ordinary electron-microscopic autoradiograph, which is examined through the layer of emulsion, thus reducing the contrast of the image, the electron-microscopic autoradiograph obtained by the proposed method is not inferior in image quality to ordinary electron-micrographs.

The grains of silver in such an autoradiograph (Fig. 3) are distributed, not above the cell but at its side, but nevertheless this localization of the grains only at the side of the nucleus of the cell under examination and their absence elsewhere in the preparation are absolute evidence of DNA synthesis in the particular cell. This important indicator of cell function is reinforced by its characteristic ultrastructural morphology, as revealed by a thin electron-microscopic section, whose contrast is not reduced by the layer of emulsion as is the case with the ordinary electron-autoradiograph.

LITERATURE CITED

1. R. Adams, Methods of Cell Culture for Biochemists [Russian translation], Moscow (1983).
2. M. L. Magalhaes, T. Serra, and M. M. Magalhaes, Tissue Cell., **19**, No. 2, 167 (1987).
3. J. Patel, M. Amherdt, and Z. Orci, Science, **127**, 1155 (1982).

ULTRASTRUCTURAL CHANGES IN AUTONOMIC NERVE GANGLIA IN EXPERIMENTAL BURN TRAUMA

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Definite difficulties arise in the evaluation of data obtained by the study of cadaveric material, especially if electron-microscopic and histochemical methods of investigation are used. These difficulties are associated primarily with postmortem changes, which may complicate the interpretation of the structural disturbances observed. Nerve cells of the vein are most sensitive to postmortem changes, and for that reason, it is recommended that material for electron-microscopic histochemical studies be collected soon after death [3, 8]. Investigations by a number of workers has shown that this period can be considerably increased [1, 2, 4-7, 9, 10]. However, no special investigations have been undertaken into the possibility of studying the state of the neuronal ultrastructure at different times after death.

In order to undertake a comparative ultrastructural analysis of intravital changes in neurons of autonomic ganglia in patients dying at different stages of burn trauma, with postmortem cadaveric changes in these ganglia, we made a special study of neural structures 4-6 h after death of animals in the control group, and also at various times after experimental burn trauma.

EXPERIMENTAL METHOD

Experiments were carried out on 15 noninbred male albino rats aged 5-7 months and weighing 230-280 g. The animals were fixed on a special table, anesthetized with ether and oxygen, and the hair cover was removed in the spinal region. Burn trauma was inflicted by applying a metal plate measuring 4 × 9 cm, heated to 80-100°C, to the depilated skin. The exposure was 5-6 sec, and as a result, a burn of the IIIa, b degree was produced on an area of 20-25% of the rats' body surface. The animals were decapitated 3, 7, and 11 days after thermal trauma, corresponding to the principal periods of burn healing. Material was taken from five control animals immediately after decapitation and 4-6 h after death. The superior cervical and stellate ganglia

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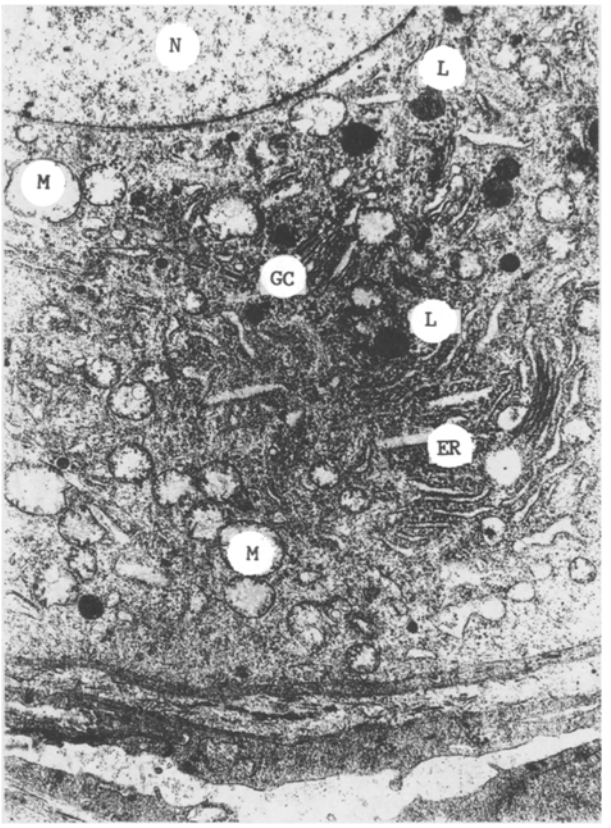

Fig. 1. Translucency of karyoplasm of nucleus (N), swelling and translucency of mitochondrial matrix (M), dilatation of tubules of endoplasmic reticulum (ER) and fragmentation of profiles and vacuoles of Golgi complex (GC). L) Lysosomes. Stellate ganglion of rat 4 h after death. Control. Here and in Figs. 2-4, 12,000×.

and the celiac ganglia of the solar plexus were studied by electron microscopy. The material was fixed in 1% glutaraldehyde solution, postfixed in a buffered in solution of osmium tetroxide, dehydrated with alcohols, and embedded in a mixture of Epon and Araldite. Sections cut on an LKB-4801A were stained with uranyl acetate, counterstained with a solution of lead monoxide and examined in the IEM-I00B electron microscope.

EXPERIMENTAL RESULTS

Electron-microscopic investigation of neurons of the autonomic ganglia of healthy rats 4 h after decapitation revealed ultrastructural changes in the nuclei, mitochondria, and other intracellular organelles. These changes, incidentally, were not destructive and were observed almost uniformly in all sympathetic ganglia. The early changes in the mitochondria are noteworthy. One of the earliest signs indicating changes in mitochondrial ultrastructure is lysis of the cristae. Considerable translucency of the mitochondrial matrix also was observed, possibly on account of its swelling. Another indicator of a change in organization of the nerve cell is associated with the reaction of the ribosomes and glycogen granules. Their number was greatly reduced in the cell cytoplasm. No appreciable changes were observed in the nuclei of the neuron. Ultrastructural changes in the lamellar complex took the form of fragmentation of vesicular and tubular elements into smaller vacuoles and cavities, and changes in the endoplasmic reticulum are expressed as moderate dilatation of the cavities and tubules and a decrease in the number of granules on their outer surfaces. Neurofilaments in the cytoplasm of most neurons were preserved and could be identified at sites of formation of the processes of the nerve cells. Fine lipid inclusions and formations of lysosomal type were found near the Golgi apparatus. The outer cell membranes had no visible disturbances. Along the outer side of the body of the neuron, processes of glial cells were in close contact (Fig. 1). As regards ultrastructural changes in nerve terminals and endothelial cells of the blood

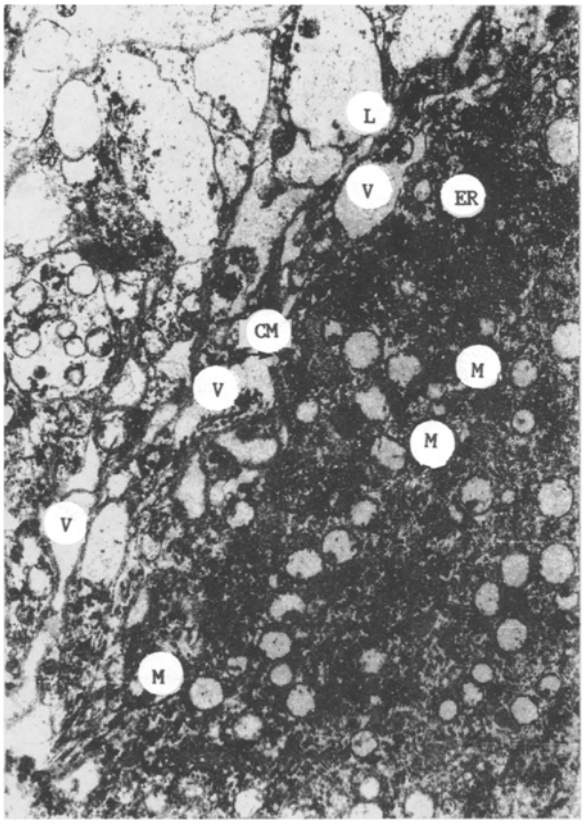

Fig. 2. Vacuolation of mitochondria (M), fragmentation of tubules of endoplasmic reticulum (ER), and destruction of outer cell membrane (CM) . Concentration of dark masses in cytoplasm of neuron of celiac ganglion of rat solar plexus 3 days after burn trauma. N) Nucleus, V) vacuoles, L) lipids.

capillaries of the superior cervical, and stellate ganglia and the celiac ganglia of the solar plexus, no differences could be detected.

Investigation of the state of the cell components of the sympathetic ganglia of the rats 6 h after death revealed an increase in the intensity of the changes. These changes were most marked in the mitochondria, nucleus, and cytoplasm. The mitochondria were swollen, their matrix translucent , and their inner membranes could not be detected. At the end of 6 h after circulatory and respiratory arrest the outlines of the nucleus became smooth, evidently because of swelling of the karyoplasm. With an increase in the duration of survival of the organs the quantity of fibrillar chromatin fell rapidly, but the granular chromatin was condensed into larger granules, which migrated toward the inner nuclear membrane. The individual layers of the nuclear membrane lost their distinct contour, their lines were blurred, and the perinuclear space was almost invisible. Individual threads of neurofilaments and dilated tubules of the endoplasmic reticulum were observed in the translucent cytoplasm. Among processes of satellite cells adjacent to the body of the neuron, extensive pale zones appeared. With an increase in the time spent by cells of the nerve ganglia in the cadaver, there was an increase in the degree of translucency of the plasm of the neurons. A striking fact is that edema and swelling of the cytoplasm were accompanied by a decrease in the number of glycogen granules and ribosomes. Changes in nerve terminals of the sympathetic ganglia were at the same level as those described above 4 h after death. Changes in processes of the Schwann cells were rather more marked. Their translucency, swelling of the mitochondria, and vacuolation of the tubules of the endoplasmic reticulum were all increased.

Electron-microscopic investigation of neurons in the sympathetic ganglia of mitochondria with a translucent matrix, which had lost their inner membranes, had the appearance of vacuoles of different sizes. Fragmented membranes of tubules of the endoplasmic reticulum, in the form of dark concentrations of granules and fragments of membranes, were distributed in different parts of the cell. The formation of large dark chromatin granules was noted in the nucleoplasm of the nucleus. The cytoplasm of the neurons was filled with concentrations of dark masses, among which there were lipid inclusions and lysosomes. Hardly any neurofilaments were visible. The outer plasma membrane of the cell in some parts had undergone lysis, and the

Fig. 3. Marked translucency of cytoplasm of Schwann cells (SC), destruction of mitochondria (M), areas of destruction of nuclear membrane (NM), and aggregation of masses of chromatin in nucleus (N) of superior cervical ganglion of rat dying on 7th day after burn trauma. L) Lysosomes, A) axon.

cytoplasm of the neurons was exposed there. Translucent processes of glial cells departed in some places from the neuron body and formed clear spaces (Fig. 2). In the terminal processes, translucency and swelling of the mitochondria and separation of the myelin membranes into layers in the myelinated axons also were observed. Under these circumstances, lysis and rupture of individual membranes could be seen. Separation of the membranes of the myelinated axon was observed around the whole perimeter of the myelin sheath. Fragmentation of tubules of the endoplasmic reticulum was observed in the translucent cytoplasm of the Schwann cells, with translucency and swelling of mitochondria and capillaries of chromatin on the inner nuclear membrane. In the lumen of the blood capillaries sludging of erythocytes and translucency of the cytoplasm of endothelial cells were noted.

The study of the ultrastructure of sympathetic ganglia of the autonomic nervous system of rats on the 7th-11th days after burn trauma revealed sharp changes both in neurons and in the endothelial cells of the blood capillaries. These changes were mainly destructive in character. Areas of concentration of destroyed membranes and tiny osmiophilic granules, remnants of destroyed cell components, were present in the cytoplasm. Attention was drawn to profound changes in the cytoplasm. Attention was drawn to profound changes in the cytoplasm of the Schwann cells, with the almost complete absence of intracellular glycogen granules and ribonuclear membranes was observed in some cells. In the translucent, swolllen processes of the Schwann cells dark inclusions of lysosomal origin were found (Fig. 3). Severe disturbances of the cell membranes also were noted in the endothelial cells of the blood capillaries. A conspicuous feature was the formation of extensive fragmented limiting membranes of the processes of the endothelial cells facing the lumen of the blood capillaries. At places of destruction of the plasma membrane, fragments of cytoplasm were released into the lumen of the capillaries (Fig. 4).

Thus profound changes in the ultrastructure of the autonomic nerve ganglia take place in burn trauma. These changes are destructive in character. One of the attributes of burns is a disturbance of the microcirculation. Disturbances observed in the endothelial cells of the blood capillaries were seen almost simultaneously with changes in the nerve cells.

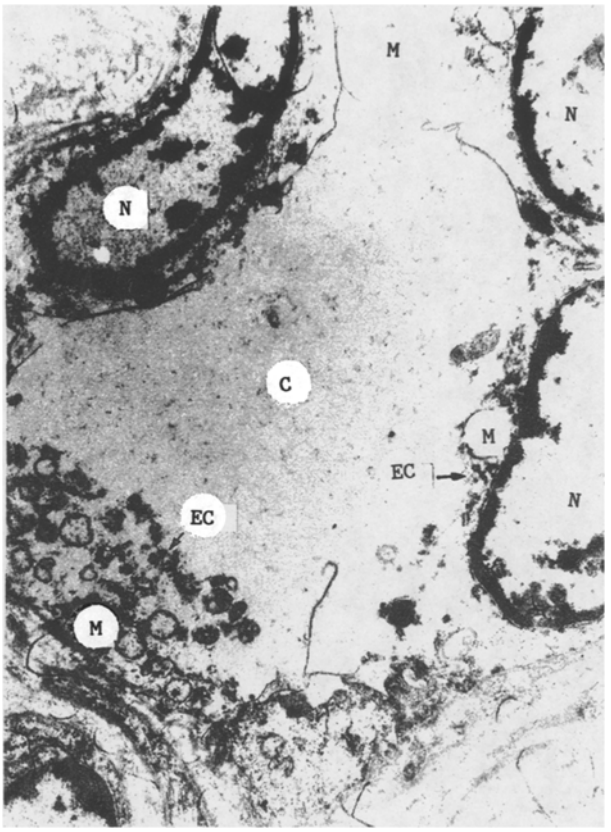

Fig. 4. Lysis of membranes (arrow) of luminal surface of endothelial cells (EC) and 11 days after burn trauma, N) Nucleus, M) mitochondria.

To conclude, comparative analysis of the character of changes in the nerve cells and blood capillaries arising after death and burn trauma revealed differences in the disturbance of their architectonics. Comparison of the results of the study of autopsy and experimental materials relating to burn trauma showed that cadaveric material is suitable for ultrastructural analysis in the early stages after death (until 6 h).

LITERATURE CITED

1. A. P. Amvros'ev and Yu. I. Rogov, Arkh. Patol., No. 2, 48 (1987).
2. B. V. Antiev and S.V. Vladimirov, Arkh. Patol., No. 11, 66 (1968).
3. V. I. Biryuzova, Yu. S. Chentsov, and V. L. Borovyagin, Electron-Microscopic Methods of Investigation of Biological Objects [in Russian], Moscow (1963), pp. 87-99.
4. N. N. Bogolepov, Ultrastructure of the Brain in Hypoxia [in Russian], Moscow (1979), pp. 75-79.
5. N. A. Shapiro, Arkh. Patol., No. 12, 57 (1968).
6. J. Carr, J. Pathol. Bact., **78**, No. 2, 533 (1959).
7. G. Richter and A. Kellner, J. Cell Biol., **18**, 195 (1972).
8. D. Robertson, Structure and Function [Russian translation], Moscow (1974), pp. 152-161.
9. A. Sanberg, et al., J. Clin. Endocrinol., **16**, No. 8, 1001 (1956).
10. W. Spannand and W. Heinemann, Dtsch. Z. Gericht, Med., **4**, 77 (1955).