CHANGES IN NEURONS IN CERTAIN PARTS OF THE RAT BRAIN DURING AMPHETAMINE-INDUCED MOTOR EXCITATION

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Changes in neurons were studied during motor excitation induced by amphetamine. At the time of maximal intensity of amphetamine stimulation after administration of small (1 mg/kg) and average (2.5 mg/kg) doses the nuclei and bodies in subcortical brain regions rich in catecholamines (caudate nucleus, dorsomedial nucleus of the hypothalamus) were increased in size, and if a high dose (10 mg/kg) was given these changes also were observed in the cortex. If a motor stereotype developed, the intracellular structure of the neurons also was altered. The view that portions of the brain rich in adrenergic structures are mainly affected by amphetamine excitation is confirmed.

TABLE 1. Area of Cross Section (arithmetic mean, in μ^2) of Cytoplasm and Nucleus of Neurons of Caudate Nucleus, Hypothalamus, and Cerebral Cortex of Albino Rats (M ± m)

Experimental conditions	Localization					
	caudate nucleus		hypothalamus		cortex	
	cytoplasm	nucleus	cytoplasm	nucleus	cytoplasm	nucleus
a) Control	83,0±0,8	56,2±0,5	76,5±0,9	50,9±0,8	324,2±3,6	139,6±1,1
b) Amphetamine, 1 mg/kg	101,4±1,2	67,5±0,6	80,8±1,4	53,2±0,8	328,1±3,8	144,3±1,2
P	<0,05	<0,05	<0,05	>0,05	>0,05	<0,05
c) Amphetamine, 2.5 mg/kg	95,9±1,2	60,0±0,6	78,5±0,9	54,3±0,7	309,2±3,7	137,3±1,8
P	<0,05	<0,05	>0,05	<0,05	<0,05	>0,05
d) Amphetamine, 10 mg/kg	94,6±1,2	58,9±0,6	82,4±1,0	52,2±0,7	370,9±4,2	159,3±1,7
P	<0,05	<0,05	<0,05	>0,05	<0,05	<0,05
e) Amphetamine, 10 mg/kg on alternate days	102,0±0,9	65,0±0,7	91,6±0,75	55,1±0,5	319,9±2,1	142,5±1,0
P	<0,05	<0,05	<0,05	<0,05	>0,05	>0,05

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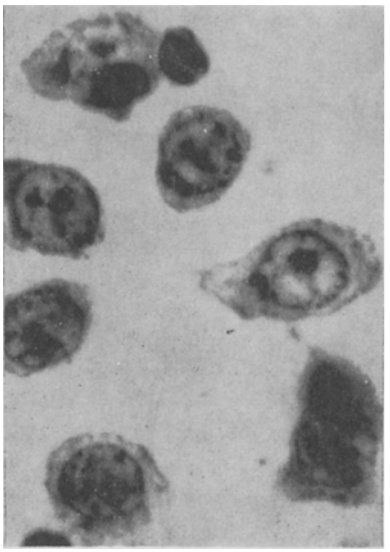

Fig. 1

Fig. 2

Fig. 1. Neurons in caudate nucleus after administration of amphetamine, 10 mg/kg. Swelling of nucleus and enlargement of intranuclear granules. Nissl, 750 ×.

Fig. 2. Large neurons in layer V of sensory motor cortex after administration of 10 mg/kg amphetamine. Swelling of nucleus and body of neuron, apical chromatolysis of Nissl's substance. Nissl, 750 ×.

An important factor in the solution of certain problems concerning the functional morphology of neurons is the use of neurotropic agents, for they allow the course of metabolic processes to be selectively modified and functional activity to be changed in particular brain systems.

In the investigation described below the drug chosen to detect the morphological picture of increased neuronal function was amphetamine, whose stimulant action on the central nervous system, in the modern view, is due to its direct action on the adrenergic systems of the brain [4, 5, 10]. In view of evidence that a cholinergic component also participates in the action of amphetamine [1, 2], the effect of the drug on cholinesterase activity also was studied.

EXPERIMENTAL METHOD

Amphetamine was injected intraperitoneally into male albino rats in doses inducing increased motor activity slightly (1 mg/kg) or considerably (2.5 mg/kg) or causing the development of a motor stereotype (10 mg/kg). These doses are conventionally described as small, average, and large. Control animals received physiological saline, under identical experimental conditions. The rats were sacrificed 2 h later at the height of the stimulant effect of amphetamine, and also 24 h after its injection. Neurons were studied in the caudate nucleus and dorsomedial nucleus of the hypothalamus, in which adrenergic structures are widely represented [12, 14], and in layer V of the sensory motor cortex, which contains few adrenergic structures [15]. The brain was treated by Nissl's method. In three rats of each series of experiments the area of cross-section of 50 neurons and their nuclei was determined by means of an ocular micrometer (magnification 420 ×). Statistical analysis of the results was carried out by the Student–Fischer method. To study changes in the ultrastructure of the cortical neurons during development of a motor stereotype, the material was fixed by Palade's method and embedded in Araldite. Sections were cut on a LKB ultratome and the material examined and photographed in a type UEMV-100B electron microscope. Cholinesterase activity was determined by the method of Karnovsky and Roots in sections through the caudate nucleus at various times after injection of a large dose of amphetamine.

EXPERIMENTAL RESULTS

Small neurons (83 μ^2), in which the large round nucleus (56.2 μ^2) was surrounded by a thin layer of cytoplasm, stained rather more intensely than the nucleus, were most numerous in the caudate nucleus of the control animals. In individual neurons partial chromatolysis was present. The nuclei of the cells were poor in chromatin and usually had one nucleolus.

Neurons in the dorsomedial nucleus of the hypothalamus were smaller than those of the caudate nucleus (76.5 μ^2). Their nucleus (50.9 μ^2) was round or oval in shape. Most cells were hypochromic with central and segmental chromatolysis.

In layer V of the sensory motor cortex, most of the large cells (328.1 μ^2) were normochromic of type. The cell nuclei were large (139.6 μ^2) and relatively poor in chromatin. The nucleolus was large and dark and situated in the center of the nucleus.

After administration of the small dose of amphetamine the intracellular structure of the neurons in these parts of the brain as a rule was unchanged in its appearance by comparison with the control. However, the mean size of the neurons in the subcortical structures was significantly increased (Table 1b).

The nuclei of the neurons also were increased in size but in the hypothalamic neurons this increase was not significant. The large cortical neurons were practically indistinguishable in size from those of the control animals. After an average dose of amphetamine (Table 1c) the nuclei and bodies of neurons in the caudate nucleus and the nuclei of the hypothalamic and cortical neurons were enlarged by a statistically significant degree. The intracellular structure of the neurons of these brain formations was similar to that observed after administration of a small dose of amphetamine.

During the development of a motor stereotype (Table 1d) the cross sectional area of the nuclei and bodies of the neurons of the caudate nucleus was greater than in the control but less than after small and average doses of amphetamine. The number of hypochromic cells showing segmental and central chromatolysis was increased. The cell nuclei stained more intensely and the intranuclear granules were increased in size, and some of them were as large as the nucleolus (Fig. 1).

In the hypothalamus the mean area of cross section of the cell bodies and their nuclei was considerable, but the increase in size of the nucleus was not significant. In the cytoplasm of the neurons chromatolysis of the Nissl's substance varied in intensity and was combined with hydropic changes.

Statistical analysis of the measurements of the nuclei and bodies of the cortical neurons revealed a significant increase in their mean values. Besides swelling of the cortical neurons chromatolysis of the Nissl's substance was detected, more especially in the region of the apical dendrite (Fig. 2). In some cases the central zone of the nucleolus was clear, the cytoplasm showed vacuolation, and considerably swollen, round cells with a much smaller quantity of Nissl's substance appeared. Under the electron microscope a decrease in the number of free ribosomes and of ribosomes located on the membranes of the endoplasmic reticulum was observed, and in individual neurons the cavities of the endoplasmic reticulum were dilated and the matrix of the mitochondria swollen and pale. In the Golgi apparatus the vacuoles were enlarged and the cisterns intact. The nuclei of the neurons were slightly swollen, the nucleoplasm uniformly distributed, and the nucleolus compact. The number of pores in the nuclear membrane was increased, the outer nuclear membrane projected with the formation of cavities in the perinuclear space.

The normal behavior of the rats was restored 24 h after injection of amphetamine. By this time the area of cross section of the cortical neurons and their nuclei was practically equal to that observed in the control rats, whereas in the hypothalamus and caudate nucleus the nuclei and bodies of the neurons were still enlarged (Table 1e) and hypochromic cells were still frequently seen. In the experimental animals no changes could be detected in the cholinesterase activity either at the height of the stimulant action of amphetamine or at various times after a single injection of the drug.

Changes in the functional state of certain brain systems during amphetamine-induced motor excitation were thus reflected morphologically as enlargement of the nuclei and bodies of the neurons, more especially in the caudate nucleus, which has the highest dopamine concentration [10], and in the hypothalamus, with a high noradrenalin level [17].

Enlargement of the neurons of the subcortical brain structures in amphetamine excitation is apparently an adaptive response aimed at maintaining function under the new conditions: an increased dopamine con-

centration [16] and a reduced noradrenalin level [9]. The absence of visible changes in cholinesterase activity suggests that in amphetamine excitation the cholinergic component undergoes no significant changes. However, it is possible that existing histochemical methods are insufficiently selective to demonstrate changes in the active cholinesterase pool in such cases.

During the development of an amphetamine stereotype the changes in the neurons persisted longer in the subcortex than in the cortex. The changes in structure of the neurons were similar to those observed in cortical neurons during physical exertion as the result of prolonged swimming [8], and in motoneurons during training in running [13]. Chromatolysis of the Nissl's substance also was observed simultaneously with an increase in size of the nuclei and bodies of the neurons.

The changes in the neurons detected in these experiments during amphetamine excitation can be explained by an increase in the number of specific intracellular structures so as to provide for an increase in function above the normal level [3, 6, 7].

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