ELECTROGENESIS OF MUSCLE FIBERS OF THE RAT MASSETER MUSCLE

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It was shown by intracellular recording of resting membrane potentials (RMP) and action potentials that the superficial layers of the rat masseter muscle contain chiefly fibers with a high MPP and small overshoot, whereas the deep layers contain mainly fibers with a low MPP but a high overshoot. The excitability of the cytoplasmic membrane of muscle fibers with different MPP levels was found to be similar with respect to its electrical parameters. It is suggested that the rat masseter muscle contains a high proportion of fast phasic fibers in its superficial layers and slow phasic fibers in its deep layers.

KEY WORDS: masseter muscle; muscle fibers.

Many skeletal muscles have been shown to consist of different types of fibers: fast, slow, and intermediate [2, 7, 11]. Under experimental conditions it is easy to modify the composition of muscle fibers, with the consequent development of changes in functional properties of the muscle as a whole [3, 9]. According to data in the literature, disturbances of this sort can be observed not only in the limb muscles [12], but also in the muscles of mastication, where changes are found in the relative proportions of the fibers and in their histochemical properties [10]. However, investigations of the pathogenetic mechanisms of disturbances of activity of the muscles of mastication require establishment of the normal composition and characteristics of muscle fibers in experimental animals. It was accordingly decided to study the parameters of the various types of fibers composing the masseter muscle in the most commonly used experimental animals, namely albino rats.

EXPERIMENTAL METHOD

Experiments were carried out on noninbred albino rats weighing 150-180 g. The animals were anesthetized by intraperitoneal injection of pentobarbital (3-4 mg/100 g body weight), the masseter muscle was dissected, and its fascia was partly removed. A hollow was formed from the skin of neighboring tissues and filled with warm mineral oil (37°C). The animal was secured to a frame for microelectrode investigation and the muscle was stretched a little by separating the upper and lower incisors by 10-12 mm with metal forceps. To record resting membrane potentials (RMP) and, at the same time, to stimulate the cell, Pyrex glass microelectrodes filled with 2.5 M potassium chloride solution, with a resistance of 4-10 M Ω , were used. The same microelectrode, connected to a bridge circuit [1], was used for intracellular testing of the cytoplasmic membrane of the fiber with depolarizing pulses. The microelectrodes were inserted into the muscle to a depth of not more than 3 mm (thickness of the muscle 4-4.5 mm). The experimental results were subjected to statistical analysis.

EXPERIMENTAL RESULTS

Because the muscle fibers in the mass of limb muscles are heterogeneous in composition [5], it was decided to study MPP of muscle fibers of the masseter muscle at depths of not more than 3 mm from the surface. RMP of 300 fibers was recorded. As the histograms in Fig. 1 show, most fibers of the rat masseter muscle have RMP of the order of 50-70 mV. There are significantly fewer fibers with a higher RMP. In view of data in the literature indicating the need to make allowance for scale when separating muscle fibers layer by layer (in the masseter muscle of the bull-toad [6], for example) eight layers, each 370 μ thick, were conventionally distinguished in the rat masseter muscle. In all cases the functional properties of the cytoplasmic membrane of the muscle fibers were tested by means of microelectrodes inserted within the cell.

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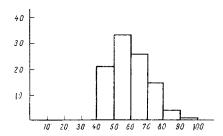


Fig. 1. Histogram of distribution of RMP of muscle fibers (n=300) of the rat masseter. Abscissa, RMP (in mV) of muscle fibers; ordinate, number of fibers (in %).

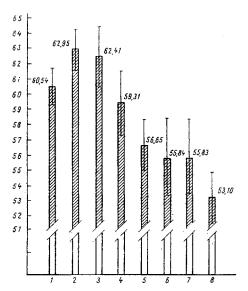


Fig. 2. Distribution of RMP of muscle fibers (n = 300) of rat masseter depending on depth. Abscissa, layers of muscle fibers; ordinate, RMP (in mV).

TABLE 1. Electrical Parameters of Cytoplasmic Membrane of Masseter Muscle Fibers

Depth of fiber from surface, mm	Statistical index	Index of membrane excitability				
		action potential, mV	overshoot, mV	rheobase current of stimulation, nA	critical level of depolarization, mV	latent period, msec
Under 1	<i>M</i> ± m	85,02±1,44	19,84±2,29	12,83±0,64 58	15,92±0,82 58	$9,43\pm0,16$
Under 2	<i>M</i> ±m	84,91 = 2,42	29,64±3,03*	13,20±0,86 23	14,94±0,94	10,45=0,05 †
Under 3	$M \stackrel{n}{=} m$ n	83,20±3,67	$ \begin{array}{c c} 25,06 \pm 3,12 \\ 10 \end{array} $	$12,34 \pm 0,99 \\ 10$	15,07±0,96 10	11,57±0,44 †

^{*}P < 0.01.

Legend. P calculated by comparison with values at depths of under 1 mm; n) number of fibers.

The amplitude of RMP of muscle fibers of the rat masseter was found to depend on the depth of the fibers (Fig. 2). The muscle fibers differed in their level of polarization: high in the first four layers, average in the next three layers, and predominantly low in the eighth and deeper layers.

[†]P < 0.001.

In view of differences found in the parameters of excitability of the cytoplasmic membrane of phasic and tonic fibers [8, 14], it was decided to study the ability of fibers of the rat masseter muscle with high, average, and low degrees of polarization to generate high-voltage spikes in response to depolarization of their membrane through an intracellular microelectrode. Since the distribution of myocytes of the various types of fibers according to degree of polarization differed at different depths in the muscle, these investigations were carried out on muscle fibers at various levels from the surface.

Despite differences in the values of RMP in the different layers of the masseter muscle, during artificial depolarization by dc pulses approximately equal parameters of excitability and ability to generate action potentials of similar amplitude were found (Table 1). Irrespective of their depth in the masseter muscle, muscle fibers in the superficial, middle, and deep layers generated action potentials of the order of 83-85 mV during passage of depolarizing currents into the cytoplasm. However, comparison of the latent period of appearance of spikes after the beginning of passage of the depolarizing currents showed that the time of action potential generation in the deeper layers of the muscle was delayed compared with the more superficial fibers, and the magnitude of the overshoot was smaller in the superficial than in the deep layers.

It can thus be concluded that mainly fibers with high RMP are located in the superficial layers of the rat masseter muscle, whereas mainly fibers with low RMP are found in the deep layers. This last allows us to assume that fast phasic fibers are distributed in the superficial layers and slow phasic fibers are found in the deep layers.

In this respect the rat masseter muscle, despite its unique morphology [4, 13], resembles mixed muscles of the limbs.

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