CONSTANT POLARIZATION POTENTIALS IN THE MEDULLA, SPINAL CORD, AND SKELETAL MUSCLE OF THE FROG AFTER DEAFFERENTATION OF THE RESPIRATORY CENTER

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The cause of the depression of respiration and the development of areflexia in the frog after exclusion of the afferent system of the vagus nerves has not been finally settled. After bilateral vagotomy or blocking confined to the aortic reflexogenic zone, some authors [4, 6] have observed irreversible respiratory arrest and areflexia, terminating in death of the frog. The results of many investigations [1-6] have shown that the activity of the respiratory center in amphibians in maintained by afferent impulses arriving mainly from the aortic reflexogenic zone, and for that reason humoral stimuli are no longer adequate for stimulating its activity after deafferentation. Investigation of the constant polarization potenials (CPP) of the frog's medulla before and after vagotomy revealed the development of hyperpolarization, against the background of which paroxysmal inspirations developed and respiration ceased. Since the hyperpolarization developed at a time of deficiency of afferent excitation, itwas described by G. N. Sorokhtin [6] as passive, in contrast to the active process of hyperpolarization inhibition. It has been concluded from the investigations of Yu. B. Temper [7] that the respiratory arrest and areflexia arising after division of the superior laryngeal branches of the vagus nerves are reversible, for the respiration and reflexes were restored fully and spontaneously within a few hours. It has also been reported that application of the cathode of a direct current to the medulla [1, 3], and stimulation of the central end of the divided vagus [4] or sciatic [5] nerves restored respiration in a period of complete apnea.

The object of the present investigation was to continue the study of the CPP not only in the medulla, but also in the spinal cord and skeletal muscle, by recording the state of the respiration, reflexes, and cardiac automatism after deafferentation of the respiratory center.

EXPERIMENTAL METHOD

Deafferentation was carried out either by applying a sponge soaked in 2% procaine solution to the dorsal surface of the bifurcation of the aorta or by bilateral vagotomy as described previously [4, 5]. The investigations were conducted in the autumn and winter on Chinese (Rana chensinensis) and grass (Rana temporaria) frogs. Their CPP were measured by the method of graded compensation, using a P-307 high-resistance potentiometer. The null indicator was a mirror galvanometer (sensitivity 10^{-9} A/mm/m). The CPP were detected by Dubois-Reymond's nonpolarizing electrodes, and the indifferent electrode was applied to the denervated and humorally isolated zone of the right leg. The active electrodes were used to record the CPP alternately from the medulla and the dorsal surface of the dura over the lumbar enlargement of the spinal cord. The third active electrode recorded the potential from the surface of the exposed gastrocnemius muscle of the left leg. Concurrently, by means of an oscillograph, the respiratory movements and the heart rate were recorded, and the reflexes to tactile and mechanical stimulation of the skin of the forelimbs were periodically tested.

EXPERIMENTAL RESULTS

After anesthesia of the aortic reflexogenic zone or bilateral vagotomy, the frogs stopped breathing and paroxysmal inspirations (gasps) appeared. Gradually the gasps became less frequent and were replaced by oscillations of the buccal diaphragm. Apnea then followed, and the reflex excitability was depressed and gradually extinguished completely. The frog appeared to be dead, and only the contractions of its heart showed that it was in a state resembling sleep. Some tens of minutes later groups of respiratory movements returned, and eventually true respiratory movements were restored. After the reappearance

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TABLE. Magnitude and Duration of Changes in Constant Polarization Potentials in the Central Nervous System and Skeletal Muscle after Deafferentation of the Respiratory Center (Mean Data).

Place of measurement	Duration (in min)			Magnitude of hyperpolarization (in mV)	Number of experiments
	1st phase of depolarization	2nd phase of hyperpolarization			
		increase	decrease		
blocking of receptors of aortic zone					
Medulla	5±0.63	23±2.02	62±5.61	1.4±0.23	13
Spinal cord	21±1.05	13±1.83	40±3.56	0.8±0.12	20
Skeletal muscle	27±4.01	34±5.21	55±6.13	1.1±0.26	36
bilateral vagotomy					
Medulla	7±1.32	19±2.50	53±5.31	1.2±0.25	10
Skeletal muscle	16±2.30	25±2.52	62±5.95	0.9±0.29	10

of respiration, the reflexes and movements of the boby returned. Parallel with the phenomena described, hyperpolarization developed in the medulla and spinal cord and also in the skeletal muscle (see Table).

At first when the sponge with procaine solution was applied to the bifurcation of the aorta, respiration was stimulated and primary depolarization developed in the medulla (Fig. 1). Respiration then stopped, coinciding with repolarization, soon changing into hyperpolarization. Against this background of an increasing electropositive potential, gasps appeared and evoked short electronegative waves, which did not interfere with the progressive increase in hyperpolarization. In the process of development of hyperpolarization in the medulla, the oscillations of the buccal diaphragm were rapidly extinguished and by the 22nd minute respiration stopped. Hyperpolarization continued to increase for a short time, and then gradually decreased. When the potential of the medulla had returned to its initial level, the respiratory movements were restored and were accompanied by slow negative waves, with a potential measuring up to -1 mV. With the return to normal respiration, the fluctuations of potential gradually became less frequent and smaller in amplitude. As the activity of the respiratory center was subsequently restored, it was no longer reflected in the curve of the CPP.

As a result of the procaine block the primary depolarization in the spinal cord was more prolonged (Fig. 1, III). It developed against the background of the gasps and continued throughout the period of residual activity of the respiratory center. Hyperpolarization began to develop in the spinal cord only when the activity of the respiratory center had ceased and the hyperpolarization of the medulla had reached a considerable level. In the period of recorery of respiration, negative waves were also recorded in the CPP. However, the smaller amplitude and the constancy of these waves indicated that they also were supraspinal in origin.

The primary depolarization arising in the skeletal muscle after the procaine block was still more prolonged and also gave way to increasing hyperpolarization (Fig. 1, II). The onset of development of hyperpolarization of the skeletal muscle coincided with the maximal level of hyperpolarization of the medulla. In the period of decline of the hyperpolarization in the medulla and spinal cord, the hyperpolarization of the skeletal muscle reached its maximum. Return of the CPP of the skeletal muscle to its initial level coincided with the restoration of reflex activity. In the experiments with bilateral vagotomy (see Table; Fig. 2) similar results were obtained.

In the control experiments division of the vagus nerves was carried out on frogs previously chordotomized at the level of vertebrae II-III. After vagotomy the reflex excitability of the hind limbs persisted throughout the experiment, whereas the cranial reflexes disappeared. Correspondingly, in all 12 of these control experiments hyperpolarization developed only in the medulla. In the spinal cord and skeletal muscle, the CPP remained at a constant level. Preliminary ligation of the femoral artery (in 5 control experiments on frogs with an intact brain) did not modify the usual course of the experiment after bilateral vagotomy. After preliminary division of the sciatic nerve (in another 5 control experiments) but with preservation of the circulation in the hind limb, vagotomy did not modify the CPP of the gastrocnemius

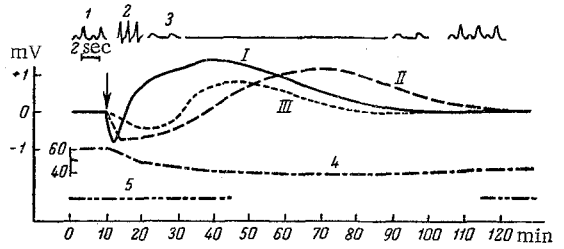

Fig. 1. Changes in constant polarization potentials (CPP) in the brain and muscle following blocking of the aortic receptors with procaine (mean results of all experiments). I - CPP of the medulla; II - CPP of the skeletal muscle; III - CPP of the spinal cord; 1) pulmonary rexpiration; 2) gasps; 3) oscillations of the buccal diaphragm; 4) heart rate; 5) presence of reflexes. Arrow - procaine block.

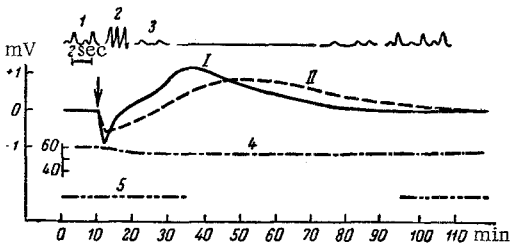

Fig. 2. Changes in the constant polarization potentials (CPP) in the brain and muscle after bilateral vagotomy (mean results of all experiments). I - CPP of medulla; II - CPP of skeletal muscle; 1) pulmonary respiration; 2) gasps; 3) oscillations of the buccal diaphragm; 4) heart rate; 5) presence of reflexes. Arrow - bilateral vagotomy.

muscle, despite the development of hyperpolarization in the medulla and spinal cord. B. D. Kravchinskii [4] showed that depression of the spinal reflexes is the result of blocking of the aortic reflexogenic zone, during which a state resembling shock spreads from the medulla down the spinal cord in a caudal direction.

The results of these experiments, together with information reported in the literature, suggest that bilateral vagotomy or exclusion of the aortic reflexogenic zone should be regarded as an excitation deficit, causing a state of passive hyperpolarization in the medulla. The constant order in the development of hyperpolarization observed in experiments with deafferentation of the respiratory center should be noted. Hyperpolarization of the medulla developed first, after a longer time interval this was followed by hyperpolarization of the spinal cord, and only after the creation of this background of hyperpolarization of the medulla and spinal cord did the skeletal muscle itself become hyperpolarized.

The temporary cessation of activity of the respiratory center evidently led to the removal of its descending tonic influences on the motor neurons of the spinal cord. It may be assumed that this marked impulse deficit evokes a state of passive hyperpolarization of the motor neurons of the spinal cord, and later in the skeletal muscle. The order of development of the hyperpolarization, starting from the respiratory center, and spreading then to the spinal cord and ultimately to the skeletal muscle, confirms this hypothesis.

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