NEW DATA ON EARLY SYNAPTOGENESIS IN THE EMBRYONIC HUMAN SPINAL CORD

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In the study of individual development of the nervous system processes of initial formation of synapses between developing neurons unquestionably occupy a central place. The earliest beginning of synaptogenesis in the anterior horns of the spinal cord (cervical division) in man is stated [1] to be the first days of the 7th week of intrauterine development (length of embryo 14 mm). However, our own observations on human embryos have clearly shown, at an even earlier age (crown-rump length 13 mm), the process of synaptogenesis in this region was already considerably advanced at this time, so that there is reason to suppose that the process must begin at an earlier period of embryonic development [2].

This paper gives the first data on this process in the anterior horns of the brachial portion of the spinal cord in human embryos at the end of the 5th and beginning of the 6th weeks of intrauterine development (length of the embryos 8 and 10 mm respectively).

EXPERIMENTAL METHOD

An electron-microscopic study was carried out on two 8- and 10-mm human embryos obtained after medical abortions. The histological structure of the embryonic spinal cord was studied by Nissl's method.

EXPERIMENTAL RESULTS

A light-optical study of transverse sections through the brachial region of the spinal cord of the two embryos revealed a wide central canal, almost completely separating the cord into symmetrical halves. It was surrounded by a thick layer of stratified ependymal ventricular epithelium in which numerous mitotically dividing cells could be seen. By this same period a general presumptive motor nucleus was forming in the basal region of the spinal cord, not yet separated into lateral and medial parts. The marginal layer was poorly developed, and only in the region of the future ventral funiculus was it a little wider, especially in the 10-mm embryos. Among the cells forming the presumptive motor nuclei, large maturing nerve cells with pale nuclei could be clearly distinguished: They were arranged in groups, and usually occupied a zone contiguous with the marginal layer.

On electron micrographs the cytoplasm of these cells appeared to be quite translucent. It surrounded the nucleus in a narrow band with definitely dispersed chromatin. Among the cell organelles, the well developed lamellar Golgi complexes must be particularly mentioned, surrounded by an accumulation of vesicles of various sizes, some of them coated. The ribosomes were present in large numbers and mainly in the form of polysomal complexes. Here and there in the cytoplasm, more especially near the poles of the nucleus, cisterns of the rough endoplasmic reticulum were starting to form. If the cells described in the 8- and 10-mm embryos are compared, in the latter the cytoplasm and, in particular, the nuclei appeared paler on the electron micrographs. In some of the better sections it was sometimes possible to see that the perikaryon of these cells was continuous without visible boundaries with a wide cell process with pale cytoplasm, which is generally described as the leading process. In young motoneurons, our observations showed, it could continue, becoming much narrower in

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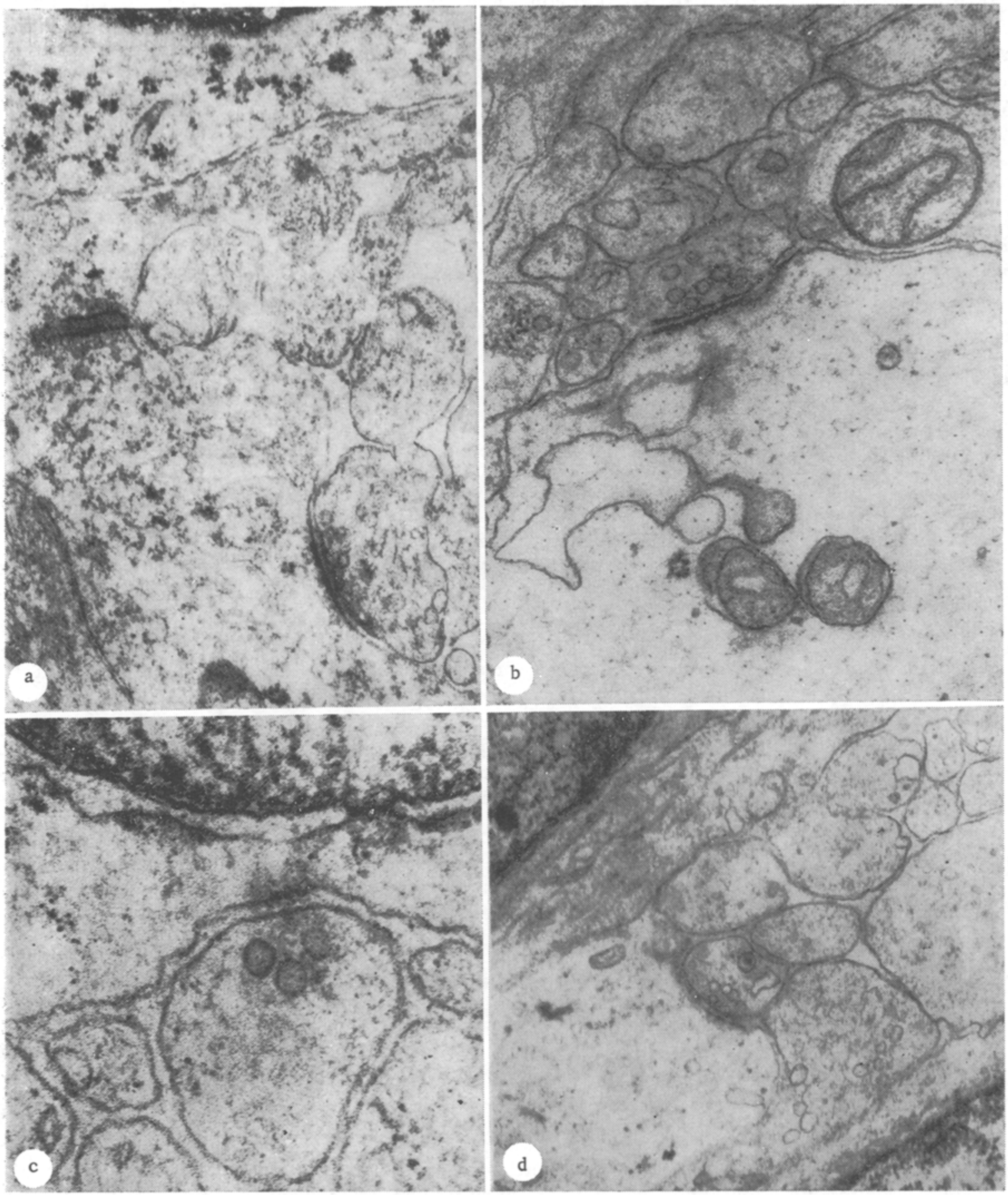

Fig. 1. Ultrastructural organization of synapses on developing nerve cells from anterior horns of brachial division of spinal cord in 8-mm (a) and 10-mm (b-d) human embryos. Magnification: a) 36,000, b) 48,000, c) 112,000, d) 36,000.

its course, into a typical axon, subsequently entering the system of the ventral roots of the spinal cord. Toward these developing nerve cells, located in regions of presumptive motor nuclei bordering on the marginal layer, also ran afferent fibers from that layer, which formed clearly defined synaptic contacts on the cell bodies or (more often) on the leading processes.

In the 8-mm human embryos synapses could be found only in the region of formation of the anterior roots, where they occupied a small region including ependymal cells and the zone of the marginal layer immediately adjacent to it. These synapses were located mainly on leading processes not far from the perikaryon (Fig. 1A). They had all the morphological properties characteristic of mature synapses: a well marked synaptic space, developed pre- and postsynaptic structures, and a collection of synaptic vesicles near the presynaptic membrane, although the number of vesicles was still small and they were not all of the same caliber. All these structural features, taken together, indicate that these synapses can be regarded as undoubtedly functionally competent forms. They were still found only in very small numbers, rarely more than two or three synapses per cross-section through the whole thickness of the formed motor nucleus and the marginal layer next to it.

In the 10-mm human embryo synapses were distributed over a much wider region. They were present not only in the ventrolateral region of the marginal layer throughout almost the whole of its depth, but they also occupied part of the intercellular neuropil in the ventrolateral regions of the motor nucleus itself. Most of the synaptic contacts in the region of these embryos, these observations showed, were accounted for by pale profiles of leading cell processes (Fig. 1B), where they could be distributed all the way through the marginal layer — close to the perikaryon and also in the most distant peripheral areas. In their structure these synapses mostly appeared asymmetrical, and in the more mature synapses the structue both of a subsynaptic complex and of a presynaptic network could be distinctly made out, although not always clearly with conventional electron microscopy. Similar asymmetry of thse synaptic contacts, according to our observations, occurred in the earliest stages of their formation.

In some cases, however, it was noted that in certain synapses on the same leading processes, besides a morphologically distinct presynaptic pole, the subsynaptic structures were hardly distinguishable. These synapses must undoubtedly possess their own individual subsynaptic receptors, differing in their morphological and functional organization from the corresponding structures of the asymmetrical synapses. It is natural to expect that these synapses themsleves must also differ, evidently, from the asymmetrical synapses in their function. Besides synapses on leading processes, axosomatic contacts also were constantly observed (Fig. 1C). They were formed by axons of relatively small caliber, which lay in close proximity to the surface of the perikaryon and could sometimes be so deeply impressed into the zone of cytoplasm surrounding the nucleus that in these cases their subsynaptic complexes were almost right up against the nuclear membrane and its pores.

Synaptic contacts often appeared also between two large pale profiles similar in structure to the leading processes of developing nerve cells. The synapses formed in this way could have a morphologically different organization: Besides definitely asymmetrical synaptic contacts, at times even with multiple active zone, so-called symmetrical forms also were found here, in which the subsynaptic structures were poorly defined.

Many of the synapses now being described were situated in the marginal layer, where, although they occurred throughout its thickenss, they were concentrated mainly in the ventrolateral region, close to the limiting neurons. Frequently they could also be found in the interneuronal neuropil of the presumptive motor nucleus, now beginning to be formed (Fig. 1D). Morphologically they appeared to be perfectly mature, with clearly developed pre- and postsynaptic structures and an accumulation of synaptic vesicles in the active zone of most of them.

Hence, at the end of the 5th and, in particular, at the beginning of the 6th week of human intrauterine development, synapses of varied structural organization and location appear in the anterior horns of the spinal cord in the brachial region, and which, by morphological criteria, must be classed as functionally competent forms. Their number is much greater in the 10-mm embryos than in 8-mm, and the area of their distribution is much wider.

This early appearance of mature synapses in the presumptive motor nucleus and in adjacent regions of the marginal layer of the spinal cord during a period of human embryonic development before any signs of spontaneous or reflex motor activity have appeared [3, 4] raises the natural question of the possible functional significance of these initial synaptic contacts. On the basis of their own observations and of data in the literature, the writers postulate that these synapses appearing so early in embryonic development play a special role in the development and differentiation of prospective neurons, and in the formation of their definitive morphological and functional status. Even at this stage it can be confidently suggested that it is through these early synapses that specific information must reach the developing nerve cells and must exert an ever-increasing influence on the differential activity of their genome, thereby directing their future cytodifferentiation and adapting them to the greatest possible degree to the tasks they will have to form when the functional systems of the developing organism have been created. The time of appearance of first synaptic contacts on the neuroblastic cell can thus be regarded as the key period in its development, when a qualitative transition from the neuroblastic stage to the stage of the juvenile neuron is completed.

LITERATURE CITED

1. N. Okado, S. Kakimi, and T. Kojima, J. Comp. Neurol., 184, 491 (1979).
2. A. A. Milokhin and I. V. Chernova, Byull. Éksp. Biol. Med., No. 4, 496 (1980).
3. W. E. Windle and J. E. Fitzgerald, J. Comp. Neurol., 67, 493 (1937).
4. J. E. Fitzgerald and W. E. Windle, J. Comp. Neurol., 76, 159 (1942).

EFFECT OF ACOUSTIC STRESS ON THE MORPHOLOGY OF THE RAT SENSOMOTOR CORTEX

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Prolonged exposure to white noise causes structural and functional changes in various formations of the CNS in man and animals. Prolonged exposure to noise leads to disturbance of motor functions also. There is clinical evidence that in persons exposed to noise changes arise in muscle tone and in reflex reactions: Tendon reflexes are increased, but the pharyngeal, palatal, and abdominal reflexes are inhibited. In animals rigidity of movements, motor stereotypy, convulsive seizures, and other disturbances may arise after acoustic stress [4, 6, 7]. There have been only isolated studies of the morphology of the motor cortex under the influence of noise, and these have been undertaken at the light-optical level [7]. Most studies of brain morphology following exposure to noise have been carried out on different formations of the auditory system [1, 3, 5, 8, 9]. The problem of the extent to which noise affects the structure of nonspecific (nonauditory) formations of the CNS, including the motor centers of the brain, has not yet been settled.

The object of this investigation was to study the morphology of the sensomotor cortex and to compare its structure and ultrastructure with changes in the structure of the auditory cortex of rats during acoustic stress.

EXPERIMENTAL METHOD

Noninbred sexually mature male rats weighing 180-200 g were used. The animals were exposed to noise for 14 h daily for 7, 14, and 21 days. The source of the noise was a GZ-12 generator of low-frequency signals, to the output of which columns with a power of 10 W were connected. The frequency band used was 250-3500 Hz and the intensity 80-90 dB above the threshold of audibility of the human ear. A special contact breaker periodically interrupted the noise. The ratio between noise and pause was 1-2 sec. The structure and ultrastructure of the sensomotor cortex were studied in 18 experimental and fixed control rats. Brain sections for light-optical microscopy were stained by Nissl's, Cajal's, and Hortega's methods in Aleksandrovskaya's modification. Pieces of brain for electron-microscopic study were fixed in a 5% solution of glutaraldehyde, then postfixed in 1% OsO_4 solution in phosphate buffer and embedded in Arladite. Sections were cut on the LKB-III Ultrotome, stained by the method in [10], and studied in the JEM-100B electron microscope.

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