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CONCEPTUAL BACKGROUNDS OF RADON BALNEOTHERAPY-RELATED LIVER RESTORATIVE MORPHOGENESIS IN EXPERIMENTAL AUTOIMMUNE DAMAGE

The experiment was conducted on nonlinear white mature male rats. Conducted lectin histochemical study, electron microscopic, morphometric and method of statistical analysis. Experimentally proved sanogenetic efficacy of radon balneotherapy autoimmune liver damage as analogue of autoimmune hepatitis, namely - braking of lymphocytic autoagression and its consequences, and especially manifestation active of restorative morphogenesis through adaptogenic and immunomodulatory (with increased of suppressor units) properties of radon.

Key words: hepatocytes, lymphocytic autoagression, radon therapy, reducing morphogenesis.

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From the standpoint of practical medicine topical is a nature and regularity of the recovery morphogenesis of organs. It is a universal basis for sanogenesis that are manifestations of individual reactivity of body, organically linked with the severity of systemic and local adaptive or compensatory reactions that occur in response to extreme factors. Reducing morphogenesis, in fact regeneration - necessary property of living should be considered as a separate phenomenon of adaptation [13, 14]. The ability to increase the adaptive properties of the body, to make adaptive stabilizing structures of the body seen in the action of natural physical factors used in balneology, particular this real effectiveness of radon water.

For a long time inefficient determined, but continues the traditional treatment of recurrent chronic diseases with developed autoimmune background. Such diseases should be considered as medical and social burden that requires frequent repetition rates of rehabilitation medical therapy. And the proportion of such pathology increased, including via polypragmasy in modern medical practice. In this issue fits known clinical form of liver disease - autoimmune hepatitis (AIH) that on the prevalence amounts to 20% of the total cases of chronic liver disease. The essence of disease is reduced to weaken immunoregulation. In the development of autoimmune reactions decisive role is played by suppressor activity of immunity. In patients decreased T-suppressor subpopulation of T-lymphocytes, so potentially autoreactive B-lymphocytes begin to react to own tissue antigens. The absence of opposition from the T-suppressor leads to uncontrolled IgG production. So, for the autoimmune process in the liver is typical the absence of suppressor activity in connection with exposure to T-suppressor autoimmune complexes that block their activity. Therefore, concerning the treatment of patients with autoimmune disorders should be considered that immunosuppressive therapy should concede methods, activating cellular immunity, raising the desired prestige of T-suppressor directed to inhibition of autoimmune process and activation of genetic basis of recovery morphogenesis. Such response of the immune system is formed in the body under the influence of term radon baths.

The aim was prove on the model of autoimmune liver damage in rats damping lymphocytic autoagression against hepatocytes and activation of their recovery morphogenesis under term of radon baths.

Materials and methods. Experiments conducted on nonlinear mature white rats - males aged 3 months with initial body weight 180-240 g. Animals were divided into 6 groups:Group 1: rats with experimental autoimmune liver damage, which did not perform any additional actions; Group 2: rats with experimental autoimmune damage that obtained radon baths under the scheme resort recommendations; Group 3: rats with experimental autoimmune damage that received baths with river water under the scheme of resort recommendations; Group 4 (control): rats that underwent radon bath under the scheme of resort recommendations; Group 5 (control, comparative): rats that received baths with river water under the scheme of resort recommendations; Group 6 (control): rats that were not subject to any additional actions. To simulate the autoimmune process in the liver of animals were injected subcutaneously complex hepatic antigen with incomplete adjuvant (by Vitebskii) in quantities and timing schemes that guarantee sustainable sensitization, eliminating the possibility of desensitization.

After 1 month from the start of sensitization of the rats in group 2 was performed baths with radon water at a temperature 37oC for 20 minutes. The content of radon in water amounted 0.5-1.5 kBq/l, which corresponds poorly radon water and provides a minimum total body irradiation [6, 8]. Similarly, rats 3rd group - baths with river water. These procedures were repeated at intervals of 1 day for 2 month (total 14 bathrooms). After 2 months performed second course of balneotherapy to thereby rats, according with radon and without radon also within one month with interval 1 day between baths (total

14 procedures). After 1, 2, 4 and 5 months after the beginning of sensitization of rats taken out of the experiment, took material for histological, electron microscopy and lectin histochemical subjects. In all cases, material was taken from the left lateral lobe of the liver. Rats of 4th and 5th control groups that obtained radon baths or baths with river water for 1 month and again after 2 months, derived from experiment to further research in the same terms as the rats from groups 1-3.

Research methods: histological, histochemical lectin (using four panels of lectins: peanut lectin (PNA); ricinus lectin (RCA); soybean lectin (SBA); lectin wheat germs (WGA)), histochemical, electron microscopic, morphometric.

Statistical analysis of the data was performed using standard package "Statistica6.1". Evaluated the accuracy of distribution characteristics for each of the received variational series. Calculated: arithmetic mean, standard deviation, average error. The reliability of differences between the values of micrometer values determined by the Student's criterion.

Results and discussion. Histological study. After 2 and 4 months after sensitization of rats of group 1 there was "invasion" of autoagressive lymphocytes, accompanied by sharply expressed degenerative changes and necrosis of hepatocytes and loss connections of hepatic plates, mainly in the central and peripheral zones of particles, which is typical for lobular hepatitis. Revealed a large number of plasma cells of varying degrees of maturity. Against the background of apparent alteration observed poorly expressed regenerative response of the liver parenchyma and stroma in the form of hypertrophy of hepatocytes in different zones of lobes, available dual core hepatocytes, often near the boundary of the plates, and fibroblast accumulation in the portal and septal areas.

In rat liver 3rd group was defined also active lymphocytic autoagression, fatty and vacuolar degeneration and widespread necrosis of hepatocytes with violation connections beams, plasma accumulation of portal tracts and parenchyma lobules in areas of necrosis of hepatocytes and congestion of active fibroblasts in portal areas. In comparison with that in the liver of rats in group 2 at the end of the 4th month after sensitization proved much smaller and not so pronounced changes as in rats of group 1. Total architectonic of hepatic lobules was saved, and on this background defined short rows of hepatocytes with degenerative and necrotic changes among which were located single cells. Weak proliferation of fibroblasts observed in portal tracts. At the end of the 5th month microscopic structure of liver recovered even more, necrosis did not detected. Only sometimes in individual hepatocytes were isolated lymphocytes. Sizes of hepatocytes increased, there are dual core hepatocytes and hypertrophied stellate macrophagocytes.

Analyses of morphometric studies have convincingly demonstrated the effectiveness of the use of radon baths in the correction of autoimmune changes in the liver. Thus, Table 1 shows the dynamics of increasing the relative amount of intact hepatocytes and decrease the relative volume of damaged hepatocytes in three areas of hepatic lobules of rats in group 2 with experimental pathology and radon correction in the projection of 100% used similar indicators in rat liver of group 1 (autoimmune damage without correction).

Table 1

The relative amount of intact and damaged hepatocytes in different zones of hepatic lobules of rats with experimental pathology and correction of radon

Terms of research	Areas of lobes	Intact hepatocytes (%)	Damaged hepatocytes (%)
2 months	Center lobular zone	104,5	81,4
	Peri lobular zone	104,6	74,8
	Intermediate zone	105,0	84,8
4 months	Center lobular zone	115,8	55,3
	Peri lobular zone	119,4	59,6
	Intermediate zone	118,9	63,9
5 months	Center lobular zone	138,5	35,5
	Peri lobular zone	139,1	37,0
	Intermediate zone	160,8	33,0

Electron microscopic at the end of 1 month from the start of sensitization in rats of group 1 in the cytoplasm of hepatocytes revealed a significant accumulation of fat inclusions and a large number of vacuoles. The structure of mitochondria changed: found compacted osmiophil matrix, cristae destruction. Number of grains with glycogen sharply reduced. After 2 months in the cytoplasm of hepatocytes determined pyknomorphous nuclei with irregular contours, extended perinuclear spaces; no nuclear pores.

After 4 months among hepatocytes with a significant amount of fat inclusions appear "dark cells". The cytoplasm of these cells has a high electron density and no nucleoli in nuclei. Near these hepatocytes observed lymphocytes and neutrophils. Defined destructive changes in sinusoid wall.

After 5 months after the beginning modeling autoimmune liver damage appear "dark" hepatocytes with deformed nuclei and nuclear membrane invagination. In the cytoplasm detected accumulation of fat inclusions, no organelles or their remains determined.

In rats of 3rd group after 2 months after the first course with river water baths ultrastructural signs of damage hepatocytes preserved. In their cytoplasm: inclusion of fat and enlightened vacuoles of various sizes; structure of mitochondria - broken. Enlightenment of bile capillaries expanded. Similar changes in rat liver in the 3rd group and after 4 months.

In rats of group 2 after one course of radon baths in the cytoplasm of hepatocytes decreases the amount of fat inclusion, the structure of mitochondria remains. In the vascular wall ultrastructural changes are not determined. After 4 months the structure of the nucleus and cytoplasm of hepatocytes not changed. Mitochondria look normal and are located in the cytoplasm in groups, there are grains of glycogen. Restores microvilli epithelium of bile capillaries. On the vascular surface of hepatocyte there are plural microvilli, indicating the capillary activity of metabolic processes that ensure the activation of intracellular regeneration of hepatocytes. After the second course of radon baths ultrastructure of hepatocytes is restored. Their cytoplasm is full of organelles, in nucleus are large nuclei, the endoplasmic reticulum membrane surface have a lot of ribosomes. Mitochondria with moderate osmiophil matrix and clear cristae. Bile capillaries limited by desmosomal contacts (Fig. 1).

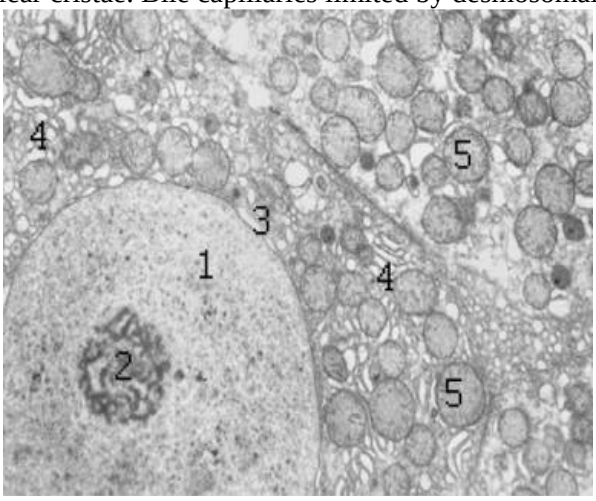

Fig. 1. The ultrastructure of rat hepatocytes of group 2 after 5 months after the beginning sensitization. Electron diffraction pattern. Increasing x 17000. Denotation: 1 - nucleus 2 - hypertrophied nucleoli 3 - local expansion of perinuclear space, 4 - well developed endoplasmic reticulum tubules, 5 - a large number of mitochondria.

This condition of hepatocytes fits their active operation. There are also "young" hepatocytes with normal organelles structure, with hypertrophied nucleoli and numerous pores in karyolemma. In the cytoplasm there are some grains or clusters of glycogen. Thus, electron-microscopic study showed that radon baths contribute to the development of intracellular regeneration and recovery ultrastructure of hepatocytes. It has been observed after the 1st course of treatment. A second course of radon baths made stabilizing effect adjustment.

According to our observations, in rats 1st and 4th groups detected accumulation of all investigated lectin receptors, both in parenchymal and vascular structures. Change of receptor activity of glycoconjugates on cell membrane in the cytoplasm of hepatocytes and macrophages in endothelial cells in the epithelium of the bile ducts and portal tracts connective tissue cells occurs gradually with the growth of pathological process.

In the WGA lectin receptors sialic acid residue cleaved from glycopolymers, so in hepatocytes and other liver structures appear absent in normal glycopolymers with final residue N- acetyl D- galactosamine.

At the same time in rats of group 2 after completing the first treatment course radon baths and even more significantly after the second course observed normalization of histological topography and quantity of lectin receptors of WGA and SBA in all structures, except for connective tissue stroma.

This dynamic especially demonstrative in the study of lectin receptors SBA. In control rats of 6th group these lectin receptors expressed on stellate macrophagocytes only. At the same time in rats of group 1 SBA lectin receptors are found in all parenchymal and vascular structures of the liver, not in the form of traces, and in large amount. Similar changes histological topography and quantity of lectin receptors PNA in rats found in 1st and 4th groups. In rats of 6th group these biopolymers absent in all cell elements of lobules, except endothelium central venous and in rats of 1st and 4th groups they are expressed in large numbers mainly stellate macrophagocytes, endothelial sinusoids in the epithelium of the bile duct and in the cells of the connective tissue of portal and periportal zones. In rats of group 2 observed normalization of receptor expression lectin PNA (differences are statistically significant, $p < 0.05$). Dynamics of changes RCA lectin receptors different by opposite direction. In rats of 6th group they are in large numbers expressed in stellate macrophagocytes in central venous endothelial lobules and other structures with the exception of hepatocytes, where their expression was insignificant.

At the same time in rats of group 1 RCA-positive amount of biopolymers is reduced in all structures and in hepatocytes and endothelial sinusoids disappear.

In rats of group 2 after the first course of radon baths increasing number of lectins RCA receptors in all structures except the cells and fibers of loose connective tissue stroma. However normal value does not reach this figure. After the second course the number of receptors lectins RCA even more increases, still remaining below normal. In the cells and fibers of the connective tissue stroma loose stored expression of these receptors.

Referring essentially of structural renewal of the liver in animals with experimental autoimmune liver damage without correction and radon balneotherapy, it must be said that the phenomenon of regeneration in damaged tissues are regulated by immune system. The last is not only a inducer that triggers the regenerative process but also as its regulator, changing the expression of intracellular or cell regeneration. T-cells in the dynamics of regeneration realize their morphogenetic potential [15]. Reduced function and / or the number of T-suppressor leads to violation of immunological tolerance and ensuring the proliferative stage regenerative process [3]. Radon baths perfectly play the role of immunomodulators, influencing the (more precisely, under the influence of alpha radiation) observed inhibition activity of Langerhans cells in the epidermis [12]. This induces the development of tolerance T lymphocytes skin. Prolonged effect relatively weak radiation causes some kind of hyperstimulation of lymphocyte multiplication and particularly T-suppressors. Electron-microscopic evidence of intracellular regeneration of hepatocytes and, in particular, identifying "young" hepatocytes means that radon immunotherapy increases morphogenetic potential of T-lymphocytes and macrophages of the liver contribute to its manifestation, which stimulates intracellular and cellular regeneration [15].

Conclusions

1. When you enter complex liver antigen (Vitebskii method) in the liver of rats developing pathological process that has structural features of autoimmune damage.
2. Radon baths are an effective method of sanogenetic correction. This is confirmed by morphological and morphometric studies. Expression lectin histochemical markers also normalized.
3. Baths with river water have no therapeutic effect. Morphological changes in the liver in this case are similar changes as without treatment in all terms of study.

Prospects for further research lies in the fact that further comprehensive *use lectin histochemical, immunological, electron microscopy, immunohistochemical and morphometric methods of research materials conducted experiments more fully define the components mechanisms of patho- and sanomorphogenesis in the dynamics of autoimmune liver damage.*

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Реферати

КОНЦЕПТУАЛЬНІ ОСНОВИ ВІДНОВНОГО МОРФОГЕНЕЗУ ПЕЧІНКИ ПІД ВПЛИВОМ РАДОНОВОЇ БАЛЬНЕОТЕРАПІЇ ПРИ ЕКСПЕРИМЕНТАЛЬНОМУ АУТОІМУННОМУ ПОШКОДЖЕННІ

Єгорова Т. М.

Експеримент проведено на білих нелінійних статевозрілих щурах-самцях. Проводили лектиногістохімічні дослідження, електронно-мікроскопічні, морфометричні та метод статистичного аналізу. Експериментально доведено саногенетичну дієвість радонової бальнеотерапії аутоімунного пошкодження печінки як аналога аутоімунного гепатиту, а саме – гальмування лімфоцитарної аутоагресії та її наслідків, і особливо, прояви активного відновного морфогенезу завдяки адаптогенним та імуномодуляторним (з посиленням супресорної ланки) властивостям радону.

Ключові слова: гепатоцити, лімфоцитарна аутоагресія, радонотерапія, відновний морфогенез.

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КОНЦЕПТУАЛЬНЫЕ ОСНОВЫ ВОССТАНОВИТЕЛЬНОГО МОРФОГЕНЕЗА ПЕЧЕНИ ПОД ДЕЙСТВИЕМ РАДОНОВОЙ БАЛЬНЕОТЕРАПИИ ПРИ ЭКСПЕРИМЕНТАЛЬНОМ АУТОИММУННОМ ПОВРЕЖДЕНИИ

Егорова Т.Н.

Эксперимент проведен на белых нелинейных половозрелых крысах-самцах. Проводили лектиногистохимические, электронно-микроскопические и морфометрические исследования, а также использовали методы статистического анализа. Экспериментально доказано саногенетическое влияние радоновой бальнеотерапии аутоиммунного повреждения печени как аналога аутоиммунного гепатита, а именно – торможение лимфоцитарной аутоагрессии и ее последствий, особенно проявления активного восстановительного морфогенеза, благодаря адаптогенным и иммунномодуляторным (с усилением супрессорного звена) свойствам радона.

Ключевые слова: гепатоциты, лимфоцитарная аутоагрессия, радонотерапия, восстановительный морфогенез.

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COULOMB INTERACTIONS AT THE SILICON WIRE-NERVOUS TISSUE INTERFACE

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In the past decade the “silicon crystal-nervous tissue” interface has been attracting huge interest due to the possibility of its application for neuro-computing and regenerative medicine. The key challenge of this research is to understand the mechanisms of the interface formation, and thus to prespecify properties of the interface and its operation. Despite numerous researches there is no clear definition of these mechanisms up to date. Here we present the study of the “silicon wire-nerve tissue” interface formed both in vivo and in vitro experiments. We have shown experimentally that there is a very good electrostatic adhesion of a nerve tissue to silicon wire in the living organism and in a medium, close to the physiological environment, as well. Strong interaction between the constituents of the interface was found to result from Coulomb mutual attraction of the oppositely charged surfaces of the nerve fiber and silicon wire. We measured the surface density of the charge at both surfaces of the interface using dual-gated SOI-nanotransistors. The surface density of positive charge at the nerve membrane and the surface density of the negative charge at the silicon wire were found to be ~$2x10^{13}$ cm^{-2} and ~$1x10^{14}cm^{-2}$, respectively. We analyzed Coulomb interactions at the interface during propagation of a nerve impulse and concluded that nerve impulse has to initiate a flexural wave in the nerve fiber and to generate an electronic surfacial wave in a space charge region of silicon wire. Moreover, the flexural wave has to provide metabolism in the nerve fiber and, hereby, vital capacity of the interface. On the other hand, the electronic wave in the space-charge region of silicon wire allows using it for extracellular recording of neuronal signal.

The interface of living-nonliving matter has become a subject of research in many fields of science. Study of the restoration of normal activity of an injured or diseased human nervous system attracts special attention. Intensive research in this area became more active several years ago [1-6]. It is evident that integration of living organisms with functional materials can create intelligent nano- and