensities, a distinct amplification of the biological effect is not evident in comparison with instances in which SHF and x-rays are employed separated. The differences which are seen in certain parameters during certain periods of the investigation are, as a rule, not pronounced, unstable, and difficult to reproduce in repeated experiments.

The tendencies at the level of the individual organs and systems in response to combined effects of low intensity SHF and soft x-rays, may vary a great deal, and in some cases, may be determined largely by the radiofrequencies, and in others, by the x-rays. Thus, in animals subjected to the combined effects of the factors under consideration at certain periods of time in the investigation, we could detect a lag in weight gain which apparently was due to SHF, since an analogous effect was detected in animals that had been exposed to radiofrequency radiation along, and in none of the experiments did these two groups of animals differ significantly from one another. At the same time, exposure to x-rays alone did not evoke changes in weight dynamics.

In the peripheral blood of animals exposed to the combined effects of low intensity SHF and soft x-rays, changes arose which were very similar to the changes elicited by isolated radiofrequency irradiation. The differences which have been noted between these groups of animals at certain periods of time during the investigations, were, as a rule, unstable and poorly reproducible on repetition of the experiments.

Studies on immunobiological reactivity over a period of six months in investigations on the effects of combined irradiation, revealed nonuniform changes in the phagocytic activity and the digestive capability of the neutrophils as well as in the bacteriocidal activity of plasma. In addition, at different times of the observations, one or the other factor predominated. In terms of the extent of changes, the results did not differ whether the factors were used in combination or singly.

It is known that the central nervous system is highly sensitive to the effects of radiowave radiation. As a result of this, we included indices in our study which reflected the functional stage of CNS. Studies on the behavioral reactions of white rats in mazes, and on the bioelectrical activity of rabbit brains by means of EEG, demonstrated that changes evoked by the combined effects of microwaves and soft x-rays did not differ in their frequency nor in the degree of the change of the alterations which were induced by the isolated factors alone.

Due attention was given to studied on the reproductive functions of animals subjected to prolonged (6-12 months) exposure to the factors. Studies were conducted on the ability of the males to fecundate females, of the females to conceive, intrauterine deaths (pre- and postimplantation), the number of progeny in the litters, as well as qualitative characteristics of the progeny (weight, body length, presence of malformations, stillbirths). The resultant data did not show any significant differences between animals exposed to a combination of the factors, or to the factors in the isolated state. Histological investigations of the organs and tissues of animals subjected to combined irradiation for various periods of time (from 3-9 months) revealed changes which were characteristic of both radiofrequency as well as x-ray irradiation. In terms of the extent of these morphological changes, they were found to correspond to those which were observed when either of these factors was employed alone (see the article by M.S. Tolgskaya, et al., in this volume).

The absence of a distinct enhancement on combination of low intensity SHF and soft x-rays suggest that at the levels which are being considered, it may be possible to set standards for each factor independently of the other. However, taking into consideration the fact that biological effects resulting from combined irradiation with SHF and soft x-rays have characteristics which are typical of both factors, it must be emphasized that hygienic standards (in terms of each factor) should have an adequate safety factor.

In evaluating the actual situation from this point of view, it should be pointed out that we have no basis for determining the actual standards of radiation safety since the intensities of the soft x-rays which were employed in our studies (25 r/week), are much greater than the permissible level. We shall only note that under these conditions we did not observe any marked changes in the majority of the organs and systems, with the exception of the male gonads. However, in the case of the latter organs, we found significant changes after prolonged exposure (6-9 months) when the total dose of exposure amounted to 675-1,000 r.

Our studies on the SHF factor were conducted at the level of the "upper" hygienic standard -- 1 mW/sq cm. Under such influence, we noted changes in terms of a number of indices -weight dynamics, blood, immunobiologic reactivity, and morphological changes -- which were not evident in the control group of animals.

Biological effects of SHF with a power flux density of 1 mW/ sq cm have also been described by other authors [2.V. Gordon, 1966; A.G. Subbota, and Z.P. Svetlova, 1970; A.I. Ivanov, and B.A. Chukhlovin, 1968; and others].

We feel that for situations in which SHF and soft x-rays are combined, a power flux density of 1 mW/sq cm for SHF cannot be recommended as the maximum permissible level since the safety factor is inadequate.

#### UDC 614.87:537.868.029.64

THE EFFECTS OF COMBINED EXPOSURE TO SHF ELECTROMAGNETIC FIELDS AND SOFT X-RAYS ON THE PERIPHERAL BLOOD

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 95-99

[Article by I.P. Sokolova]

[Text] Long-term experiments conducted with a combination of low intensity SHF fields (1 mW/sq cm power flux density), and soft x-ray radiation (25 r/week), led to a decrease in leukocyte (due to a decrease in the absolute number of the lymphoid elements) and erythrocyte counts, and an increase in the number of reticulocytes. Analogous effects were evident in animals exposed only to microwave irradiation.

The literature indicates that both SHF as well as soft x-ray irradiation may elicit definite changes in the peripheral blood [I.A. Kitsovskaya, 1964; N.V. Tyagin, 1971; A.N. Liberman, 1966].

In recent years a number of studies have been published dealing with the effects of combinations of these factors, usually at high intensities (K.V. Nikonova and N.D. Khramova, 1968]. Furthermore, it has been noted that the effects due to a combined exposure are more pronounced than those elicited by either factor alone.

The object of our investigation was to elucidate the effects on the peripheral blood of combined exposure to 10 cm microwaves and soft x-rays ( $E_{eff} = 13.5$  kev) under conditions of a long-term experiment.

Studies conducted with 10 mW/sq cm SHF and 50 r/week x-ray irradiation, show that a combination of these two factors evoked a much more pronounced leukopenia than they did when employed alone (Figure 1).

Therefore, our data are in agreement with those published earlier, but which dealt with higher intensities.

The major portion of the study dealt with long-term exposure to combined irradiation of low intensities: 1 mW/sq cm SHF for 1 hour/day, and 25 r/week x-ray radiation.

The studies were conducted on white rats of the same weight, sex, and age. The animals were divided into four groups: one group was exposed to microwaves alone, another group was exposed to x-rays, a third group was exposed to combined irradiation, and a fourth group served as controls. The peripheral blood of the animals was analyzed for the numbers of leukocytes, erythrocytes, reticulocytes, thrombocytes, hemoglobin levels, and the leukocyte formula. These determinations were conducted once every two weeks.

Three series of experiments were conducted lasting from six to nine months. In the present article we present generalized data for all three series.

Let us consider the leukocyte counts in the peripheral blood of the animals. Certain variations may be evident in the leukocyte levels when the animals are exposed to x-rays alone. In one of the experiments, we encountered a relatively stable decrease after a month and a half of irradiation. However, in two other groups, the leukocyte levels were close to the control levels, and only on isolated days -- 3 out of 20 determinations -- differences between these groups were statistically significant.

In animals exposed to radiowaves alone, a decrease in the number of leukocytes was noted by the 2.5 - 3.5 month and remained depressed until the end of the experiment. Animals exposed to combined irradiation showed changes similar to the group exposed to microwaves alone, i.e. a stable decrease in peripheral blood leukocytes became evident from 2.5 - 3.5 months. There were no significant differences between these two groups of animals with the exception of one experiment in which, after three months of combined irradiation, a **pr**onounced leukocytosis was noted which, at a subsequent period of time (3.5 months) was replaced by leukopenia (Figure 2).

Analysis of the leukocyte formula with determinations of the absolute number of cells, showed that a decrease in the number of leukocytes in both groups was due to the decrease in the absolute number of lymphocytes (Figure 3). Let us now turn our attention to peripheral blood erythrocytes. In one series, the animals exposed to x-rays alone showed a significant decrease in the erythrocyte counts in comparison with a control group of animals after three months of irradiation. This effect was not clearly reproduced in other experiments. At different periods of time a moderate decrease in the number of erythrocytes could be encountered, but as a rule, it was of short duration, and was subsequently replaced by normalization of the erythrocyte levels.

The erythrocyte levels in the animals exposed to combined irradiation and to microwave irradiation alone, showed significant differences from the erythrocyte levels in the control animals after  $2\frac{1}{2}$  to  $4\frac{1}{2}$  months in every series. Furthermore, there were no differences between the animals of these two groups (Figure 4).

Hemoglobin levels in all groups of animals were similar, and only toward the end of the experiment  $(5\frac{1}{2})$  and 7 months) did we note a moderate decrease in hemoglobin in every group that was exposed to some form of radiation.

Irradiation caused a significant increase in the numbers of reticulocytes (Table 1).

The data in Table 1 show that after three months of irradiation reticulocyte counts were increased in all animals exposed to some form of radiation. However, these changes were most pronounced and similar in their degree in the animals exposed to combined irradiation and to SHF alone. After 3 and a half months, the group of animals exposed to combined irradiation differed to a statistically significant degree both from the control animals as well as from animals which were exposed to x-rays alone.

Thrombocyte levels did not undergo significant changes in the experimental animals.

Therefore, exposure to low intensities of SHF fields (1 mW/ sq cm power flux density), and soft x-ray radiation (25 r/ week) results in a decrease in the leukocyte (due to a decrease in the absolute number of lymphoid elements) and erythrocyte counts, and an increase in the number of reticulocytes.

Analogous changes were evident in animals exposed to microwaves alone. The differences between these two groups which were evident at different periods of time, were poorly expressed, unstable, and could not be reproduced.





Figure 1. Peripheral Blood Levels of Leukocytes (10 mW/cm<sup>2</sup>, 50 r/week)

Key:

1. Control 2. SHF

3. X-rays

4. Combined exposure

5. Backround

6. Time of exposure, months



Рис. 2. Содержание лейкоцитов в периферической крови (1 мВт/см<sup>2</sup>, 25 Р/неделя)



Key	i
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- 1. Control 2. X-rays
- 3. SHF

- 4. Combined exposure
- 5. Backround
- 6. Exposure time, months







- 1. X-rays 2. Control
- 2. 0011010
- 3. SHF
- 4. Combined exposure
- 5. Backround
- 6. Irradiation time, months



Рис. 4. Содержание эритроцитов в периферической крови (1 мВт/см², 25 Р/неделя)

Figure 4. Peripheral Blood Levels of Erythrocytes (1 mW/cm<sup>2</sup>, 25 r/week)

- Key:
- 1. X-rays
- 2. Control

3. SHF

- 4. Combined exposure
- 5. Backround
- 6. Irradiation time, months

ĺ		(2) Срок облучения, месяцы						
	грулпа животных (Г)	0,5	1,5	3	3,5	4,5	5,5	7
(3)	I. Рентген	18±1,9	14±1,4	18±1,2	16±1,3	16 <b>±0,8</b>	18±0,9	19±1,4
(4)	2. СВЧ	21±2,4	16±1,5	19±1,3	18±1,1	18±1,1	21±0,9	23±1,2
(5)	облучение	19±2,0	17±1,2	$17 \pm 1.5$	20±1,6	19±1,0	23±0,7	24±1,1
(6)		19±1,6	18±1,4	$14 \pm 1.0$	12±0,8	13±0,7	12±0,9	14±0,8

Содержание ретикулоцитов в крови, 0/00

# Table 1. Blood Reticulocyte Levels, %

1. Animal group

- 2. Irradiation time, months
- 3. X-rays 4. SHF
- 5. Combined exposure 6. Control

#### UDC 614.87:537.868.029.64

THE EFFECTS OF COMBINED EXPOSURE TO MICROWAVES AND SOFT X-RAYS ON IMMUNOBIOLOGICAL REACTIVITY OF ANIMALS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 100-102

Article by I.P. Sokolova

[Text] Studies conducted on experimental animals exposed to combined irradiation with low intensity SHF and soft x-rays, demonstrated that the induced immunobiological changes were analogous to, and no more pronounced than, those seen when either of these physical factors was employed alone.

At the present time, the literature is replete with data indicating that microwaves elicit changes in the immunobiological responses of an organism. Thus, A.I. Ivanov [1962] found a decrease in the phagocytic activity of neutrophils on irradiation of volunteers with 1,000-3,000 mW/sq cm microwaves. This change was not persistent, and after a day, this activity returned to the initial level.

B.A. Chukhlovin [1963] demonstrated that the decrease in the phagocytic activity of leukocytes, and in the bacteriocidal properties of blood serum in animals under the influence of continuous SHF field, depends on the parameters of the latter.

Ye. I. Smurova [1966] in studies on irradiation of white rats with centimeter waves at 10 mW/sq cm power flux density, noted increase in the ingestive capabilities of neutrophils with respect to E. coli, which was subsequently followed by inhibition of this activity.

Phasic changes in the phagocytic activities of leukocytes under the influence of radiofrequency band electromagnetic irradiations has been noted by Ye. G. Tkachenko and V.S. Padalka [1965], as well as by B.A. Chukhlovin [1966, 1968]. On the basis of the data in the literature, it is also known that the state of an organism in the course of immunological reactions, are also greatly influenced by other physical factors.

Thus, Ye. P. Shuvalova, et al. [1960] have noted poorly differentiated changes in the phagocytic activity of neutrophils in medical personnel of radiology sections who had been exposed for long periods of time to low doses of roentgen rays.

The object of our investigation consisted of studying the effects of combined exposure to low intensity SHF and soft x-ray radiation on immunobiological reactivity under conditions of a long-term experiment.

A 10 cm band generator was used to generate SHF while operating in the pulse mode. Roentgen rays were generated by RUM-7 (RUT60-20-1) equipment. The effective energy of radiation in our experiments was 13.5 kev.

The experiments were conducted on 48 white rats. The animals were divided into four groups: one group was exposed to SHF irradiation with a 1 mW/sq cm power flux density for one hour every day, another group was exposed to x-rays at intensities of 25 r/week, a third group was exposed to a combination of these factors, and the fourth group of animals served as controls.

Phagocytic activity and plasma bacteriocidal activity were determined by the method of V.M. Berman and Ye. I. Slavskaya as modified by A.P. Volkova and V.I. Ternov which made it possible to evaluate the intensity of phagocytosis, not only in terms of ingestion, but also in terms of the digestive capabilities of the neutrophils. In our experiments, the phagocytic and bacteriocidal activities of the blood were studied on a 24-hour agar culture of E. coli strain 675.

The effects of these physical factors alone, and in combination, were studied over a period of 6.5 months (Figure 1).

The data in Figure 1 indicate that significant differences were present in the natural resistance of the experimental groups.

In the animals exposed to microwaves alone, phasic changes in the ingestive function of the neutrophils was evident during the first three months, and subsequently, was followed by persistent decrease in this function -- initially this took the form of a decrease in the number of phagocytizing cells, and later was also characterized by a decrease in the number of ingested microbes (phagocytic index). Even more pronounced changes occurred in the digestive function of the neutrophils. A decrease in the digestive function was evident after 1.5 months, and remained depressed throughout the course of the experiment. Simultaneously, blood plasma bacteriocidal activity decreased. Consideration of the entire data for the period of irradiation showed that the group of animals exposed to SHF alone showed the most pronounced changes in the digestive activity of neutrophils, and in the bacteriocidal activity of the plasma (Table 1).

Exposure to x-rays alone resulted in a moderate decrease in the ingestive function of neutrophils (difference between this and the control group became statistically significant only after months of exposure).

The digestive activity of the neutrophils of this group of animals after a month of exposure, significantly exceeded control values, and after a limited decrease after two months of irradiation, a new rise in digestive activity was evident, which after four months, was replaced by persistent decrease in the digestive index.

Exposure to a combination of SHF and x-rays over a period of six and a half months resulted in variable changes in the ingestive and digestive functions of the neutrophils, and in the bacteriocidal activity of plasma. Furthermore, depending on the time of the observation, and the function which was being evaluated, either microwaves or x-rays seemed to be the more dominant factor. The extent of changes observed on combined irradiation, did not exceed those when either factor was employed alone.

Therefore, studies on the immunobiological reactivity of animals exposed to a combination of SHF field and soft x-ray irradiation, did not reveal any peculiar characteristics. Changes in the functions which reflected immunobiological reactivity under conditions of exposure to both factors, proceed analogously to those seen when either factor is employed alone, and do not exceed the latter case in terms of degree.



Цинамика поглотительной и переваривающей способности по показателям % фагоцитоза и % переваривания

Figure 1. Dynamics of Ingestions and Digestion in Terms of % Phagocytosis and % Digestion

- 1. % phagocytosis 2. Control
- 3. X-rays
- 4. SHF

- 5. Combined exposure
- 6. % Digestion
- 7. Backround
- 8. Irradiation time, months

# Table 1. Over-All Indices of Immunobiological Responsiveness During Irradiation Period

Суммарные показатели иммунобиологической реактивности животных за период облучения

	(1) Группа животных	(2) % фагоцитоза	(З) Индекс фагоцитоза	ина ж перевари- вания	Индекс пере- варивания	6) Бактерицид- ность плазмы
(7) (8)	1. Рентген 2. СВЧ	58±1,09 57±1,15	$0,94 \pm 0,034$ $0,96 \pm 0,042$	$55 \pm 1,20$ $49 \pm 1,29$	0,52±0,021 0,43±0,017	19 <b>±0,69</b> 13 <b>±0,41</b>
(9) (10)	з. Комоинирован- ное облучение 4. Контроль	$59 \pm 1.08$ $60 \pm 1.06$	0,96±0,033 1,00±0,034	$52 \pm 1.46$ $55 \pm 1.42$	$\begin{array}{c} 0,51 \pm 0,022 \\ 0,54 \pm 0,025 \end{array}$	17±0,47 20±0,60

- 1. Animal group
- 2. % phagocytosis
- 3. Phagocytic index
- 4. % digestion
- 5. Digestion index
  6. Plasma bacteriocidal activity
- 7. X-rays
- 8. SHF
- 9. Combined exposure
- 10. Control

### UDC 614.87:537.868.029.64

PATHOANATOMICAL CHARACTERIZATION OF CHANGES INDUCED IN EXPERI-MENTAL ANIMALS BY COMBINED IRRADIATION WITH MICROWAVES AND X-RAYS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 103-108

[Article by M.S. Tolgskaya, K.V. Nikonova, and R.S. Vorontsov]

[Text] Studies on the morphological changes on the organs and tissues in animals exposed to a combination of SHF and soft x-rays, showed that the nature and the degree of change correspond to those observed when either physical factor is employed alone. When high intensity irradiation is employed the x-rays exert the dominant influence, while at low intensities, the histologic picture of the organs and tissues is characterized by changes characteristic of both factors.

The object of the present study was to evaluate the morphological changes in tissues of animals exposed to a combination of microwaves (10 cm band) and soft x-ray irradiation ( $E_{eff} = 13.5$  kev).

The study was conducted with white rats and mice (a total of 94 animals).

The animals were sacrificed by decapitation. The material obtained for investigation was fixed in 12% neutral formalin or 80% alcohol and imbedded in celloidin and paraffin. Sections of the internal organs were stained by hematoxylin-eosin with sudan for the fatty tissues; sections of the brain were stained by the method of Nissl. Cutaneous receptors were studied by silver impregnation of the sections by the method of Bilschowsky and Gross, while the levels of ribonucleoproteins (RNP) in the epidermis and its derivatives were studied histochemically by the method of Brashe [transliterated].

Comparative evaluations were made of the histology of the

organs and tissues of animals exposed to a combination of SHF and soft x-rays, and animals exposed to either of these physical factors alone.

The resultant data were in excellent agreement with the results made on the functional state of the various systems, and point to the interrelationship between the nature and the degree of morphological changes during combined exposure, and the intensities of the physical factors.

Thus, exposure to high intensity combination (250-2,500 r single x-ray exposure, 40-10 mW/sq cm multiple SHF exposures) elicited pathomorphological changes in the animals which were similar to those encountered with x-rays alone. Below, we shall present morphological data on animals that were sacrificed after a single exposure to x-rays at a dose of 250 r followed by microwave irradiation for two weeks.

Animals exposed to combined irradiation or x-ray irradiation alone, showed evidence of perivascular and pericellular edema in the brain, and some swelling of the neurons. In the liver, x-rays evoked dystrophic changes in the hepatocytes. In addition, the liver cells are swollen and have a transparent, seemingly "dispersed" protoplasm. On SHF irradiation, the liver cells were not effected, and there was only a limited hyperplasia of histiocytic elements. On combined exposure the same type of proteinaceous dystrophy of the liver cells is evident that is seen when x-rays are employed alone, but there is also a moderate hyperplasia of the reticuloendothelial elements. Exposure to x-rays alone and to combined irradiation elicits changes in the lungs which are characterized by moderate congestion, perivascular edema, and a sharp decrease in the lymphoid elements, while irradiation with the radiofrequency waves alone, elicits moderate hyperplasia of the lymphoid elements around the vessels and the bronchi. X-ray irradiation also evokes a sharp decrease in the lymphoid elements of the spleen which appears to be an advanced stage of atrophy. The spleen does not undergo changes under the influence of SHF, and the Malphigian bodies are distinct, with an adequate quantity of lymphoid elements. Under the influence of SHF irradiation, the bone marrow retains the usual numbers of cellular elements, while in animals exposed to x-rays alone, or to combined exposure, the cellular elements decrease.

The greatest differences were noted in the testes. The seminiferous tubules retained the normal amounts of germinal epithelium distributed in layers when exposed to microwave irradiation. Occasionally, there was desquamation of large cells (spermatocytes) into the lumen of the tubules. In the case of x-irradiation, the epithelium of the tubules was, in most cases, necrotic, disorganized, and desquamated into the lumen; occasionally the tubules contained necrotic rose masses in the center which were surrounded by membrane without any germinal epithelium. Certain tubules were dilated, empty, and lined with one tc two layers of degenerated cells. Spermatocytes were absent. Combined irradiation induced analogous changes in the testes. Without a special enumeration it was impossible to detect any differences between the effects exerted by x-rays alone, or the combined exposure.

Thus, histologic investigations did not reveal any specific changes elicited by exposure to a combination of 10 mW/sq cm SHF, and 250 r soft x-ray radiation. The changes which were noted were similar to those obtained with x-rays alone in combination with compensatory-adaptive phenomena of certain stages which were due to the effects of SHF.

Analogous conclusions came from morphological studies on animals that had undergone long-term exposure to a combination of SHF and x-rays at intensities of 10 mW/sq cm and 50 r/week, respectively, and which were sacrificed 2 and 10 months after initiation of exposure. In this case changes due to the x-rays were less pronounced, and the compensatory-adaptive response of the RES was much more pronounced.

Subsequent investigations dealt with the analysis of the morphological changes under exposure to chronic combined irradiation with SHF and soft x-rays which we arbitrarily designated as being of "low" intensities -- daily one hour exposure to SHF with a 1 mW/sq cm power flux density in combination with x-ray radiation at an intensity of 25 r/week. Pathomorphological changes were studied at 3. 6, and 9 months.

After three months of exposure, dystrophic changes in the internal organs of the animals were not noted. Exposure to microwaves alone resulted in limited hyperplasia of the lymphoid elements of the lungs, and of the reticuloendothelial elements in the liver. The bone marrow was active and contained large numbers of cells. Multiplication of the interstitial elements occurred in the testes as well as an increase in the number of the cells of the germinal epithelium, a portion of which was desquamated into the lumen of the seminiferous tubules. In the skin, stimulation of the receptors was evident in the form of marked argentophilia, thickening, swelling, and increased volume of the axoplasm, as well as marked tortuosity of the nerve fibers. Histochemical methods revealed a decrease in the level of RNP in the epidermis and its derivatives.

On exposure to x-rays alone the testes showed initial stages of dystrophic changes in the germinal epithelium of individual seminiferous tubules (disorganization of the layers of germinal epithelium, occasionally limited desquamation, and in isolated tubules, precipitation of protein masses, and a decrease in the number of sperms). In addition, initial changes were evident in the sternal bone marrow (along with active bone marrow small foci were evident in which the active elements had been replaced by fatty tissue) and limited deposits of hemosiderin in the spleen, intestinal submucosa, and the liver interstitium. The receptor organs in the skin were not changed. RNP layers in the upper layers of the skin were normal, and large quantities of plasma cells and mast cells were present in the subcutaneous tissue.

Histologic studies of the organs and tissues of the animals exposed to combined irradiation showed features which were characteristic of both physical factors. In addition to hyperplasia of the lymphoid and the reticuloendothelial elements, stimulation of the cutaneous receptors, and a decrease in RNP of the upper skin layers, there were also initial localized dystrophic changes in the sternal bone marrow, increased deposition of hemosiderin in a number of organs, and the appearance of plasma and mast cells around the dermal vessels. In the testes, the stimulatory effect of SHF overlapped the effects due to x-rays (along with dystrophic changes in some of the tubules, a marked thickening of the cellular layer was seen in other tubules).

The changes noted above become more prominent as the duration of exposure is extended to six and nine months. However, even after nine months of irradiation, the initial dystrophic changes in the neurons of the cortex, the muscle fibers of the heart, and in the hepatocytes, do not exceed the changes which are evident when either physical factor is employed alone. As before, SHF elicits marked hyperplasia of the reticuloendothelial elements in the liver and the spleen of the lymphoid elements in the lungs, and of the microglia in the brain; SHF is also responsible for marked changes in the receptors of the skin, and for the decrease in the RNP of the epidermis and its derivatives. X-ray radiation is responsible for the increased numbers of mast and plasma cells in the subcutaneous tissues, and of the mast cells in the cellular tissue around the spleen. as well as for the increase in the deposition of iron-containing pigments in the lymph nodes of the lungs and the spleen. It may be assumed that these changes are due to increased permeability of the blood vessels. Exposure to x-rays alone for a period of nine months, elicits marked dystrophic changes in the germinal epithelium of the testes, which results in almost complete obliteration of some seminiferous tubules. In other tubules, the dystrophic changes are accompanied by an abnormal regeneration with the appearance of giant cells.

In case of combined irradiation, we must also add certain changes which are due to SHF such as homogenization and interstitial edema under the capsule, and multiplication of the interstitial elements with the formation in individual cases of tumor-like proliferation of the interstitial elements. It should be pointed out that this process is focal in nature, and this obviously is responsible for the fact that in combined exposure, we did not find a deterioration in the function of the testes.

Consequently, on the basis of the results obtained in three series of histological investigations conducted with different intensities of irradiation, we are led to the conclusion that morphological changes in the organs and tissues of animals exposed to a combination of SHF and soft x-ray irradiation, correspond in their nature and extent to the changes seen when either of these factors is employed alone. The effects due to x-rays seem to predominate when high intensity combined irradiation is employed, while the histological picture of organs and tissues of the animals exposed to a low intensity combination of SHF and soft x-rays, contains features characteristic of both types of factors.



Figure 1. Comparative Characteristics of Histochemical Changes in the Skin With Different Exposures. Brashe Stain , X 500. a) Mast and Plasma Cells in the Subcutaneous Tissues on X-Ray Irradiation (25 r/Week for Six Months). b) Decreased Levels of RNP in the Epidermis and Its Derivatives on Microwave Exposure (1 mW/sq cm, Six Months). c) Decreased Levels of RNP in the Skin Epidermis and Its Derivatives; Plasma and Mast Cells in the Subcutaneous Tissue on Combined Exposure to Both Factors.



Figure 2. Changes in the Sensitive Fibers of Receptor Zones. Bilschowsky-Gross Stain, X 500. a) Microwave Irradiation, 1 mW/sq cm, for Nine Months. b) Exposure to a Combination of Microwaves and Soft X-Rays (1 mW/sq cm and 25 r/Week) for Nine Months

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Figure 3. Comparative Characteristics of Testicular Changes. Hematoxylin-Eosin Stain, X 250. a) Moderate Desquamation of Germinal Epithelium Into Tubules on Exposure to 1 mW/sq cm Mirowave Irradiation. b) Desquamation, Disorganization, and Accumulation of Protein Masses in the Lumens on Exposure to 25 r/Week X-Rays. c) Wasting of Individual Tubules and Desquamation of Germinal Epithelium on Combined Exposure.



Figure 4. Stimulation of the RES Elements in the Liver of White Rats After Six Months of Exposure. Hematoxylin-Eosin Stain, X 250. a) Microwave Irradiation (1 mW/sq cm). b) Combined Irradiation (1 mW/sq cm; 25 r/Week)

UDC 614.87:537.868.029.64

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EXPERIMENTAL STUDIES ON THE BIOLOGICAL EFFECTS EVOKED BY COMBINED EXPOSURE TO MICROWAVES AND HIGH AIR TEMPERATURE

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 109-114

[Article by K.V. Nikonova]

[Text] Acute and chronic experiments were conducted to determine the biological effects of exposure to both SHF and heat. The combination of SHF and high air temperature was found to evidence a more potent biological effect in terms of a number of indices than when either of these factors was employed alone.

In practice, exposure to radiofrequency electromagnetic fields often takes place under adverse meteorological conditions. Furthermore, the combination of radiowaves and high air temperature is of particular interest since we are dealing with two physical factors which apparently are similar in their mechanism of action.

The literature contains isolated studies dealing with the combination of SHF and heat. Thus, A. Ya. Loshak [1965] noted more pronounced changes in the state of health of individuals exposed to SHF under conditions of a hot climate. I.J. Holland, et al. [1961] studied the combination of microwaves and heat, and found a greater loss in weight and decreased tolerance of microwaves in dogs. Individual reports on the relationship between the biological effects of SHF fields and meteorological factors, can be found in the studies of N.V. Tyagina [1960], Deichmann, et al. [1959], and Michaelson, et al. [1961]. Certain authors (Mumford, et al.) have pointed to the necessity of setting standards for SHF which give due consideration to the temperature factor.

On the one hand, the above considerations point to the current importance of combined exposure to electromagnetic radiofrequency

waves and heat, and on the other hand, to the fact that this problem has not been studied adequately.

In the present article we present results of chronic and acute studies on the biological effects of exposure to a combination of SHF and heat. SHF was generated by a 10 cm band generator operating in the pulse mode. A specially constructed heat chamber was used to provide the requisite temperature conditions. The latter consisted of an 80 x 80 x 80 cm wooden box, inside of which two perforated partitions made of organic glass were located. At the bottom of the chamber, between the external wall and the perforated partitions, two nozzles were located for introducing and removing air. Air was heated by means of an electric heater containing an electric coil. Air was admitted into the chamber from the heater by means of a connecting hose. The presence of the perforated partition assured the uniform distribution of the air within the chamber. The air was discharged into the working room, or out of the window. The chamber had an observing window and the door made of organic glass for observing the animals.

SHF intensities employed in our experiments covered 80-10 mW/ sq cm, and the air temperature was selected according to the experiment with a maximum of  $38-40^{\circ}$ C.

Survival studies were conducted on white mice and rats employing SHF of 80 and 60 mW/sq cm power flux densities at 14, 22, and 38-40°C. The resultant data are presented in Table 1.

The data presented in Table 1 show that the average survival time of white mice exposed to an 80 mW/sq cm power flux density SHF at 40°C was  $28.3 \pm 1.8$  minutes, and that all of the animals died between the 17th and the 41st minute of the experiment; in the group of animals exposed to the same intensity of radiofrequency irradiation at 22°C, some of the animals survived 2 hours of irradiation, and the average survival time was  $63.3 \pm 7.6$  minutes. The effects of SHF were even less pronounced when the air temperature was  $14^{\circ}$ C. A similar relationship was found to apply to white rats when 60 mW/sq cm power flux density SHF was employed.

The data show that death of animals under SHF fields of equivalent power flux densities, occurs more rapidly the higher the environmental temperature.

In order to obtain a clearer understanding of the more rapid death of animals exposed to a combination of SHF and heat, we conducted studies on rats exposed to 60 mW/sq cm power flux density SHF, and  $40^{\circ}$ C. The temperature response of the animals was studied at 15 and 22 minutes, since the animals begin to die at about 25 minutes. Body temperatures were determined with a universal electrothermometer TEMP-60. The results showed that at 15 minutes, there was no significant difference between the animals exposed to SHF, and those exposed to a combination of SHF and heat.

However, at 22 minutes, the increase in body temperature was more rapid on combined exposure due to decreased heat loss as a result of dysfunction of the compensatory mechanism, and this apparently was responsible for the more rapid death of the animals (Table 2).

Further studies dealt with a combination of 10-15 mW/sq cm power flux density SHF and heat ( $38-40^{\circ}$ C). In this situation, the increase in body temperature did not exceed that when either factor was employed alone, and consisted of  $0.5 - 0.7^{\circ}$ C.

A single 30 minute exposure to this type of combination decreased physical endurance of white mice which was expressed in their decreased swimming time (Table 3); long-term exposure to this combination elicited more pronounced changes in the weight curve, and in the vascular tone of the white rats.

Changes in the body weight of rats over a four month period with a 30 minute daily exposure period to the different factors is given in Figure 1.

The data in Figure 1 indicate the absence of a significant difference between the body weight of animals exposed to SHF or heat alone, in comparison with a control group. However, combined exposure evidences a certain lag in the gain of weight. The difference between this and the control group is statistically significant beginning with the third week, and only toward the end of the experiment do the differences minimize which apparently is due to adaptation.

Arterial pressure was determined on the rat tails by means of a plethysmograph. Background levels for blood pressure were obtained for a period of one month prior to the experiments, and were quite similar for each group with an average value of 81.7 - 89 mm Hg.

The results of determinations of arterial pressure over the four month period of irradiation are given in Figure 2.

The results presented in Figure 2 show that the curve representing changes in the vascular tonus of rats undergoing combined exposure, differ qualitatively from the other curves. While exposure to SHF or heat alone elicited a hypotensive effect, the changes observed on combined exposure were biphasic in The first phase occurred between the second to the nature. fifth week and consisted of increased arterial pressure. This was followed by conditional normalization which on the 13th week of irradiation, was replaced by a hypotensive effect. How can one explain this qualitatively different character of change of arterial pressure of animals undergoing combined exposure? Which type of effect reflects a more pronounced change? 0nlv an indirect answer can be provided to these questions.

Z.V. Gordon [1969] in her studies on the effects of various SHF bands on arterial pressure found, as a rule, that hypertension was evoked by high intensities of irradiation, while low intensities were frequently characterized by a hypotensive effect. These data indicate that combinations constitute a more powerful factor than when either of these factors was employed alone. SHF intensities that by themselves do not induce elevation of arterial pressure, in combination with heat, influence the vascular tonus to the same extent as obtained with high intensity SHF.

Conclusions regarding the amplification of the biological effect by a combination of SHF and heat, were also supported by functional studies on the state of the central nervous system of animals. Below we shall present results of studies on the conditioned reflex activity of mice employing the maze method, and on the bioelectrical activity of rabbit brain.

A conditioned avoidance reflex was developed in mice employing a T-shaped maze. This required that mice placed in the initial portion of the maze would run to a safe part of the maze before the extinction of a 5 sec electrical stimulus. The reflex was considered to be established when the animal made ten error-free runs in succession. After two stages of testing in which the strength of the stimulus was evaluated, the animals were exposed to a month and a half of irradiation. During that time the state of the conditioned reflex was tested four times. Determinations were made on the number of reinforcements required to maintain this reflex, and the reflex The resultant data demonstrated that the group with time. combined exposure, and a background of irradiation, required a greater number of reinforcements to achieve 10 correct responses than did the remaining groups. The data in Table 4 demonstrate that the group of animals with combined exposure

differed to a significant degree both from the control group as well as from the groups that were exposed to either of these physical factors alone.

EEG studies were performed on rabbits. The same animals were used to determine the PEG response to SHF or heat alone, and to the combination. Biocurrents were obtained from the occipital and the sensory motor areas of the brain. The recordings were conducted with a Hungarian encephalograph MB 5202/A. In addition to biocurrent recordings before and after the exposure studies were conducted on the response to sound and light as well as on the adaptation reaction to light flashes of different frequencies.

Analysis of the resultant data showed that in most cases combination of SHF and high air temperatures resulted in more pronounced effects than when either factor was employed alone. This indicates that synergism prevails between the effects of SHF and temperature used in combination at these intensities.

In conclusion, a better appreciation of the interaction of these factors, and resolutions of the problems pertaining to the setting of hygienic standards, will require more detailed evaluation of the biological effects evoked by the combination of heat and SHF intensities which in themselves do not have thermal properties.

# Table 1. Survival of Animals

Вид жи- вотного (1)	Воздействие (2)	Кол-во животных (3)	Погибло в течение 2 ч наблюде- (14) <sup>8</sup> )	Средняя про- должитель- ность жизни,
(6)(	(7 §0 мВт/см <sup>2</sup> при 40°С	18	18	28,3±1,8
нп	(8)0 мВт/см <sup>2</sup> при 22°С	18	14	63,3±7,6
M u	(9) <sup>с</sup> <sub>мВт/см<sup>2</sup> при 14°С</sub>	18	9	106,5±4,5
	( <b>150)</b> действие тепла 40° С	12	2	113,0±5,4
(11)	(1260) мВт/см <sup>2</sup>	12	12	4 <b>3,3±</b> 3,2
· · · · ·	13) мВт/см <sup>2</sup> при 20°С	12	3	111,5±4,7
К	(14) здействие тепла 38-40° С	12	12	105±6,3

- 1. Species
- 2. Effect
- Number of animals
   Expired during the 2 hr of observation
   Mean survival, minutes
   Mice

- 7. 80 mW/sq cm at  $40^{\circ}$ C 8. 80 mW/sq cm at  $22^{\circ}$ C 9. 80 mW/sq cm at  $14^{\circ}$ C 10. Heat effect,  $40^{\circ}$ C

- 11. Rats 12. 60 mW/sq cm at  $40^{\circ}$ C 13. 60 mW/sq cm at  $20^{\circ}$ C 14. Heat effect,  $38-40^{\circ}$ C

# Table 2. Temperature Response of White Rats

	(1) Воздействие	Экспо- зиция,	Температурный прирост, °С
(4) (5) (6) (7)	Комбинированное (60 мВт/см <sup>2</sup> +40°С) 60 мВт/см <sup>2</sup> 40°С Контроль	15	$1,5\pm0,15$ $1,9\pm0,22$ $0.44\pm0,1$ $0.25\pm0,09$
(4) (5) (6) (7)	Комбинированное (60 мВт/см <sup>4</sup> +40°С) 60 мВт/см <sup>2</sup> 40°С Контроль	22	$3,5\pm0,14$ $2,8\pm0,3$ $0,86\pm0,15$ $0,23\pm0,14$

Key:

- 1. Irradiation Invariation
   Time of Exposure
   Temperature gain, <sup>o</sup>C
   Combined (60 mW/sq cm + 40<sup>o</sup>C)
   60 mW/sq cm
   40<sup>o</sup>C
   200 c
- 7. Control

# Table 3. Physical Endurance of White Mice

	Возлействие	(1)	Кол-во жнвотных	Средняя продол- жительность пла- вания
4) 5) 6) 7)	Комбинарованно СВЧ Тспло Контроль	e .	12 12 12 12 12	$(-)7$ $119.6 \pm 11.5$ $139.6 \pm 15.8$ $142.1 \pm 11.3$ $153.2 \pm 6.8$

- 1. Irradiation
- 2. Number of animals
- 3. Mean duration of swimming
- 4. Combined
- 5. SHF
- 6. Heat
- 7. Control

## Table 4. Results of Conditioned Reflex Studies on Mice Employing the Maze Method

#### Таблица 4

Результаты исследования условнорефлекторной деятельности мышей по лабиринтной методике

		- (?	1		
	Возлействие	Количество полкреплений, не обходимое для достижения критерия упроченности			
	(1)	Φon ( )	Облучение		
5) 6) 7) 8)	Комбинированное . СВЧ Тепло Контроль	$ \begin{array}{c} ( ) \\ 8 \pm 1,52 \\ 7 \pm 1,32 \\ 7 \pm 1,56 \\ 6 \pm 1,21 \\ \end{array} $	(4+) 7,1±1,0 4,2±0,8 3,0±0,45 2,6±0,38		

- 1. Exposure
- 2. Number of Reinforcements Required for Permanence Criterium
- 3. Backround
- 4. Irradiation
- 5. Combined 6. SHF
- 7. Heat
- 8. Control


Figure 1. Weight Dynamics of Animals

Key:

- 1. Weight gain, gm 2. Control 3. Heat 4. SHF

- SHF + heat Irradiation, weeks 5.



Figure 2. Changes in the Arterial Pressure Under Different Factors.

Key:

- Arterial pressure, mm Hg
   Control

  - 3. 4. SHF
  - SHF + heat

  - 5. Heat 6. Backround
  - Irradiation, weeks 7.

#### UDC 614.87:537.868.029.64

RESULTS OF EXPERIMENTAL STUDIES ON ELECTROMAGNETIC IRRADIATION WITH LOW INTENSITY USE, SW, AND MW

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 115-118

[Article by P.P. Fukalova, M.S. Bychkova, M.S. Tolgskaya, I.A. Kitsovskaya, A.P. Volkova, and N.K. Demokidova]

[Text] Threshold values have been established for the biological effects of ultrashort waves (USW), short waves (SW), and medium waves (NW), in laboratory experiments on the basis of an analysis of the quantitative relationship between the effects and the incident energy. The results of this investigation were used in setting hygienic standards.

Previous investigations employing USW bands (69.7, 155, and 191 MHz) at threshold (nonthermogenic) intensities corresponding to 150, 50, and 20 V/m and with SW (14.88 MHz) at an intensity of 20-50 V/m demonstrated changes in individual organs and systems [1966, 1968]. We did not regard these changes as significant since they were not marked and were reversible in nature; most of them appeared after prolonged irradiation.

The morphological changes were dominated by fine changes in nervous structures (receptors, brain cells) and proliferative changes in various organs.

K.V. Nikonova and M.S. Tolskaya [1964] conducted studies with SW bands (500 kHz) at intensities of 1,800 V/m and noted a small hypotensive effect and moderate morphological changes.

The object of the present study was to study the biological effects of USW, SW, and MW at intensities which were significantly below the threshold for nonthermogenic effects (on an order and greater). Studies with USW were conducted at frequencies of 69.7, 155, and 191 MHz with different durations of irradiation (four to nine months for each band) which differed in the intensity and duration of each session of irradiation. The experimental animals were irradiated with the 69.7 MHz band at intensities of 2.5-12 V/m for 1-4 hours in chronic experiments (white mice and rats), and for 30 minutes in acute experiments (rabbits).

Studies were conducted on the effects of these USW bands on the weight, physical endurance, blood pressure, vascular permeability, EEG, the threshold of neuromuscular excitability, and immunobiological reactivity; morphological studies were also performed.

In both cases we encountered either a complete absence of any type of an effect (weight dynamics, threshold of neuromuscular excitability, and vascular permeability), short-term changes, or a tendency toward a change in the functional state (endurance, pressure, immunobiological reactivity, and morphological changes in the internal organs).

Studies of blood pressure at intensities of 10-12 V/m evidenced moderate but statistically significant change in the form of a short-term increase and subsequent decrease in pressure.

Studies on the immunobiological reactivity showed a decrease in the phagocytic function of neutrophils after ten months of irradiation without any changes in the state of natural immunity (these data are presented in greater detail in the appropriate article of this compendium).

Irradiation with every USW band at an intensity of 2.5 V/m for four months did not show any changes in weight dynamics, physical endurance, threshold of the neuromuscular excitability, blood pressure, and so forth, when done on a long-term basis.

EEG studies were conducted on rabbits in acute experiments (30 minute period of irradiation) to determine the effects of 69.7 and 191 MHz USW at intensities of 12 and 5 V/m.

In the majority of rabbits, both bands at an intensity of 12 V/m elicited deactivation which was expressed in the form of slow synchronous fluctuations, a slowing of the activity, and ar increase in the number of spindles; less frequently this consisted of total inhibition of electrical activity.

There were no significant changes in the bioelectrical activity of the brain at an intensity of 5 V/m.

Intensities of 2.5 and 5 V/m did not elicit any morphological changes. At intensities of 10-12 V/m, insignificant changes were noted in the internal organs in comparison with controls; no changes were evident in the nervous system or the vessels.

Therefore, data obtained in the investigations on USW bands (69.7, 155, and 191 MHz) at intensities of 10-12 V/m, demonstrated insignificant short-term changes in blood pressure, immunobiological reactivity, EEG, and morphology; similar changes were not encountered when the intensities were less than 10 V/m (i.e., 5 or 2.5 V/m).

Studies on the biological effects of SW (14.88 MHz) and SW (500 kHz) bands were conducted with intensities of 70 and 600 V/m. respectively, for four hours per day over a period of eight to nine months.

Studies on the effects of SW and MW dealt with weight dynamics, physical endurance, blood pressure, vascular permeability, EEG, the threshold of neuromuscular excitability, studies on the hypophysis-adrenal cortex system, certain aspects of protein and electrolyte metabolism, immunobiological reactivity, and morphological factors.

Studies on the effects of SW over a period of eight months did not show any changes in physical endurance, blood pressure, vascular permeability, or the threshold of neuromuscular excitability. However, the irradiated animals (rats) showed a lag in the gain of weight after 1, 2, 7 and 8 months (the difference in comparison with controls was statistically significant).

Studies on the hypophysis-adrenal cortex system showed a significant decrease in the activity of true erythrocyte cholinesterase and the number of eosinophils after one and two months of irradiation without any further changes later.

A decrease in the activity of specific cholinesterase may indicate changes in the functional state of the diencephalic region. The decrease in eosinophils indicates patency of the glucocorticoid function of the adrenals. No changes apparently take place in the function of the adrenal medulla since the data indicate absence of any changes in the epinephrine and norepinephrine levels in the adrenal glands.

An increase in total nitrogen and chlorides in the urine was evident along with an increase in diuresis at the end of the period of irradiation which may be related to certain changes in the functional state of the adrenal cortex. This is supported by a decrease in weight of the adrenal glands at this time (see the appropriate article in this compendium).

Immunobiological investigations revealed phasic changes (activation and inhibition) of the phagocytic and bacteriocidal function of the blood, and an insignificant depression of natural immunity.

EEG studies were conducted at intensities of 70 and 50 V/m.

At 70 V/m, rather clear-cut but moderate changes were evident which primarily consisted of activation.

At 50 V/m, either no changes were evident, or they consisted of fluctuations within the normal physiological range.

Morphological studies after eight months of irradiation showed moderate changes in the receptors of the skin and in the interoreceptors of the internal organs, a small increase in the number of mast cells in the connective tissue along blood vessels, and limited dystrophic changes in the nervous system and the internal organs.

Therefore, chronic exposure to 70 V/m SW over a period of eight months, showed either no changes in terms of some indices (physical endurance, blood pressure, vascular permeability, and the threshold of neuromuscular excitability) and in terms of other indices, moderate and in most cases, short-term changes which disappeared on subsequent irradiation (lag in weight gain, changes in the hypophysis-adrenal system, insignificant decrease in natural immunity, and moderate morpholo-Fical changes, and EEC changes).

Exposure to 600 V/m EW for eight months did not evoke any changes in the weight dynamics, blood pressure, vascular permeability, and the threshold of neuromuscular excitability.

Studies on the functional state of the hypophysis-adrenal cortex system demonstrated a limited decrease in cholinesterase after two months and a tendency toward an increase in the number of eosinophils after three months (without any subsequent changes).

Studies on the immunobiological reactivity at different periods of time showed a decrease in the phagocytic activity of neutrophils along with compensatory activation of the bacteriocidal properties of blood plasma, and a very mild change in natural immunity after ten months of irradiation. EEG studies on changes in background activity and response to sensory stimulation were conducted at intensities of 600 and 400 V/m.

With an intensity of 600 V/m a rather clear-cut bivariant shift was noted with approximately equal frequency of activation and deactivation (the difference was statistically insignificant). At 400 V/m, either weak or moderate activation was seen, and at lower intensities of the field, significant changes in EEG were not apparent.

Morphological investigations conducted after eight months of irradiation did not show any significant changes in comparison with the controls.

Therefore, MW did not evidence a biological effect in terms of most of the parameters investigated.

The results of experimental studies on electromagnetic irradiation with low intensities USW,  $\mathbb{CW}$ , and NW, supplement our understanding on the principles which govern biological effects in relation to the level of irradiation with these bands.

These data have been employed in setting hygienic standards.

# UDC 614.87:537.268.029.64

CHANGES IN CERTAIN PROTECTIVE REACTIONS OF AN ORGANISM UNDER THE INFLUENCE OF SW IN EXPERIMENTAL AND INDUSTRIAL CONDITIONS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 119-123

Article by A.P. Volkova and P.P. Fukalova

[Text] Studies have been conducted on the effects of short waves (SW) on changes in the immunobiological reactivity of experimental animals, and of people, under industrial conditions.

The present study dealt with investigations on the effects of SW on the state of natural immunity of experimental animals and of people under industrial conditions.

In laboratory experiments, one group of outbred white rats were exposed to 14.88 MHz SW with intensity of 2250 V/m (threshold nonthermogenic intensity) for one hour. A second group of animals were irradiated for four hours with an intensity of 100 V/m. A third group served as controls. The state of the natural immunity was evaluated in terms of the phagocytic activity of blood neutrophils [A.P. Volkova and V.I. Ternov, 1965] and plasma bacteriocidal properties [O.G. Alekseyeva, 1961] As the test microbe we employed a 24 hour agar culture of E. coli strain 675 as a one billion + suspension. At the end of the experiment the animals were infected with this microbial strain, and the nature of the inflammatory reaction was used to evaluate the state of natural resistance.

The resultant data are presented in Figures 1, 2 and 3 (the indices were calculated as a percentage of the control data taken as 100).

AW resulted in marked activation of the phagocytic activity of neutrophils. These changes were more pronounced when irradiation was conducted with 100 V/m intensity. The index of digestion in the second group of animals increased threefold (P < 0.01). By the second month, activation was replaced by sharp inhibition, and again the lowest values for this index were encountered when low intensity -- 100 V/m -- was employed (0.14  $\pm$  0.01 against 0.5  $\pm$  0.03 in the control group). At later stages, changes in the phagocytic activity of neutrophils depended on the intensity of radiation. Over a period of three to ten months, the animals that had been exposed to irradiation at an intensity of 2,250 V/m evidenced extremely low phagocytic activity. At the end of the experiment the phagocytic activity of this group had decreased by 56% (0.16 + 0.4 and 0.36  $\pm$  0.05 in the control); the second group demonstrated a decrease in phagocytosis by 25% (0.27  $\pm$  0.04).

Plasma bacteriocidal activity in this and in the second group, was at a low level beginning with the second month. Nevertheless, such a distinct linear relationship between the bacteriocidal activity and the intensity was not noted, as was the case in studies on phagocytosis. Only at the end of the experiment did the activity in the first group decrease by 11%, while in the second group, it was at the same level as the control value.

Evaluation of the animals a month and a half after irradiation had been terminated, showed that the phagocytic activity was at a low level. In the first and the second group, it stood at 0.3  $\pm$  0.02, while in the controls, it was 0.5  $\pm$  0.09 (0.05 < P > 0.02). It is obvious that recovery of phagocytic activity did not occur. Bacteriocidal properties of plasma in both cases did not differ from the control data.

Within nine months of the termination of the experiment, the experimental and control animals were infected with the strain of E. coli which had been used for the phagocytic studies. E. coli, in an amount of 1.4-1.6 billion microbial cells in a volume of 0.1 ml physiological solution, administered beneath the aponeurosis of the foot of a posterior extremity. The degree of inflammation was evaluated in terms of a scale, and the observations were conducted for a period of two weeks. Figure 3 illustrates the course of the inflammatory process. In both the first and the second groups the nature of the reaction differed significantly from the reaction in the control animals. The course of the inflammatory reaction was more pronounced in those rats that had been exposed to more intense irradiation. Thus, in that group four days after infection, the degree of the inflammatory reaction was evaluated as  $6.6 \pm 0.5$  units, while in animals exposed to 100 V/m, it was 5.0  $\pm$  0.01, and in the control animals, it was 4.0  $\pm$ 0.3; at the end of the period of observation, the corresponding values were  $1.2 \pm 0.02$ ,  $0.5 \pm 0.001$ , and in the control,  $0.2 \pm 0.01$ .

Differences in the nature of the inflammatory response in the first and the second group, in comparison with the control group, were statistically significant (P < 0.01).

Therefore, our experiments have demonstrated that prolonged exposure over a ten month period to SW electromagnetic energy at an intensity of 2,250 V/m for one hour, or 100 V/m for four hours, leads to a change in natural immunity which is more pronounced when the higher intensity is employed.

Studies on the effects of SW on the protective forces of man were conducted under industrial conditions.

Four stations are in operation at the radio center where we conducted our investigations. At three of these stations, after the safety measures recommended by the Institute of Labor Hygiene and Occupational Diseases of the Academy of Medical Sciences USSR had been implemented, the intensity of irradiation did not exceed the maximum permissible level (up to 20 V/m). At the fourth station where the safety recommendations had not yet been implemented, the field intensities significantly exceeded the maximum permissible levels, and amounted to scores of V/m and greater.

The studies were conducted on 123 clinically healthy individuals (radio engineers and radio technicians) who serviced the radio transmitters.

All of the individuals in the study live in the same residential area and enjoy approximately the same living conditions.

In our investigation, in addition to a study of the phagocytic and bacteriocidal function of the blood, we studied the numbers and the nature of the autoflora in the mouth cavity [0.G. Alekseyeva, 1965], as well as the bacteriocidal properties of the skin [N.N. Klemperskaya and G.A. Shal'nova, 1966]. The results of these observations showed that prolonged exposure to SW electromagnetic energy elicits certain changes in the state of some of the protective factors of the organism (Figure 4). Thus, employees at the fourth station (54 men) who were exposed to intensities which exceeded the maximum permissible levels (shaded columns on Figure 4) evidenced changes in the phagocytic activity and the numbers of autoflora in the mouth cavity. The number of microbes on the mucous membrane of the mouth cavity was, on the average,

1,791  $\pm$  109, the phagocytic activity was 0.3  $\pm$  0.03, the bacteriocidal activity of the skin was 70  $\pm$  7.5, and the bacteriocidal property of the plasma was 33  $\pm$  1.7. Therefore, work with SW emitters at intensities exceeding the maximum permissible levels, leads to a sharp (two-fold) inhibition of the phagocytic activity of neutrophils, and a high (four-fold) seeding of the mucous membrane of the mouth with autoflora microbes, in comparison with normal indices [N.E. Klemperskaya and G.A. Shal'nova, 1966]. Consequently, the same trend in changes was observed as was seen with the experimental animals, namely, inhibition of certain factors of natural immunity.

Among the workers at the three stations (69 men) where the levels of irradiation did not exceed maximum permissible levels, the indices under study were virtually within the normal range; the autoflora counts were  $469 \pm 65$ , the phagocytic activity was  $0.7 \pm 0.05$ , skin bacteriocidal activity was  $73 \pm$ 5.9, and plasma bacteriocidal activity was  $27 \pm 1.2$  (unshaded columns in Figure 4). These findings indicate that it is possible to use immunobiological methods under industrial conditions in order to evaluate the effectiveness of sanitary and hygienic measures.





- Key: 1. Recovery
  - 2. Irradiation, months



- Figure 2. Changes in the Bacteriocidal Function of Blood Plasma Under the Influence of Various Intensities of the SW Band
- Keyı

1. E = 2250 V/m 2. E = 100 V/m 3. Recovery

- 4. Irradiation, months



Figure 3. Inflammatory Response in Animals Exposed to Different Intensities of the SW Band

- Key:

- 1. Scale 2. SW 2250 V/m 3. SW 100 V/m 4. Control
- 5. Observation time, days



- Figure 4. The State of Barrier Functions in Workers at Radio Transmission Centers
- Keyı
- Intendisty above maximum permissible level
   Intensity not exceeding maximum permissible level
   Skin bacteriocidal activity
   Flora of the mouth mucous membrane

  - 5. Phagocytosis
     6. Blocd plasma bacteriocidal activity

#### UDC 614.87:537.868.029.64

CERTAIN PRINCIPLES GOVERNING THE EFFECTS OF MICROWAVES ON K+ AND Na+ TRANSPORT IN HUMAN ERYTHROCYTES

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 124-131

[Article by V.M. Shtemler]

[Text] Microwaves have been observed to affect the rate at which K+ and Na+ are transported into human erythrocytes. The rate of transport was studied in relation to the parameters of exposure -- frequency, intensity, and duration -- as well as the dynamics of recovery of the rate of transport after irradiation was terminated. The possible mechanisms responsible for the observed effects have been discussed. Consideration was also given to the possible relationship between these results, and the problem of the direct effects of microwaves on excitable structures.

Numerous investigations on the biological effects of radiofrequency electromagnetic fields (EMF), have shown that the central and the peripheral nervous systems of humans and animals are highly susceptible to the effects of EMF. EMF has also been demonstrated to influence the excitability of isolated nerve cells, as well as other excitable cells [M.S. Bychkov, 1960, 1967, and articles in this compendium; A.S. Presman, 1963, 1964, 1965; Yu. I. Kamenskiy, 1964, 1968; P.G. Bogach, 1972; V.I. Mirutenko, 1972].

On the other hand, it is known that the ability of nerves and muscles to generate action potentials and to conduct excitability is due to the assymetrical distribution of K+ and Na+ between the intra- and the extracellular media. This assymetry in distribution is due to the fact that the cell membrane shows selective permeability to certain ions, as well as to active transport of ions against the gradient of the electrochemical potential for which the energy is provided by the metabolism of the cell. Consideration of the accumulated data leads to the conclusion that one of the possible mechanisms of action of EMF on excitable systems may consist of the effects of the field on the permeability of the membrane of the nerve cells, and in part, on the permeability of the potential-forming K+ and Na+, which in turn will influence their excitability.

Studies on the mechanism of action of EMF on the ionic permeability of membranes is important in elucidating the mechanism of direct action of EMF on excitable cells. Furthermore, up to the present time, this phenomenon has not been studied adequately.

In dealing with microwaves with respect to this problem, we are familiar only with the studies of E. Sh. Ismailov [1970, 1971] on human erythrocytes, who demonstrated a reversible change in the K gradient under the influence of microwaves in terms of the K+ and Na+ concentrations in the internal and external erythrocyte medium.

The purpose of the present investigation consisted of the following:

-- a study of the effects of microwaves on the rate of unidirectional flux of K+ and Na+ inside erythrocytes by means of a radioactive label,

-- a study of the relationship between the nature of this effect and frequency,

-- a study of the relationship between this effect and the intensity and duration of exposure,

-- determination of the threshold intensity for this effect, -- a study of the recovery dynamics following termination of irradiation.

An erythrocyte suspension was exposed to monitored irradiation in one of the two instruments indicated in Figure 1 and Figure 2: a wave-guide instrument (2340 MHz) or a coaxial instrument (900-1600 MHz). The suspension to be irradiated was placed inside the wave-guide or the coaxial instrument in polyfluoroethylene cuvettes as indicated in Figures 1 and 2. Both instruments made it possible to regulate and measure the average power flux density which was incident to the object, as well as the power input in the solution, and made it possible to conduct irradiation under conditions of constant temperature (37.0  $\pm$  0.2°C).

The experiments were conducted with washed human donor erythrocytes which had previously been equilibrated with Ringer's solution at  $37^{\circ}$ C, and which contained tris HCl buffer, pH 7.4, glucose, and the usual electrolytes. Determinations were conducted on unidirectional fluxes of K+ and Na+ from the external medium into the erythrocytes from the rate of intake of radioactive isotopes  $^{42}$ K, and  $^{24}$ Na or  $^{22}$ Na during a 20-30 minute incubation of the erythrocytes in the radioactive medium at  $37^{\circ}$ C.



- Figure 1. Block Diagram for Wave-Guide Instrument for Dose Irradiation of Solutions
- Key: 1. Microwave generator (Luch-58)
  - 2. Cable wave-guide adapter
  - 3. Alternating attenuator
  - 4. Measuring line
  - 5. Guiding couples
  - 6. Low power meter M4-2
  - 7. Impedance transformer
  - 8. Micromotor with mixing rod
  - 9. Polyfluoroethylene cuvette with solution
  - 10. Short-circuiting device which also serves as a water jacket for thermostating the cuvette
  - 11. Differential thermocouple
  - 12. Water ultrathermostat



Figure 2. Block Diagram for Coaxial Instrument for Dose Irradiation of Solutions

- Key: 1. Microwave generator (LMS-541)
  - 2. Ferrite fan
  - 3. Alternating attenuator
  - 4. Measuring line
  - 5. Single loop impedance transformer
  - 6. Dry air thermostat
  - 7. Coaxial object chamber
  - 8. Polyfluoroethylene cuvette with solution
  - 9. Low power meter
  - 10. Differential thermocouple

# Table 1. The Effects of the Frequency of Microwave Radiation on Changes in the Rate of K+ and Na+ Transport

	· ·	(1)	10	-/			(r) Na+		
		(1) Чзсто- та, МГц	(2) Чис- ло оды- 10В	()) Поток К+ внутрь мэкв К+ литр эритро- цитов-час М±т	(4) Изменение, % к конт- ролю	t	Поток Na+ внутрь мэкв Na+ литр эритро- цитов. час М±т	(4) Изменение, <sup>95</sup> х конт- ролю	t
(6) (7)	Опыт Контроль	898	6 6	$1,31 \pm 0.04$ $1,52 \pm 0.02$	-14±3,0	4,7	$2,83 \pm 0,07$ $2,55 \pm 0,07$	11±3,9	2,8
(6) (7)	Олыт Контроль	1009	4 4	1,25±0,04 1,49±0,03	$-16 \pm 3,4$	4.8	$2.87 \pm 0.06$ $2.63 \pm 0.06$	9±3,2	2,8
(6) (7)	Опыт Контроль	1:81	6 6	$1,29 \pm 0,05$ $1,55 \pm 0,05$	-17 <del>±</del> 4,6	2,7	$2.70 \pm 0.06$ $2.50 \pm 0.05$	8±3,0	2,6
(6) (7)	Опыт Контроль	1579	5 5	$1,28 \pm 0.05$ $1,52 \pm 0.03$	-16±3,9	4,1	3,02±0,07 2,67 <b>±0,07</b>	13±3,7	3,5
(6) (7)	Олыт Контроль	2340	4 4	1,30±0,03 1,50±0,04	-13±3,3	3,9	$2,95 \pm 0.06$ $2,68 \pm 0.06$	10±3,2	3,1

(8) М — среднеарифметическое; т — среднеквадратическая ошибка среднего; t — коэффициент Стыодента.

Key:

- 1. Frequency, MHz
- 2. Number of experiments
- 3. K+ influx
  (meq K+/liter erythrocytes hr)
  M + m
- 4. Change, % of control
- 5. Na+ influx (meq Na+/liter erythrocytes.hr) M + m
- 6. Experiment
- 7. Control
- 8. M--arithmetical mean; m--standard error of mean; t--Student's coefficient

Depending on the purposes of the investigation, the influx of K+ and Na+ was determined either directly during the time of irradiation, or after irradiation had been terminated (after effect). In order to compare the results obtained under different conditions, we expressed the absolute influx of ions as meq K+ (Na+)/ liter erythrocytes.hour. All experiments were conducted in parallel with controls under conditions which were identical, with exception of exposure to microwaves.

The results of these experiments showed significant changes in the rate of K+ and Na+ transport into the erythrocytes under the influence of irradiation with microwave EMF in relation to the appropriate thermal control. The nature and the degree of change depended on the conditions of the experiment.

The Relationship Between Ion Transport and EMF Frequency

Five frequencies in the microwave band (898, 1009, 1181, 1579 and 2340 MHz) were randomly selected to test the effect of EMF frequencies on ion transport. In this series of experiments we determined the influx rates of K+ and Na+ immediately after irradiation was terminated. The experimental conditions were as follows: 50 mW/cu cm incident power density, 30 min irradiation,  $37^{\circ}$ C, and 5 x 10<sup>6</sup> cells/mm erythrocyte concentration.

The results of the studies on the effects of frequency on K+ and Na+ transport are presented in Table 1. Following microwave irradiation, in each case the influx of K decreased by approximately 15% and the influx of Na ions increased by approximately 10% with respect to the controls. These changes in the rates of K+ and Na+ transport should lead to a decrease in the K-Na gradient between the internal and the external medium of the erythrocytes. On the other hand, it is known that the influx of K+ into the erythrocytes proceeds against the concentration gradient and is an active process [J. Funder et al., 1967], i.e. it is almost completely depressed by inhibitors of glycolysis, and in part, by ouabain . Influx of K+ into the erythrocytes is passive along the concentration gradient. Thus, EMF effects both active and passive components of ion transport. However, within the limits of experimental accuracy, this effect does not significantly depend on the frequency of radiation.

The Effects of Intensity and Duration of Irradiation on Changes in Transport Rates

The next series of experiments was concerned with the relationship between the rate of K+ influx and the intensity (1-100 mW/ cu cm) and duration (20-120 min) of exposure to a 2340 MHz field. Determination of the transport rates was conducted during irradiation at 37°C with a thermostatic control to compensate for the heating of the erythrocyte suspension due to heat production of



Figure 3. Dependence of Changes in the Rate of K+ Influx Into Erythrocytes on the Intensity and Duration of Irradiation

EMF. The relationship between influx and the intensity and duration of irradiation is rather complex in nature.

The results of these series of experiments are presented in Figure 3. Six curves are presented in Figure 3 on the relationship between the change of potassium ion influx and the duration of irradiation at fixed incident power densities (1, 10, 30, 50, 80 and 100 mW/cu cm). The ordinate represents the effect as a percent of the thermal control. The results are represented by points with vertical lines which reflect the standard deviation of the mean. Each point represents the average value of 4 to 5 experiments.

At an intensity of 1 mW/cu cm a small change in the rate of K+ transport which is close to being significant, occurs only after 120 min exposure. A significant change in the rate of transport appears at intensities of 10 mW/cu cm and higher. It is evident, therefore, that the threshold intensity for this effect lies in the region of 1-10 mW/cu cm.

It is evident from Figure 3 that the rate of transport fluctuates with time and, furthermore, that at intensities below 80 mW/cu cm not only does the value change, but also its sign . For all intensities, beginning with 10 mW/cu cm, initially a decrease in K+ influx occurs which reaches a maximum low value (approximately 20%) in about 20-30 min after initiation of irradiation, and is then followed by a recovery phase with a change in sign to a positive value, and thereafter, a negative phase comes into play again.

For convenience of analyses of this effect in terms of the parameters of exposure, we attempted to approximate the experimental data with analytical expression in the form of a function with two variables (intensity and duration). A satisfactory approximation was reached with the following function:  $\Phi(I,t) =$ 

-0,0013  $\cdot$  t(1+20) - (23-0,14  $\cdot$  t) sine  $\frac{2\pi t}{66+0,951}$ , where I is the intensity of irradiation (incident power density), and t represents the duration.

The coefficients for this function were calculated by means of a BESM-4 computer on the basis of experimental data by the method of least squares [Yu. V. Kemnits, 1964].

The graphic profiles of this function at fixed intensities are indicated in Figure 3 by solid lines. It is evident that at intensities ranging from 10-100 mW/cu cm, and 20-120 min exposure, this function represents in an adequate manner the experimental results.

Consideration of the analytical expression for the relationship between the change in the rate of transport and time at fixed intensities shows that this relationship may be interpreted as the sum of two different processes: a linear decrease in the rate of transport under the influence of irradiation with time [the term 0.0013  $\cdot$  (1+20)] and autoregulation of this parameter in response to the effects of irradiation in the regime of overregulation with damping amplitude [the term (23-0.14  $\cdot$ t) sine  $\frac{2\pi t}{66+0.95 \cdot 1}$ ]. In Figure 4, the two component functions are indicated by the dashed line, and the solid line represents the sum function at fixed intensities. The linear component of the change in the transport is indicated by the dashed line also in Figure 3 for the corresponding intensity values. The effects of intensity on the transport, is also reflected in the increase in the slope of the linear component, as well as the increase in the period of autoregulation.



Figure 4. Graphic representation of  $\phi$  (I,t) as a sum of two components

Unfortunately, at the present time we do not possess concrete chemical data on the nature of these processes which bear responsibility for these two types of changes in the rate of transport. That problem requires special investigations, which, however, were not a part of the present study. Apparently, the two components of the transport changes are related to the effects of microwaves on two different systems of the cell which regulate membrane transport.

We may assume that the changes in the first system which are linearly related to the absorbed energy dose -- this is obvious from the expression for the linear component which contains the product I.t that is equal to the absorbed dose -- appear to be the results of microthermal effects. The microthermal effect may be related to selected absorption of electromagnetic energy by "ice-like" membrane of bound water which hydrates the surface of the erythrocyte membrane. This process should lead to a change in the phase state of the membrane water -- melting of the ice-like membrane -- as a result of which we may expect significant changes in the transport properties of the membrane. The possibility of selective absorption of EMF energy in heterogenous systems at the boundary of phases with sharply different conductivities and dielectric constants has been suggested on numerous occasions in the literature, and has received experimental confirmation on model systems [Esaux, 1933; V.T. Tomberg, 1961; P.O. Vogelhut, 1969]. The physical basis for the absorption of microwave energy by the layer of bound water at the surface of the membrane could either be due to relaxation of the molecules in the hydration layer -- the characteristic frequencies should lie in the region between the characteristic frequencies for pure ice ( $\sim 10^4$  Hz) and of liquid water ( $\sim 10^{10}$  Hz) -or to increased ionic conductivity in the ionic atmosphere (double electrical layer) which arises around fixed charges on the surface of the membrane. On the basis of this, it becomes comprehensible why there was no clear-cut relationship between the effect which we encountered and the frequency of the microwaves.

It is obvious that changes in the second system do not depend on the dose. This is apparently a relatively complex biochemical system which is responsible for protecting the cell against damaging influences. A living cell (including erythrocytes) possesses a definite hierarchy of regulatory mechanisms which maintain cellular homeostasis, i.e. the maintenance of various variables -- primarily the composition -- within certain limits. Under the influence of injurious factors, the cell is capable of mobilizing its chemical resources at a given moment for correcting the changes in the ionic balance of the cell.

Variations in the rate of transport are apparently due to a kinetic mechanism which itself is determined by the rate constant of individual stages in the multistage process in the biochemical system -- which controls the transport -- in the presence of positive and negative feedbacks. As an example of such a complex biochemical system in the erythrocytes, we may consider the enzyme system on the membrane which are responsible for ionic transport (membrane ATPase and others) as well as the closely related system of anaerobic (glycolysis) and aerobic (pentose cycle) oxidations which provide energy for the transport processes [I. Rongust, 1969; R. Whittam, 1964; N.M. Mirsalikhova, 1970].

The Dynamics of Transport Rate Restitution Following Termination of Irradiation

Typical restitution dynamics following termination of irradiation are given in Figure 5. Both curves were obtained under the following conditions of irradiation: N = 2340 MHz and 50 mW/cu cm intensity. The upper curve corresponds to 30 min exposure, and the lower curve to 100 min exposure. As is evident from the upper curve, after 30 min exposure to microwaves, restitution of the initial rate of K+ transport occurs which also prevails as a result of autoregulation of the transport under conditions of over-regulation with amplitude attenuation. Complete recovery takes place in about 90 min. Longer exposure at this same



Figure 5. Change Dynamics in the K+ Influx Rate Following Termination of Irradiation

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intensity (lower curve) evoked disruption in autoregulation and recovery did not occur throughout the course of this experiment.

Apparently changes in the rate of transport are due to partial destruction of the erythrocyte membrance which leads to its decreased resistance. Relatively weak and short term exposures evoked reversible changes in the membrane structure, while more powerful and prolonged periods of exposure elicited irreversible damage.

As was noted at the beginning of the article, one of the current problems in biophysics consists of studying the primary mechanisms of action of EMF by which this physical factor acts directly on excitability of neurons and other excitable calls. Consequently, it is of interest to review the connections between the results of the present study and this problem.

Although the erythrocytes are not excitable cells, they possess a plasma membrane, the structure and properties of which are essentially identical with the properties of the membranes of excitable cells. Erythrocytes as a result of their relative simplicity in terms of structure and convenience, provide an excellent biological model for investigating ion transport across membranes.

The excitability of a cell is known to depend on the size of the membrane potential (MP). A decrease, or an increase in MP, leads to an increase or a decrease in the excitability of a cell, res-Dectively. According to the membrane theory, the size of MP is determined by the stationary non-equilibrium process of ion defusion across the membrane, as well as by active transport against the concentration gradient. Any change in passive or active rates of transport leads to a disturbance in the overall relationship, and alters the total ionic flux across the membrane and, therefore, changes its polarization (i.e., MP). The depolarizing effects of microwaves have been detected in a number of investigations conducted on isolated excitable preparations [M.S. Bychkov, and Z. Ye. Moreva, 1960; A.S. Presman, 1963; A.S. Presman and S.M. Rappoport, 1965; S.M. Zubkova, 1967; P.G. Bogach, and V.I. Mirutenko, 1972; V.I. Mirutenko and P.G. Bogach, These studies have shown that changes in polarization of 1972 cellular membranes and MP due to irradiation, occur very rapidly within a time interval during which no significant changes in the intra- and extracellular electrolyte concentration could take place. This indicates that, in any case, the primary response to irradiation is due to changes in the rate of transport, and not to a change in the ionic concentration. It is also of interest that in practically all studies dealing with the effects of microwaves on the excitability of isolated cells during prolonged irradiation, as well as after its termination, changes in excitability were marked by fluctuation [M.S. Bychkov,

and Z. Ye. Moreva, 1960; S.M. Zubkova, 1967; S.M. Zubkova, and V.A. Nikonova, 1969, 1970; P.G. Bogach, and V.I. Mirutenko, 1972; V.I. Mirutenko, and P.G. Bogach, 1972]. The periods of these fluctuations correspond in terms of the order of magnitude (scores of minutes) with the periods of fluctuations in the rate of K+ transport across the erythrocyte membrane. It is possible that the variability in the changes of excitability and cellular MP is due directly to changes in the rate of transmembrane transport of ions responsible for potential formation which arise as a result of autoregulatory mechanisms in the cell.

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# UDC 614.87:537.868.029.64

#### THE EFFECTS OF MICROWAVES ON ACTOMYOSIN ATPase ACTIVITY

## Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 132-136

## Article by V.M. Shtemler

[Text] In vitro experiments have demonstrated a significant decrease in ATPase activity of actomyosin of rabbit muscle under the influence of microwave irradiation. Below we shall consider the possible mechanisms responsible for this.

The investigations of a number of authors [Y.A. Engelhard, 1957; Szent-Gyorgyi, 1951] have demonstrated that the structure of actomyosin which consists of fibrillar actin and myosin, is highly labile and may undergo confirmational changes as a result of small changes in the ionic strength or pH, and that various conformational states of actomyosin correspond to different levels of ATPase activity [S.E. Shnol' et al., 1957, 1959, 1961, 1964], and that furthermore, under certain conditions (ionic strength, pH, dilution, and so forth), conformational transformations may occur spontaneously. The present study dealt with the effects of microwaves on the structure of actomyosin and its ATPase activity under conditions of in vitro experiments.

#### Methods

The experiments were conducted with freshly prepared preparations of actomyosin from rabbit muscles in 0.6 M KCl and 0.03 M MgCl2 solutions. ATPase activity was determined from the rate of hydrolysis of ATP during incubation with actomyosin at 23°C, pH 8.0, and 3 mg/ml ATP concentration. The incubation time ranged from 10 to 40 min depending on the protein dilution. Hydrolysis was terminated by the addition of trichloroacetic acid which denatured and precipitated the protein, after which the mixture was filtered via a paper filter, and the amount of inorganic phosphate obtained from the ATP was determined in the filtrate spectrophotometrically from the absorption of a phosphorus-molybdenum complex at  $\lambda = 390$  nm [M.N. Kondrashova et al., 1965].

Two types of experiments were conducted; in the first variant the ATP was incubated with actomyosin which had already been irradiated, and in the second variant, irradiation was conducted during the incubation of the enzyme-substrate mixture. The solutions of actomyosin and the actomyosin-ATP incubation mixture, were exposed to graded doses of irradiation in a coaxial apparatus (V.M. Shtemler, subsequent volume). Most of the experiments were conducted with a frequency of 350 MHz. The structure of the field in the coaxial line corresponded to a transverse electromagnetic wave of TEM [transliteration] type with a variable gradient of the electric field along the radius. The potential of the E field in the internal conductor was about 18 V/cm, and in the external conductor about 5 V/cm; the mean power flux density was about 250 mW/sq cm. At a frequency of  $\sqrt{2}$  = 350 MHz, only a small fraction of the incident energy (3-5%) was absorbed by the irradiated solution. Specific power absorption in all experiments was maintained at  $5 \pm mW/cu$  cm. The duration of exposure was 20 min. The temperature of irradiated actomyosin solutions increased by 0.2 to 0.3°C. By means of a thermostat, the temperature was maintained at a constant level  $(23 + 0.2^{\circ}C)$ during the entire period of incubation, as well as during the time of irradiation or after irradiation.

Results and Discussion

A series of experiments were conducted on the determination of ATPase activity of actomyosin that had been irradiated. Characteristic results obtained with three different protein preparations are presented in Table 1. It became evident that the susceptibility of actomyosin to irradiation rested on a number of factors. Thus, actomyosin preparations obtained at different times differed significantly in their properties -- activity, homogeneity, and susceptibility to irradiation. Apparently, dertain uncontrolled factors play a role inthe method used to obtain the protein. Furthermore, as is evident from data in Table 1, in each case the field exerted an effect only on the fresh preparations. After two hours of storage from the time of its preparation, the sensitivity of the protein to the field decreased. Unfortunately, it is not clear which properties of the protein are responsible for its loss of susceptibility to irradiation. In certain cases, simple mechanical treatment of the protein in a manual homogenizer, completely abolished its sensitivity to EMF (Experiment 44). A significant decrease in ATPase activity was noted in 14 out of 21 experiments (66% reproducibility); furthermore, the decrease in enzyme activity varied from -4 to -40% in different experiments, despite identical conditions of irradiation. The wide disparity in the observed changes in activity is apparently due to the high ability of actomyosin. It is possible in addition, that the effect of EMF on ATPase activity of actomyosin is also dependent on the ionic strength of the protein solution and the dilution.

However, in our experiments, these factors did not seem to evidence any influence.

Although our experimental conditions were similar to those which according to S.E. Shnol' et al., are responsible for its continuous fluctuations of actomyosin ATPase in solution, spontaneity alone cannot be used to explain the fluctuations observed by us in view of their uniformity (always a decrease in enzyme activity), since in the case of spontaneity, both an increase and a decrease would have been expected.

In a second series of 12 experiments in which the same parameters of irradiation were employed, i.e., irradiation of the enzymesubstrate mixture of actomyosin-ATP, changes in ATPase activity in comparison with the control were not noted which apparently was due to the fact that ATP was bound to the active center of the actomyosin molecule and thereby preserved its specific confirmation [S.E. Shnol', 1965]. Consequently, one may assume that the effects of EMF are due to its influence on the active center of ATPase activity in the actomyosin molecule.

On the basis of these data it appears that irradiation with EMF of a frequency  $\neq$  = 350 MHz, is capable of influencing the structure of actomyosin and its ATPase activity under certain conditions.

The present study was not concerned with a concrete molecular mechanism responsible for the changes in ATPase activity since that would require specialized in-depth studies; this is further complicated by the fact that the high lability of actomyosin solutions, and rather poor reproducibility of the results, make it difficult to provide a quantitative analysis of the relationship between the noted effects and the parameters of irradiation.

However, we may suggest that a nonresonant mechanism is responsible for the changes in ATPase activity since the selection of  $\gamma = 350$  MHz employed in our experiments, was largely random in nature. In addition, in vitro irradiation of myosin, one of the proteins of the actomyosin complex at a frequency of  $\gamma = 2400$  MHz [V.S. Ulashik et al., 1970] also resulted in a change in ATPase activity and in the number of titratable SHF groups.

This cannot be explained on the basis of an integral increase in the temperature of the solution since the experiments were conducted with a heat control and the temperature was maintained constant with an accuracy of  $\pm$  0.2°C. Even in the absence of a thermostatic control, the maximum increase in the temperature of the solution over a 20 min period does not exceed about 1.5°C at a specific power absorption of 5 mW/cu cm. A temperature increase of 1.5°C does not evoke a change in ATPase activity.

One of the possible explanations for this phenomenon may come from the concept of S.E. Shnol' [1965, 1967] on the nature of variation in ATPase activity in actomyosin solutions, and the

involvement of intermolecular water in this process. According to this concept. changes in ATPase activity are due to local confirmational changes of the hydrophobic regions of the nonhelical portion of the molecule at the active center. Here an important role in the confirmational transformations of actomyosin (and in general, proteins) is assigned to intermolecular water which is in the immediate vicinity of the surface of the molecule. Around apolar groups of the protein molecule, water has a tendency to form "hydrophobic" structure, and around polar groups, water exists in a "hydrophilic" structure. Any change in the configuration of a protein molecule which results in a change in the relationship of the polar and the apolar groups on its surface, could be accompanied by a change in the relationship and spatial distribution of hydrophobic and hydrophilic water structures. It is also assumed that there is mutual interaction between water structure and the molecule. In the light of current data on the importance of water in living tissue, there is no doubt as to its structure-forming role with respect to the biopolymer macromolecules [M.N. Vol'kenshteyn, 1965; V.B. Ptitsyn, 1967: N.A. Askochenskaya et al., 1972]. Consequently, the confirmational state of a macromolecule may not only determine confirmational structure of water, but may be a consequence of the latter.

In the light of this conception, it would appear that the effects of radiation on actomyosin structure is due to the effects of radiation on the state of water in the actomyosin gel. Actually, water in the actomyosin solution may exist in various confirmational states (phases). Despite the energetic similarity of these phases, transformation from state to another may proceed slowly due to the presence of an activation energy barrier. In this sense, water actually exists in a nonequilibrium state, i.e., as a rule, it appears to be either a "over-cooled" or "over-heated" liquid. Any factors which "catalyze" structural alterations in water, may induce corresponding confirmational changes in protein macromolecules. It is possible that EMF is such a "catalyzing" agent.

Изменение АТФазной активности актомиозина под влилнием облучения									
	(1) Препарат бел. (2) ка. Срок хра- цения до нача. No спытов		Условия и <b>(13</b> аления онытов $\left(350 \text{ МГи}, 5\frac{\text{MBT}}{\text{см}^3}, 20 \text{ мия}\right)$ Ношая си-			(4) АТФазная актив- ность (з условных еданицах) и сред- всява гратичиая	(5) % изменения отно- сительно контро- ля. Коэффициент Стьюдента г.		
	ла опыта		ла (КСІ), моль	Разведе- ние	раллель- ных проб	ошнбка среднего М≘т	Достоверность различий р		
(12)	II Один день.	<b>(9)</b> Сблучение № 4	(6) <sub>0,09</sub>	<b>(7)</b> 1:5	(§)	0,272 <u>:</u> ±0,021	$(10)_{t=1,2}$		
		Контроль	<u> </u>	<u> </u>	5	0,244±0,009	Не достоверно		
(12)	II Один день	Облучение № 5	0,19	1:5	5	$0,179 \pm 0,006$	$(10)_{t=0,2}$		
		Контроль			5	0,181±0,006	не достоверно		
(12)	II Один день	Облучение № 6 Контроль	0,47	1:5	5	0,171±0,006	$(10)_{t=1,5}$		
					5	0,159±0,005	Не достоверно		
(13)	II Два дня	Облучение № 7 Контроль	0,09	1:5	5	0,537±0,010	(10)=0,85		
					5	0,585±0,047	Не достоверно		
(13)	II Два дия	Облучение № 8	0,19	1:5	5	0,481±0,067	$(10)_{t=0,5}$		
		Контроль			5	0,530±0, <b>0</b> 50	Не достоверно		
(13)	Ц Двадня	Облучение № 9 Контроль	0,47	1 : 5	5	0,444±0,056	(10) <i>t=</i> 0,8		
					5	0,489±0,002	Не достоверно		
(12)	IV Один день	Облучение № 17 Контроль	<b>0</b> ,05	1:4	5	0,125±0,002	-4.5% t=2.95		
					5	0,131±0,001	p>95%		
(12)	IV Один денъ	Облучение № 18 Контроль	0,20	(11) не раз- веден- ный	5	0,170±0,009	$(10)_{t=0,0}$		
(					5	0,170±0,008	Не достоверно		
(12)	IV Один день	Облучение	0.45		5	0,126±0,0013	-6,6%		
(12		Контроль	•		5	0,135±0,0025	p>95%		
(1)	IV Лна лня	Облучение № 20	0,45		10	0,289 <b>±0,0025</b>	11.6%		
(13)	дла дня	Контроль			10	0,327±0,0025	p>99,3%		
(13)	IV ) Два дня	Облучение № 21 Коптроль	<b>0</b> ,10	1:1	5	0,216±0,0015	-4.0%		
					5	$0,225\pm0,002$	p>99%		
(13)	IV ) <sup>Два</sup> дня	Облучение № 22 Контроль	0,25	16 11).	5	0,424±0,004	-4,5%		
				រវាណី	5	0,444±0,006	p>95%		
(14)	1V Три дня	Облучение № 23 Контроли	0,50	не раз- веден-	5	$0.372 \pm 0.004$	$(10)^{=1,0}$		
. 1	34			(11)		FUUID	пе достоверно		

			(3)		(4)	Продолжение (Б)	
(1) Препарат бел- ка. Срок хра- сения до нача-	(2) Na obsitob	Условая (550 МГ Иошая си-	проведени и, 5 <sup>м91</sup> , 2 ем <sup>1</sup>	ч оцытев 20 мня ) Число па-	АТФарная актичность (в условных слиницах) и сред- неквадратичная опнибка среднего	46 изменения отно- сительно-контро- ля. Коэфанциент Стьюдента I. Достоверность	
	- (0)		HHE	иых проб	M ± m	различий р	
1V Три дня	Облучсние № 24	0,70	(0)	(0) -	0,281±0,005	<i>t</i> =-1,8	(10)
(14)	Контроль			5	$0,304 \pm 0.011$	Не достоверно	
IV Истыре дня	Облучение № 25	1,00		5	0,203±0,007	t=0,3	
(15)	Контроль			5	$0,206 \pm 0,006$	Не достоверно	(10)
	Облучение	0,50		7	0,310±0,004	1=0.2	
(15)	Контроль			7	0,311±0.003	Не достоверно	(10)
VII	нь № 35 Контроль	0,1	1:9	10	$0,406 \pm 0,008$		
(12)				9	0,634±0,014	p>99,9%	
	Облучение	0,1	1:9	12	0,423±0,012		
(12)	Контроль			12	0.570±0,009	p>99,9%	
VII	Облучение	0.1	1:9	10	0,414±0,007	-22%	
(13)	Контроль	итроль		10	0,534 <u>+</u> 0,028	p > 99%	
V11 ) Два дня	Облученис ′№ 38		(16) обрабо- тан в	10	0,634±0,004	t=1,0	
(13)	Контроль	0,1	1:9 гомоге- нера- торе	10	0,626±0,006	Не достоверно	(10)
	Облучсние № 39	0.1	1:9	10	0,460±0,010	t=1,69	
(14)	Контроль			10	0,434±0,012	Не достоверно	(10)

Table 1. The Effects of Radiation on Changes in ATPase Activity of Actomyosin

Key: 1. Protein preparation. Duration of storage before experiment

- 2. No. experiment
- 3. Experimental conditions (350 MHz, 5 mW/cm<sup>3</sup>, 20 min)
- 4. ATPase activity (in arbitrary units) and the standard error of the mean, M + m
- 5. % change with respect to the control. Student's t coefficient. Significance of difference
- 6. Ionic strength /KCl/, mole
- 7. Dilution
- 8. Number of parallel tests
- 9. Irradiation No. Control
- 10. Insignificant
- 11. Undiluted
- 12. One day
- 13. Two days
- 14. Three days
- 15. Four days
- 16. Treated 1:9 in homogenizer

UDC 614.87:537.868.029.64

THE DEPENDENCE OF THE TEMPERATURE RESPONSE TO MICROWAVE IRRADIATION ON THE INITIAL FUNCTIONAL STATE OF THE CNS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 137-140

Article by Ye. A. Lobanova

[Text] Investigations have been conducted on the temperature response of white rats to microwave irradiation on the initial functional state of the CNS, changes in which were induced by preliminary treatment with neurotropic agents. Chlorpromazine and acetylcholine were found to decrease the hyperthermic effect of microwaves and shorten the period of recovery, while epinephrine and atropine had the opposite effects.

In the present study we were concerned with the importance of the initial functional state of the CNS in the formulation of a response on the part of an organism to microwave (MW) irradiation of thermogenic intensity according to thermometry data. Changes in the functional state of CNS were achieved by preliminary administration of pharmacologic agents which were selected on the basis of the effect they exert on various structures in the nervous system.

The experiments were conducted on 200-250 gm white rats (90 animals). Rectal temperature was measured by an electric thermometer. The animals were irradiated in a polystyrene cage consisting of six cells. Six rats were irradiated at one time. In each group (12 rats), control measurements of temperature were performed two times. Subsequently, the temperature response was determined at one week intervals:

-- after the administration of the pharmacological agents, -- after irradiation with 10 cm MW with an intensity of 40 mW/ sq cm for 30 min,

-- after irradiation following preliminary administration of the pharmacological agent.

Temperature determinations were conducted every 15 minutes for 1.5-2 hours as in the first control investigations. Pharmaco-

logic agents were administered subcutaneously 15 minutes before irradiation in the following doses: 5 mg/kg chlorpromazine, 1mg/kg epinephrine, 10 mg/kg atropine, 60 mg/kg acetylcholine, 2 mg/kg histamine, 10 mg/kg caffeine, and 7.5 mg/kg bemegride.

As shown with the studies involving the administration of the adrenolytic chlorpromazine -- which possess hypothermal effects (Figure 1a) -- the temperature response to MW irradiation did not differ from the response observed when radiation alone was employed. However, after irradiation was terminated, body temperature fell sharply, and by the 60th minute of the recovery period was  $0.9 \pm 0.1^{\circ}$ C lower than the initial temperature, while in irradiation without the agent, the temperature was close to the initial level ( $\Delta t = 0.3\pm0.2^{\circ}$ C). The impression was created that MW temporarily undermined the effects of chlorpromazine. In fact, consideration of the data obtained when the agent was administered 45 minutes before irradiation instead of 15 minutes, confirmed this impression.

Despite the initial decrease in body temperature by 0.5°C under the influence of chlorpromazine, MW irradiation elicited a marked hyperthermal response ( $\Delta t = 3.3\pm0.2$ °C) which even exceeded the response obtained when the preparation was administered 15 minutes prior to exposure to MW ( $\Delta t = 2.4+0.2$ °C).

There is no general agreement on the mechanism of hyperthermic action of chlorpromazine. According to some authors [S. Courvoisier, J. Fournel, and B. Ducrot, 1953], it is due to the effects of chlorpromazine on metabolism, while others [J.Cheymol, and C. Levassort, 1955; D. Hopkin, 1955; W. Finkelstein, 1954] hold that decreased oxygen consumption by tissues occurs only with very high doses of chlorpromazine. There are also fairly convincing data which indicate that the hypothermal effects of chlorpromazine are due to its action on the thermoregulatory centers.

A completely different hyperthermal effect due to MW was obtained in animals injected with epinephrine. Preliminary injection with epinephrine significantly magnified the effects of MW, and delayed recovery (Figure 1b). This effect could be explained in two ways: on the one hand, constriction of cutaneous vessels by the injection of epinephrine lead to a redistribution of blood flow into the internal organs which in turn resulted in increased rectal temperature, which reflects the average body temperature. The heat loss via the skin leads to overheating of the organisms under exposure to MW irradiation. On the other hand, it is possible that epinephrine stimulates the thermoregulatory centers, in part the nuclei of the posterior hypothalamus, which lead to increased heat production, and prevent loss of heat by the tissues; in this manner also, the temperature response to MW irradiation would increase.
Interesting data were obtained with preliminary administration of atropine, an M-cholinolytic agent (Figure 2a). Atropine had virtually no effect on temperature change during MW irradiation, but exerted a significant effect on the course of temperature recovery after irradiation had been terminated.

By the 45th minute of the recovery period, the rectal temperature was  $1.7 \pm 0.3^{\circ}$ C higher than the starting temperature; at the corresponding period of time in the atropine untreated animals, it was only  $0.2 \pm 0.1^{\circ}$ C higher than the initial temperature. Atropine is known to have a selective effect on the M-cholinoreactive system, as a result of which the tonic effects of parasympathetic nerves are abolished. However, it is also felt that atropine may possess a central cholinolytic effect in addition to a peripheral one [S.V. Aniskov, P.P. Denisenko, 1962; N.V. Savateyev, 1957]. Consequently, if we may assume -- and the reasons for this are indicated above -- that on irradiation with MW the tone of the parasympathetic branch of the autonomic nervous system decreases, then it is easy to explain the effects of atropine.

This view is supported by data obtained in studies on the temperature response to MW irradiation on animals that had received acetylcholine. Administration of acetylcholine significantly changed the temperature response to MW irradiation, and shortened the recovery period (Figure 2b). The favorable effects of acetylcholine may have been due to its direct vasodilatory action on arteries and arterioles, as a result of which heat loss increased and body temperature decreased. In addition, a stimulatory effect of acetylcholine on reactive structures in the brain cannot be excluded, such as the nuclei of the posterior hypothalamus, which increased heat loss by tissues as a result of vasodilation which, in turn, also decreases microwave hyperthermia.

We also studied the effects of histamine on the hypothermic effects due to MW irradiation since, according to the literature data, histamine decreases the volume of the circulating blood as a result of capillary dilation, and loss of blood plasma into the tissues across the capillary wall. However, administration of histamine did not alter the response to MW irradiation to any significant extent.

The use of a cerebral cortical stimulant (caffeine), and a stimulant of the formations of the limbic system (bemegride), did not alter the temperature response to MW irradiation from that seen when the drugs were not employed; only a slower recovery period was noted after irradiation ceased. In addition, in studies on the resistance to MW irradiation (14 mW/sq cm for 2 hours), bemegride at the same dose significantly increased the life time of the "irradiated" animals, while caffeine at higher doses, decreased the life time of these animals. The heating of animal tissues and increase in body temperature on irradiation with 10 cm MW depends not only on the incident electromagnetic energy and its transformation into heat energy, but also on a possible indirect effect of MW on the brain [A.S. Presman, 1968]. Since each of the tested agents possessed not only peripheral effects, but also central effects, it is very difficult to differentiate the different factors which modified the reaction of the organism to MW irradiation. Nevertheless, it is evident that the initial functional state of the CNS, determines to a large extent the response to irradiation, and the course of recovery.

It should be emphasized that substances that block both adreno- and cholinoreactive structures in the nervous system (chlorpromazine and atropine) had virtually no effect on hypothermal effect due to MW, but did significantly alter the course of recovery. On the other hand, substances which stimulated these structures, (epinephrine and acetylcholine) changed both the extent of microwave hyperthermia, and the course of the recovery. Both adrenolytic agents, chlorpromazine and acetylcholine, decreased the temperature response to MW exposure, while both cholinolytic agents, atropine and epinephrine, magnified the response. On the basis of this, it appears that under the thermogenic conditions of MW irradiation, the tone of the sympathetic branch of the autonomic nervous system predominates.



Figure 1. Temperature Response to MW Irradiation 15 min After Treatment With Chloropromazine (a) and Epinephrine (b). Ordinate represents △t in <sup>O</sup>C (M + m). Abscissa represents study time in minutes. 1) Control Determinations. 2) Response to Agent. 3) Response to MW Irradiation. 4) Response to WM Irradiation After Administration of the Agent.

Key: 1. Change in body temperature, degrees 2. Time, min

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Figure 2. Temperature Response to MW Irradiation 15 min After Administration of Atropine (a) and Acetylcholine (b). Ordinate: At in C (M + m). Abscissa: Investigation time in Minutes. 1) Control Determinations. 2) Response to Agent. 3) Response to MW Irradiation. 4) Response to MW Irradiation After Administration of the Agent

Key:	1.	Change	in	body	temperature,	degrees
	2.	Time, r	nin			

## UDC 614.87:537.868.029.64

INVESTIGATIONS ON THE SUSCEPTIBILITY OF ANIMALS TO MICROWAVE (MW) IRRADIATION FOLLOWING TREATMENT WITH PHARMACOLOGIC AGENTS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 141-143

Article by Ye. A. Lobanova

[Text] This article deals with studies on the sensitivity of animals (white rats) to MW irradiation (14 mW/sq cm for 2 hours) after the administration of 15 pharmacological agents. Only narcotics (chloral hydrate and sodium barbital) and the analeptic bemegride, decreased susceptibility to MW, as indicated by the longer survival of the "irradiated" animals. Substances which stimulate and inhibit adreno-, cholino-, and serotonin responsive structures in the brain, as well as caffeine and pentylenetetrazol, increased the susceptibility of animals to MW.

In recent years a great deal of attention has been devoted to studies on the reactivity of an organism, and the various factors that may influence it one way or the other. However, this interesting problem has been little studied with respect to microwaves. There are only the data of individual authors which indicate that the response of an organism to MW of nonthermogenic and thermogenic intensities, depends on the functional state of the organism [A.G. Subbota, 1958; Ye. A. Lobanova, 1960, 1968; M.I. Yakovleva, 1966; V.A. Pukhov, 1965].

We studied the effects of thermogenic MW on animals with different functional states of the CNS induced by the use of pharmacologic agents with very well known mechanisms of action. We studied the general state of the animals, the survival time under "irradiation", and the temperature response. In addition, we studied the effects of 15 pharmacological agents on the sensitivity to MW irradiation of more than 300 white male rats weighing 200-250 gms. The agents were introduced subcutaneously (with the exception of chloral hydrate and sodium barbital which were introduced into the stomach) 30 minutes before or immediately prior to irradiation. The animals were irradiated in polystyrene cages with 10 cm MW with an intensity of 40 mW/sq cm; duration of irradiation was limited to two hours.

The clinical picture induced by MW irradiation consisted of a series of sequential stages. At the beginning of irradiation, the animals sat quietly in their cells and cleaned themselves. After 10-15 minutes, they became restless and attempted to open the door of the cage. At this time their noses, ears, and paws became red. Body temperature increased by  $1.2 \pm 0.2^{\circ}$ C. Subsequently depression set in, and the rats showed little movement, and only occasionally changed their position in the cell. Marked hyperemia was evident, as well as copious sanious discharges from the nose. The rectal temperature increased by  $4.5 \pm 0.2^{\circ}$ C. During this prolonged stage, there were convulsive twitches of the head and extremities. Following this, the animals lay down on the side , and death ensued.

Therefore, in the clinical picture it was evident that a state of excitation was replaced by a state of inhibition.

In view of the second more prolonged stage of depression, we employed central stimulants, such as caffeine, which primarily acts on the cerebral cortex, and pentylenetetrazol and bemegride, which act on the subcortical structures, and in the view of many authors, also on the formations of the limbic system. In view of the mechanisms of action of these substances, we felt that they might alter the susceptibility of the animals to MW irradiation. The experiments show that 10-50 mg/kg of caffeine did not have any effect on the resistance of the animal to MW, and at a dose of 100 mg/kg, somewhat increased the susceptibility of the animals to irradiation.

The "subcortical" stimulant, pentylenetetrazol, was ineffective at a low dose (20 mg/kg), and at a higher dose (60 mg/kg), led to a more rapid death (52  $\pm$  10') than in the control (74  $\pm$  5'); furthermore, death ensued more rapidly the earlier was the appearance and intensity of the convulsions.

The use of the antidepressant bemegride (7.5 mg/kg) which possesses highly interesting and multifaceted mechanism of action [Ya. Yu. Bagrov, 1968] significantly lowers the susceptibility of the rats to MW irradiation; out of the 12 rats, 5 survived for 110 ± 6', and there was not a single case with chronic convulsions. In the control groups, all animals died with an average survival time of 81 ± 11'.

At the dose employed, bemegride constituted an exception to the analeptics which were tested and decreased susceptibility to MW. It is difficult to say whether this is due to activation of the hippocampal structures, which, like the nonspecific thalamic system, exert an inhibitory regulatory function which they protect the cerebral cortex from pathogenic effects of MW, or whether this is due to some other factors. Since we had obtained negative results with the analeptics we assumed that perhaps prolongation and amplification of CNS inhibition will result in increased resistance to MW. Both chloral hydrate (200 mg/kg), and sodium barbital (200 mg/kg) increased resistance to MW irradiation with survival times under irradiation of 101  $\pm$  4', and 97  $\pm$  11', respectively, for comparison with a control survival time of 71  $\pm$  4'. It appears that the positive effects of the narcotics were due to a blocking effect on the ascending activating impulses of the reticular formation.

However, the use of the vegetolytic chlorpromazine (5-10 mg/kg) which also blocks activating influences from the reticular formation of the midbrain and the posterior hypothalamus, not only did not decrease susceptibility to MW irradiation, but, on the contrary, enhanced it. The survival time of the animals decreased depending on the dose (5-10 mg/kg), and the time at which the substance was administered (immediately before irradiation, or 30 and 90 minutes before irradiation was commenced), by 16-25% in comparison with the controls.

Administration of epinephrine (0.2-0.5 mg/kg) and ephedrine (40 mg/kg) immediately before irradiation, increased the susceptibility of animals to MW, with a corresponding decrease in the survival time by 35% and 25%, respectively, in comparison with the controls.

Consequently, both inhibition and stimulation of the adrenoresponsive structures in the brain, lead to a similar effect.

An analogous result was obtained with the use of substances which inhibit or stimulate cholinoreactive structures. Thus, atropine (10-20 mg/kg) an M-cholinolytic agent, decreased resistance to MW by 13-25%. Administration of acetylcholine (60 mg/kg), and nicotine (3 mg/kg) also decreased resistance to irradiation by 21 and 26%, respectively.

An attempt to use a combination of acetylcholine (60 mg/kg) and epinephrine (0.5 mg/kg) did not lead to a positive result, since in this case, the sensitivity of the animals to irradiation increased.

The use of neostigmine (0.1 mg/kg) -- a substance with peripheral anti-cholinesterase activity -- did not effect susceptibility to MW.

Stimulation of serotonin responsive structures by the administration of serotonin (10 mg/kg) and mexamine (10 mg/kg) immediately before irradiation, did not influence the susceptibility of animals to MW, while the administration of indopan [transliteration], (20 mg/kg), a monoamine oxidase inhibitor, significantly increased susceptibility of the animals, and their survival under "irradiation" was  $31 \pm 5$ ', while the control survival time was  $87 \pm 9$ '. Stimulation of the adrenals by the administration of exogenous ACTH (3 units) four hours before irradiation, did not alter the susceptibility of rats to MW irradiation; ascorbic acid levels in the adrenals of these animals were lower  $(145 \pm 9.3 \text{ mg}\%)$  than in control animals  $(204 \pm 5.8 \text{ mg}\%)$ .

Therefore, the data indicate that only the sedatives chloral hydrate and sodium barbital, and the analeptic bemegride, decreased susceptibility of the animals to MW irradiation, and increased their survival. The other substances that were tested, either did not change or increased their susceptibility to MW. It is difficult at the present time to determine whether the favorable effects of narcosis are due to depression of external respiration, gas exchange, depression of basal metabolism, inhibition of tissue metabolism, hypothermia, or blocking of the ascending activating impulses from the reticular formation. This can hardly be explained on the basis of hypothermia since chlorpromazine, which possesses a marked hypothermic effect, inhibited microwave hyperthermia only on short-term exposure (15 minutes), but did not influence MW on longer exposure. The effects of the narcotics cannot be ascribed to the blocking of ascending activating influences from the reticular formation of the midbrain and posterior hypothalamus, since chlorpromazine which possesses analogous properties, did not decrease susceptibility to MW. One may also consider increased tone of the brain system which are responsible for the inhibitory regulatory function. It is then that the positive effects of bemegride may be understood.

In conclusion, it should be noted that under terminal conditions of MW irradiation, the administration of substances which inhibit or stimulate adreno- cholino-, and serotonin responsive structures in the nervous system, should lead to unequivocal results as a result of the dysfunction in the sympathetic and parasympathetic branches of the vegetative nervous system, as a result of the breakdown and exhaustion of the adaptive compensatory mechanisms of the organism.

## UDC 614.87:537.868.029.64

PRINCIPLES OF NEUROPHYSIOLOGICAL INVESTIGATIONS OF MICROWAVE BIOEFFECTS AND CHANGES IN ELEMENTARY EXCITABLE STRUCTURES ON EXPOSURE TO VERY LOW INTENSITY IRRADIATION

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 144-149

Article by M.S. Bychkov

[Text] Studies were conducted on the methodologic limitations of electrographic studies of EMF, which are due to the appearance of artifacts resulting from SHF-induced currents. Results are described of electrophysiological studies of the effects of a single short-term exposure to microwaves with a power flux density (PFD) on the order of 5  $\mu$ W/sq cm on elementary excitable structures which have demonstrated a change in the membrane potential, threshold of excitability, latent and refractory periods, rate of conduction, amplitudes of the action potentials in frog muscle fibers, synaptic delays at the myoneural junction, and impulse activity of individual neurons in the nervous ganglia of the medicinal leech.

Experimental studies on the bioeffects of radiowave SHF employing neurophysiological methods commenced in the 50's [M.S. Bychkov, 1957, 1959], and in subsequent years, a large number of electrophysiological studies have appeared which have dealt with the phenomena and mechanisms of action of this factor on the nervous system.

All of these studies were conducted on the basis of differentiation between the thermal and the so-called nonthermal effects of radiofrequency EMF. Naturally, the greatest interest has been accorded to the nonthermal effects, i.e., bioeffects of SHF exposure of low intensities which are of special interest in medicine, space biology, biophysics, genetics, cybernetics, bionics, agrobiology, and other scientific disciplines.

It is understood, for example, in the case of the centimeter band, that low intensity PFD does not exceed 10 mW/sq cm, i.e., the value which is regarded as representing a threshold for the thermal effect on various systems including the nervous system.

It should be mentioned that despite the relatively large number of neurophysiologic studies in this field, only a small portion of them deal with low intensities, and studies with very low intensities (for example, up to 100  $\mu$ W/sq cm inclusive) are very infrequent.

In speaking of the limited number of studies conducted by neurophysiologists on low intensities, it should be pointed out that as a rule, electrophysiologic studies on CNS are limited to EEG, i.e., studies which are limited to the electrical activity of the cortex alone, and consequently total (slow) activity which makes it difficult to evaluate intersystem relationships, and fundamental neurophysiological processes, including the mechanism of action of the factor under consideration.

On the other hand, since neurophysiological investigations of the biological effects of SHF are directed almost exclusively toward the CNS, they neglect the effects which are exerted on elementary excitable structures, in part the isolated substrate, the cellular structures, and so forth, which also constitutes a serious gap.

Finally, we must consider the unavoidable methodological limitations of electrography in EMF due to possible (and uncertain PFD unavoidable) effects on the biological object of electromotive force (artifact), which is induced by the lead electrodes, and which arises as a result of nonlinear (for induced SHF currents) events at the input circuits of the biopotential amplifier, and at the "electrode-tissue" contact layer.

Without going into details, we should note that to prove this type of induction in SHF field (for example, by means of polarization) which may attain subthreshold (in the biological sense) values at certain PFD, is virtually impossible. However, even at relatively low intensity (on order of 1 mW/sq cm and less) -- careful control must be exercised to eliminate artifacts which may mask or imitate EMF bioeffects in those situations in which it is impossible to avoid the presence of pick-up electrodes within the field.

As a result of this, in a large number of experimental situations, -- primarily in those in which the PFD is greater than 1 mW/sq cm -- we cannot study the development of various effects during irradiation, but are limited to a comparison of initial background with the after effects, and thereby we stand to lose the most valuable type of information.

In studies on the biopotentials of isolated structures (as well as in recording spike activity of cortico neurons (we employed glass microelectrodes, tip diameter of about 1-2  $\mu$ m, 5 to 10 Mohm resistance, filled with 2.5-3 M KCl) which in distinction to metallic electrodes, do not distort the field for all practical purposes, and yield reliable PFD data.

In those situations when the pick-up electrodes were located in a field, determinations were made of the possible stimulating "induction" potential which was compared with the threshold of excitation of the cellular structures. Under these conditions, "induction" did not exceed 5 µV (PFD up to 100 µW/sq cm inclusive) when metallic electrodes were employed in studies of a number of brain structures, and was virtually nonexistent when glass electrodes were employed. In addition to measurements of "induction", special control experiments were conducted in which the electrodes were employed to apply high frequency oscillations to the biological objects from a generator which was employed in these experiments, which had an amplitude known to exceed (2-3 fold) the measured "induction" for a relatively long period of time (one hour or more with exposure in the major experiments from 1 to 30 minutes). In this manner, artifacts in functional changes were eliminated, and studies could be made of the effects due to "pure" irradiation.

In the present article we shall limit ourselves to a review of the effects of microwaves on certain elementary excitable structures which are of interest on their own accord.

A frog nerve-muscle preparation was used to study a number of physiological parameters at the cellular level with PFD on the order of 5  $\mu$ W/sq cm.

Studies on the membrane potential (MP) of muscle fibers showed a significant increase in MP (by 30-37% of the initial value). The increase in MP occurred largely during the first five minutes, and on longer exposure (up to 30 minutes), significant changes did not take place.

Therefore, SHF field exercised a hyperpolarizing effect at least on the muscle fibers under the present conditions of irradiation (including both the centimeter and the decimeter bands).

In addition, a rapid increase in the threshold of excitability of the muscle fiber was evident (on the average of 19% during 1 minute), as well as a statistically significant prolongation of the latent period by an average of 1 msec (initial values were on the order of 4 msec).

Table 1 data are presented on changes in the action potential (AP) of the muscle fibers during indirect stimulation on exposure to decimeter radiowaves (1 minute exposure), and in Table 2, data on changes in the velocity of impulse conduction and absolute refractory periods of the muscle fibers; in Table 3 data are presented on synaptic delay under the same conditions of exposure to EMF.

The microelectrode data demonstrated that inhibition results when a neuromuscular preparation is irradiated at negligible PFD (5  $\mu$ W/sq cm) for a very short period of time.

In principle, a similar effect was obtained when these parameters of exposure --  $5 \mu W/sq$  cm,  $5 \mu$  minute exposure -- were employed in studies on spontaneous activity of individual neurons in the nervous ganglion of the medicinal leech. In addition, certain features became evident which should be discussed in greater detail.

The experiments were conducted on isolated and unisolated ganglia. In the first case, studies were conducted on the direct effects of irradiation (primary physiological effects) on the neurons and interneuronal connections in the ganglia without complications due to reflex impulsation from irradiated periphery. In working with an unisolated ganglion, we obtained first of all, an integral effect of irradiation of the entire leech, and secondly, we dealt with natural currents of impulses in distinction to the somewhat artificial conditions which prevail when studies are conducted on isolated ganglia.

As a rule, in the unisolated ganglia, we obtained a significant inhibition of impulse activity within one to two minutes of irradiation which indicated decreased excitability of the neurons. An example of this is provided by Figure 1. Figure 1 demonstrates that before irradiation, simultaneous recordings were obtained of impulse currents for at least three neurons, and that two minutes after irradiation, only isolated impulses could be detected in the background noise. These results are presented in Figure 2, which consists of a long-term recording obtained before and during a five minute period of irradiation of an isolated Toward the end of one minute of irradiation, an upganglion. surge phase in impulse activity set in (right half of frame d). This phase persisted during the second and the third minute (frames d, e and theleft part of frame f up to the point of the x mark, represent a continuous recording) reached a stage of culmination at the beginning of the fourth minute (the right portion of frame f), and began to weaken at the end of this time (frame g); subsequently, it was replaced by an increasing inhibitory effect during the course of the fifth minute of irradiation (frames h, i and j which constitute a continuous recording during the fifth minute of irradiation), and immediately after termination of irradiation which isindicated by the arrow.

The inhibition of neuronal activity under these conditions was found to be rapidly reversible (maximum recovery was achieved within 12 minutes). Figure 3 shows complete restoration of the background activity of a neuron of an isolated ganglion 12 minutes after irradiation (frame 4) which had elicited marked inhibition of impulse activity (frame 3). Comparison of frames 1 and 2 in Figure 3 shows absence of changes in this neuron during preliminary exposure to SHF oscillations through the microelectrode, which exceeded the value of the possible "induction" (control).

It may be assumed that the difference in the dynamics of the effects in the isolated and the nonisolated ganglia are due to the participation of regulatory mechanisms of the entire organism in the latter case.

Table 1. Changes in the Amplitude of AP (20 Hz Frequency Stimulus) of Optimal and Maximal Rhythms of the Frog Muscle Fiber

		(2) Амплитуда ПД			(39nT)	имальный ј	DHTM	(Даксимальный риты		
	(1) Серня	Среднеарифме- тическая и и пределы ко- лебаний, % к исходной ве- личене б	у при сопо- лавлении с Оригродем	Сритерий	, Гастота 113- Оменений, %	Г при сопо- кставлении с Кгонтролем	Критерий	Частота из- менений, 96	г при сопо- ставлении с контролем	БКритерий
(9) (10)	Коктрольная Опытная	100 (99—103) 79 (51—86)	< 0,01	Вил- коксо- на (11	(8) 10 58	<b>(0)</b> <0,025	Точ- ный метод Фн- шера (12)	20 84	< 0,025	Точ- ный метод Фи- шера (12)

Key:

l. Series

- 2. AP amplitude
- 3. Optimal rhythm
- 4. Maximal rhythm
- 5. Arithmetical mean and limits of fluctuation,  $\mathcal R$  of initial value
- 6. P in comparison with control
- 7. Criterium
- 8. Frequency of change, %
- 9. Control
- 10. Experimental
- 11. Wilcoxon
- 12. Fisher accurate method

# Table 2. Changes in the Velocity of Excitability Conduction and the Absolute Refractory Period of the Frog Muscle Fiber

	(2) Скорость проведения				(3) Рефрактерный период			
	(1) Серия	Среднеарифме- тическая и пределы коле- баний. % к ис- ходной вели- чипе	р при со- поставле- ния с кон- тролем	Критерий	Среднеарифме- тическая н ирелелы коле- баний, ж ко- ходной вели- чине	р при сс- ноставле- нии с кон- тролем	Критерий	
		(4)	<b>-(</b> 5 <b>)</b>	(6)		(5)	(6	
(7)	Контрольная	100 (98—102)			99 (97—102)	1		
( <sup>8</sup> )	Опытная	75 (64—89)	< 0,01	Вилко- колиа (9)	150 (94—176)	<0,05	Вилко- ксона	

Key:

- Series
- 2. Conduction velocity
- 3. Refractory period
- 4. Arithmetical mean and limits of fluctuation, % of initial value
- 5. P in comparison with control
- 6. Criterium
- 7. Control
- 8. Experimental
- 9. Wilcoxon

Table 3. Changes in Synaptic Delay (Myoneural Transmission in the Frog)

	(1) Серня	Среднеарнфмотн- ческая и проделы колебаний, % к исходной величинс (2)	р при сопо- ставлении с контролем	Критерий	
(5) (6)	Контрольная Опытная	193 (90—110) 140 (100—167)	<0,01	(4) Вилкоксона	(7)

Key:

- 1. Series
- 2. Arithmetical mean and fluctuation limits, % of initial value
- 3. P in comparison with control
- 4. Criterium
- 5. Control
- 6. Experimental
- 7. Wilcoxon



Figure 1. Spontaneous Spike Activity of Neurons of an Unisolated Nervous Ganglion of a Medicinal Leech. 1) Backround. 2) Second Minute of Irradiation



Figure 2. Spontaneous Neuronal Spike Activity of an Isolated Ganglion of the Medicinal Leech. a-c)Continuous Recording; Arrow on Section c Indicates initiation of Irradiation. d-f) Continuous Recording; d is the end of 1st min of exposure; e the 2nd and 3rd min; f the 3rd and the beginning of the 1sth. g) End of 1sth min. h-j) Continuous Recording During the 5th min of Exposure and After its Termination (Indicated by Arrow)



Figure 3. Spontaneous Activity of a Neuron of an Isolated Ganglion of the Medicinal Leech. 1) Backround. 2) 5th Minute of Contact Exposure to SHF Oscillations (Control). 3) 1st min After 5 min Exposure. 4) 12th min.

#### UDC 614.87:537.868.029.64

THE PROBLEM OF GLIO-NEURONAL RELATIONSHIP IN THE RAT CEREBRAL CORTEX DURING LONG-TERM EXPOSURE TO MICROWAVES

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 150-153

Article by I.M. Kazbekov and Ye. A. Lobanova

[Text] Cytometric studies on the glio-neuronal relationship in the rat brain demonstrated that in the presence of relatively weak (but significant) changes in the dimensions of the cytoplasm and the nuclei of neurons, highly significant changes take place in the perineuronal glia. The greater reactivity of the neuroglia may be regarded as a protective reaction to microwave exposure which do not elicit changes in the neurons.

Earlier physiological and morphological investigations have demonstrated that changes in conditioned reflex activity of animals that had been subjected to long-term exposure to microwaves are due to changes in the axosomatic and axodendral connections in the brain of rats, in the absence of marked changes in the neurons themselves [Ye. A. Lobanova, and M.S. Tolgskaya, 1960].

In view of the great interest which is attached at the present time to studies of the glio-neuronal relationships, and the limited data which pertain to changes in this system under the influence of weak alternating factors [M.M. Aleksandrovskaya, 1966; Yu. A. Kholodov, 1966; Svaetichin, 1965], we decided to investigate this unique metabolic and functional system under conditions in which animals sustained chronic irradiation with microwaves.

The glia is known to consist of astro- and oligodendrocytes (microglia which contains mesodermal elements is not strictly part of the neuroglia). The astrocytes participate in water and metabolite exchange and in the transport of ions, while the oligodendrocytes provide the immediate surrounding of the nerve cells and act as their satellite [Friede, 1965; Galambos, 1965].

Numerous communications are available on the accumulation by both the astrocytes and the oligocytes of various enzymes and mediators which possess certain specificity in terms of both types of glia. It is held to some extent that the oligoglia have a predominant role in the oxidation of glucose, while the astrocytes are of importance in protein synthesis [Dimova and Gerebtcoff, 1966]. The relatively greater metabolic activity of the satellite glial elements in comparison with other glial elements is indicated by the fact that the satellites contain higher concentrations of DNA [Viola M. Pia, 1963].

Extensive data have accumulated which point unquestionably to the fact that in the glio-neuronal system, it is the glial elements that respond earlier by quantitative, qualitative, and structural changes to various stimuli than do nerve cells.

The nature of the mechanisms responsible for the interaction of glial and neuronal elements in normal and pathological states, has not yet been resolved according to the data in the literature. Data are available which indicate that the glia maintain the functional stability of the synaptic apparatus of the neurons [Purpura, 1961], abolishes the slow negative potential of the brain cortex which arises on direct electrical stimulation and on its interaction with morphine during its application to neuroglia [A.I. Roytbak, 1965, 1967]. M.M. Aleksandrovskaya [1966, 1968] treats the activation of neuroglia (reimpregnation, proliferation) under the influence of small doses of noxious agents as a defensive reaction.

In the present investigation, we studied the state of the glioneuronal system in the brain cortex of rats exposed to 10 cm microwaves at an intensity of 10 mW/sg cm for 60 minutes/day over a 6 month period.

The dimensions (areas) of the neurons and their nuclei in the third and the fourth layers of the brain cortex (in the postcentral zone) were conducted cytometrically. The dimensions of the cytoplasm were obtained by subtraction of the nuclear dimension from the neuronal dimension. Cytometry was accompanied by the simultaneous enumeration of the perineural glial elements (satellites) which consist of oliodendroglial cells. We limited our attention to the perineural glia which is capable of mobility and proliferation and is intimately related to the neurons, taking into consideration the importance of the satellites in the metabolic function of the nerve cells, as noted above.

The cytometric data and quantitative changes in the perineural glia were studied on preparations that had been stained with gallocyanin and chromium alum. Three irradiated rats and three control animals were sacrificed at each of the six times of investigation: after 1, 2, 3, 4, 5, and 6 months. In each case,

the dimensions of the neurons and their nuclei were determined on five sections (10 neurons per section). Consequently, the cytometric data for each time period were based on 150 neurons. In each section, the number of satellites/10 neurons were determined.

The resultant data showed that dimensions of the nuclei and the cytoplasm of the neurons in the third layer did not differ significantly between the experimental and the control animals.

In the fourth cortical layer, we determined certain differences between the groups which obviously could be evaluated as an indication of greater reactivity of the neurons in this layer (Figure 1). The cytometric data indicated an increase in the dimensions of the nuclei and to an even greater extent, of the cytoplasm, during the initial stages of the investigation; electron microscopy was reflected as cloudy swelling of the nucleus in the cytoplasm; in isolated cases the cytoplasm of the reurons was found to be vacuolized. The same picture was encountered in subsequent periods of investigation. Furthermore, cytometry revealed a significant increase in the cytoplasmic area of the neurons in the irradiated animals, in comparison with the control rats. Temporal comparison of the relative and not the absolute indices. i.e., differences in the cytoplasmic dimensions of the experimental and the control animals (Mdifference  $\pm m$ ), demonstrated a certain attenuation of these differences toward the end of this experiment (Figure 2).

The limited degree of fluctuation in the indices under study was found to correspond to disruptions in the conditioned reflex activity of rats which were most pronounced during the initial stages of microwave irradiation that had the same parameters. These dysfunctions consisted of prolonged latent periods of the conditioned reflexes, frequent loss of positive conditioned reflexes, occasionally absence of natural reflexes to food, and weakening or intensification of differential inhibition.

Determination of the satellites in the fourth layer of the postcentral cortex, showed that their number increased significantly in the experimental group (Figures 3 and 4), particularly during the first four months, after which their numbers decreased. The latter coincides in the decrease in the cytometric indices of the nuclei and the cytoplasm of the neurons after six months of irradiation. It should be pointed out that despite relatively unremarkable changes in the cytometric indices, the numbers of perineural gliocytes in the irradiated group increased several fold in comparison with the control group.

The data which have been obtained indicate that on exposure to microwaves the glio-neuronal system functioned as one unit, and

the greater changes in the glia can obviously be regarded as a protective reaction against the microwaves which themselves did not elicit significant changes in the neurons. This is in agreement with the conclusions of other authors that the reaction of the neuroglia and changes in the glio-neuronal system under the influence of weak alternating physical factors, represents a unique nonspecific adaptive reaction [M.M. Aleksandrovskaya, 1966, 1969; and others].



Figure 1. Changes in the Dimensions (Areas) of the Nuclei (a) and Cytoplasm (b) of Neurons of Layer IV of the Rat Cerebral Cortex During Prolonged Exposure to Microwaves (Confidence Limits With 98% Probability).

Key: 1. Nuclear area, mu<sup>2</sup> 2. Cytoplasmic area, mu<sup>2</sup>



Figure 2. Change Dynamics in the Dimensions (Areas) of the Cytoplasm of Irradiated and Control Groups and Confidence Limits at 98% Probability During the Investigation. Abscissa: Time of Investigation (Months). Ordinate: Area Differences in mu<sup>2</sup>.



- Figure 3. Change in the Number of Perineural Oligodendrogliocytes in Layer IV of the Cerebral Cortex During the Investigation. Abscissa: Time of Investigation (Months). Ordinate: Number of Satellites. 1) Irradiated Group, 2) Control Group.
- Key:

1. Number of satellites



Figure 4. Increase in the Number of Perineuronal Glial Elements in Layer IV of the Cerebral Cortex of the Rat After Three Months of Irradiation (Lower Section of the Photograph). Gallocyanin Stain, X600.

#### UDC 614.87:537.868.029.64

STUDIES ON THE REPRODUCTION AND TESTICULAR MICROSTRUCTURE OF MICE EXPOSED TO MICROWAVES

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 154-159

Article by A.N. Bereznitskaya and I.M. Kazbekov

[Text] Chronic irradiation of mice with microwaves of nonthermal intensity evokes diffuse changes in the testes and a decrease in the litter size of healthy females mated with irradiated males.

Experimental studies have shown that radiofrequency electromagnetic waves may elicit various malfunctions in the reproductive system [S.F. Gorodetskaya, 1963; V.A. Povzhitkov et al., 1961; G.K. Timeskova, 1966; L. Minecki and L. Cieciura, 1966; V.K. Tkach, and S.F. Gorodetskaya, 1969; T.P. Blinkova et al., 1961; I.D. Boyenko et al., 1967; R.I. Kruglikov, 1967; I.A. Piontkovskiy et al., 1970; Yu. A. Spasokukotskiy and S.F. Gorodetskaya, 1972; V.A. Zhuravlev, 1972; and others].

In recent years, this problem has attracted the attention of a wide circle of investigators. Studies on individuals exposed to radiowaves under industrial conditions have shown that males frequently have complaints of a sexual nature [N.N. Goncharova, 1972; A.M. Palladin et al., 1962]. Experiments on animals indicate that the male sexual system is less susceptible to this factor [S.F. Gorodetskaya, 1960, 1963; V.K. Tkach and S.F. Gorodetskaya, 1969].

The object of the present study was to determine the degree of the susceptibility of the male sexual system in mice exposed to nonthermogenic microwaves.

Etudies were conducted on the reproductive function and morphological changes in the testicles of white outbred mice exposed to microwaves ( $\lambda$  10 cm) of subthermal intensity (10 mW/sq cm). The animals were irradiated for two hours/day. These investigations were conducted on 59 males that were divided into four groups depending on the condition of irradiation.

Sexually mature group I mice were exposed to microwave irradiation for five months, group II mice were irradiated antenatally, group III mice were irradiated antenatally and then for five months once they achieved sexual maturity, and group IV mice served as controls.

Data on the reproductive function of the irradiated mice are presented in Table 1.

Mice exposed to microwaves retained their fertility. There was a statistically significant decrease in the number of progeny of healthy females mated with irradiated males belonging to groups I and III. The number of abnormal individuals (stillborn and nonviable) in the litters of irradiated males was small and exceeded to only an insignificant extent the percentage of such individuals in control litters. There was a statistically significant difference in the increase of the percentage of debilitated progeny, and in group III animals an increase in the number of stillbirths. Postnatal mortality did not exceed that in the controls, and only among the progeny of group III males was it somewhat higher.

The development of the progeny resulting from the mating of females with irradiated males was followed over a three week postnatal period. The development of the young mice was evaluated in terms of weight, and the times at which the eyes and ears opened, and a hair cover appeared.

Postnatal ontogenesis of the progeny of the irradiated males is represented in Table 2.

The newborn progeny of the irradiated males did not differ from control mice in terms of weight and body dimensions. Later their weight was somewhat greater than that of control animals. The progeny of the females that had been mated with group III irradiated males weighed an average of 3 gms less than the control animals on the 21st postnatal day. The postnatal ontogenesis of the progeny of the irradiated males did not differ to any practical extent from the control group with the exception that opening of the eyes occurred somewhat later.

Therefore, studies on the reproductive function of male mice exposed to microwaves antenatally (delayed sequelae), during the postnatal period, as well as antenatally and postnatally, revealed poorly delineated changes which were reflected in the decrease of the number of progeny, and an increase in the number of defective individuals. The most pronounced changes were noted in mice exposed to microwaves both antenatally and postnatally. Microscopic studies of the testes were conducted on 10 mice from each group.

The weight coefficients of the testes are presented in Table 3.

The testes were fixed in formalin and celloidin sections were stained with hematoxylin and eosin. In the case of each animal 100 seminiferous tubules were examined.

For 1,000 tubules in each group of animals, enumeration was made of the number of tubules with limited desquamation of the germinal epithelium, marked desquamation, atrophy of the epithelium, presence of giant cells.

The results of the microscopic studies of the testes are presented in Figure 1.

The most marked changes (in terms of every index), were noted in the testes of group III animals (irradiated antenatally and postnatally). Tubular atrophy -- although infrequent -- was observed only in group III animals. In addition, this group contained the highest incidence (58 tubules) of moderate epithelial desquamation. The formation of giant cells was noted with an almost equal frequency in group III animals. Pronounced desquamation of germinal epithelium as a pathologic sign was encountered very infrequently, and the differences between the groups were insignificant.

It is important to note that despite the presence of certain pathological changes in the control animals, changes in the experimental groups of animals were much more pronounced and statistically significant.

Morphological changes in the testes are reflected in the microphotographs (Figures 2-5).

Therefore, studies on the microstructure of testes and the reproductive capacity of mice exposed to 10 mW/sq cm microwaves, revealed moderate changes in the gonads (increased number of seminiferous tubules with desquamated germinal epithelium, formation of giant cells, decreased weight coefficients of the testes), and a decrease in the litter size produced by healthy females mated with irradiated males.

In group II mice, which had been exposed to microwaves during embryonic development, changes were noted after irradiation (at the age of 6 months) which were similar to the changes noted in group I mice that had been exposed for a long period of time (5 months) when they were sexually mature. This fact, along with a decrease in the number of progeny in experimental mice of every group, may indicate that microwaves exert a mutagenic effect. Control experiments on the irradiation of mice during both the antenatal and the postnatal periods (group III) confirmed the results obtained with the above groups of animals. The most pronounced changes were noted in group III mice which indicated summation of the changes which occurred during embryonic and postnatal irradiation.

At the present stage of the investigation, it would appear to be too early to attempt a comparison of the relative susceptibilities of the male and the female sexual systems to microwaves. A certain degree of specificity is noted in the pathological changes in male and female mice exposed to microwaves. In the females, microwaves decreased fertility to the extent that permanent sterility may appear, and result in the appearance of a large number (up to 50%) of defective progeny (small litter size, delayed rate of postnatal ontogeny, and high perinatal mortality). However, the litter size of irradiated females frequently does not change [A.N. Bereznitskaya, I.M. Kazbekov, and T.Z. Rysina, 1972]. Males exposed to microwaves retain their fertility, the number of defective progeny is small, and postnatal development and mortality of the progeny are virtually the same as in the controls. Change in the reproductive function is primarily expressed as a decrease in the number of progeny, It is possible that the pathogenesis of the changes in the males and females exposed to microwaves reflects a different mechanism of action.

Table 1. Results of Studies on the Reproductive Capacity of Rats Exposed to Subthermal Intensities of Microwaves During the Antenatal and Postnatal Periods

(1)	(2)		(6	) Облучен	ке		
Показатели	Контроль Матизиці	Г группа М±тднф	t	II групка М = тдиф	t .	III Группа M±m <sub>диф</sub>	t
Количество самцов (7) Количество доленженных	12	14		15		· 18	
к ним самок . (.8) .	24	28		30		36	
Из них дало потомство % Количество петомков (10	(9)20 84,3±7,6 150	22 78,6±7,5 143	0,4	23 76,6±7,5 155	0,6	26 72,2 <u>+</u> 7,5 161	0,6
Среднее количество по- томков в пометах (11	)7,5±0,35	6,5 <u>+</u> :0,38	2,0	6,7±0,39	1,7	6,2±0,41	2,4
соличество мертворож- денных . (12.) . %	4 2,6±1,33	6 4,2±1,67	0,7	5 3,2±1,41	0,3	12 7,4±2.06	1,9
количество осларленных особей	17 11,3±2,6	$29 \\ 20,2\pm3,4$	2,1	87 23,9 <u>-⊢</u> 3,42	3	31 19,2±3,1	1,9
Постнатальная смертность (14).	23 15,3±2,9	29 16,8±3,1	0,4.	36 23,2 <u>+</u> 3,4	1,8	40 24,8±3,4	2,1

Key:

- 1. Index
- 2. Control,  $M + m_{diff}$ 3. Group I,  $M + m_{diff}$ 4. Group II,  $\overline{M} + m_{diff}$
- 5. Group III, M ± m diff
- 6. Irradiation
- 7. Number of males
- 8. Number of admitted females
- 9. Number of females with litters
- 10. Number of progeny
- 11. Average litter size
- 12. Number of stillbirths
- 13. Number of debilitated individuals
- 14. Postnatal mortality

Table 2. Results of Studies on Postnatal Ontogenesis of Progeny Resulting Male Mice Exposed to Microwaves

	(1)	(2)		(3)	Облучен	н <del>е</del>		
	Показатели	Контро́ль <i>М</i> ± т <sub>диф</sub>	1 группа М±т <sub>диф</sub>	t	II группа М±т <sub>диф</sub>	ť	1)1 группа М≞ <sup>т</sup> диф	t
(7)	Количество потомков . Краниокаудальные раз-	150	( <u></u> 4) 143		(5)		(6) 161	
(0)	мышат, мм	28,60±0,44	$29,20\pm0,60$	0,7	28,60+0,25		28,50+0,25	~
(9)	Новорожденных . (10)	$1,44 \pm 0,07$	$1,40 \pm 0,08$	0,4	1,38±0,05	0,7	1,38 <b>±0,</b> 07	0,6
	⊑ 3-й (1.1.),., ⊒	2,17+0,15	$2,72 \pm 0,23$	2,0	2,58±0,1	0, <b>3</b>	2,46±0,1	1,6
(12)	Гжо 5-й	3,34±0,21	3,56 <u>+</u> 0,22	0,5	3,80±0,27	0,6	3,30 <b>±0,1</b> 3	0,6
	er ou ⊔	7 <b>,7</b> 1±0,58	7,60±0,44	0,2	7,40±0,47	0,3	8,33±0, <b>0</b> 9	1,0
	<sup>1</sup> <sup>1</sup> <sup>1</sup> <sup>1</sup> <sup>1</sup> <sup>1</sup> <sup>1</sup> <sup>1</sup> <sup>1</sup> <sup>1</sup> <sup>1</sup>	12,5 <u>+</u> 0,43	11,96 <b>±0,</b> 8	0,6	11,3±0,7	1,8	9,74±0,42	4,6
(13) (14) (15)	Отлипание ушек, дии Опушение, дни Прозрение, дпи	3,54±0,2 6,11±0,22 13,6±0,16	$3,58\pm0.31$ 5,80±0,16 13,9±0,16	0,1 1,2 1,1	$3,60\pm0.1$ $6,30\pm0.21$ $14,1\pm0.21$	0,1 0,6 2,0	3,70±0,07 6,20±0,19 14,4±0,2	0,3 0,3 2,8

Key: 1. Index
2. Control, M + m diff
3. Irradiation
4. Group I, M + mdiff
5. Group II, M + m diff
6. Group III, M + m diff
7. Litter size
8. Craniocaudal dimensions of mouse neonates, mm

9. Weight, gm

. .

Continued Table 2.

- 10. Neonates Key:

  - 3rd, 5th, etc.
     Days after birth
  - 13. Ear opening, day
  - 14. Hair cover, day
  - 15. Eye opening, day
- Table 3. Results of Studies on Weight Coefficient of Mouse Testes Following Exposure to Microwaves

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	(1) Группы животных	(2) Количество животных	Вес семенников (правого + левого) по отношению в вссу тела живот- ного М±тдиф	t
(3)	I II II1 Контроль	75 62 36 ô4	$0,670 \pm 0,009$ $0,660 \pm 0,011$ $0,620 \pm 0.015$ $0,730 \pm 0,015$	3,5 6,0 5,2

Key:

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- 1. Animal group
- 2. Number of animals
- 3. Testes weight (right + left) in relation to body weight, M + m diff

4. Control



Figure 1. The Conditions of Cells of the Spermatogenous Epithelium in the Seminiferous Tubules of Murine Testes Exposed to Microwaves.
A) Limited Desquamation of Germinal Epithelium. B) Marked Desquamation of Germinal Epithelium. C) Atrophy of Germinal Epithelium Cells. D) Presence of Giant Cells

Key: 1. Number of cases





Figure 2.

Figure 3.

- Figure 2. Giant Cells in the Seminiferous Tubules of Group II Mouse; Limited Epithelial Desquamation; Hematoxylin-Eosin; X 600
- Figure 3. Limited Desquamation of Germinal Epithelium in a Group I Mouse; Hematoxylin-Eosin; X 350



Figure 4.

Figure 5.

Figure 4. Marked Desquamation of Germinal Epithelium in the Testicle of a Group III Mouse; Hematoxylin-Eosin; X 350

Figure 5. Testicle of a Control Mouse; Hematoxylin-Eosin ; X 350

## UDC 614.87:537.868.029.64

#### EMBRYOTROPIC EFFECTS OF MICROWAVES

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 160-164

Article by A.N. Bereznitskaya and T.Z. Rysina

[Text] The embryotropic effects of nonthermogenic microwaves have been analyzed. The conclusions have been reached that microwaves may evoke developmental anomalies and increase embryonic and postnatal death on the basis of studies on the embryonic material and the pathology of the internal organs of feti. At the present time, any deviation from the normal during embryogenesis is regarded as a developmental anomaly, and the causative agents are regarded as teratogens [N.N. Konstantinova, 1960; P.G. Svetlov, 1962; I.C. Gerbelle, 1962; L.S. Persianinov, 1963; P.N. Aleksandrov et al., 1965; A.P. Dyban, 1970; and others].

Developmental anomalies may be general or local and anatomical or functional. According to N. Brock and T. Kreybig [1964], lag in development and small body weight and dimensions should be regarded as developmental anomalies. Early and later embryonic death also belongs in the category of developmental abnormalities.

Embryotropic effects may be uncovered by a variety of methods ranging from visual examination of external anomalies to analyses of histological, histochemical, and biochemical deviations in the tissues. However, even a very careful morphological examination of the fetus does not always uncover all of the malfunctions since the effects of various factors become apparent only during postnatal development. Thus, for example, small doses of ionizing radiation do not elicit noticeable deviations in embryos. However, during the postnatal period serious CNS malfunctions become evident [A.I. Piontkovskiy, 1964].

The object of the present investigation was to study the embryotropic effects of microwayes. The experiments were conducted on 77 white outbred mice. Females were exposed to microwaves  $(\lambda 10 \text{ cm}, 10 \text{ mW/sg cm power flux density}, 2 \text{ hr exposure/day})$ from the 1st to the 15th day of pregnancy. The appearance of a vaginal plug was taken as indicating the first day of pregnancy. For analysis of external and internal fetal developmental abnormalities, 21 of the irradiated mice and 21 of the control mice were sacrificed on the 15th day of pregnancy. The number of corpora lutea in the ovaries, as well as the number of living and dead embryos were recorded. On the basis of the difference in these three indices, determinations were made of the pre- and postimplantation embryonic deaths. The feti were weighed, examined macroscopically, the craniocaudal dimensions were measured, and they were fixed in Bouin's solution. Subsequently, the feti were studied by the method of Wilson I. Wilson, 1964, 1965

Microwaves of the intensity employed here increased embryonic mortality. The average number of living feti in females irradiated during pregnancy was  $6.95 \pm 0.13$  and  $7.8 \pm 0.08$  in the control females. Preimplantation death of zygotes obtained in the irradiated females reached  $13.06 \pm 2.39\%$ , and  $5.0 \pm 1.6\%$  in the control group; postimplantation mortality of the embryos was respectively,  $15.6 \pm 2.76\%$ , and  $6.47 \pm 1.88\%$ . Intrauterine death of embryos was noted in one irradiated female.

A large number of the embryos obtained from irradiated females evidenced hematomas in various locations on macroscopic examination, as well as cyanosis and a pale icteric skin. In addition, the size of the feti was markedly decreased (body weight  $633 \pm 0.74$  mg vs  $811 \pm 1.56$  mg in the control, and respective craniocaudal measurements of  $17.7 \pm 0.1$  mm and  $19.9 \pm 1.4$  mm).

Subthermal microwaves (10 mW/sq cm) evoked dysfunction in the process of embryogenesis. Examination of the feti for pathology of the internal organs showed that developmental anomalies were noted in the control group  $(37.06 \pm 4.48\%)$ , however, they were more frequent  $(56.16 \pm 3.89\%)$  in embryos that had been exposed antenatally to microwaves. Frequently, individuals were encountered with two or more type of anomalies of embryogenesis.

The spectrum of anomalies was rather extensive and included lesions of both the individual organs (eyes, liver, kidneys) as well as systems (cardiovascular, nervous, urogenital). Most frequently, both in the control and the experimental groups, we encountered adhesion of the diaphragm to the liver, unilateral and bilateral hydronephrosis, changes in the cardiovascular system, and frequently hematomas were present, particularly in the peritoneal cavity. Such embryogenic anomalies as an increase in the ventricles of the brain (hydrocephaly), and general underdevelopment of the embryos (small size, absence of eyelids, underdevelopment of the external ear) in the control group were completely absent, while in the irradiated embryos they accounted for 45.8% of the total number of developmental anomalies. These anomalies are illustrated by the microphotographs presented in Figures 1-3.

Some of the females exposed to microwaves during pregnancy had natural births. We studied the duration of pregnancy, the weight dynamics and the postnatal ontogenesis of the progeny. Data on the delivered progeny are given in Table 1.

Results show that there is a certain degree of agreement between the data obtained on the embryos and evaluation of the delivered progeny. The average litter size in the irradiated females was smaller than in the control females, and the same type of relationship prevailed between the living feti for the control, and the experimental groups. Body weights and dimensions of the neonates (in percentages) corresponded to fetal weight and dimensions. Postnatal ontogenesis of the mice irradiated antenatally lagged behind that of the control animals for the first few days. Later the difference became minimized and was statistically insignificant. The high postnatal mortality among the irradiated mice corresponded to the greater number of weak and debilitated individuals. In the control group, postnatal mortality was  $15.4 \pm 2.83\%$ ;  $36.7 \pm 5.5\%$  of the irradiated mice did not survive to the 21st day. The high mortality rate occurred largely during the first week of life (neonatal mortality). It is obvious that the difference in the weight dynamics and postnatal ontogenesis between the irradiated and control mice became minimized as a result of the death of the weakest neonates during the first few days after birth.

The state of the CNS was evaluated in the antenatally irradiated mice by means of their behavior in a T-shaped maze. Prior to the experiments the mice were weighed. The weight of the irradiated mice was lower  $(29.57 \pm 0.44 \text{ gms})$  than of the control animals  $(31.05 \pm 0.33 \text{ gms})$ .

The desired reflex developed much more rapidly in the control animals than in the irradiated mice. The number of trials required for the development of a reflex varied greatly, and on the average, was higher for the irradiated mice (19.45  $\pm$  2.0 against 12.7  $\pm$  1.67 in the controls).

The permanence of the reflex was tested one week and one month after it had been developed. Every animal was subjected to 20 tests. The number of positive responses were enumerated.

After a week, the percentage of mice which gave only a positive response was 2.5 times higher in the control animals than in
the irradiated group  $(20.8 \pm 8.3\% \text{ against } 7.5 \pm 4.3\%)$ . After one month, most of the animals gave a certain number of negative responses, and only one control mouse gave only positive responses. The number of negative responses out of 20 attempts in the irradiated mice, was almost twice as high as in the control animals  $(36.6 \pm 2.5\% \text{ against } 19.2 \pm 4.0\%)$ .

Resultant data indicated that some time after antenatal irradiation the mice weighed less, and one of the primary nervous processes was weakened -- the process of excitability. The results of our investigations confirm the conclusions of A.I. Piontkovskiy, et al [1970] who, in their studied on rats exposed to microwaves antenatally, found changes in the rates of postnatal ontogenesis, and delay in the development and alterations of conditioned avoidance reflexes, as well as inferior retention of such reflexes.

The totality of the data indicate that 10 mW/sq cm microwaves possess embryotropic effects. Furthermore, one must not forget that human and animal embryos differ in their susceptibility to noxious agents. Certain agents, such as thalidomide, for example, possess low toxicity in terms of laboratory animals, but have been shown to be very powerful teratogens for the human embryo. On the other hand, certain other substances (pyrimethamine) do not possess embryotoxic properties on the human embryo, but alter the process of embryogenesis of animals [V.I. Bodyazhina, A.P. Kiryushchenkov, et al., 1962; E.A. Kosmachevskaya, and N.A. Chebotar', 1968; A.P. Dyban, 1970; and others]. Therefore, our investigations only point to the potential embryotropic danger of microwaves. A definitive decision as to the danger of radiowaves for the human embryo can come only from a comparison of clinical and experimental data.

## Table 1. Studies on Progeny Born to Females Exposed to Microwaves During Pregnancy

4		۵. 	· · · · · · · · · · · · · · · · · · ·	
	(1) Показатели	(2) Контроль М±м_м5	(3) Облучение Лі±т <sub>анф</sub>	t
(4)	Количество самок	22	15	
(6)	лин Количество потомков	20,00±0,13 162	21,00 <u>+</u> 0,19 <b>98</b>	3,0
(7)	Среднее количество нотомков в по- мете	7,36 <u>+</u> 0,48	$6,53\pm0,74$	1,0
(0)		0±0.67	$2,04 \pm 1,41$ 28	1,3
(9)		$4,89 \pm 1,62$	28,57±4,55	4,9
(10	Кранпокаудальный размер новорож- денных, мм	$30,43\pm0,35$	28,90±0,48	2,6
(11	иес, г: Новорожденных . (.12) · · · · · · · ·	1,34±0.05	1,13±0,1/1	2,7
	≦ 3-A (13)	2,34	2,18	-
	на ГГЖ 5-й	3,26	3,04	_
(14	ре од 13-й	6,85	6,29	-
	북 더 21-й	8,76	8,42	_
(15) (16)	Сроки отлипания ушек, дин Сроки опушения, дин	3,57±0,17 5,71±0,17 13,85±0,17	4,41±0,22 6,27±0,28 14,25±0,26	3,5 1,8 1,3
(18)	Постнатальная смертность (количе- ство потомков, погнбинк к 21 дию)	25 15,4±2,83	<b>36</b> 36,7±5,5	.3,4

. . .

Key:

- 1. Index 2. Control, M + mdiff
- 3. Irradiation, M + mdiff
- 4. Number of females
- 5. Gestation, days
- 6. Number of progeny
- 7. Mean litter size
- 8. Number of stillbirths
- 9. Number of debilitated

Table 1. Continued

- Key: 10. Craniocaudal measurements of neonates, mm
  - 11. Weight, gm
  - 12. Neonates
  - 13. 3rd, 5th, etc.
  - 14. Days after birth
  - 15. Ear opening, days
  - 16. Hair cover, days
  - 17. Eye opening, days
  - 18. Postnatal mortality (number of progeny that died by the 21st day)



Figure 1. Second Fetal Section. a) Female Irradiated During Pregnancy; Absence of Eyelids; X 9. b) Control Female, X 12



Figure 2. Third Fetal Section. a) Female Irradiated During Pregnancy; Hydrocephalus; Brain Edema; X 10. b) Control Female, X 12.



Figure 3. Eighth Fetal Section. Female Irradiated During Pregnancy. a)Unclosed Abdominal Wall With Extrusion of an Intestinal Loop and Part of the Liver. b) Hepatic Hemorrhage. a) and b) At X 9

## UDC 614.87:537.868.029.64

THE EFFECTS OF RADIOWAVES ON THE GROWTH OF ANIMALS

Moscow O BIOLOGICHESKOM DEYSTVII ELEKTROMAGNITNYKH POLEY RADIOCHASTOT in Russian 1973 pp 165-168

Article by N.K. Demokidova

[Text] Experimental studies yielded results on the effects of high frequency (HF) and ultrahigh frequency (UHF) bands on a number of indices reflecting the state of the growth function of the adenohypophysis. There were no specific signs of stimulation of the latter. It has been assumed that the increase in body weight of young rats exposed to radiowaves in the presence of water, chloride and nitrogen retention, is due to a depression of metabolism which is mediated via the endocrine system.

Early studies on the biological effects of UHF waves reported that this band affected growth and development of animals [M.P. Derevyagin, 1936; K.P. Golysheva and N.M. Andriyasheva, 1937; M.A. Tikhonova, 1948; and others].

Later, Z.V. Gordon, et al. [1945] and Ye. A. Lobanova [1960], found an increase in the body weight of young rats exposed to SHF of thermal intensities on short-term multiple exposures.

It remains unclear, however, whether these authors observed a true stimulation of animal growth. Thus, M.P. Derevyagin explained the increase in the weight of pups under the influence of UHF as adiposity, and M.A. Tikhonova observed elongation of skeletal bones in young rabbits.

It is known that the growth of an organism is achieved by the growth hormone or the somatotropic hormone (STH) which is released by the secretory acidophilic cells of the anterior portion of the hypophysis, and which enhances the anabolic phase of protein metabolism in the presence of the appropriate complement of other hormones. Hormones produced by the adrenal cortex [S.M. Leytes and N.N. Lapteva, 1967] and the thyroid gland [Ya. M. Kabak and Ye. B. Pavlova, 1949; N.K. Demokidova and Ya. M. Kabak, 1957; N.K. Demokidova, 1958; R.D. Ray et al., 1954] are also important in the growth processes.

In the present study we attempted to investigate the effects of radiowaves on animal growth utilizing a number of modern methods. We investigated the dynamics of body weight, diuresis, and the urinary concentration of total nitrogen and chlorides. After the animals had been sacrificed, we determined the width of the tibial cartilage, and the weight coefficients of endocrine glands. Total nitrogen was determined by the method of Kjeldahl with isothermic distillation of ammonia in Conway dishes, and chloride was determined by the method of Votochek [B.I. Zbarskiy et al., 1954]. We also prepared several histological sections of the anterior portion of the hypophysis (Heidenhein's stain). In individual experiments we also determined the activity of alkaline phosphatase [A.A. Pokrovskiy, 1964], and the levels of NEFA in blood plasma [V.P. Dole, 1956].

In the experiments we employed 199 rats, most of which were males.

We studied the effects of UHF (69.7 MHz at intensities of 5.12 and 48 V/m in the electromagnetic field), and HF (14.88 MHz, 70 V/m, 0.185 A/m).

Data in the literature indicate that at a definite period in life, rats reach a plateau as far as growth goes, but after administration of STH they are capable of resuming growth, and significantly gain in weight [H.M. Evans and M.E. Simpson, 1931]. It is also known that the anabolic effects of STH are most pronounced in terms of dissimilatory changes in protein metabolism. As a result of this, we investigated the effects of UHF, both on adult and infant rats.

UHF Band. Irradiation of adult male rats with UHF (12 V/m for 1 hr/day for 1.5 months) resulted in a constant tendency toward an increase in body weight. The difference between the irradiated and the control animals at the end of the experiment amounted to 13 gms, or 42%. After the animals had been sacrificed a statistically significant increase in the weight of the hypophysis was found (P < 0.001). However, histologic study of the adrenohypophysis did not show a difference in the number of acidophilic cells between the irradiated and control rats. The width of the tibial cartilage in the irradiated animals was somewhat smaller than in the control rats (120 µm in the control, and 107 µm in the experimental animals). A tendency to decrease was observed on the part of the alkaline phosphatase activity of blood serum on the basis of two determinations conducted during the course of the experiment.

Therefore, despite indications of increase in weight, we did not find signs which would indicate stimulation of growth, and such changes as decrease in size of the cartilage and a decrease in the activity of alkaline phosphatase suggest activation of the adrenal cortex. Repetition of the experiment under the same conditions showed that one week and one month after initiation of irradiation there was a significant increase in the weight of the adrenal glands and the hypophysis (Table 1), as well as a decrease in blood plasma NEFA.

We studied the effects of UHF (5, 12, and 48 V/m) on infantile rats employing exposures of different lengths. We did not find any significant changes in body weight or in the weight of the endocrine glands. Only in the group of animals exposed to the greatest amount of incident energy (48 V/m, 4 hr/day, for 1.5 months) did we observe a statistically significant decrease in the weight of the thyroid gland, and a significant increase in the weight of the adrenal gland. In virtually every experiment, there was a tendency for the tibial cartilage to decrease in size.

HF Band. Table 2 demonstrates that irradiation of rats -- beginning with the infantile age -- with the HF band led to a significant increase in weight gain (the difference between the mean value for the irradiated and control animals amounted to 41 gms).

Under the influence of HF, there was a significant decrease in diuresis and inthelevels of total nitrogen and chlorides in the urine at the end of the experiment (Table 2).

After the animals had been sacrificed, a significant decrease in the weight of the thyroid glands and the adrenal glands was observed (Table 3).

However, two determinations on body length during the course of the experiment, did not show any significant differences between the irradiated and the control animals.

Therefore, under these experimental conditions, radiowaves did not stimulate the growth of rats. The increase of body weight of the animals under the influence of HF in the presence of water, chloride, and total nitrogen retention in the organism may be explained on the basis of depressed metabolism due to a certain degree of functional inhibition of the thyroid gland (and possibly, the adrenals) whose participation in this response is suggested by the decrease in its weight.

Table 1. The Effects of UHF on Weight Coefficients of Endocrine Glands in Adult Male Rats

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(1) Группа	(2) Длительность облучения	Весевий козффициент эндокринных желез		
жиротных		гилофиз	щитовид- ная железа (5)	надпочеч- лики (6)
Контроль Олыт (7)	Две недели (8)	3,12±0.28 3,97±0,22 p<0.05	8,4±0,82 8,3±0,35	$13.8 \pm 0.80$ $17.3 \pm 1.00$ p < 0.05
Контроль Опыт (7)	Один месяц (9)	3,31±0,10 3,71±0,13 p<0,05	_	13,6±0,52 16,3±0,99 <i>p</i> <0,05

Key:

1. Animal group

2. Length of exposure

. .

- 3. Weight coefficients of endocrine glands
- 4. Hypophysis
- 5. Thyroid gland
- 6. Adrenals
- 7. Control
  - Experimental
- 8. Two weeks
- 9. One month

Table 2. The Effects of the HF Band on Certains Aspects of Metabolism

(1) Группа животных		Длятельность облу- (2) чепня	(3) Суточное содержание, мг		
			(4) <sup>хлориды</sup>	(50 <sup>6щна азот</sup>	(6)
Контроль Опыт	(7) (8)	4 Фон (10)	13,2±1,4 17,3±1,5	$\begin{array}{c c} 40,9 \pm 10,0 \\ 47,7 \pm 6,2 \end{array}$	$3,1\pm0,47$ $4,1\pm0,30$
Контроль Опыт	(7) (8)	, Один месяц (9)	$21,1\pm2,2$ $14,8\pm1,6$ p<0,05	$84,4\pm11,3$ $72,4\pm12,2$	4,1±0,69 3,9±0,37
Контроль Опыт	(7) (8)	 Два месяца (11	) $23.8\pm2.5$ $23.6\pm1.8$	206,7±28,6 159,1±16,5	6,7±0,77 4,6±0,69
Контроль Опыт	(7) (8)	Четыре месяца (1	22.5±3,5 2)17,1±1,7	$158,3 \pm 16,1$ $152,0 \pm 14,4$	10,1±1,19 8,5±0,72
Контроль Опыт	(7) (8)	-ч Восемь месяцев (13)	13,8+2.3 $6,4\pm1,2$ p<0,05	$244.6\pm 21.8$ 109.6±18.6 p < 0.00.1	$8.7 \pm 0.96$ $3.6 \pm 0.79$ p < 0.01

Key:

- 1. Animal group
  - 2. Length of exposure
  - 3. Daily concentration, mg
  - 4. Chlorides
  - 5. Total nitrogen
  - 6. Diuresis, ml
  - 7. Control
  - 8. Experimental
  - 9. One month
  - 10. Backround
  - 11. Two months
  - 12. Four months
  - 13. Eight months

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Table 3. The Effects of HF on Body Weight and Length of Rats and the Weight Coefficients of the Endocrine Glands



Key:

1. Animal group 2. Body weight, gm

3. Body length, cm

4. Weight coefficients of the endocrine glands

5. Initial

6. Before sacrifice

7. After three months

8. After sacrifice

9. Hypophysis

10. Adrenals

11. Thyroid

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