MORPHOLOGY AND PATHOMORPHOLOGY

Apoptosis in Cells of Rat Sympathetic Ganglia during Early Ontogeny: Electron Microscopic Study

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Postnatal ontogeny of sympathetic ganglia includes both proliferative processes and programmed cell death. Electron microscopy helps to evaluate the intensity and the relationship between these processes.

Key Words: *sympathetic ganglia; postnatal ontogeny; apoptosis*

Early postnatal ontogeny of sympathetic ganglia is characterized by inhibition of neuroblast proliferation. At this term some differentiating nerve cells undergo cell death. The role of nerve growth factor in this process is well studied [1,4]. These processes in nerve cells are followed by mitoses and apoptosis of glial Schwann cells [5]. It is known that definitive population of Schwann glia originates from two sources. Myelin-forming cells more earlier leave the glial proliferative pool. Cells not forming myelin retain mitotic activity for a longer time [3].

We investigated morphological aspects of the relationships between the loss of sympathetic neurocyte and their processes and death and proliferation of glial cells not forming myelin.

MATERIALS AND METHODS

The study was carried out on L_{III}-L_{IV} sympathetic ganglia of Wistar rats aged 1-14 days. The animals narcotized with Nembutal (40-60 mg/kg) were perfused through the left ventricle with 0.1 M phosphate buffer (pH 7.2) containing glucose and heparin and then with 2.5% glutaraldehyde on Millonig buffer. The ganglia were removed, incubated in fixative for 3 h, washed with 5% glucose on the same buffer (pH 7.2), postfixed in 1% osmic acid (pH 7.2) for 2 h, dehydrated, and embedded in epon-araldite mixture. Sections (50 nm) were prepared on a Reichert ultratome, contrasted with uranyl acetate and lead citrate, and examined under a Hitachi-HU-12 electron microscope.

RESULTS

Apoptosis in differentiated neuroblasts affects virtually all cell components. The initial phases of chromatin condensation presumably involve the nucleolus and perinucleolar matter and only later involve the nucleoplasm under the karyolemma. Cytoplasm organelles undergo more complex rearrangements. Cisterns of the endoplasmic reticulum are either dilated or contain electron dense material. Only few mitochondria look unchanged, while the majority have clarified matrix. Abundant compact lysosomes are characterized by decreased electron density. Solitary nerve endings, which cannot be regarded as presynaptic, are surrounded by neuroblasts. The neuropile adjacent to the neuroblast contains profiles of neuronal processes undergoing destructive changes (mainly pronounced condensation of their internal structure) (Fig. 1, *a*).

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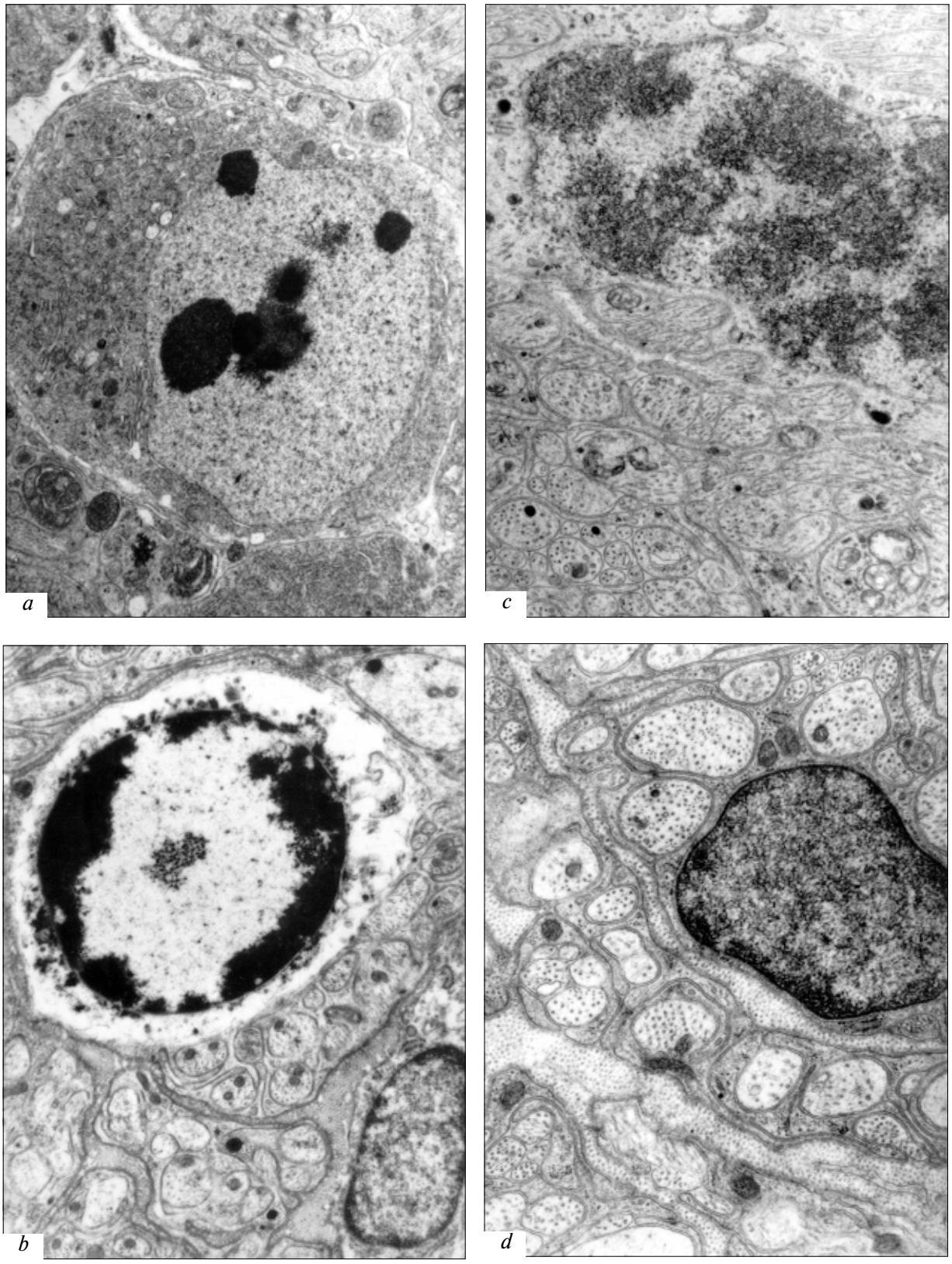

Fig. 1. Ultrastructure of sympathetic ganglia in rats aged 1 day (*a*), 1 (*b, c*) and 2 weeks (*d*). *a*) initial stage of neuroblast apoptosis. Degenerative changes in processes, ×9000; *b*) Schwann cell apoptosis, ×13,100; *c*) prometaphase of Schwann cell mitosis, ×16,900; *d*) structure of intraganglionic bundle of myelin-free figures, ×22,500.

Neuroblast and neurocyte death is paralleled by the loss of part of perineuronal glia cells. Typical apoptosis is observed in Schwann cells (SC). Acquisition of atypical structure by the nucleolus precedes extension of the zone occupied by heterochromatin under the karyolemma. Intact nuclear pores lose selective permeability, as a result of which nuclear material released into the cytoplasm forms the so-called protuberances. The cytoplasm of SC loses organelles. Cytoplasmic edema develops. Few axial cylinders are not yet completely included into Schwann membrane (Fig. 1, *b*).

Proliferative activity of perineuronal glial cell population compensates for its natural loss. During the analyzed period mitose are observed as often as apoptosis. Proliferation of SC is not associated with loss of preexisting contacts with neuronal processes. Axial cylinders surrounded by dividing lemmocytes look mainly intact (Fig. 1, *c*).

All these processes result in the formation of definitive structure of nerve ganglia, which is completed two weeks after birth. SC occupy a number of neuronal processes sufficient for their survival. The borders of zones belonging to individual lemmocytes are separated by collagen fibers, and axial cylinders gradually acquire round contours (Fig. 1, *d*).

It is now impossible to evaluate quantitatively the ratio of apoptotic cells and lemmocytes entering mitosis for two reasons: mitosis and apoptosis are short [2], and the postnatal time course of SC takes only two weeks [4]. However certain conclusions can be made on the basis of three-component scheme of relationships between degenerating neuronal processes and the number of lemmocytes in apoptosis and mitosis. The order of events is presumably as follows: neurocyte death — degeneration of processes — apoptosis of respective SC — replacement of dead lemmocytes by SC entering mitosis later.

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