migrates immediately after the leading active cells [3, 8]. Unfortunately, it is impossible at present to reproduce the precise sequence of events leading to migration. It is not clear, for instance, whether the initial stage of this process is a change in the intercellular junctions (loss of AGI) or whether these changes arise through active movement of peripheral cells.

The main conclusion from this investigation can be taken to be the discovery of definite correlation between loss of AGI, the appearance of bundles of prekeratin and actin, and expression of AFP synthesis. It is therefore possible to include all these features in a single marker complex of "embryonalization" of the hepatocyte.

For a more detailed study of relations between all these features and processes of reorganization of the structure of the liver, simpler systems reproducing this process in tissue culture are probably necessary.

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ULTRASTRUCTURE OF NEURONS AND INTERNEURONAL CONNECTIONS IN THE SENSOMOTOR CORTEX OF PROGENY OF ALCOHOL-ADDICTED RATS

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KEY WORDS: neurons; interneuronal connections; alcohol intoxication; progeny.

There is clinical evidence of the high degree of risk that parents who are chronic alcoholics will produce physically and mentally defective children. However, the morphological basis of alcohol-induced brain damage in the progeny has not been adequately studied. Experiments on animals have demonstrated the negative effects of simultaneous alcoholic intoxication of females and males on physical development and structure of the higher levels of the motor system in the progeny [9, 10] and have revealed significant disturbances of the ultrastructure of neurons and interneuronal connections of the caudate nucleus under these conditions [14]. There are no data in the literature on changes in ultrastructure of the sensomotor cortex of the progeny of animals following simultaneous alcohol intoxication of females and males.

The aim of this investigation was to study the ultrastructure of neurons and interneuronal connections in the sensomotor cortex of the progeny of alcohol-addicted rats.

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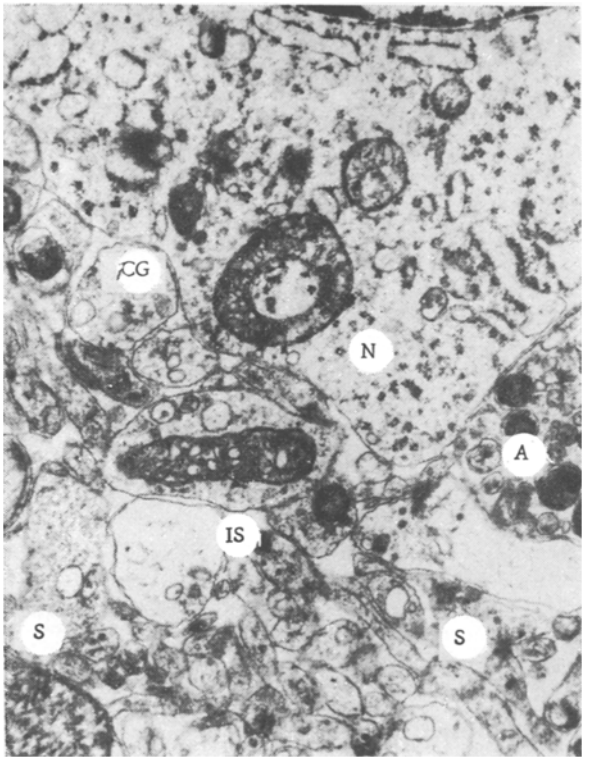

Fig. 1. Ultrastructure of cytoplasm of immature neurons (N), wide intercellular spaces (IS) in neuropil, cones of growth (CG), immature synapses (S), and destruction of axon terminal (A). Magnification 14,400.

EXPERIMENTAL METHOD

Experiments were carried out on 12 female and four male albino rats. Instead of water, the animals were given 15% alcohol solution for 1 month and 25% alcohol solution for 3 months to drink. The rats were then mated and the females continued to consume alcohol during pregnancy and lactation until the young rats acquired vision. The sensomotor cortex of experimental young rats aged 21 and 30 days and of intact animals of the same age was investigated. Material was processed by the formula adopted in the Laboratory of Brain Ultrastructure, Brain Institute, Academy of Medical Sciences of the USSR. The sections were stained with uranyl acetate and lead citrate and studied in the HV-600 electron microscope (Hitachi, Japan).

EXPERIMENTAL RESULTS

Signs of delayed development were found in the sensomotor cortex of the progeny aged 3 weeks.

The nuclear chromatin was located near the inner nuclear membrane, the perinuclear cistern was dilated, and the lamellar complex underdeveloped. The cytoplasm contained a few ribosomes, polysomes, and mitochondria, sometimes ring-shaped (Fig. 1). Wide intercellular spaces also were observed in the neuropil, with cones of growth, unevenness of axonal and dendritic profiles, varicosity of dendrites, poor development of the spinous apparatus, and few synaptic vesicles in the presynaptic process. Degenerative changes also were observed in the neurons and interneuronal connections, in the form of appearance of vacuoles, membranes and myelin-like bodies in the nucleus and cytoplasm of the neurons, edema and destruction of individual cisterns of the rough endoplasmic reticulum and lamellar complex, and also intensification of osmiophilia of some neurons to the extent of their conversion into dark pycnomorphic cells. At the same time there were neurons which showed little change. Osmiophilic oligodendrocytes surrounded by numerous swollen processes of the astrocytic glia also could be seen. In the sensomotor cortex the ultrastructure of mitochondria and of cell bodies and processes was significantly changed. While mitochondria with a dark matrix and

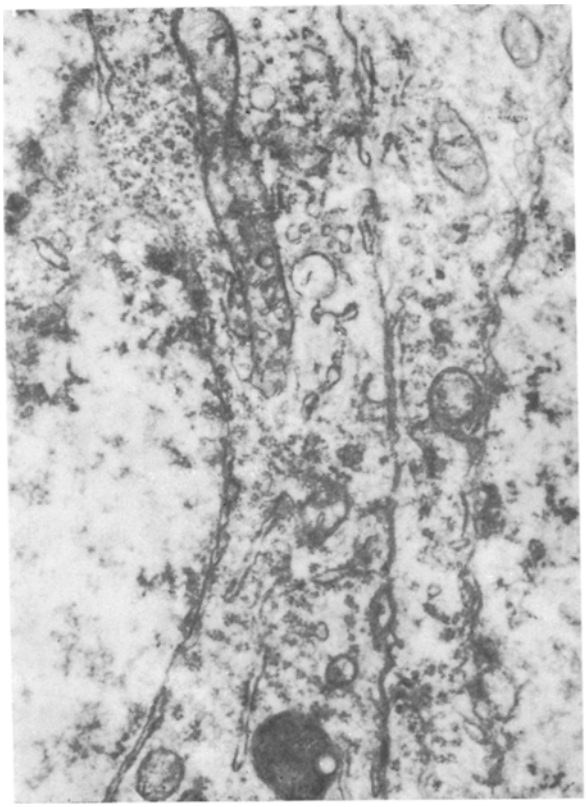

Fig. 2

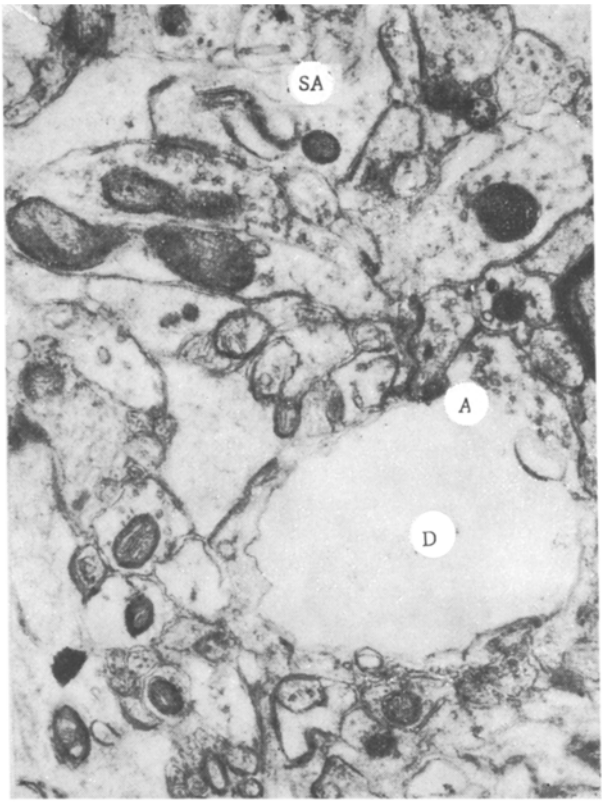

Fig. 3

Fig. 2. Tears of cytolemma in zone of junction of two cortical neurons. Magnification 28,000.

Fig. 3. Single complex of dendrite with pale type of modification (D) and axon terminal (A), swollen sacs of spinous apparatus (SA). Magnification 28,000.

vacuolation of the cristae were present, the majority of mitochondria were swollen, with a reduced number of cristae, and edematous organelles with the appearance of large vacuoles and curiously shaped formations with small remnants of cristae on their inner membrane. Besides altered mitochondria, a reduced number of microtubules and neurofilaments was observed in the dendrites, with the appearnce of numerous vacuoles and vacuole-like cavities. Occasionally dark dendrites were seen. Changes in the axons were less marked than in the dendrites. Only in some of them could large pale vesicles of different shapes be detected, with a small number of synaptic vesicles. Axon terminals were present in a state of destruction (Fig. 1).

In 30-day-old experimental rats swelling and edema of individual cisterns of the laemllar complex, and focal or, less frequently, total clearing of the matrix of the mitochondria, with destruction of cristae, were preserved in some neurons. In other neurons there were marked features of a reparative character: displacement of the nucleolus toward the nuclear membrane, invagination of that membrane, hyperplasia of the vesicular component, and the presence of small and hypertrophied mitochondria. Activation of the nucleus was observed, with the formation of deep invaginations of the nuclear membrane, in which numerous ribosomes and polysomes were concentrated, together with the formation of elements of the rough endoplasmic reticulum from the perinuclear cistern. Paired neurons with a considerable area of contact between their cytolemmas also were found. The latter were most frequently twisted with local pairs, where the cytoplasm of the two neurons fused. In the zone of junction of the neurons different cytoplasmic organelles were concentrated, including hypertrophied mitochondria and large lysosomes (Fig. 2). Sometimes in the paired neurons displacement of the nucleolus toward the nuclear membrane or its invaginations, filled with numerous ribosomes and polysomes and with single cisterns of the rough endoplasmic reticulum, could be seen. The axon terminals contained various numbers of synaptic vesicles. In some terminals, more often with a small number of synaptic vesicles, large, pale vesicles, round or rod-shaped, could be distinguished; sometimes large mitochondria with spherical swellings, almost free from cristae, and with small vesicles in the remaining translucent part of the mitochondria,

were distinguished. Dendrites were more seriously damaged, especially large ones. Focal or complete disappearance of microtubules and neurofilaments, translucency of the dendroplasm, and vacuoles and vacuole-like structures were observed in them. Dendritic profiles showing changes of pale type, and free from organelles, could be fused with the adjacent axon, to form a single complex as a result of destruction of their limiting membranes (Fig. 3). In some areas of vacuolated dendrites the integrity of the membrane was disturbed and the axon terminal, with multiple synaptic vesicles, appeared to open into a dendrite. Sometimes the dendrolemma was destroyed in a zone in which the adjacent axon had no synaptic vesicles. The ultrastructure of the axo-spinous synapses and, in particular, of the spinous apparatus, which consisted of single or several swollen sacs, was considerably changed (Fig. 3).

Alcoholic intoxication of females and males thus causes significant disturbances of the structural organization of the sensomotor cortex in the progeny. Besides delayed maturation of neurons and interneuronal connections, degenerative changes also are found in them; these changes were more marked in rats aged 3 weeks, especially those with signs of retardation of physical development. This was observed also in the caudate nucleus of the progeny in the same model of alcohol intoxication [14]. Disturbances of ultrastructure of the sensomotor cortex observed in the progeny of alcohol-addicted animals are evidently largely determined by cerebral hypoxia. Evidence of cerebral hypoxia is given by the presence of dark neurons and dendrites [2, 5, 6] and by a marked disturbance of mitochondrial ultrastructure. Similar changes in the mitochondria have been found in experimental hypoxia [2] and cerebral anoxia [5, 6], in hypokinesia [8], and in the brain of old animals [1, 8]. Considerable changes in dendrites and spines, detectable at both light-optical [9, 10] and submicroscopic levels, are evidence of disturbance of primary integration of information reaching the neurons and of the synaptic mechanisms of brain activity in the progeny of alcohol-addicted rats. In experimental animals aged 30 days, reparative changes become more evident. Repair of cytoplasmic organelles and the formation of new organelles in some cortical neurons take place in accordance with the principle of intracellular reparative regeneration [11-13], and the appearance of paired neurons is regarded as a manifestation of a compensatory character [3]. The formation of a single dendrite—axon complex as the final stage of invagination of an axon terminal or synaptic contact into a dendrite, allowing direct release of transmitter into the affected dendrite, may also be regarded as a compensatory reaction. Invagination of axon into dendrite has been observed by other workers under different experimental conditions, and this phenomenon is regarded as a compensatory manifestation [2, 4, 7].

Despite the presence of features of reparative processes, degenerative changes in neurons and, in particular, in dendrites are still preserved in the experimental animals at the age of 1 month; this fact must be taken into account when the pathogenesis of alcohol brain damage is considered in children born to alcohol-addicted parents.

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