CHANGES IN FREQUENCY OF MOTOR END-PLATE MINIATURE POTENTIALS AFTER TRANSIENT TETANIC STIMULATION OF THE MOTOR NERVE

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Intracellular recordings were made of motor end-plate miniature potentials (MEPP) from muscle fibers of isolated nerve-muscle preparations of the cutaneous pectoris and sartorius muscles of the frog. Neuromuscular transmission was not blocked. The frequency of the MEPP rose after transient (500 msec) stimulation of the motor nerve at frequencies of 50 or 200 Hz. After stimulation at 200 Hz the frequency of the MEPP rose to a greater degree than after stimulation at 50 Hz, and took longer to return to its initial level. The lower the initial frequency of the MEPP, the greater the increase in their frequency.

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A method which can be used to test the state of the neuromuscular apparatus as it changes during rhythmic activity is to study the reaction of the preparation to short tetanic stimulation of the nerve at different frequencies [1-3]. To shed light on the mechanisms of phenomena taking place in the neuromuscular apparatus during rhythmic activity, posttetanic changes in the electrical response of the myoneural junction and, in particular, changes in the frequency of miniature motor end-plate potentials (MEPP) can be investigated with advantage. The frequency of the MEPP reflects the state of presynaptic structures [10].

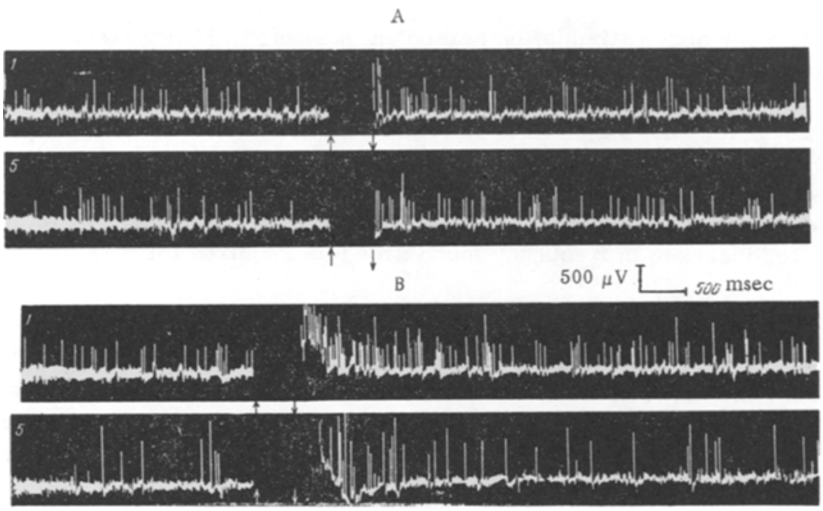

Fig. 1. Increase in MEPP frequency after stimulation of motor nerve at 50 Hz (A) and 200 Hz (B). Preparation of cutaneous pectoris muscle. 1) Result of 1st, and 5) of 5th successive stimulus applied during experiment (stimuli applied every 15 sec). Arrows denote stimulation on and off.

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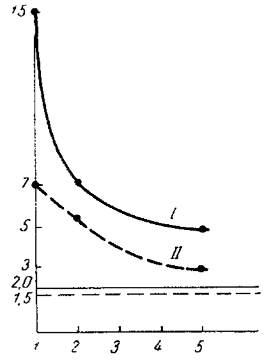

Fig. 2. Relationship between degree of increase in MEPP after nerve stimulation and frequency of stimulation. I) Stimulation at 200 Hz; II) at 50 Hz. Here and in Fig. 3, MEPP frequency (pulses/sec) is plotted along ordinate and time (in sec) after stopping stimulation along the abscissa.

Hitherto changes in MEPP frequency after nerve stimulation [5, 7-9, 11] have been investigated during blocking of neuromuscular transmission produced by an excess of magnesium ions and a deficiency of calcium ions in the physiological saline. In view of facts indicating that the character of posttetanic potentiation depends on the presence of a transmission block, the factor producing the block, and its depth [4, 5], we decided to carry out a series of experiments to investigate the effect of tetanization of the motor nerve on MEPP frequency without blocking neuromuscular transmission.

EXPERIMENTAL METHOD

Experiments were carried out on isolated nerve-muscle preparations of the common frog: 25 experiments on a preparation of the cutaneous pectoris muscle and a series of experiments on a preparation of the sartorius muscle. Potentials from the synaptic region of the muscle fiber were recorded intracellularly by means of "floating" microelectrodes with impedance of 20-30 MΩ, prepared as described by Woodbury and Brady [12]. An ac amplifier with cathode follower at the input were used. The potentials were recorded with a type N-102 loop oscillograph. The nerve was stimulated with square pulses (0.5 msec, 50 or 200 Hz, duration 500 msec) through silver electrodes. To detect changes in MEPP frequency arising immediately after nerve stimulation, the stimulation was applied during a period of continuous recording of the MEPP, using high amplification. For this reason, the deflections of the oscillograph beam corresponding to electrical activity produced by tetanization of the nerve went beyond the edge of the film on these oscillograms.

EXPERIMENTAL RESULTS

In all experiments after nerve stimulation ceased an increase in MEPP frequency was observed (Fig. 1). Comparison of the oscillograms obtained during nerve stimulation at 50 Hz (Fig. 1, A) and 200 Hz (Fig. 1, B) shows that the degree of increase in MEPP was higher after stimulation at 200 Hz. This is also demonstrated by the graph showing changes in MEPP frequency after tetanization of the nerve (Fig. 2). For instance, in one experiment during the 1st second after stimulation at 200 Hz (Fig. 2, I) the frequency of the MEPP was 7.5 times higher than initially, while after stimulation at 50 Hz (Fig. 2, II) it was only 4.7 times higher. After stimulation at 200 Hz the MEPP frequency took longer to return to its initial level, i.e., after a greater increase in frequency more time was required for a return to normal than after a smaller increase.

The character of the increase in MEPP frequency depended not only on the frequency of stimulation, but also on the initial MEPP frequency (before stimulation). With a lower initial MEPP frequency the degree of its increase (the ratio between MEPP frequency after and before stimulation) as a rule was higher than when the initial frequency was lower. This relationship is illustrated by the graph in Fig. 3. Each curve represents the mean results of 6 experiments with a similar initial MEPP frequency. In the experiments used for plotting curve 1 in Fig. 3, a and 3, b, the initial frequency (shown on the graph by lines parallel to the abscissa) did not exceed 1 pulse/sec, while in the experiments used to plot curve II, it exceeded 1/sec. Although the absolute MEPP frequency after simulation was higher in experiments in which the initial frequency was higher (curve II in Fig. 3, a and b), comparison of the MEPP frequency before and after nerve stimulation shows that in the case of a low initial MEPP frequency in the experiments with tetanization of the nerve at 50 Hz it was increased by 23 times (from 0.3 to 7 spikes/sec), compared with an increase of only 4 times (from 3.5 to 14 spikes/sec) when the initial frequency was higher. In experiments with tetanization of the nerve at 200 Hz the MEPP frequency after stimulation increased by 100 (from 0.2 to 20 spikes/sec) and by 10 times (from 2.6 to 26 spikes/sec), respectively. The existence of this relationship between the initial MEPP frequency and the degree of its increase after tetanization of the nerve suggests that the possibility of liberation of mediator from the nerve endings is greater during rhythmic activity of the neuromuscular apparatus than in a resting state, and that this possibility is about equal in synapses of the same muscles in different individuals of the same species. In fact, whereas the

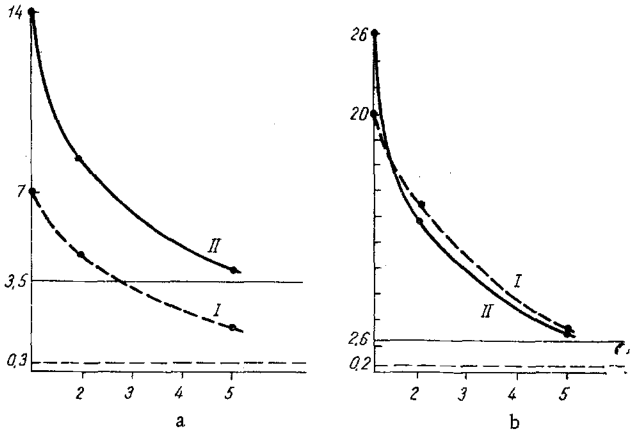

Fig. 3. Ratio between MEPP frequencies before and after nerve stimulation. a) After stimulation at 50 Hz; b) after stimulation at 200 Hz. Remainder of explanation in text.

difference between the initial MEPP frequency levels in the experiments used to plot the graph in Fig. 3 was considerable, after stimulation of the nerve the MEPP frequencies became somewhat closer.

The experimental results showed that the increase in MEPP frequency after stimulation of the motor nerve is typical not only of a blocked myoneural junction when the level of mediator liberation from the nerve ending is preliminarily lowered, but is also evidently a normal property of the junction. The dependence of the degree of increase in MEPP and the rate of decrease in their frequency after stimulation on the frequency of tetanization of the nerve is similar in character in the presence [5] and absence of a block to transmission. The increase in MEPP frequency after tetanic nerve stimulation presumably reflects a process of reorganization associated with mobilization of mediator [6] and with restoration of supplies of mediator ready for liberation from nerve endings.

LITERATURE CITED

1. M. A. Kamenskaya and M. V. Kirzon, Nauchn. Dokl. Vyssh. Shkoly, Biol. Nauki, No. 2, 43 (1965).
2. M. V. Kirzon, Investigation of the Mechanism of Sympathetic Action on Skeletal Muscle and Related Phenomena of Temporary Abolition of Fatigue [in Russian], Doctorate Dissertation, Moscow (1948).
3. M. V. Kirzon and M. A. Kamenskaya, Dokl. Akad. Nauk SSSR, 162, No. 2, 472 (1965).
4. T. N. Oniani, Problems in Comparative Physiology of the Neuromuscular Apparatus [in Russian], Tbilisi (1964).
5. M. Braun, R. F. Schmidt, and M. Zimmerman, Pflüg. Arch. ges. Physiol., 287, 41 (1966).
6. J. C. Eccles, The Physiology of Nerve Cells, Baltimore (1957).
7. J. I. Hubbard, Nature, 184, 1945 (1959).
8. J. I. Hubbard and R. F. Schmidt, Nature, 196, 378 (1962).
9. J. I. Hubbard, J. Physiol. (Lond.), 169, 641 (1963).
10. B. Katz, Proc. Roy. Soc. B., 155, 455 (1962).
11. A. W. Liley, J. Physiol. (Lond.), 132, 650 (1956).
12. J. W. Woodbury and A. J. Brady, Science, 123, 100 (1956).