MORPHOLOGICAL SUBSTANTIATION FOR ACUTE IMMOBILIZATION STRESS-RELATED DISORDERS OF ADAPTATION MECHANISMS

MORFOLOGICZNE UZASADNIENIE MECHANIZMÓW ADAPTACYJNYCH NAGŁEGO ZNIERUCHOMIENIA W ZABURZENIACH ZWIĄZANYCH ZE STRESEM

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ABSTRACT

Introduction: Nowadays, an individual is being constantly accompanied by stresses in his/her everyday life. Stress reactions, produced in the process of evolution, have become the organisms' response to emergency action or pathological factors and are the important link in adaptation process. However, the adverse course of stress reaction can lead to derangement of the adaptation mechanisms in the body and become the element of the pathogenesis of various diseases.

The aim: The study was aimed at morphological substantiation of derangement of adaptation mechanisms in white Wistar rats caused by the acute immobilization stress.

Materials and methods: 40 Wistar white male rats of 240-260 g body weight aged 8-10 months were involved into study. 20 laboratory animals were assigned to the main group and the rest 20 rats formed the control (II) group. Experimental stress model was simulated by immobilization of rats, lying supine, for 6 hours. Morphological examination of heart, lungs and kidneys was carried out after animals' decapitation, which proved the derangement of rats' adaptation mechanisms caused by the acute immobilization stress.

Results: It has been established that six-hour immobility of rats, lying in the supine position, led to the development of destructive phenomena, hemorrhagic lesions and impaired hemomicrocirculation. Microscopically, the acute immobilization stress causes significant subendocardial hemorrhages, plethora of vessels of hemomicrocirculatory flow with dysdiemorrhysis, myocardial intersticium edema in the heart. Histologically, immobilization-induced trauma causes significant hemodynamic disorders, spasm of arterioles and considerable venous hyperemia, concomitant with microthrombosis in kidneys; at the same time dystrophic lesions and desquamation of epithelium of renal tubules has been observed in renal corpuscles.

Conclusion: The abovementioned structural changes can contribute to origination and development of multiple lesions, demonstrating the morphologically grounded role of the acute immobilization stress as the factor for disorders of adaptive mechanisms.

KEYWORDS: stress, morphology, rats.

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INTRODUCTION

Nowadays, an individual is being constantly accompanied by stresses in the everyday life. Stress reactions, produced in the process of evolution, have become the organisms' response to emergency action or pathological factors and are the important link in adaptation process. However, the adverse course of stress reaction can lead to derangement of the adaptation mechanisms in the body and become the element of the pathogenesis of various diseases [1]. Therefore, study of the stress problem is the topical issue for researchers to date, and the analysis of organisms' adaptive capacities is crucial in current biological and medical sciences. Adaptive ability of the organism is limited; in this way according to experimental studies, even six-hour immobility can cause a number of morphofunctional alterations in the body of white Wistar rats [2-5].

THE AIM

The paper was aimed at morphological substantiation of the derangement of the adaptation mechanisms of white Wistar rats induced by six-hour acute immobilization stress.

To reach the objective of the study morphofunctional alterations, occurred in the heart, lungs and kidneys of white Wistar rats after six-hour immobility on the back have been investigated.

MATERIALS AND METHODS

40 Wistar white male rats of 240-260 g body weight aged 8-10 months were involved into study. 20 laboratory animals were assigned to the main (I) group and exposed to simulated acute immobilization stress. The rest 20 rats formed the control (II) group, which were housed in the regular conditions of vivarium at Ukrainian Medical Stomatologial Academy and were not involved in any other tests or experiments.

The acute immobilization stress was simulated by six-hour immobility of rats, lying on the back with fixed extremities from 9 a.m. to 3 p.m. The rats were killed on an empty stomach by decapitation under thiopentone anesthesia after 2 hours after immobilization. Euthanasia of rats from control group was made on an empty stomach by the similar method.

Fragments of organs (the heart, lungs and kidneys) were fixed in 10% neutral formalin and after dehydration in spirits of the ascending densities they were embedded into

paraffin according to conventional technique. Microtome specimens were stained by hematoxylin and eosin according to Hart Van Gizon and Mallory.

To prepare the semi-thin sections pieces of lungs were fixed in 4% glutaraldehyde in phosphate buffer, pH 7,4 and osmium fixative, dehydrated in spirits of the ascending densities and embedded into EPON-812. The semi-thin sections were stained with 0,1% toluidine blue.

The study was performed in compliance with the requirements of international principals of the "European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes" (Strasbourg,1985) and corresponding Law of Ukraine "For the Protection of Pet Animals" (No.3446-IV, 21.02.2006, Kyiv) [6-7].

RESULTS

Macroscopic analysis of the investigated organs of rats from Group I has shown that their lungs, in contrast to controls, were plethoric, and numerous foci of hemorrhages under the pleura and mucous membrane of the intrapulmonic bronchi have been found. Macroscopic examination of the heart and kidneys of rats, which were subjected to stress, showed no differences from the controls.

Microscopic analysis of the respiratory part of lungs of rats from Group I showed the foci of destruction of the alveolar walls. Numerous interalveolar enlarged pores have been detected in the thinned alveolar septa. Individual alveolocytes lost interconnection with the basal membrane, leading to its exposure; they have been found in the lumens of alveoli, forming the cellular conglomerations at various stages of destruction. Fibrin, red blood cells, destroyed and undamaged alveolar macrophages were also found in the cavities of alveoli. Clusters of alveolar macrophages have been found locally, and their cytoplasms were tightly filled with phagocytic material. Destructive changes in the intrapulmonic bronchi were demonstrated by the vacuolization of epithelial cells' cytoplasm and loss of epithelial layer integrity. Cellular detritus and red blood cells have been found in the lumen of the bronchi. Proper plate of bronchial mucosa was edematous – collagen and elastic fibers were fluffed up by amorphous substance. Vessels of the lungs' hemomicrocirculatory flow were plethoric with signs of blood stasis; morphological signs of their walls' destruction with numerous hemorrhages into lung tissue have been found (Fig.1).

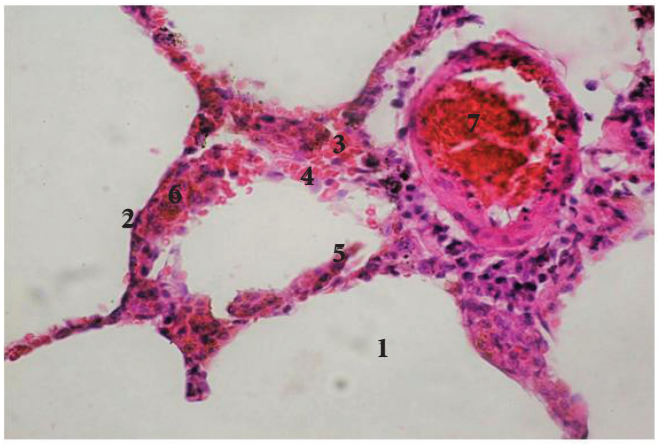

Fig.1. Rat lungs after simulation of the experimental model of acute immobilization stress. Microimage. Hematoxylin and eosin stain. Lens:40: Ocular lens:10: 1 – lumen of the dilated alveolar; 2 – alveolocyte; 3 – capillar; 4 – diapedetic hemorrhage; 5 – desquamated epithelium; 6 – alveolar macrophage; 7 – thrombotic masses in the lumen of arteriole.

Occurrence of the abovementioned alterations in rats from Group I was proved by the findings of morphometric study: acute immobilization stress causes significant

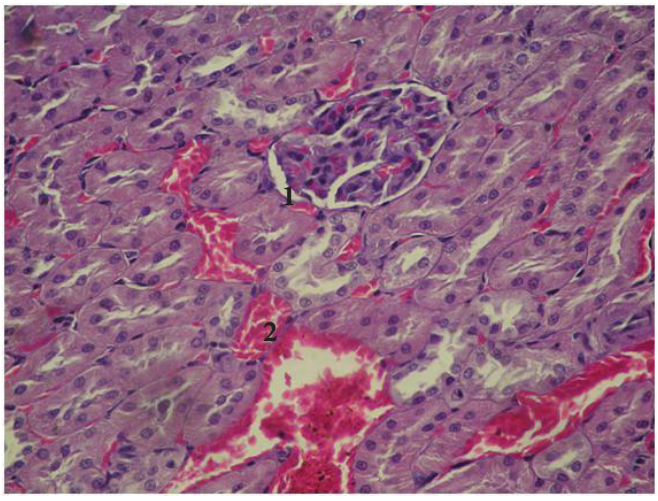

Fig.2. Rat myocardium after simulation of the experimental model of acute immobilization stress. Microimage. Hematoxylin-eosin stain. Lens:40: Ocular lens:10: 1 – plethora of myocardial vessels; 2 – hyperhydration of the myocardial intersticium.

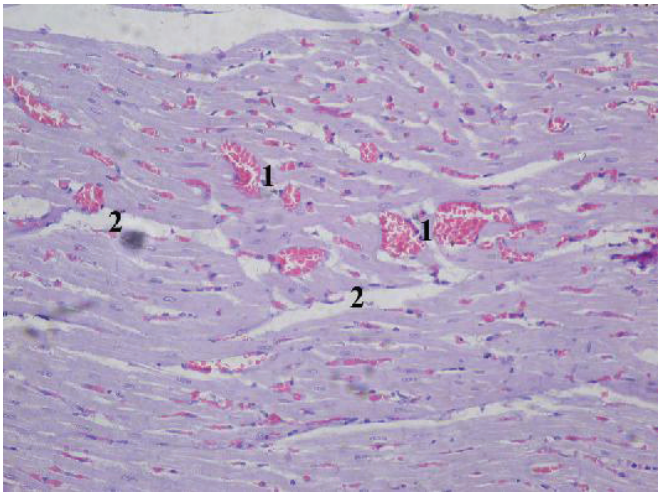

Fig. 3. Kidney of rat, subjected to experimental acute immobilization stress. Microimage. Hematoxylin and eosin stain. Lens:40: Ocular lens:15: 1 – enlarged Bowman-Shumlanski's capsule; 2 – venous plethora.

enlargement of alveoli (by 63,8%, p<0,01, in the left lung and 70,6%, p<0,01, in the right one), thinning of the alveolar septa (by 42,3%, p<0,01, on the left and 40,4%, p<0,01, on the right), thickening of epithelial layer of the intrapulmonic bronchi mucosa (in the smaller bronchi: by 85,9% (p<0,01) in the left lung and 87,4% (p<0,01) in the right lung; in the medium sized bronchi: by 65,7% (p<0,01) on the left and 65,9% (p<0,01) on the right). The mean values of the diameter of the capillary lumen in animals of the main group were two times higher as compared with controls and accounted for 7,31±0,71 and 3,62±0,25 μm on the left, and 7,64±0,69 and 3,68±0,22 μm on the right, respectively (p<0,01). In the left lung the diameter of the venule lumen was enlarged from 19,01±1,14 μm by 34,9% and reached 25,64±1,76 μm (p<0,01), and in the right – from 18,72±1,07 to 26,08±1,59 μm, i.e, by 39,3% (p<0,01).

Histological study of the heart of rats from the experimental group showed that acute immobilization stress affected its tissue. In all parts of the heart changes in endocardium first were manifested by its thickening and significant subendocardial hemorrhages with local phenomena of desquamation of endothelial cells. Myocardial tissue was plethoric, and the vessels of the hemomicrocirculatory flow were dilated with the phenomena of blood stasis. Signs of edema have been found in the connective tissue of myocardial intersticium (Fig.2).

Microscopic study of kidney specimens of rats from Group I showed that the acute immobilization trauma led to dilatation of lumen and thickening of walls of vessels of hemomicrocirculatory flow of this organ. Renal veins had signs of apparent plethora with the phenomena of microthrombosis. Lumens between the vessels of glomerules and capsules of renal corpuscles were dilated. The cytoplasm of the proximal renal tubules epithelium contained inclusions in the form of fine eosinophilic granules, sometimes with the formation of homogeneous hyaloid drops. Locally, these epithelial cells were separated from the wall, around which a pericanalicular edema was observed (Fig. 3).

No significant changes in heart, lungs and kidneys of animals from the control group were detected.

DISCUSSION

Morphological studies show that six-hour immobility of rats, lying in the supine position, causes the apparent stress-reaction in the body of laboratory animals that leads to significant morphofunctional alterations, but not to the development of favorable adaptive responses.

According to the concept of the pathogenesis of diseases by the theory of adaptation, health is considered as the manifestation of the optimal adaptation of the body with support of its homeostasis to the surrounding changes [8]. General nonspecific adaptation syndrome is based on activation of the adrenergic and hypothalamic-pituitary-adrenal stress-realizing systems of the body, resulted in hypertrophy of the adrenal glands with increased secretion of glucocorticoids and catecholamines. Under the influence of catecholamines, especially the adrenaline, phospholipases are activated that initiate the process of peroxidation of lipids of biomebranes. This can lead to derangement of the regulatory mechanisms and occurrence of significant morphofunctional disorders in the body which can become the element of the pathogenesis of multiple pathological syndromes [9, 10].

Recently, comprehensive studies of the progress and violations of the mechanisms of adaptive responses that occur in the body under the influence of various stressors have gained a significant relevance. Notwithstanding the considerable number of publications on this issue, scarce papers with morphological substantiation of pathogenic mechanisms of immobilization stress-induced violations of the adaptive mechanisms have been found to date; medical publications are more concerned about its pathophysiological, biochemical and clinical aspects [11-17].

The study shows that acute immobilization stress leads to the development of destructions, hemorrhagic alterations and impairment of hemomicrocirculation in the respiratory part of the rats' lungs and intrapulmonic bronchi. Microscopically, the acute immobilization stress is accompanied by the significant subendocardial hemorrhages, plethora of vessels of cardiac microcirculatory flow with phenomena of blood stasis and edema of myocardial intersticium. Histological study of specimens of kidneys showed that immobilization trauma caused significant hemodynamic disorders, spasm of arterioles and prominent venous plethora with phenomena of microthrombosis; dystrophic changes and desquamation of epithelium of renal tubules has been detected in renal corpuscles of rats from experimental group.

The abovementioned structural changes can promote origination and development of multiple lesions, demonstrating the morphologically grounded role of the acute immobilization stress as the factor for disorders of adaptive mechanisms.

CONCLUSION

The study shows that acute six-hour immobilization stress causes the derangement of the adaptation mechanisms in white Wistar rats, accompanied by the apparent structural and functional changes in the heart, lungs and kidneys of laboratory animals.

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