The State of Peroxidation Processes in the Basal Nuclei of the Brain in Conditions of an Altered Photoperiod

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The state of peroxidation processes in the basal ganglia (caudate nucleus, globus pallidus, nucleus accumbens, amygdalar complex) of the rat brain was studied in conditions of an altered photoperiod. The results showed that changes to the normal photoperiodicity led to metabolic changes in these brain structures. In conditions of constant light, the basal nuclei showed increased lipid and protein peroxidation processes, with increases in the intensity of fibrinolysis and proteolysis and decreases in the activities of antioxidant defense system enzymes. In conditions of constant darkness, there were changes in fibrinolytic and proteolytic processes which were in different directions in different structures, along with decreases in free-radical processes on the background of the accumulation of modified proteins.

KEY WORDS: peroxidation processes, basal nuclei, altered photoperiod, constant darkness, constant light.

One of the most significant and constant environmental factors acting on living organisms is the photoperiod. Regular alternation of day and night have had the result that living beings have developed innate periodic programs (circadian rhythms) for the metabolism of substances and, correspondingly, functional parameters. Changes in the photoperiod induce changes in measures of an organism’s activity, producing mismatch in biological rhythms and a state of desynchronosis, which may lead to the appearance of new pathology or the deterioration of existing pathology.

Synchronization of circadian rhythms with the external “light–dark” rhythm in mammals involves two morphological formations – the suprachiasmatic nuclei of the hypothalamus and the epiphysis [3, 5]. Both the suprachiasmatic nuclei and the epiphysis influence the state of the body via the activities of other brain formations with which they interact. These formations include the hypothalamic centers regulating endocrine activity, as well as the basal ganglia of the forebrain and limbic structures [1, 10]. The places and roles of these parts of the brain in the body’s chronoperiodic system have not been definitively established. Studies using a variety of behavioral models (forced swimming, daily locomotion, reproductive behavior) have demonstrated the involvement of the striopallidal system and amygdala in forming the circadian rhythms of behavioral reactions and changes in behavioral measures in response to different photoperiods [2, 4, 22, 26, 27]. The data obtained in these studies provide evidence that the functional activity of the basal nuclei is constantly modulated by the photoperiod. However, these studies did not identify which metabolic shifts resulted from changes in the photoperiodicity in these brain structures, and constitute the essential basis of changes in their functional state.

Thus, the aim of the present work was to study the effects of alterations to the photoperiod of the intensity of metabolism in the basal nuclei, which are morphologically complex brain structures with various functions, responsible for the development of varying pathologies. The criteria used for assessing the level of metabolism were universal “marker” assessments of the pathological influences of the study factors on the body, i.e., measures of peroxidation processes.

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METHODS

Studies were performed on 48 juvenile male rats. Photoperiod changes were modeled weekly using different light regimes: 1) the natural alternation of the light and dark phases, i.e., the standard photoperiod; 2) constant illumination; and 3) constant darkness. This model of photoperiod alterations yields marked changes in biochemical measures, as keeping animals in constant light or darkness corresponds to modeling of the states of epiphyseal hypo- and hyperfunction respectively; on the other hand, such “extreme” types of photoperiod changes (exclusion of one of the phases of the light cycle) allows clear definition of the directions of the effects of shortening and lengthening the illumination period on the study parameters in the basal nuclei.

The following brain structures were selected for study: the caudate nucleus, globus pallidus (pallidum), nucleus accumbens (accumbens), and amygdalar complex (amygdala). Assignment of the amygdala to the basal nuclei was based on classical neuroanatomy [15]. More recent physiology regards it as part of the limbic system. Nonetheless, all the basal nuclei were found to be in tight morphological relationships which, being functionally specific, are nonetheless involved in the mechanisms of a variety of general processes [21, 23, 24].

Brain homogenates were prepared in 0.05 M tris-HCl buffer (pH 7.4). Samples of structures were obtained by pooling materials from two animals. The state of peroxidation processes was assessed in terms of the activity of free-radical processes, the major enzymes of antioxidant defense, and tissue fibrinolysis and proteolysis. The activity of free-radical processes was assessed in terms of the levels of lipid and protein peroxidation. Lipid peroxidation processes were assessed in terms of the content of primary products, i.e., diene conjugates [6], and secondary products, i.e., malondialdehyde [17]. The level of oxidation of modified proteins was evaluated in terms of the content of neutral and basic aldehyde- and ketone dinitrophenylhydrazone detected at 370 nm (neutral) and 430 nm (basic) [12]. The state of the antioxidant defense system was evaluated in terms of the activities of the major antioxidant enzymes: superoxide dismutase [20], catalase [11], and glutathione peroxidase [18]. The intensity of tissue fibrinolysis in brain structures was measured using the reaction with azofibrin, assaying total fibrinolytic activity as well as enzymatic and non-enzymatic fibrinolytic activity. Proteolytic activity was measured using the reaction with azo compounds. The intensity of proteolysis was measured using azoalbumin, azocasein, and azocollagen [18]. Experimental results were analyzed by variational statistics run on Statistica 5.0 [7] and Student’s t test. Relationships between changes in measures were assessed using correlation analysis with non-parametric Spearman rank correlation coefficients, r. The significance of the effects of study factors on peroxidation processes was assessed using variance analysis. In single-factor analysis, the independent factors were: constant darkness, constant light, altered photoperiod. The dependent variables were the biochemical measures studied in the basal nuclei. Changes were regarded as statistically significant at \( p \leq 0.05 \).

All experimental manipulations were performed with consideration of ethical norms for animal studies and met the requirements of the Animal Protection Society.

RESULTS

Results obtained from studies of peroxidation processes in the basal nuclei of animals kept in conditions of an altered photoperiod showed that alterations to normal photoperiodicity led to metabolic changes in the brain structures studied here.

Animals kept in conditions of constant light showed intensification of liperoxidation processes in individual structures due to increases in diene conjugate levels, e.g., 19.5% in the nucleus accumbens and 10.9% in the globus pallidus (Table 1). The significance of the effects of constant light on diene conjugate concentrations in these structures was supported by factorial variance analysis (\( F_{1,11} = 15.17, p = 0.0025 \) in the accumbens, \( F_{1,11} = 7.96, p = 0.02 \) in the pallidum). At the same time, in conditions of constant darkness, malondialdehyde contents in the nucleus accumbens and amygdala decreased significantly, by 17.9% (\( F_{1,11} = 13.53, p = 0.004 \)) and 10.4% (\( F_{1,11} = 9.25, p = 0.011 \)), respectively.

Changes in the photoperiod had greater effects on the concentrations of protein peroxidation products in the basal nuclei (\( F_{1,82} = 21.02, p = 0.00002 \) for neutral products; \( F_{1,82} = 49.07, p = 0.00 \) for basic products).

In conditions of constant darkness, the levels of neutral protein peroxidation products increased in all study structures from those seen with a normal photoperiod: by 24.4% (\( F_{1,12} = 17.32, p = 0.0013 \)) in the nucleus accumbens, by 25.3% (\( F_{1,12} = 17.63, p = 0.0012 \)) in the caudate nucleus, by 52.6% (\( F_{1,12} = 19.03, p = 0.001 \)) in the pallidum, and by 68% (\( F_{1,12} = 40.45, p = 0.00004 \)) in the amygdaloid complex. These conditions also provoked increases in the concentrations of basic protein peroxidation products: by 64.7% (\( F_{1,12} = 42.06, p = 0.00003 \)) in the nucleus accumbens, by 34.1% (\( F_{1,12} = 20.34, p = 0.0007 \)) in the caudate nucleus, by 16.5% (\( F_{1,12} = 7.60, p = 0.017 \)) in the globus pallidus, and by 64.0% (\( F_{1,12} = 41.16, p = 0.00003 \)) in the amygdala. Constant illumination increased the level of protein oxidation in the nucleus accumbens by 17.6% (neutral: \( F_{1,12} = 8.37, p = 0.014 \)) and 57.1% (basic: \( F_{1,12} = 119.79, p = 0 \)) in the caudate nucleus by 55.2% (basic: \( F_{1,12} = 86.09, p = 0.00001 \)), and in the globus pallidus by 12.1% (basic: \( F_{1,12} = 7.42, p = 0.020 \)).

Alterations in the photoperiod affected measures of antioxidant defense in the basal nuclei (Table 2). In conditions of constant darkness, superoxide dismutase and cata-