

venous sections of the capillary were significantly greater than initially starting from the 11th day and until the end of pregnancy (Table 1, Fig. 2). Changes in the number of attached ribosomes correlated with the time course of changes in RER during pregnancy. We found significant changes in the numerical density and absolute number of attached ribosomes in the endothelial cells of the arterial and venous sections of the capillaries during pregnancy. These parameters were significantly higher than the control levels by the 11th day of pregnancy, and they increased even more by its end (Table 1, Fig. 3). These data on changes in numerical density and absolute numbers of free ribosomes in the endothelial cells of the arterial and venous sections of the capillaries are evidence of a gradual decline in the values of these parameters during pregnancy.

Investigation of the ultrastructural organization of capillary endotheliocytes in the iliac LN of animals with a hemochorial type of placentation revealed a combination of adaptive structural changes: an increase in the bulk density and absolute number of mitochondria, the surface density, and the total surface area of the inner membrane of the mitochondria (evidence of increased activity of transmembrane transport by means of carriers), and dilatation of the tubules and cisterns of the endoplasmic reticulum and of GC. Toward the end of pregnancy the area of cross section of the endothelial cells and their nucleus and cytoplasm is increased, microvesicular transport is activated, and transendothelial channels are formed. The ultrastructural changes in the capillary endotheliocytes of LN-regional relative to the uterus during physiological pregnancy in rats are adaptive in character and evidently tend to reduce congestive phenomena in the system of the inferior vena cava, thereby improving the utero-placental circulation.

LITERATURE CITED

1. Yu. I. Borodin, N. A. Sklyanova, Yu. I. Sklyanova, and S. A. Patrusheva, *Arkh. Anat.*, **90**, No. 4, 18 (1986).
2. E. R. Weibel, *Morphometry of the Human Lungs*, Springer-Verlag, New York (1963).
3. E. P. Voityuk, *Functional Morphology of the Lymphatic System* [in Russian], Novosibirsk (1981), pp. 22-25.
4. N. L. Garmasheva and N. N. Konstaninova, *Pathophysiological Basis of Protection of Human Intrauterine Development* [in Russian], Leningrad (1985).
5. V. A. Shakhlamov, *Capillaries* [in Russian], Moscow (1971).
6. M. Gilbert and A. Leturque, *J. Dev. Physiol.*, **4**, No. 4, 237 (1982).

ELECTRON-AUTORADIOGRAPHIC DATA ON RNA SYNTHESIS AND CHROMATIN STRUCTURE IN RAT CEREBRAL CORTICAL CELLS

E. Ya. Sanovich and V. P. Tumanov

UDC 616.831-008.93:577.216.3:
[616.831-018.13:576.315.42]-076.4

KEY WORDS: neuron; chromatin; electron-microscopic autoradiography.

One of the first electron-autoradiographic studies [5], aimed at determining regions of RNA synthesis in the nucleus more accurately than can be done by light microscopic autoradiography, revealed that synthesis takes place in dispersed (eu-), but not in condensed (hetero-) chromatin. These data were subsequently confirmed by numerous experiments [4, 6]. A detailed study of RNA synthesis in the nucleus by various methods [2] showed that rRNA is synthesized in the nucleolus whereas mRNA and tRNA, used for protein synthesis, are synthesized in dispersed chromatin. The following theoretical hypothesis is based on these findings: cells producing much protein must synthesize RNA intensively outside the nucleolus,

Department of Pathological Anatomy, M. F. Vladimirkii Moscow Regional Clinical Research Institute. Department of Pathological Anatomy, A. V. Vishnevskii Institute of Surgery, Academy of Medical Sciences of the USSR, Moscow. (Presented by Academician of the Academy of Medical Sciences of the USSR D. S. Sarkisov.) Translated from *Byulleten' Éksperimental'noi Biologii i Meditsiny*, Vol. 105, No. 1, pp. 85-87, January, 1988. Original article submitted June 5, 1987.

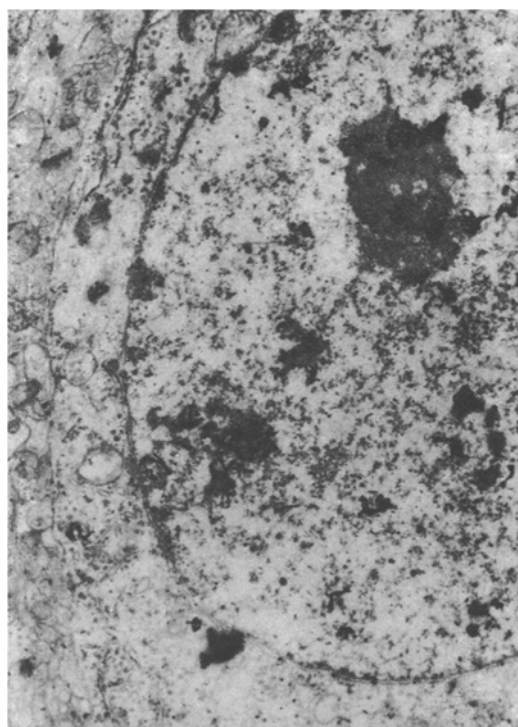


Fig. 1. RNA synthesis in nucleus of neuron. Nucleoli are most intensively labeled. Large centers of heterochromatin located alongside them (arrows) contain no label. 15,000 \times . [Arrows omitted in Russian original - Publisher].

they must have a large area occupied by euchromatin and a small area occupied by heterochromatin. Cells producing little protein must synthesize RNA correspondingly more slowly and must contain an extensive zone of heterochromatin in their nucleus. Differences in the structure of chromatin in neurons, astrocytes, oligodendrocytes, microglia, endotheliocytes, and leukocytes are well known [7] and by using electron-microscopic autoradiography the validity of the above hypothesis can be tested experimentally.

EXPERIMENTAL METHOD

The investigation was conducted on serial sections through neurons from layer V of the cerebral cortex of albino rats weighing 180 g. Under ether anesthesia, using a special needle, the RNA precursor 5- ^3H -uridine (specific activity 26 Ci/mmol) was injected into the region of the cutaneous sensory-motor zone (area PAM). Labeled uridine (50 μCi) was dissolved in 0.05 ml of Ringer's solution. Pieces of tissue 3 h after injection of ^3H -uridine were fixed in 2.5% glutaraldehyde solution made up in phosphate buffer (pH 7.4). During the next 24 h the preparations were washed with buffer with frequent changing of the solution and then postfixed in 1% OsO_4 solution. After dehydration in alcohols the material was embedded in Epon and light microscopic autoradiographs were prepared on semithin sections, and the results of their analysis determined the region chosen for cutting ultrathin sections. Serial electron-microscopic preparations were produced with the aid of type M emulsion [2]. After exposure for 1 month the preparations were developed and examined in the JEM-100B electron microscope. The intensity of RNA synthesis was determined by measuring the density of grains of silver above the nucleus. For this purpose the number of grains of silver located above the nucleus was divided by the area of the nucleus. Since the labeling density, with the method used, largely depended on the distance of the brain region studied from the site of injection of ^3H -uridine, only neighboring cells of different types were investigated.

EXPERIMENTAL RESULTS

The regular pattern mentioned above, namely activity of euchromatin in relation to RNA synthesis and the inertness of the heterochromatin was clearly revealed in the electron-

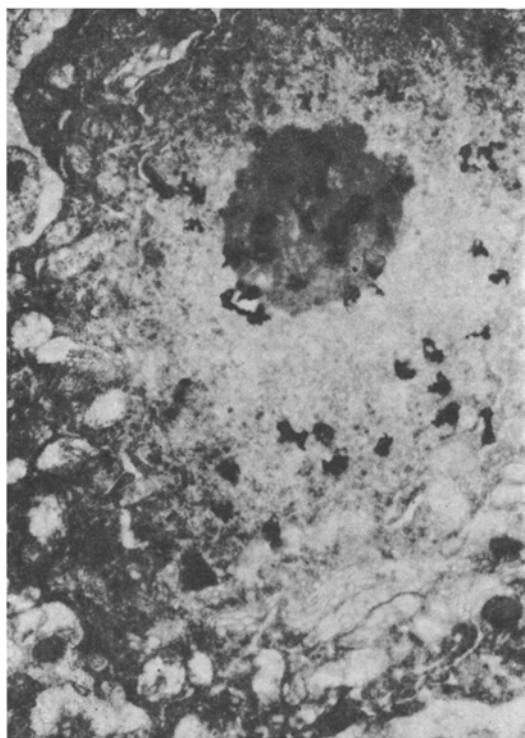


Fig. 2

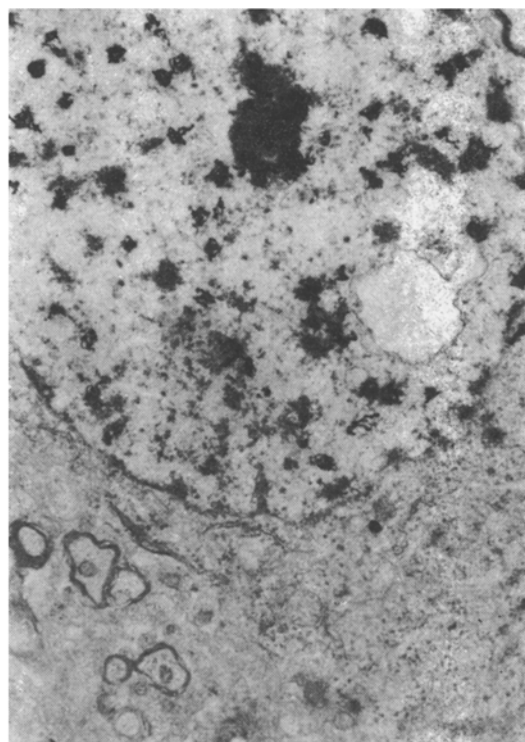


Fig. 3

Fig. 2. Incorporation of ^3H -uridine into brain cells. Neurons (N) and astrocyte (A) located side by side differ significantly in the concentration of grains of silver above their nuclei. [Labels omitted in Russian original - Publisher].

Fig. 3. Large concentration of grains of silver visible above nucleus of neuron (N); only four grains of silver are present above the nucleus of the adjacent oligodendrocyte (O). 8000 \times . [Labels omitted in Russian original - Publisher].

microscopic autoradiographs. This property of the genome was revealed particularly clearly in the nucleolus (Fig. 1).

Examination of the electron-microscopic preparations showed that the neurons had the highest euchromatin/heterochromatin ratio. This ratio was appreciably lower in the astrocytes and oligodendrocytes, mainly on account of a belt of heterochromatin lying along the inner nuclear membrane. This belt was even wider in the nuclei of the microglia and endothelial cells. The euchromatin zone was minimal in neutrophils, which were frequently found in the brain.

Corresponding to the difference in the chromatin structure in the types of cells mentioned above, there was a sharp difference in the intensity of their RNA synthesis, obvious even without quantitative analysis (Figs. 2 and 3). Quantitative analysis of the electron-microscopic autoradiographs revealed the following values of labeling density: neuron 80.2 ± 4.7 , astrocytes 23 ± 2.0 , oligodendrocytes 12.7 ± 2.1 , microglia 7.6 ± 0.8 , and endothelial cells 6.3 ± 0.9 ; the neutrophils contained no label.

This investigation thus proved experimentally that the intensity of RNA synthesis is higher in neurons, i.e., in cells containing more widely dispersed chromatin, and confirmed once again the general principle of the unity of structure and function with particular respect to chromatin or, in other words, to the cell genome. An important conclusion regarding the characteristics of function of the neuron, and its great superiority over other types of cells found in the brain in its level of RNA synthesis, can also be drawn from these results. This not only confirmed existing data showing that large quantities of protein are produced in the neuron [1, 3], but also filled in their essential details. High levels of dispersion of chromatin and of RNA synthesis in the neuron can be interpreted in the light of these results as evidence that this cell produced, not so much a large total mass of protein, as a large number of individual proteins, i.e., the diversity and complexity of the protein composition of neurons. This view may be substantiated by comparing the neuron with

the plasma cell. The latter is not inferior to the neuron in the quantity of protein it produces (the high degree of development of the rough endoplasmic reticulum in the plasma cell is well known), but the bulk of the protein produced by the plasma cell is of only one type, namely a concrete immunoglobulin. This narrow specialization is combined with a high degree of condensation of chromatin in the plasma cell. The opposite character of the structure of chromatin in the neuron and the intensive incorporation of ^3H -uridine in the zone where, as has been shown, mRNA is synthesized, are evidence of derepression of a much greater part of the genome in the neuron than in other cells of the body, and of the many different kinds of proteins produced by the neuron.

LITERATURE CITED

1. O. N. Dolgov, A. B. Poletaev, and V. V. Sherstnev, *Usp. Fiziol. Nauk*, 11, No. 3, 47 (1980).
2. D. S. Sarkisov, A. A. Pal'tsyn, and B. V. Vtyurin, *Electron-Microscopic Autoradiography of the Cell* [in Russian], Moscow (1980).
3. V. V. Sherstnev, *Vestn. Akad. Med. Nauk SSSR*, No. 2, 47 (1981).
4. S. Fakan and W. Bernhard, *Exp. Cell Res.*, 67, 129 (1971).
5. V. C. Littay, V. G. Allfrey, and J. H. Frenster, *Proc. Natl. Acad. Sci. USA*, 52, 93 (1964).
6. G. Moyne, *Cytobiologie*, 15, 126 (1977).
7. A. Peters, S. Palay, and H. Webster, *The Fine Structure of the Nervous System: the Neurons and Supporting Cells*, Saunders, Philadelphia (1976).

BRAIN MORPHOLOGY AND FUNCTION IN RATS WITH VARIED DEGREE OF NEUROLOGIC RECOVERY AFTER SYSTEMIC CIRCULATORY ARREST

M. Sh. Avrushchenko, T. L. Marshak,
and E. A. Mutuskina

UDC 616.12-008.315-008.66-07:
[616.831-091+616.831-008.1

KEY WORDS: clinical death; brain; Purkinje cells; nucleolus; rat.

The most difficult and important task in modern resuscitation practice is the full restoration of brain activity [9]. In connection with this task, the choice of the method of assessment of changes developing in the brain after clinical death assumes particular significance. We know that analysis of the morphology and function of Purkinje cells (PC) in the cerebellar cortex, which are highly sensitive to anoxia, provides a means of determining the role of changes in the general density of the population and its composition in the pathogenesis of postresuscitation brain damage, of assessing the role of different types of cells in the maintenance of population homeostasis, and of revealing some mechanisms of repair processes developing in the brain after clinical death. Investigation of the PC population is essential not only because of the important role of cerebellar injury in the formation of postanoxic encephalopathy [2], but also because the state of the PC population depends on the duration of ischemia and correlates with the degree of recovery of the neurologic status of revived animals and with changes in other brain regions [4]. In recent years a new experimental model of clinical death has been developed, namely systemic circulatory arrest caused by ligation of the vascular bundle of the heart in rats [6]. No morphological investigations of the nervous system of rats subjected to clinical death of this etiology have hitherto been undertaken.

The aim of this investigation, using an approach developed previously, was to assess the state of the brain in rats differing in their degree of neurologic recovery after systemic circulatory arrest caused by ligation of the vascular bundle of the heart.

Institute of General Resuscitation, Academy of Medical Sciences of the USSR. Laboratory of Cytology, Institute of Developmental Biology, Academy of Sciences of the USSR, Moscow. Translated from *Byulleten' Éksperimental'noi Biologii i Meditsiny*, Vol. 105, No. 1, pp. 87-90, January, 1988. Original article submitted June 10, 1987.