A.V. Gerasimov, V.P. Kostyuchenko, E.Yu. Varakuta, S.V. Logvinov

THE EFFECT OF LIGHT ON THE FORMATION OF OSMIOPHILIC BODIES IN PINEALOCYTES AND CALCIFICATION OF THE PINEAL GLAND

DOI 10.25789/YMJ.2023.81.04

УДК 611.814.53:612.4.09:616-008.61/.63

In order to verify the assumption that changes in the content of osmiophilic bodies (OB) in pinealocytes in rats are a morphological marker not only of shifts in secretory activity and cal-cification of the pineal gland responding to round-the-clock lighting, but also reflect violations of pineal biorhythm, by methods of light and transmission electron microscopy, microrentgenospec-tral analysis in an experiment on rats, exposed to 48-hour exposure to bright light, the numerical density of single and grouped OB, their chemical composition was estimated. It is concluded that the grouped OB with the material of disposed mitochondria and calcifications are a residual manifestation of desynchronosis. Their content increases at an earlier time after exposure than the total content of various types of OB.

Keywords: pineal gland, exposure to light, rats, ultrastructure, chemical composition.

Introduction. In the northern latitudes beyond the Arctic Circle, such a phenomenon as a polar day is observed. In connection with the active development of the Arctic, the northern sea route, the study of the effects of round-the-clock lighting on the body is becoming increasingly relevant. The pineal gland plays an importance role in the regulation of daily biorhythms. Modeling the effect of 24-hour illumination with 48-hour exposure to bright light on white rats, we showed that hyperilluminated animals develop temporary desynchronosis of the daily activity of the pineal gland, which resulted in an increase in the content of calcified cells in pinealocytes [3, 4]. During the study, a complex of morphological markers of functional activity of the gland was considered [7], which made it possible to assess, among other things, the state of the mitochondrial apparatus, which is the exclusive site of synthesis of the main pineal hormone melatonin [10]. The specific volume of mitochondria in the cytoplasm of most large light pinealocytes (type IB) significantly exceeds the value of the indicator characteristic of all other types of cells of a neural nature, including hypothalamic neuroendocrine [1]. Mitochondria in pinealocytes, in addition, exhibit a diurnal fusion-division rhythm, and are subject to mitophagy [11]. Disposal is accompanied by the formation of single and grouped into aggregates from [2]. It is assumed that the utilization of damaged mitochondria is associated with the formation of OB accumulating in small light pinealocytes (type IA), dark pinealocytes (type II) and degenerating pinealocytes (type III), and that the formation of grouped OB is a consequence of a violation of the daily rhythm of mitochondrial fusion.

The aim of the study was to analyze the formation, chemical composition and changes in the numerical density of single and grouped OB in pinealocytes of the pineal gland in rats after the cessation of round-the-clock illumination with bright light.

Materials and methods of research. The work was performed on 80 mongrel white male rats weighing 180-200 g in accordance with the rules of laboratory practice (Order of the Ministry of Health of the Russian Federation No. 267 dated 06/19/2003). The illumination of animals in the daytime (from 8 to 20 hours) was 200 lux. The effect of round-the-clock lighting was modeled by placing rats in hyperilluminated cells for 48 hours (6 lamps LB-40, illumination 3500 lux). Experimental and control animals were withdrawn from the experiment at 11-12 hours of the day after 24 hours, 10, 30 and 180 days after the cessation of exposure to bright light. The gland was taken after decapitation of rats under ether anesthesia, fixed in 2.5% glutaraldehyde at 0.2 M cacodilate buffer (pH 7.4), post-fixed in 1% osmium tetraoxide solution, dehydrated in ethanol, poured into epon. The sections were made using LKB-III ultratome (Sweden), contrasted with uranyl acetate and lead citrate, and examined in a JEM-100 CX II transmission microscope (JEOL, Japan). The electron microscope JEM-210 (JEOL, Japan) and the energy dispersive spectrometer Oxford Instruments X-Max (Great Britain) were used for the microrentgen spectral analysis. In sections of the gland with an area of 0.06 mm2, the numerical density FROM was calculated using a 368-point test system. The data was processed using the software package "Statistica for Windows", version 7 (StatSoft Inc., USA).

Results and discussion. In rats, in type IA pinealocytes, along with OB, mitochondria with inclusions of osmiophilic material and grains of calcifications are detected, which are isolated by cisterns of the endoplasmic reticulum with the formation of autophagolysosomes. Autophagolysosomes filled with amorphous osmiophilic material contain lamellar structures, calcified grains, lipids. It is not so common in autophagolysomes to observe complete digestion of the recycled material, as well as the presence of calcified lamellar material in the intercellular space. In the daytime, when mainly small mitochondria are detected in the zone of the inactive Golgi complex of light pinealocytes in control animals, some of them, thanks to the folds of the outer mitochondrial membrane, can interact with each

GERASIMOV Aleksander Vladimirovich – professor of the department of histology, embry-ology and cytology of the Siberian State Medical University of the Ministry of Health RF, MD, associate professor, e-mail: av-gerasimov62@yandex.ru; **KOSTYUCHENKO Vitta Petrovna** – senior teacher of the department of histology, embryology and cy-tology of the Siberian State Medical University of the Ministry of Health RF, PhD, e-mail: vittacos@yandex.ru; **VARAKUTA Elena Yurievna** – head of the department of human anatomy with the course of topographic anatomy and operative surgery of the Siberian state medical university of the Ministry of Health RF, MD, e-mail: varelen@rambler.ru; **LOGVINOV Sergey Valentinovich** – head of the department of histology, embryology and cytology of the Siberian state medical uni-versity of the Ministry of Health RF, MD, professor, e-mail: s_logvinov@mail.ru

other and with OB, turning into osmiophilic clear-cut structures (Fig. 1).

In larger numbers, grouped and single OB (up to 7.5 microns in size) are found in control and experimental animals in pinealocytes of type II and III, as well as in apoptotic corpuscles. The OB is involved in the genesis of concretions, which is consistent with the data of other authors and is considered as a physiological process, and calcification of the contained OB (its amorphous type) itself is a reflection of aging and/or degenerative condition of the pineal gland [5, 6, 9]. Only sometimes at the site of pinealocyte death in rats, crystals with a size of 1.2 × 0.2 microns and rosettes of crystals are organized (Fig. 2).

In terms of the ratio of calcium and phosphorus, the crystals are close to hydroxyapatite [8]. Calcium, phosphorus, and sulfur are rich not only OB wich utilize mitochondria, but also in organelles capable of melatonin synthesis themselves, depositing, in addition, Ca^{2+}, involved in oxidation and phosphorylation, containing substances that include cysteine, for example, glutathione (Fig. 3).

After the cessation of round-the-clock illumination, the numerical density of grouped pinealocytes in the cytoplasm increases and exceeds the control by 10-180 days. The increase in the total numerical density is measured by 30-180 days (Fig. 4). Since hyperilluminated rats developed desynchronosis with suppression and then inversion of the exact rhythm of pineal gland activity [3], it can be assumed that osmophilic material and calcifications accumulated in small mitochondria with inhibited activity of IB pinealocytes activate utilization organelles with similar changes. The OB are formed, type IB cells acquire morphological features of type IA cells, the fusion of small mitochondria into large ones synthesizing melatonin is complicated. Small mitochondria without signs of osmiophilia and with osmiophilic material are more often combined after the cessation of exposure into clear-cut structures, often in combination with OB, therefore, after 24 hours in the phase of inversion of the circadian rhythm, there is no significant increase in the specific volume in the cytoplasm of pinealocytes of mitochondria in hyperilluminated rats [4]. In large mitochondria, not only the enlightenment of the matrix is noted, but also the deposition of osmiophilic material. The specific volume of mitochondria in pinealocytes in rats increases 10 days after the cessation of light exposure at the peak of the adaptive response to

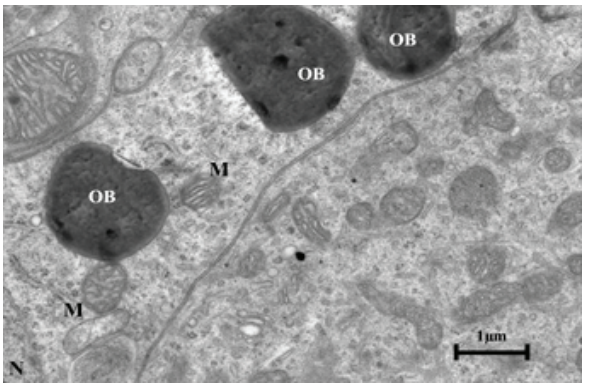

Fig. 1. Grouping of small mitochondria (M) and OB (N – pinealocyte nucleus)

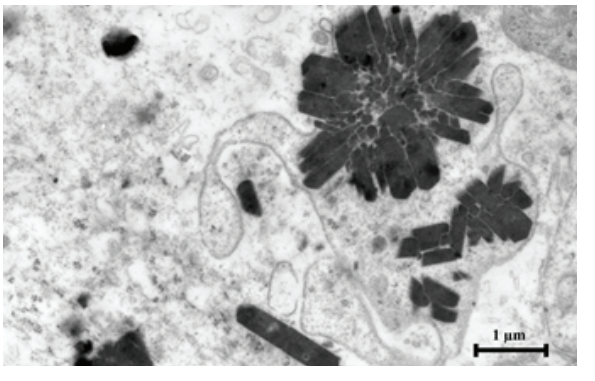

Fig. 2. Crystals at the site of pinealocyte death

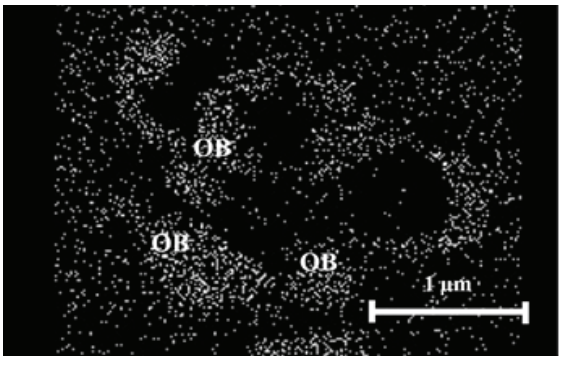

Fig. 3. Phosphorus distribution in the pinealocyte cytoplasm according to the data of dispersion microrentgen-ospectral analysis

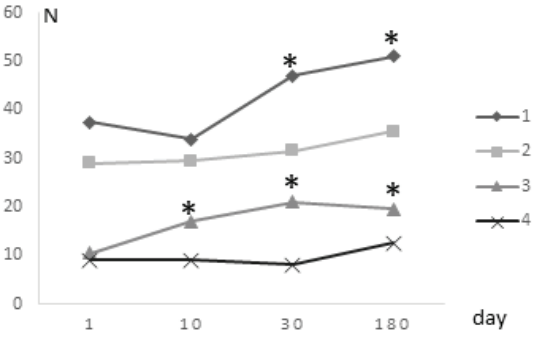

Fig. 4. Changes in the numerical density (N) of in the cytoplasm of pinealocytes in rats after exposure to light: 1 – total N in the experiment, 2 – total N in the control, 3 – grouped N in the experiment, 4 – grouped N in the control, asterisk – significant differences with the control

stress [4], which obviously activates mitophagy, the utilization of large mitochondria with signs of damage of the "dark" type with the formation of single OB. This may explain the earlier increase in the numerical density of grouped OB compared to the increase in the total numerical density of OB.

Conclusion. Thus, the peculiarity of the morphofunctional organization of the pineal gland, associated with the site of synthesis of the main pineal hormone melatonin in the mitochondria of pinealocytes, causes their frequent fusion with daily frequency, a significant specific volume in the cytoplasm, widespread calcification and osmiophilia of the mitochondrial matrix, utilization in the composition of OT, in place of which hydroxyapatite crystals or concretions of amorphous type. Violation of the circadian rhythm of mitochondrial fusion increases the content of grouped OT in pinealocytes, which can be considered as a residual manifestation of desynchronosis when adapting to round-the-clock lighting.

Reference

1. Gerasimov A.V. Funktsionalnaya morfologiya neyronov suprakhiazmaticheskikh yader gipota-lamusa krys posle kombinirovannogo vozdeystviya rentgenovskogo izlucheniya i sveta [The functional morphology of suprachiasmatic nuclei neurons of rat hypothalamus after combined influence of X-irradiation and light exposure]. Radiation biology. Radioe-cology. 2003; 43(4): 389–395 (In Russ.).]
2. Gerasimov A.V., Kostyuchenko V.P., Logvinov S.V. [et al.]. Ultrastrukturnye osoben-nosti pi-nealotsitov shishkovidnoy zhelezy gryzunov v vozrastnom aspekte [Ultrastructur-al specialfeatures of pinealocyтes of the pineal gland of rodents in the aspect dependent on age]. Fundamental research. 2013; 12(3): 449–452 (In Russ.).] URL: https://fundamental-research.ru/ru/article/view?id=33426.
3. Logvinov S.V., Gerasimov A.V., Kostyuchenko V.P. Morfologiya epifiza pri vozdeystvii sveta i radiatsii v eksperimente [Epiphysis morphology under light and radiation exposure in experiment]. Bulletin of Siberian Medicine. 2003; 2(3): 36–41 (In Russ.).] DOI: org/10.20538/1682-0363-2003-3-36-43.
4. Logvinov S.V., Gerasimov A.V., Kostyuchenko V.P. Ultrastruktura pinealotsitov u krys pri vozdeystvii sveta i radiatsii [Pinealocyte ultrastructure in rats exposed to light and ra-diation]. Morphology. 2004; 125(1): 71–75 (In Russ.).
5. Humbert W., Pévet P. Calcium content and concretions of pineal glands of young and old rats. A scanning and X-ray microanalytical study. Cell Tissue Res. 1991; 263(3): 593-596. DOI: 10.1007/BF00327294.
6. Humbert W., Pévet P. Calcium concretions in the pineal gland of aged rats: an ultrastruc-tural and microanalytical study of their biogenesis. Cell Tissue Res. 1995; 279(3): 565-573. DOI: 10.1007/BF00318168.
7. Karasek M. Morphofunctional aspects of the mammalian pineal gland / M. Karasek, R.J. Reiter // Microsc. Res. Tech. 1992; 21: 136–157. DOI: 10.1002/jemt. 1070210206.
8. Kodaka T., Mori R., Debari K, Yamada M. Scanning electron microscopy and electron probe microanalysis studies of human pineal concretions. Microscopy. 1994; 43: 307–317.
9. Krstić R. Pineal calcification: Its mechanism and significance. J. Neural Transm. Suppl. 1986; 21: 415–432.
10. Suofu Y., Li W., Jean-Alphonse F. [et al.] Dual role of mitochondria in producing mela-tonin and driving GPCR signaling to block cytochrome c release. Proc. Natl. Acad. Sci. USA. 2017; 114: 7997–8006. DOI: 10.1073/pnas.1705768114.
11. Tan D.-X, Manchester L.C., Qin L., Reiter R.J. Melatonin: A mitochondrial targeting molecule involving mitochondrial protection and dynamics. Int. J. Mol. Sci. 2016; 17: 2124. DOI: 10.3390/ijms17122124.

I.D. Ushnitsky, T.V. Alekseeva, E.Yu. Nikiforova, M.I. Solovyova, I.L. Savvina

STRUCTURAL CHARACTERISTICS OF DENTAL ANOMALIES IN SCHOOLCHILDREN DUE TO THE VAULT HEIGHT OF THE HARD PALATE WITH DIFFERENT DEGREES OF SEVERITY OF CONNECTIVE TISSUE DYSPLASIA

DOI 10.25789/YMJ.2023.81.05

УДК 616.315-007.24

USHNITSKY Innokentii Dmitrievich – MD, Professor, Head of the Department of Therapeutic, Surgical, Orthopedic Dentistry and Pediatric Dentistry of the Medical Institute of the M.K. Ammosov North-Eastern Federal University, e-mail: incadim@mail.ru; **ALEKSEEVA Tatiana Vasilievna** – Head of the children's dental department of the Yakutsk specialized dental center, e-mail: tanina2708@gmail.com; **NIKIFOROVA Ekaterina Yurievna** – senior lecturer of the department of therapeutic, surgical, orthopedic dentistry and pediatric dentistry of the Medical Institute of Ammosov North-Eastern Federal University, e-mail: feay88@mail.ru; **SOLOVYOVA Marianna Innokentievna** – PhD in Biology, Associate Professor, Biology Department, Institute of Natural Sciences, M.K. Ammosov North-Eastern Federal University, e-mail: cmi79@mail.ru; **SAVVINA Irina Lvovna** – Candidate of Pedagogical Sciences, Associate Professor of the Department of Foreign Languages in Technical and Natural Sciences, Institute of Foreign Philology and Regional Studies, M.K. Ammosov North-Eastern Federal University, e-mail: sil26@list.

Nowadays a high level of CTD is prevailed, which has local and general phenotypic signs. At the same time, despite its wide study, the problems of diagnosis, treatment, prevention and comprehensive rehabilitation of its manifestations in the oral cavity and maxillofacial region remain unresolved. In this connection the research aim is to solve these problems, which have an extremely important practical, scientific and theoretical significance in dentistry, and also in medicine. The **research** o**bjective** is to study the structure of dentoalveolar anomalies of schoolchildren due to the hard palate vault height at different degrees of connective tissue dysplasia severity. **Materials and methods**. A clinical and craniometric study was performed in 964 children and adolescents aged from 12 to 15 years old diagnosed with CTD. The CTD severity was determined by the method of T. Milkovska-Dmitrova and A. Karkashev (1985). In this case, anomalies of occlusion, dental deformities and anomalies in the teeth position were determined taking into account the height of the hard palate vault at different CTD severity. The vault height of the hard palate was determined by the method of Ushnitsky I.D. et al. (2018). **Results**. The examined patients most frequently revealed a medium degree of severity, then a mild degree and less often a severe degree in the CTD structure severity. At the same time, in the structure of occlusal anomalies associated with distal occlusion, deep traumatic incisal overlap and underdevelopment of the upper jaw, dental arch anomalies including occlusion of the maxillary and mandibular dental arches, shortening of the maxillary and mandibular dental arches, displacement of the upper and lower central tremes, dental position anomalies such as close position of incisors, vestibular position of upper canines, protrusion of upper incisors, retrusion of upper and lower incisors, primary adentia, dystopia of upper canines are determined by increasing their prevalence in school children due to the height of the hard palate vault depending on CTD severity.