REVERSIBILITY OF CHANGES IN CORTICAL NEURONS IN EXPERIMENTAL ALCOHOL INTOXICATION

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Numerous investigations have shown significant disturbances of the structural organization of the brain in chronic alcohol intoxication. However, the possibility that morphological changes arising under these circumstances are reversible has not yet been adequately studied. According to data of computer tomography, in patients with chronic alcoholism 6 weeks and up to 1 year after the ending of alcohol intake, in some case the degree of cerebral atrophy, especially cortical decreases [9, 10]. Experiments on animals have shown that changes in neurons during alcohol intoxication for 20 days are fully reversible [3]. During alcohol intoxication for 3 months they persist for 7-10 days, especially in deep brain structures [7]. The time course of the morphological changes in various brain cells after alcohol intake is discontinued depends on the duration of alcoholization. During alcohol intoxication lasting 1 month the structure of the blood vessels and nerve and glial cells is virtually restored 4 weeks after abstinence from alcohol, whereas during poisoning for 3 months, at the same period after discontinuing alcohol intake complete recovery of neuronal structure is not observed and proliferation of the glia remains [2]. During alcohol intoxication for 6, 9, and 12 months, the pathological process continues even a long time after abstinence. In the investigations cited above the response of the dendrites and the spines covering them, which play an important role in the primary processing of impulses reaching the neuron and in the formation of interneuronal connections, was not studied.

The object of this investigation was to study the dynamics of changes in cortical neurons after the ending of chronic alcohol intoxication, paying particular attention to the structure of dendrites.

EXPERIMENTAL METHOD

The model of chronic alcohol intoxication was developed in the Department of Higher Nervous Activity, M. V. Lomonosov Moscow University [1]. The brain of 15 rats receiving 35% ethyl alcohol solution with sugar (150 g/1000 ml) instead of water for 2 months was taken for morphological investigation. The food ration included penicillin in a dose of 10,000 U daily and 0.1-0.15 g of polyvitamins. The animals were killed 2 weeks and 40 and 60 days after discontinuing alcohol consumption. The test material consisted of the sensomotor cortex. The brain was treated by the method of Nissl and Golgi (in I. V. Viktorov's modification). Besides qualitative analysis, methods of morphometry were used. The area of cross section of the bodies and nuclei of 50 large pyramidal neurons were measured in three rats of each experimental series, under a magnification of 280, followed by statistical analysis by Student's test. The number of nerve cells (with a nucleus, and hyperchromic) was counted in two or three rats of each series in 15 grids (magnification 500). The area of the grid with this magnification was 0.04 mm^2, and its volume for a section 10 μ thick was 0.0004 mm^3. Knowing the volume of the grid and the mean number of nerve cells in it, their number in 0.01 mm^3 of tissue of layer V of the sensomotor cortex could be calculated.

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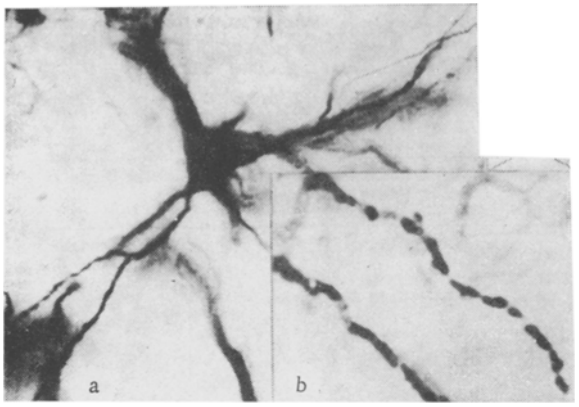

Fig. 1. Neurons in layer V of sensomotor cortex 2 weeks after withdrawal of alcohol: a) reduced thickness and irregularity of impregnation of individual branches of basal dendrities, 300 ×; b) regions of basal dendrites under higher power, 900 ×. Golgi.

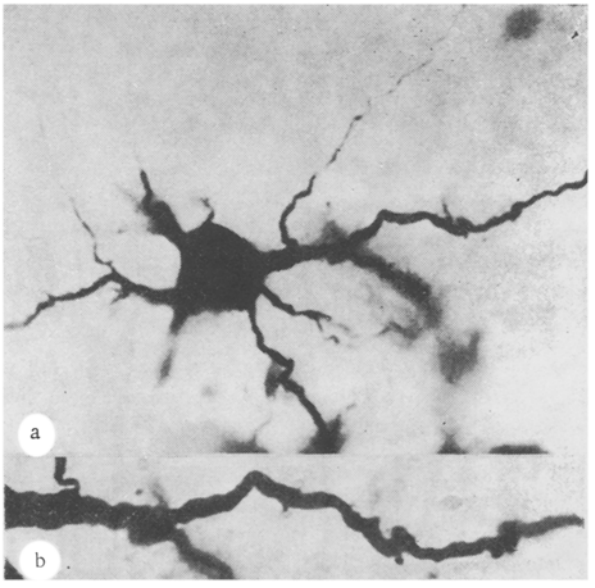

Fig. 2. Neurons in layer V of sensomotor cortex 40 days after withdrawal of alcohol. a) Unevenness of outline and course of dendrites, irregularity of impregnation of individual dendritic branches; b) region of apical dendrite with few spines, 900 ×. Golgi.

EXPERIMENTAL RESULTS

Two weeks after withdrawal of alcohol, just as after 2 months of alcohol poisoning [4], the predominant findings were swelling of the bodies, nuclei, and processes of the cells combined with chromatolysis of the basophilic substance. The area of cross section of the body (334.22 ± 12.3 μ^2) and nucleus (145.44 ± 3.74 μ^2) of the large neurons in layer V was on average a little greater than after poisoning for 2 months (292.74 ± 17.46 and 134.71 ± 4.04 μ^2 respectively), but these differences are on the borderline of significance (Table 1). The presence of karyocytolysis, of cell ghosts, and of neuronophagy and the decrease in the number of neurons in 0.01 mm^3 of layer V to 586.62 compared with 692.25 after poisoning for 2 months, indicate continuing damage to cerebral cortical cells even after withdrawal of alcohol. The basal dendrites of neurons in the lower layers of the cortex were almost without spines, individual branches were reduced in thickness and irregularly impregnated (Fig. 1a, b), and the apical dendrites pursued an undulating course, which is observed after more prolonged alcoholization [5], and is evidence of deafferentation of the dendrites and disturbance of synaptic mechanisms of brain activity in the early stages after the end of alcohol intoxication.

TABLE 1. Area of Cross Section of Body and Nucleus of Large Neurons in Layer V of Sensomotor Cortex after Withdrawal of Alcohol (in μ^2)

Experimental conditions	Body of neuron	Nucleus of neuron
Alcohol intoxication for 2 months	292,74±17,46	134,71±4,04
After withdrawal of alcohol		
2 weeks	334,22±12,3 T=1,94	145,44±3,74 T=1,95
40 days	355,03±7,86 T=4,74	159,56±4,75 T=3,98
60 days	348,61±14,83 T=2,43	165,61±5,4 T=4,58

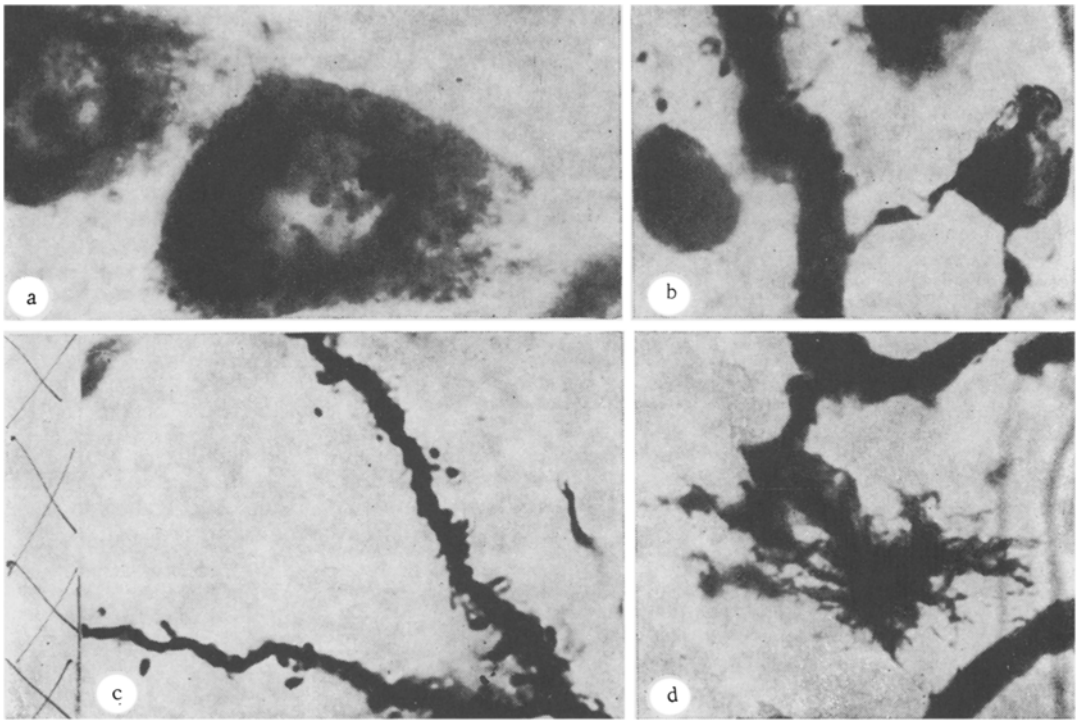

Fig. 3. Structure of nerve and glial cells in layer V of the sensomotor cortex 60 days after withdrawal of alcohol. a) Swelling of processes of large neurons. Nissl; b) diversity of structure and impregnation of spines. Golgi; c) glio-dendritic "contact." Golgi; d) structure of perivascular astrocyte. Golgi, 900 ×.

No significant changes in the structure of the sensomotor cortical neurons were observed 40 days after withdrawal of alcohol. The mean number of neurons per unit volume of layer V of the cortex (604.0) was virtually indistinguishable from that 2 weeks after withdrawal of alcohol (586.62). However, after 40 days there was a statistically significant increase in the area of cross section of the body (355.03 ± 7.86 μ^2) and, to a lesser degree, of the nucleus (159.56 ± 4.75 μ^2) of the large pyramidal neurons of layer V (Table 1), one feature of repair processes [4, 8, 11], which are based on activation of nuclear and cytoplasmic structures [6]. Meanwhile, in some neurons in the lower layers of the cortex, unevenness of outline of the dendrites and a decrease in thickness of individual branches were combined with the presence of a few spines (Fig. 2a). Some of them were hardly visible, others had a hyperimpregnated head or were completely hyperimpregnated (Fig. 2b). Neurons of the upper layers contained a fair number of dendritic spines. Partially impregnated oligodendrocytes, which could form glio-dendritic "contacts," were stained black by Golgi's method. Their formation in the early stages of alcohol intoxication is regarded as a manifestation of compensation [4, 5].

The intercellular structure of most neurons was restored 60 days after withdrawal of alchohol. Cells at this stage had a large nucleus and nucleolus, sometimes displaced toward the nuclear membrane, and with basophilic material uniformly distributed throughout the cytoplasm. However, the apical dendrites of many large cells were thickened, and sometimes the initial portions of the basal dendrites could be seen (Fig. 3a). The area of cross section of the large neurons in layer V was a little less than at the previous time, on average $348.61 \pm 14.83\ \mu^2$, but the area of the nucleus was increased to $165.61 \pm 5.4\ \mu^2$ (Table 1), reflecting its activation (an increase in the mean volume of nuclei is considered to be evidence of intensification of RNA synthesis in cells [11]). At this period the deficit of neurons was more marked, for their mean number in 0.01 mm^3 of layer V was 507.75, appreciably less than after the end of 2 months intoxication and in the earlier stages after withdrawal of alcohol. After 2 months of alcohol deprivation the structural organization of dendrites of neurons in the upper layers of the cortex was similar to that for intact animals. In the lower layers of the cortex, besides neurons with altered dendrites there were cells whose tertiary dendritic branches were covered with rod-like hyperimpregnated spines, spines with a hyperimpregnated head, flask-shaped spines with a pale central zone, and spine-like outgrowths (Fig. 3b). Among the oligodendrocytes there were some which were unevenly impregnated, and also black cells which could form "contacts" on dendrites of cortical neurons (Fig. 3c). Enhancement of the impregnation properties of glial cells of this type is evidence of their increased metabolism, for under normal conditions they never stain black [12]. Pericapillary astrocytes had numerous vascular pedicles (Fig. 3d), facilitating the intensified metabolism.

These observations are evidence of the long-term nature of processes leading to restoration of neuronal structure after the end of intoxication for 2 months and, in particular, of the dendritic system, complete recovery of which is not observed even 2 months after withdrawal of alcohol.

LITEARTURE CITED

1. L. G. Voronin, K. A. Nikol'skaya, and Sh. K. Sagimbaeva, Dokl. Akad. Nauk SSSR, 228, No. 5, 1240 (1976).
2. L. Sh. Galeeva, in: First All-Union Congress of Forensic Medical Experts [in Russian], Karaganda (1976), pp. 586-588.
3. V. A. Il'in, The Nervous System in Alcoholism [in Russian], Kishinev (1959).
4. É. N. Popova, Zh. Nevropatol. Psikhiat., No. 12, 1674 (1979).
5. Ē. N. Popova and G. A. Shchelkalina, Zh. Nevropatol. Psikhiat., No. 4, 539 (1980).
6. D. S. Sarkisov, Regeneration and Its Clinical Significance [in Russian], Moscow (1970).
7. S. A. Sarkisov and G. N. Krivitskaya, in: Morphology of the Basic General Pathological Processes in the Nervous System and Their Organospecific Particular Features [in Russian], Moscow (1972), pp. 40-41.
8. N. E. Yarygin and V. N. Yarygin, Pathological and Adaptive Changes in Neurons [in Russian], Moscow (1973).
9. P. L. Carlen, A. D. Wilkinson, and L. T. Kiraly, Neurology (Minneapolis), 26, 355 (1976) (Abstract).
10. P. L. Carlen, G. Wortzman, K. C. Holgate, et al., Sciences, 200, 1076 (1978).
11. J. E. Edstrom, J. Neurochem., 5, 43 (1959).
12. M. Scheibel and A. Scheibel, in: Biology of Neuroglia, W. F. Windle, (editor) Springfield (1958), pp. 5-23