STRUCTURAL AND FUNCTIONAL ORGANIZATION OF NUCLEI OF THE DORSAL COLUMNS AFTER UNILATERAL SECTION OF THE MEDIAL LEMNISCUS

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The writers showed previously that kinesthetic afferent influences spread toward the forebrain not only along the lemniscal system [2]. After unilateral hemisection of the tegmentum mesencephali, including the corresponding medial memniscus, in animals it is still possible for conditioned instrumental reflexes to the contralateral forelimb to be formed. These observations raised the question of additional afferent pathways, besides the lemniscal system, for the spread of kinesthetic sensation. It has been suggested that there are three possible structures which may conduct these influences: spino-cerebello-thalamic tracts, spino-reticulo-thalamic tracts, and extralemniscal connections of nuclei of the dorsal columns (NDC). To assess these possibilities, it was necessary to isolate each of these structural connections by blocking the other two coexisting connections, and to investigate its effectiveness in the conduction of afferent influences to the forebrain.

The structural stability and functional mobility of NDC after section of the corresponding contralateral medial lemniscus were studied in the investigation described below.

EXPERIMENTAL METHOD

Experiments were carried out on five adult cats weighing 2.5-3 kg in which the half of the tegmentum mesencephali which receives fibers from the medial lemniscus was divided unilaterally [1]. One year after the operation the nuclei of the funiculus gracilis and funiculus cuneatus were investigated under optical and electron microscopes. The nerve cells were drawn by means of the RA-4 drawing apparatus in sections 20 μ thick stained by Nissl's method. For the electron-microscopic investigation the animals were perfused with 2.5% glutaraldehyde in phosphate buffer. After fixation and dehydration in alcohols in the usual way the brain tissue was embedded in Epon. Thin sections, cut on an LKB Ultrotome (Sweden), were stained with uranyl acetate and lead citrate. Electron micrographs were obtained with NI-II (Hitachi, Japan) and Tesla BS-540 (Czechoslovakia) microscopes. NDC were identified with the aid of semithin sections 2-3 μ thick, stained with a 1% solution of toluidine blue in 1% borax. Conditioned instrumental defensive reflexes "to each forelimb" were formed in all the animals after the operation in two stages.

One year after the operation, an acute electrophysiological investigation of the animals was undertaken. Under pentobarbital sodium anesthesia the skin and fascia covering the posterior half of the cranium and the upper third of the neck were divided with a sagittal incision. The muscles covering the occipital bone and the posterior surface of cervical vertebra I were removed, the fibro-fascial membrane was excised, and the posterior surface of the medulla and the 1st cervical segment of the spinal cord were exposed. With the aid of a stereotaxic holder, active nichrome electrodes were applied to the surface of the medulla above NDC. The reference electrode was fixed to the divided cervical muscles. The two forelimbs were stimulated with square pulses through laid-on bipolar electrodes from an electrostimulator (from Nihon Kohden, Japan). Evoked potentials (EP) were recorded on an oscilloscope (Nihon Kohden) and on magnetic tape, by a recorder from Brüel and Kjaer (Denmark). The electrophysiological data were analyzed by M-6000 computer.

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Fig. 1. Frontal section at caudal level through nuclei of funiculus gracilis and funiculus cuneatus of a cat 1 year after unilateral hemisection of tegmentum mesencephali on the right side. Loss of neurons can be seen in both nuclei on the side contralateral to hemisection. Nissl's method. 100 ×.

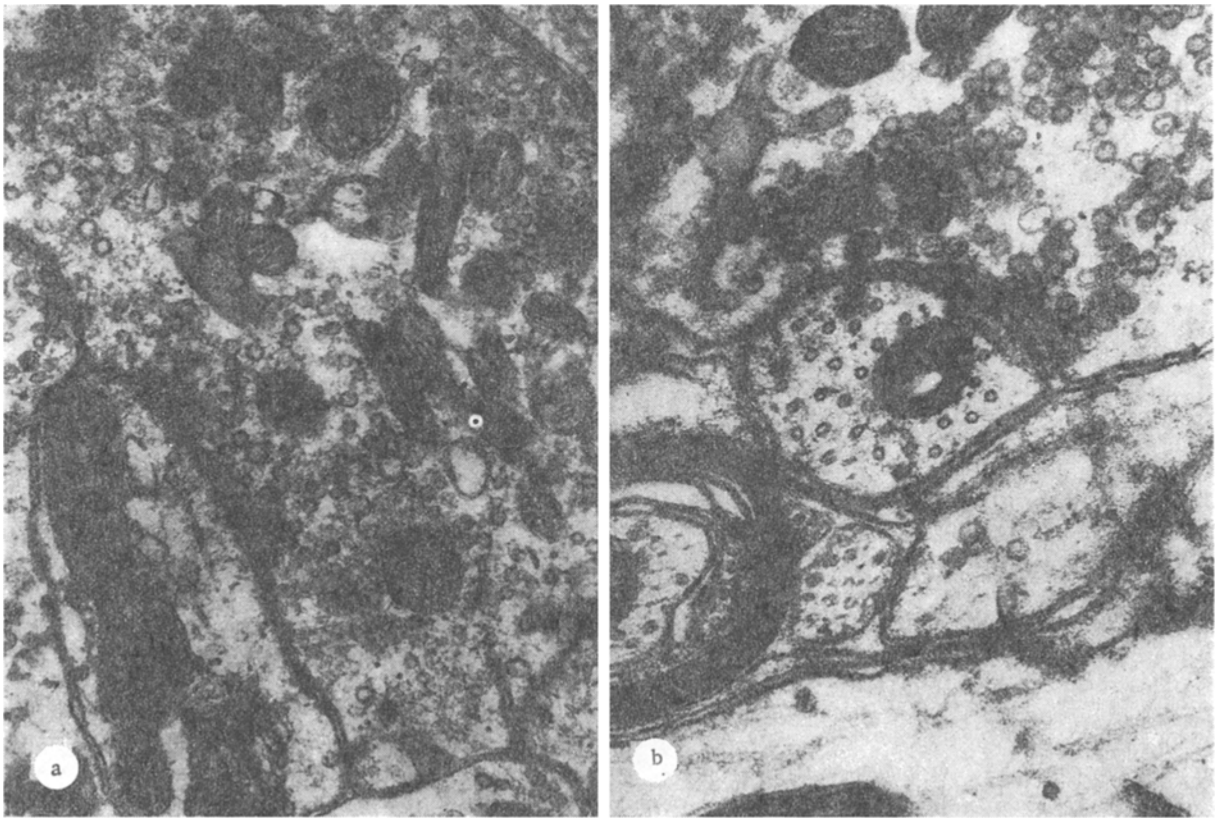

Fig. 2. Structure of synapses of nuclei of funiculus gracilis and funiculus cuneatus: a) axodendritic synapse on a large synaptic terminal; nucleus of funiculus gracilis on side contralateral to operation, 1 year after hemisection. 30,000 ×; b) intact axodendritic synapse in nucleus of funiculus cuneatus of a cat 1 year after operation, on side contralateral to hemisection. 40,000 ×.

EXPERIMENTAL RESULTS

In animals surviving division of the connections of the medial lemniscus for 1 year, degenerative changes in neurons of the nuclei of the funiculus gracilis and funiculus cuneatus were virtually invisible. Compensatory and restorative reactions predominated in this area. Neurons responsible for reactive adjustments in response to injury gradually restored the normal structure. No cell ghosts were present, and proliferation of the neuroglia was observed. Preserved neurons were distributed diffusely. Among them there were neurons 2 or 3 times the normal size, and cells with few nuclei. Both usually stained more brightly than ordinary neurons. They were few in number, they were found not in every section, and they were present mainly at the caudal level of NDC (Fig. 1).

Electron-microscopic investigation of NDC at the same time after the operation confirmed the data of light microscopy, showing that processes of destruction in the nerve cells were virtually complete. Meanwhile, in individual areas of neurons and their processes, reorganization was not yet complete. This was shown by activation and hypertrophy of the Golgi apparatus and by an increase in the number of free ribosomes in the cytoplasm of the nerve cells, and also the appearance of glycogen granules in processes of the neurons and oligodendroglia. Lysosomes and lipofuscin granules were present in the cytoplasm of some nerve cells.

The study of the synaptic organization of NDC after destruction of their ascending connections showed integrity of synapses of glomerular type, including the presence of axo-axonal synaptic contacts in them; according to some workers [5, 8], it is these contacts which are responsible for presynaptic inhibition, connected with ascending inhibitory pathways of the spinal cord [3]. Under these circumstances nerve endings showed near-normal ultrastructural features (Fig. 2a, b).

The cytoarchitectonic and electron microscopic study of nuclei of the funiculus gracilis and funiculus cuneatus 1 year after destruction of the medial lemniscus thus showed that cells in the nuclei did not disappear completely; more of them were preserved, moreover, at rostral levels of the nuclei. This is in agreement with data in the literature [6, 7] indicating the existence of extralemniscal structural connections of the rostral zones of NDC. Further evidence of this is given by integrity of the synapses typical of the normal organization of NDC, and which may evidently belong both to reorganized synaptic inputs and to systems unaffected by the operation.

However, our morphological observations on preservation of some of the neurons in NDC on the side contralateral to hemisection required corresponding assessments of their function. For this purpose EP in these experimental animals were recorded from the surface of the medul-

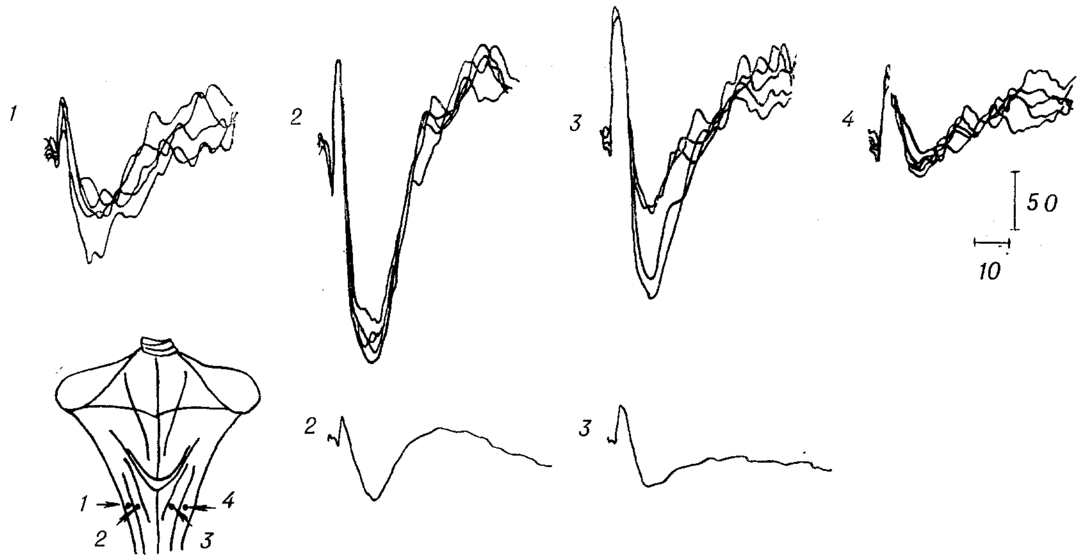

Fig. 3. Superposed EP at symmetrical points of the medulla at the NDC level during stimulation of ipsilateral forelimb (top row) and averaged EP (bottom row). Scheme on left shows recording points. Calibration: 10 msec, 50 μV.

la over NDC and analyzed. We know [4] that in respone to stimulation of the forelimb nerves, a negative-positive wave complex with a latent period of about 3 msec is recorded in NDC. The present experiments confirmed these observations on intact animals as regards the negative-positive sequence of components of the response and its latent period (Fig. 3). However, in these observations the duration of the second positive component of the response to forelimb stimulation was shorter. In cats 1 year after unilateral hemisection of the tegmentum mesencephali the negative-positive complex of the response in NDC on the side contralateral to the operation was preserved. The latent period of EP in the focus of maximal activity was 2.5-3.0 msec; it consisted of a small (under 30 μV) first positive wave, a negative wave with an amplitude of up to 75 μV, with a duration of 4-7 msec, and a large second positive wave with amplitude up to 200 μV, about 30 msec in duration. The whole negative-positive response thus developed within the interval until 40 msec. Moreover, no marked asymmetry was present in the distribution of EP during stimulation of the forelimb in the corresponding halves of the medulla. According to data in the literature [3-5], a specific negative-positive complex is recorded in NDC in response to stimulation of nerves of the ipsilateral forelimb. The first negative component of this response was associated by investigators with postsynaptic depolarization of lemniscal elements in these funicular formations. The second positive component was linked with presynaptic depolarization of terminals of ascending spinal pathways on lemniscal cells. Considering these workers' observations and hypotheses, it might be expected that after division and subsequent degeneration of the medial lemniscus, the cellular sources of the medial lemniscus would be lost in the contralateral NDC, and that asymmetry would be observed in the distribution of EP in these nuclei, during stimulation of the corresponding forelimb or of its nerves.

However, in the present investigation total loss of projection cells of the lemniscal system in NDC did not take place during the year after division of the contralateral medial lemniscus. Electron-microscopic investigations revealed preservation of the cells and of their synaptic organization. Meanwhile, this structural asymmetry of the psychoarchitectonic picture of NDC was not reflected in the distribution of afferent stomatosensory spinal functional projections to these structures. During electrical stimulation of the forelimb ipsilateral relative to the NDC which have lost their lemniscal connections, not only did the EEP in them not disappear, but they were comparable in temporal and amplitude characteristics with evoked responses in NDC on the opposite side.

In animals with unilateral division of the lemniscal system, descending or ascending asymmetrical influences which equalize the functional asymmetry of NDC evidently also are activated.

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