AGE CHANGES IN VASOMOTOR REFLEXES AND ULTRASTRUCTURE OF SYMPATHETIC NEURONS IN RATS AFTER CHEMICAL DESYMPATHIZATION

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The number of cells in the sympathetic ganglia of rats was reduced by means of guanethidine to 30% (group 1) and to 1% (group 2) of normal. In rats aged 2 months pressor responses to asphyxia and to stimulation of the femoral nerve were absent. In the animals of group 1 (but not of group 2) recovery of the reflexes was observed at the age of 4 months. An increase in the number of neurofibrils was demonstrated in the neurons surviving guanethidine treatment, indicating growth of the axon of these cells. Investigation of responses to the indirect sympathomimetic tyramine revealed an increase in the number of effector sympathetic endings at the periphery at the age of 4 months in the animals of group 1. It is suggested that restoration of reflex responses in the animals of this group at the age of 4 months took place on account of growth and branching of the axons of the surviving nerve cells, as a result of which the density of the effector innervation at the periphery was restored.

KEY WORDS: chemical desympathization; blood pressure; neurofibrils; tyramine.

After administration of certain substances (6-hydroxydopamine, guanethidine) into newborn animals the number of nerve cells in the sympathetic ganglia falls sharply [3, 5]. This phenomenon has been called chemical desympathization. The writers showed previously [1, 2] that elevation of the blood pressure in asphyxia and after stimulation of the central end of the divided femoral nerve is not observed after desympathization. However, further investigation showed that the changes in blood pressure, although not arising in desympathized animals at the age of 2 months, can again be observed at the age of 4 months.

The object of this investigation was to study this phenomenon.

EXPERIMENTAL METHOD

Experiments were carried out on 56 rats, of which two groups were desympathized. The rats of group 1 (21) received guanethidine in a dose of 20 μg/g daily for two weeks starting on the first day after birth. Histological investigation showed that the number of cells in the stellate ganglion of these animals was reduced to 30% of the control level. The method of treating the ganglia in order to plant the cells was described earlier [1]. The animals of this group will subsequently be described as partially desympathized. The animals of group 2 (nine rats) received guanethidine daily for 4 weeks after birth. The number of nerve cells remaining in the stellate ganglion of these animals did not exceed 1% of the control. This group of animals will subsequently be described as completely desympathized. The control group consisted of 26 animals. The rats were investigated at the ages of 2 and 4 months.

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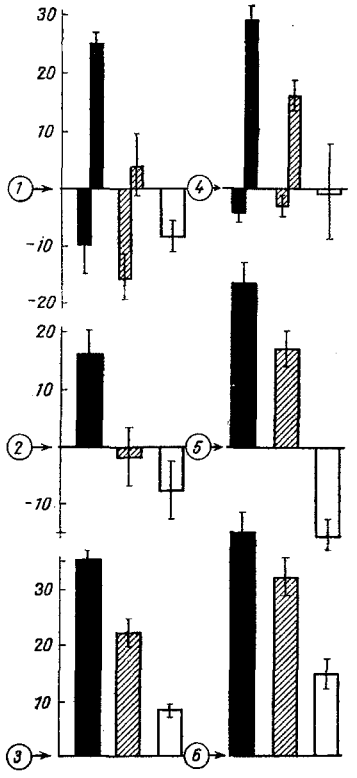

Fig. 1. Changes in arterial pressure responses to asphyxia (1, 4), to stimulation of the central end of the divided femoral nerve (2, 5), and to administration of tyramine (3, 6) in experimental (black columns), partially desympathized (obliquely shaded columns), and completely desympathized (unshaded columns) rats aged 2 (1, 2, 3) and 4 months (4, 5, 6). Ordinate, response of arterial pressure (in mm).

Experiments with measurement of the arterial pressure were carried out under urethane anesthesia (500 mg/kg). The blood pressure was measured in the carotid artery by an electromanometer. Two types of reflex responses of the blood pressure were studied: the response to asphyxia (compressing the tracheotomy tube for 15 sec) and the response to electrical stimulation of the femoral nerve (1 msec, 10 Hz, 10 V, for 10 sec).

For investigation in the electron microscope the stellate ganglia of the experimental and control rats were fixed in 2.5% glutaraldehyde and postfixed in 2% OsO_4. The material was embedded in Durcupan. Sections were cut on the KV ultramicrotome and stained by Reynolds' method [8].

EXPERIMENTAL RESULTS

A marked rise of blood pressure was observed in the control animals of both age groups in response to both procedures – asphyxia and stimulation of the femoral nerve. In the control animals asphyxia evoked a biphasic response: an initial short (3-5 sec) fall of pressure followed by a longer and greater pressor phase of the response. Only a pressor response was observed to femoral nerve stimulation. In the partially desympathized animals at the age of two months the depressor phase in response to asphyxia was deeper and longer than in the control, but the pressor phase was considerably reduced and was not observed in all the animals. In the completely desympthathized animals no elevation of the pressure in general took place during asphyxia. In both groups of desympathized animals no pressor response was observed at the age of two months to femoral-nerve stimulation (Fig. 1). By the age of four months significant changes took place in the character of the reflex responses in the partially desympathized animals. The pressor phase of the response to asphyxia was considerably increased. It was observed in all the animals studied. Stimulation of the femoral nerve also evoked elevation of the blood pressure. The reflex responses in this group of animals were indistinguishable qualitatively from the control, although they remained reduced (Fig. 1). By the age of four months the pressor-reflex responses had not beeen restored in the completely desympathized animals (Fig. 1).

What mechanisms are responsible for the recovery of the reflex responses of the partially desympathized animals at the age of four months? The answer cannot be an increase in the number of neurons in the ganglia. Counting the neurons in the stellate ganglion showed that there was no increase in their number in either group at four months compared with the age of two months. This result is in agreement with the widely held view that differentiated nerve cells of warm-blooded animals have lost their ability to divide.

Results permitting definite hypotheses regarding the causes of this observed recovery of the reflexes with age to be put forward were obtained by comparative electron-microscopic investigation of the residual neurons in the partially desympathized and control animals aged from one week to four months. Attention was drawn to an increase in the number of microfibrils in the cytoplasm of the neurons in the partially desympathized animals (Fig. 2). Similar accumulations of microfibrils in the perikarya of nerve cells are known to arise during stimulation of the growth of their axons [6, 7, 9]. Perhaps in the neurons surviving chemical desympathization intensive growth began, with ramification of the axon, as a result of which the density of innervation at the periphery increased. This process could be the cause of the observed recovery of reflex responses at the age of four months.

Further evidence in support of this hypothesis was obtained in experiments to study responses to the indirectly acting sympathomimetic tyramine. Vasoconstriction observed as a result of the action of tyramine is known to arise through the liberation of catecholamines from sympathetic endings under the influence of this substance [10]. The more endings remaining at the periphery, the greater is the effect of ty-

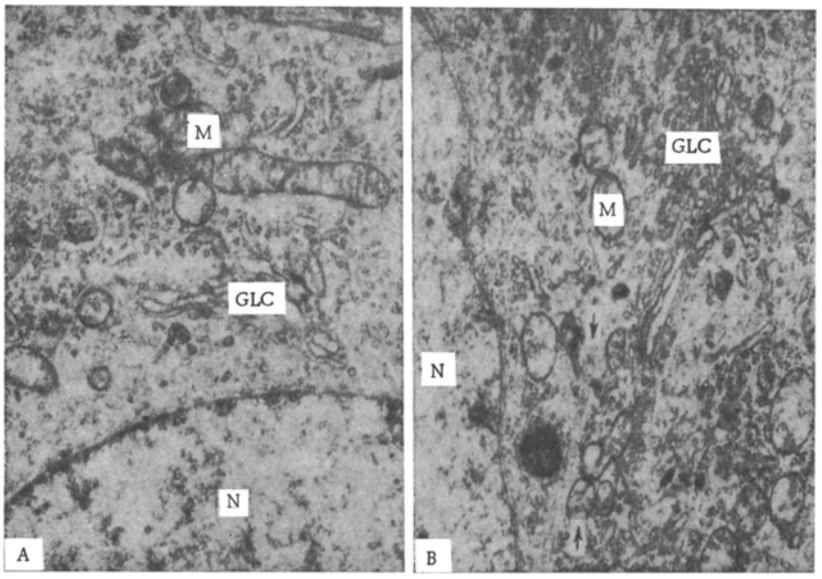

Fig. 2. Increase in number of microfibrils (arrows) in cytoplasm of sympathetic neurons of rats aged 1 month: A) control, B) partial desympathization by guanethidine. N) Nucleus of nerve cell; M) mitochondria; GLC) Golgi lamellar complex, 5000 ×.

ramine, other conditions being the same. However, it must be pointed out that the magnitude of the pressor response observed following administration of a given dose of tyramine cannot be regarded as a precise measure of the number of sympathetic endings on the smooth muscle of the blood vessel, for after desympathization the sensitivity of the effector organ to the mediator is increased [10].

Investigation of pressor responses to a standard dose of tyramine injected intravenously (1.5 μg/g) showed that their magnitude at the age of two months was considerably smaller in the partially and completely desympathized animals than in the controls. By the age of four months the response to tyramine in the partially desympathized animals was increased and had become comparable to the response in the control (Fig. 1). A small increase in the response to tyramine (but by no means reaching the control level) also was observed in the partially desympathized animals, indicating the presence of certain compensatory powers in this group of animals. However, the fact is regarded as important that the most effective recovery both of reflex responses and of responses to tyramine was observed in the partially desympathized animals. It was in that group that a considerable number of cells remained in the symptathetic ganglia, and these cells could be the potential source of restoration of the density of innervation at the periphery as a result of growth of their axons. This explanation does not rule out the possible participation of other mechanisms in the formation of the phenomena observed – for example, processes in the effector organs themselves, leading to an increase in their sensitivity to the mediator.

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