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SEX FEATURES OF INTERMITTENT HYPORBARIC HYPOXIA ACTION ON OXIDATIVE MODIFICATION CONTENT AND ON PRODUCTS OF LIPID PEROXIDATION IN THE GUM TISSUES OF RATS UNDER THE PHOTOPERIOD OF DIFFERENT DURATION

Abstract. *We have established in experiments on albino rats of different ages and sex that a systemic continued intermittent hypobaric hypoxia causes a slight decrease in the intensity of lipid and protein peroxidation and a significant reduction in enzyme activity in the gum tissues. Antioxidant-prooxidant index indicates the superiority of antioxidant activity in the tissues of the gums, and, respectively equal, to the strengthening of the resistance processes in these tissues.*
Key words: *age, rats, hypoxia, gums, photoperiod.*

The condition of epithelial tissues and mucous membranes covering the mouth largely depends on the systemic organismal factors, including the influence of hormonal factors, which have drawn an increasing attention. Effects of estrogen and progesterone of both endogenous origin and hormones of replacement therapy and contraceptives on the periodontal tissue of the female body in different ages including puberty period have been described in details [1]. When the body is affected by damaging factors, including bacteria, hypoxia and others, exept LP, an activation of oxidative modification of proteins (OMP) occurs. After the oxidative modification proteins become highly sensitive to proteolysis. Therefore, the determination of the total proteolytic activity (TPA) has been widely used in modern experimental dentistry [2, 3]. The systemic prolonged intermittent hypobaric hypoxia caused a slight decrease in the intensity of lipid and protein peroxidation and a significant decrease in activity of antioxidant enzymes in the gum tissues of immature male rats [4].

As the research of recent years has shown, hypoxia is seen not only as a damaging factor. Intermittent hypoxia becomes more practical as a factor for strengthening resistance under conditions of hypoxic hypoxia. It is based on the double function of ROS, which are formed under the influence of hypoxia. On the one hand, excess formation of ROS causes an oxidative stress, destroying structures of cells, including lipids and membranes, proteins and DNA. On the other hand, various processes, caused by low or moderate concentrations of ROS protect cells from oxidative stress caused by the same ROS and contribute to the restoration or maintenance of "redox balance" ("redox homeostasis") [5].

Objective: To determine functional properties of gum tissue of rats depending on age and gender and performance of the systemic intermittent hypobaric hypoxia and photoperiod of various duration.

Materials and methods. The study was conducted on 42 albino nonlinear sexually mature and immature male rats and sexually mature and immature females. The research was performed in compliance with the main provisions of GLP (1981), Rules of work using experimental animals (1977), Council of Europe Convention on the Protection of Vertebrate Animals used in experiments and other scientific purposes of 18.3.1986, EEC Directive №609 of 24.11.1986 and MHP of Ukraine №281of 01.11.2000.

We developed an original technique with simultaneous using the long-term intermittent hypoxic exposure to changes in photoperiod with different directions. We used a hypobaric hypoxia which was equivalent to an altitude of 4000 m above sea level, which was created in a transparent flow-type pressure cell by sucking air out with a vacuum compressor. The rate of "lifting" the animals to a given value was 24 km / h. Sessions of hypoxia lasting 2 h. were conducted every day from 9.00 to 11.00 for 14 days against the background of three modes of

lighting: natural light, typical for spring-summer period with duration of light period of day 15 hrs., continuous round clock lighting with intensity of 500 lux and permanent round clock darkness. The changed lighting mode was introduced a day before the first session of hypoxia, and the animals stayed 15 days under the changed photoperiod.

On the next day after the last session of hypoxia the animals were taken out of the experiment by decapitation under a light ether anesthesia. Euthanasia was performed in the morning from 9.00 to 12.00 for all groups of animals. After the exsanguination, the cold gum tissue samples were weighed on a torsion balance and homogenized in 1.2 ml of cooled TAE buffer and in 2 ml of chilled borate buffer. The homogenate was frozen and stored in a freezer at - 20˚C until it was used in a laboratory research.

Results and discussion. Under the normal lighting sex differences in the contents of OMP products were found in the gum tissues. For instance, the content of main oxidation-modified proteins in males was by 9.0% higher compared to females ($p < 0.05$), and the neutral ones were the same in both sexes.

Under the influence of intermittent hypobaric hypoxia such differences became even more pronounced. The immature female rats responded less actively on 14-day hypoxia than males. The females tended to have an increase in the main OMP products by 25.0% ($p > 0.05$), whereas the males experienced significant reduction in both neutral products ($p < 0.03$), and the main OMP ($p < 0.05$). The response of the animals to hypoxia is an important test (factor) for the ability of the gum tissue to withstand external hazards. Hypoxia is one of such natural factors. Reducing the partial pressure of oxygen leads to the production of reactive oxygen species (ROS), to formation of free radical compounds that have strong damaging effects on cells and tissues. At the same time, we know that the ROS have a dual function: their excessive production leads to oxidative stress and damage cellular structures; their low or moderate number is necessary for the formation of the defence, including the antioxidant one [5].

Considering the above, the results of the effects of hypoxia on the contents of OMP products in gum tissues of female rats can be regarded as a failure of the antioxidant system to resist the increasing content of OMP products, while the defence system in males turned out to be more effective. To some extent it is consistent with the existing idea that testosterone has a protective effect on the gums and periodontal tissues in general [1]. Since our results on the effects of hypoxia on the content of OMP products in gum tissues were obtained in immature animals, in which the testes do not produce testosterone yet, then, perhaps, this protective ability is a property of a male body that is genetically determined.

Changes in photoperiod of keeping animals duration have a significant impact on the content of oxidation-modified proteins in the tissues of the gums, provided the impact of intermittent hypobaric hypoxia on the animals. This is especially evident in a series of experiments, when hypoxia was used under 15 day darkness to which the animals were exposed (stimulation of pineal melatonin production). The females of this series of experiments had a significantly lower content of OMP products in the gum tissues (the main one by 22.9% and the neutral one by 16.5%) compared with animals that were only exposed to hypoxia ($P < 0.05$). Under the similar conditions of experiments the content of OMP products in the gum tissues of male rats remained the same. However, a combined effect of hypoxia and darkness reduced significantly the content of OMP products in the tissues of the gums (main OMP by 21.9% neutral one by 23.2% ($p < 0.05$)) in male rats compared to intact animals, kept in conditions of normal light and barometric pressure. In females OMP process in the gum tissues under conditions of combined effect of hypoxia and darkness was the same as in the intact animals. We have not found sexual features as to the content of OMP products in the gum tissues in the experiments while using the action of hypoxia against the background of 15-day lighting ("physiological" hypophys-ectomy).

The results led us to the need to introduce differentiation factors that cause activation of lipid peroxidation and oxidative modification of proteins into two groups in both sexes: 1) hormonal ones, caused by respective sex hormones available in the relevant sex; 2) non-

hormonal ones, which occur and act on the gum tissues in immature animals.

Conclusions. There are some gender features as to the defense system response in the gum tissue of sexually immature animals. The female animals respond with a significant reduction in products of lipid peroxidation and activity of antioxidant enzymes, deterioration of general condition of gum protection system and the lack of changes in content of oxidation-modified proteins in the gums to continued lighting. Males respond to light much less actively than females by a reduction of diene conjugates alone and the activity of antioxidant enzymes, with decreasing content of oxidation-modified proteins and proteolytic activity of the growth of collagen. Under long darkness sex differences in physiological state of the system to protect the gums appear even more clearly. Females respond to the darkness by reducing the content of diene conjugates in the gums, as well as malondialdehyde awhile the antioxidant enzyme activity and the content of oxidation-modified proteins remain unchanged and the integral indicator of the antioxidant-oxidative status is high. Males in the dark respond by high levels of lipid peroxidation products in the gums; the activity of antioxidant enzymes remains unchanged, and the levels of oxidation-modified protein and antioxidant-oxidant index reduce.

Prospects for further research. A study of gender and age features in the response of the gum tissues on hypoxia based on stomatological practice as to various lesions depending on sex and age.

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