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KEY WORDS: denervation; digastric muscle; muscle fibers; neuromuscular spindles.

Besides their specific receptor function, skeletal muscle afferents also have a neurotrophic influence on muscle fibers [1, 3]. After division of the sensory roots of the spinal cord and of other afferent nerves regional dystrophic changes develop in skeletal muscles. However, besides denervation of sensory nerve endings in muscles, such an operation is also followed by considerable disturbances in cholinergic and histaminergic nervous influences on blood vessels and the microcirculation, which play an important role in the genesis of the dystrophic changes in the tissues in the region of deafferentation [3, 7].

The aim of this investigation was to determine the more precise character of denervation changes and, in particular, of the electrogenic properties of the membrane of various kinds of fibers in the rat digastric muscle (DM), which does not naturally contain muscle spindles [8, 9, 12].

### EXPERIMENTAL METHOD

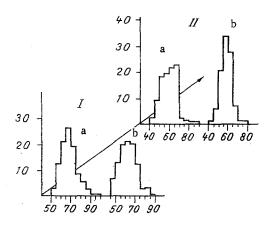
Experiments were carried out on noninbred albino rats weighing 160-210 g. Under sterile conditions, under pentobarbital anesthesia (4 mg/100 g body weight) segments (3-4 mm in the anterior and 1-2 mm in the posterior bellies) of branches of the V and VII nerves innervating the corresponding belly of the left DM, were excised. The wound was then closed in layers. The animals were used in acute experiments on the 3rd-5th day after denervation. Under pentobarbital anesthesia (4 mg/100 g body weight) the corresponding belly of DM was dissected and part of the fascia removed. A bath was formed from the skin and adjacent tissues and filled with warm mineral oil at 37°C. The animal was fixed to a frame for microelectrode investigations, the jaws were fixed with metal clamps secured to the upper and lower incisors, and the corresponding belly of DM was stretched by a glass hook attached to the intermediate tendon. Resting membrane potentials (RMP) were recorded and the cell simultaneously stimulated by means of Pyrex glass microelectrodes filled with 3M KCl solution (resistance 4-10 M $\Omega$ ). The same microelectrode, connected to a bridge circuit [2], was used for intercellular testing of the cytoplasmic membrane of the fiber with depolarizing pulses. The amplitude of the action potential (AP), the critical depolarization level (CDL), rheobase values of stimulating currents, and the latent period (LP), magnitude, and duration of the negative after-potential were investigated. The microelectrodes were inserted into the muscle to a depth of not more than 2.5 mm (the thickness of both bellies is about 3 mm). The experimental results were subjected to statistical analysis by Student's test.

## EXPERIMENTAL RESULTS

A layer by layer study of the fiber composition of the two bellies of DM showed that myocytes with high (70-90 mV) and average (60-70 mV) levels of RMP (Fig. 1, I,a) predominated in the anterior belly, which is a flat muscle, and muscle fibers of different types were uniformly distributed by depth. In the posterior belly, however, which is spindle-shaped, myocytes with average and low (40-60 mV) RMP levels predominated (Fig. 1, I, b) and fibers in different layers differed quite clearly in the magnitude of their RMP and AP (Fig. 2).

When these results are compared with those of histochemical investigations [4, 6] considerable similarity can be seen: in both cases mainly fast glycolytic fibers with high values

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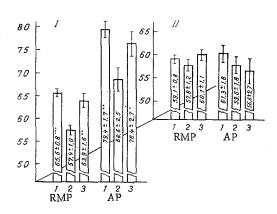


Fig. 1

Fig. 2

Fig. 1. Histograms of distribution of RMP of muscle fibers of rat DM under normal conditions (I) and after denervation (II). Abscissa, RMP, mV; ordinate, number of fibers, %; a) anterior belly; b) posterior belly.

Fig. 2. Layer by layer distribution of RMP and AP of muscle fibers of posterior belly of rat DM under normal conditions (I) and after denervation (II). Abscissa, layers of muscle fibers: 1) 0.01-1.00 mm, 2) 1.01-1.70 mm, 3) 1.71-2.50 mm; ordinate, RMP and AP, mV.

TABLE 1. Electrophysiological Characteristics of Fibers of Anterior and Posterior Bellies of Rat DM under Normal Conditions and after Denervation (M  $\pm$  m)

Experimental conditions	RMP	AP, mV	CDL	Rheobase value of stim- ulating current, nA	LP, msec	Negative after-potential	
						amplitude, mV	duration, msec
Anterior belly							
Norma1	$68,8\pm0.7$	$ 80,9\pm1,8 $	$12,94\pm0,37$	$ 8,12\pm0,52 $	6,79±0,30	$6,03\pm0,35$	$6,17\pm0,21$
Denèrvation	$ \begin{vmatrix} (177) \\ 60,4\pm0,9*** \\ (87) \end{vmatrix} $	$\begin{bmatrix} (49) \\ 58,1\pm 1,0*** \\ (54) \end{bmatrix}$	(46) 10,06±0,43*** (36)	$ \begin{array}{c c}  & (77) \\  & 4,18 \pm 0,34 *** \\  & (25) \end{array} $	$ \begin{array}{c} (91) \\ 7.59 \pm 0.64 \\ (29) \end{array} $	$ \begin{array}{c} (32) \\ 5,68 \pm 0,21 \\ (44) \end{array} $	$ \begin{array}{ c c c c c c c c c c c c c c c c c c c$
Posterior belly							
Normal	$63,1\pm0,8$ (231)	$76,4\pm1,5$	$14,03\pm0,28$ (54)	$8,98\pm0,59$ (66)	$4,78\pm0,19$	7,84 $\pm$ 0,30	$6,61\pm0,33$
Denervation	$58,7\pm0,8**$ (82)	$\begin{bmatrix} 57,7 \pm 1,3 *** \\ (37) \end{bmatrix}$	$12,36\pm0,42**$ (25)	$8,11\pm0,61$ (33)	$ \begin{array}{c c}  & (65) \\  6,54 \pm 0,68 * \\  & (40) \end{array} $	$ \begin{array}{c c} (53) \\ 5,33\pm0,38*** \\ (26) \end{array} $	$ \begin{array}{ c c c c c c c c c c c c c c c c c c c$

<u>Legend.</u> \*P < 0.05, \*\*P < 0.01, \*P < 0.001. Number of muscle fibers tested shown in parentheses.

of RMP and AP predominated in the anterior belly, whereas oxidative fibers with a mainly aerobic type of metabolism, and with lower values of RMP and AP than glycolytic fibers, predominated in the posterior belly.

In the next part of the investigation the same parameters were studied 3-5 days after denervation of the anterior or posterior belly of DM. It will be clear from Fig. 1 and Table 1 that under these circumstances the fiber composition of both bellies of DM was significantly altered. The histograms in Fig. 1 show a sharp decrease in the number of highly polarized fibers in both anterior and posterior bellies, with a corresponding increase in the number of myocytes with average and low levels of RPM.

Confirmation that after denervation it is mainly the fast, highly polarized muscle fibers that are affected is given by data on the changes in the layer by layer structure of the posterior belly of DM after denervation (Fig. 2). It will be clear from Fig. 2 that after denervation differences between the muscle layers disappeared; the greatest decrease in levels of RMP and AP was observed in layers 1 and 3, i.e., in the outer layers, composed of biers with high and average levels of polarization, and a much smaller decrease was observed in the inner layer, consisting mainly of fibers with average and low levels of polarization.

A study of the parameters of excitability of the cytoplasmic membrane of DM fibers (Table 1) showed that after denervation of the corresponding belly typical denervation changes developed in it, for the existence of the intermediate tendon between the bellies prevents any possibility of crossed innervation. These changes were manifested as a fall in the levels of RMP, AP, and CDL (in both bellies), the rheobase values of the stimulating current (in the anterior belly), and the amplitude of the negative afterpotential (in the posterior belly), and as an increase in LP (in the posterior belly) and duration of the negative after-potential (in both bellies). It can be concluded from analysis of the character of these changes that they were virtually indistinguishable from disturbances of the electrogenic properties of fibers of other muscles which have both sensory and motor innervation [5, 10, 11].

These results as a whole are evidence that neurotrophic influences on muscle fibers in muscles opening the mouth are affected mainly by terminals of efferent nerves.

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# ERYTHROCYTE RESPONSE AND METABOLIC CHANGES IN LONG-TERM EXPERIMENTAL HYPERCATECHOLAMINEMIA

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Recent investigations are evidence of increased interest in the study of the mechanisms of involvement of the blood system in adaptation to stress and to extremal factors [1, 3, 13]. Changes in erythrocyte metabolism and the role and participation of enzymes in its disturbances have received careful study [14]. The view is now held that mechanisms of individual resistance to stress must be investigated at molecular and cellular levels, and that blood cells must be an informative object for study in this connection [12].

Considering the leading role of the sympathicoadrenal system in adaptation processes, it was decided to study the response of the erythron and metabolic shifts on a model of hyper-catecholaminemia developed in the writer's laboratory [11].

### EXPERIMENTAL METHOD

Experiments were carried out on 17 mongrel dogs of both sexes weighing 15-20 kg. Under thiopental sodium anesthesia chronic catheterization of the aorta and superior vena cava was

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