- 11. O. H. Lowry, N. J. Rosebrough, A. L. Farr, et al., J. Biol. Chem., 193, 265 (1951).
- 12. D. Mayer, M. Stohr, and L. Lange, Cytobiologie, 15, 321 (1977).
- 13. M. Nilsson and T. Berg, Biochim. Biophys. Acta, 497, 171 (1977).
- 14. S. J. Pilkis, Methods Enzymol., 42, 31 (1975).
- 15. K. Weigand, I. Otto, and R. Schopf, Acta Hepato-Gastroenterol., 21, 245 (1974).

SYMPATHETIC INFLUENCES ON SKELETAL MUSCLE STUDIED BY RECORDING MITOGENETIC RADIATION

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Sympathetic stimulation is known to restore the working capacity of the fatigued neuro-muscular apparatus (the Orbeli-Ginetsinskii effect). The sympathetic nervous system exerts its influence through several points of application, with the aid of mechanisms located in pre- and postsynaptic regions [6]. Each stage of neuromuscular transmission is coupled with conformational and energetic reorganizations of pre- and postsynaptic structures [1].

We know from the character of mitogenetic radiation that changes in the functional state of the neuromuscular synapse also are connected with reorganizations of nonequilibrium molecular assemblies (constellations) of the sarcoplasm [3].

The aim of this investigation was to study radiation of the frog gastrocnemius muscle when achieved by sciatic nerve stimulation and restoration of ability to contract by simultaneous stimulation of the sympathetic chain [5].

## EXPERIMENTAL METHOD

Experiments were carried out on male frogs (Rana temporaria L.) in the fall and winter. The frog nerve-muscle preparation was fatigued by the method described previously [5]. To reproduce the Otbeli-Ginetsinskii effect in the frogs the lateral sympathetic trunk was dissected and the seventh and eighth ganglia of the sympathetic chain were laid on electrodes. Electrical stimuli were applied to the sympathetic nerve with a frequency of 15-20 Hz, duration 20-50 msec, and amplitude 0.7-1 V. The gastrocnemius muscle was brought into a state of incomplete fatigue by direct stimulation, for the sympathetic effect is manifested more clearly against this background [4].

A yeast culture, the technique of working with which was described previously [2], was used as a radiation detector. The intensity of ultraviolet chemiluminescence was judged by the increase in the number of young buds in the experimental specimen compared with the control (P=0.99). Mitogenetic radiation was recorded in the early phases of development of the sympathetic effect, with exposures of 3, 5, and 8 sec: during the first seconds of sympathetic stimulation, in the maximal phase of development of the sympathetic effect, and after discontinuation of sympathetic stimulation.

## EXPERIMENTAL RESULTS

The results of experiments to stimulate the sympathetic nervous system against the back-ground of prolonged motor stimulation are given in Table 1. During the first 3 sec of sympathetic stimulation the number of budding yeast cells differed significantly from the control. With an increase in exposure (5 and 8 sec) no significant differences were observed compared with the control. The intensity of radiation fell after discontinuation of sympathetic stimulation.

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TABLE 1. Increase in Number (in %) of Budding Yeast Cells in Experiment Compared with Control ( $M \pm m$ , n = 6)

Exposure,	Resting state	Indirect stim- ulation	State of fatigue	Beginning of sympathetic stimulation (latent period)	ment of sym-	Discontinuation of sympathetic stimulation
3 5 8	$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	$32,0\pm5,5$ $18,5\pm3,1$ $23,0\pm4,6$	$6,0\pm2,5$ $9,5\pm2,1$ $30,0\pm2,4$	22,0±5,9 11,7±4,3 7,3±3,5	$47,4\pm3,8$ $44,2\pm3,2$ $53,0\pm6,3$	27,2±3,8 12,7±3,0 20,1±4,9

The writer previously examined changes in mitotic radiation of the frog gastrocnemius muscle at rest and in a state of active contraction [5].

The similarity between the results obtained with muscles in two different states — the resting state and combined stimulation of the sciatic nerve and sympathetic chain — will be evident. In both cases radiation was found; its intensity during the experiment did not depend on the length of exposure. However, the intensity of the radiation was higher during combined stimulation than at rest or during indirect stimulation of the gastrocnemius muscle (Table 1).

To discover the relationship between exposure and intensity of radiation an experiment was carried out in which threshold exposures of 3 sec were repeated three times with intervals of 1.5 min. The difference between the mean values obtained was not statistically significant. Comparison of the intensity of radiation using exposures of different length (3, 5, and 8 sec) confirmed the relationship of indirect proportionality between exposure time and intensity of radiation observed previously [2], when lengthening of exposure increased the deviation from normal.

It was concluded from the data given in Table 1 that in the resting state and during sympathetic stimulation the neuromuscular system can regulate and compensate any deviations which arise. Judging by the intensity of radiation, compensatory regulation of the neuromuscular synapse is manifested particularly clearly during sympathetic stimulation. This conclusion is in full agreement with views on the modulating effect of the sympathetic system on muscle.

The relatively long latent period of action of the sympathetic nervous system and the late appearance of radiation can as yet be only the subject of conjecture. When radiation of the muscle during the first 10-15 sec after the beginning of sympathetic nerve stimulation was compared with radiation of the previously fatigued muscle, changes were found: in the latter case it was weak (Table 1). Predominance of prepolymer conformational states characterized by weak power of fluorescence can be postulated in the fatigued muscle. Similar changes toward prepolymer conformations may perhaps also spread to synapses, lowering their activity. The commencing sympathetic stimulation leads to a flash of radiation, which is quenched after a few seconds. The subsequent dark period is evidently connected with active structural changes increasing the lability of the substrate gradually, and leading after about 1 min to a balanced dynamic state, linked at this time with intensive radiation. Interaction between muscle and nervous system is intensified, synaptic regions are activated, and contraction develops.

The data described above confirm once again the dynamic and nonequilibrium state of the structural-energetic organization of the sarcoplasm and the importance of its reorganization during changes in its functional state.

## LITERATURE CITED

- 1. R. N. Glebov and G. N. Kryzhanovskii, Functional Biochemistry of Synapses [in Russian], Moscow (1978).
- 2. A. G. Gurvich and L. D. Gurvich, Mitogenetic Radiation [in Russian], Moscow (1945).
- 3. A. A. Gurvich, The Problem of Mitogenetic Radiation as an Aspect of Molecular Biology [in Russian], Leningrad (1968).
- 4. M. V. Kirzon, "The study of the mechanism of sympathetic action on skeletal muscle and related phenomena of temporary abolition of fatigue," Dissertation for the Degree of Doctor of Medical Sciences, Moscow (1948).
- 5. N. N. Lazurkina, Byull. Éksp. Biol. Med., No. 11, 43 (1982).
- 6. I. M. Rodionov, Usp. Fiziol. Nauk, 10, No. 4, 52 (1979).