

TETANIZED SINGLE CONTRACTIONS OF A NEUROMUSCULAR FROG PREPARATION AFTER PANCREATECTOMY

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The phenomenon of the single tetanized contraction has important theoretical significance. It represents the simplest case of summation excitability, which plays an important role in the activity of the central nervous system, and is regarded as the simplest peripheral model of dominance [13].

Single tetanized contractions (STC) are observed when a single wave of excitation passes through an area of sub-threshold or threshold tetanization [2]. The response of contraction to this has the nature of brief tetany, evidenced by an increase in the height and duration of the single contraction.

The development of the single tetanized contraction, apparently, is connected with a certain stage of the parabiologic condition, which is characterized by low lability, low accommodation and high inertness of the flowing excitation [8, 10]. Corresponding to this, the tetanized single contraction was observed to be dependent on the degree to which the trace processes are evident and, in particular, on the residual traces of low-voltage potentials, and also a connection is observed between the latter and the tetanized single contraction and the phase of exaltation [1, 4, 7, 11, 12].

Investigations directed at the clarification of the concrete physico-chemical factors which ensure the conditions necessary for the development of the tetanized single contraction have special interest.

The problem of the present work is the study of the STC under varying chemical conditions of the tissues caused by the preliminary excision of the pancreas. In accord with a number of investigations [5, 6], this operation has an effect on the formation of a chemical agent-acetylcholine, which is of great significance in the activity of the autonomic nervous system.

EXPERIMENTAL METHODS

The experiments were carried out on a neuromuscular preparation of the gastrocnemius muscle of a frog (*Rana ridibunda*). The sciatic nerve was placed on two pairs of electrodes. Sub-threshold tetany was produced by an induction current through the electrodes nearest the muscle, by rhythmic stimulation with single inductive shock every 2 seconds through the more distant electrodes. The preparation was in a humid chamber during the experiment, the muscular contraction was registered under isotonic conditions. The degree to which the tetanized single contractions were evident and their nature were observed, as well as the speed with which the tetanized single contractions developed, according to the time at which they appeared during the experiment.

The experiment was begun 15-20 minutes after the preparation, in a number of cases the preparation was first kept in Ringer's solution from 2-3 to 24 hours in order to ensure the regular reproduction of the tetanized single contraction on each preparation [3].

The pancreas was removed under sterile conditions with ether anesthesia several (4 to 17) days before the experiment. In all, 100 experiments were set up.

EXPERIMENTAL RESULTS

In preparations of frogs which had not been operated, under conditions of constant stimulation of the nerve by single inductive shocks, STC did not appear at once and occurred in far from all the experiments. Usually, the appearance of STC was observed at the 20-30th minute of the experiment, when single contractions already decreased substantially (Fig. 1a). By the 30th minute of the experiment, contractions were observed in 20-30% of the cases. Before complete fatigue developed, hindering further observations, single tetanized contractions were observed in 40-50% of the experiments*.

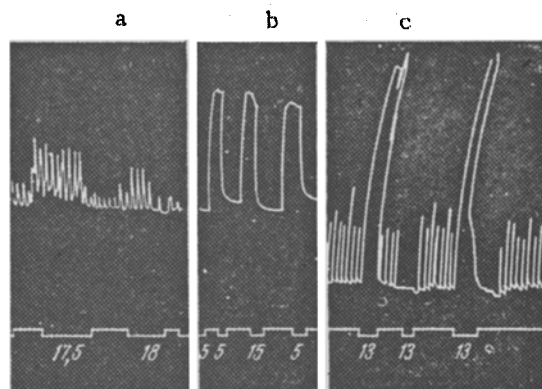


Fig. 1. Myogram of single tetanized contractions (STC).

a) STC at the 30th minute of the experiment; b) equalizing phase, corresponding to the development of STC (tetanic current of 5, 15 and 5 cm between induction coils, in turn); c) STC in a preparation which was kept for 22 hours beforehand in Ringer's solution. Tetany did not occur without single contractions (second stimulus). Single stimuli once every 2 seconds.

In connection with the relatively small number of positive experiments and with the development of fatigue, which hindered observation, we first kept the preparations in Ringer's solution for 2-4 hours. STC were observed considerably earlier on such "surviving" preparations, beginning with the 5-10th minute of the experiment, and had a more evident nature. By the 30th minute they were observed in 70-80% of the experiments. When the "survival" times were extended-after the preparations had been kept for 24 hours - STC were observed during the very first minutes in all the experiments. We noted that in these experiments the STC became so evident that they lost the nature of single contractions and represented a more or less extensive continuous tetany, which decreased only gradually (Fig. 1c).

Prolonged preservation of the preparation in Ringer's solution, represents in its physiological action a very slight, delicate alteration which causes, however, the development of parabiosis with the phases characteristic of it. The equalization phase is especially prolonged and clearly evident (Fig. 1b), which is connected with the presence of STC and the development of the equalization phase was observed by L. V. Latmanizovaya [8] before us.

Preliminary pancreatectomy substantially affected the dynamics of the development of single tetanized contractions. The effect of the excision depended on the length of time after the operation. The clearest changes were observed on the 5-9th day after pancreatectomy. At these lengths of time, ETC already appeared at the 5-10th minute, i.e., 2-3 times earlier than in unoperated frogs, and by the 30th minute they had occurred in 70-75% of the cases. The increase in the duration of the STC and their acquisition of the nature of short tetanies (see Fig. 3a, b, c) was typical. The degree to which the effects of single tetanized contractions were evident and their acquisition of the nature of tetany indicate the very considerable degree to which the lability of the nerve is lowered as a result of the excision of the pancreas.

It should be observed that the muscular contractions were still great when the single tetanized contractions appeared. This indicates that the excision of the pancreas affects the functioning of the neural transmission more than it does the muscle and myoneural junction.

The described changes were, however, temporary: after the 10-11th day, the development of single tetanized contractions in operated frogs again slowed. In Fig. 2 are presented comparable curves of the development of single tetanized contractions in unoperated frogs [1], on the 5-9th day after pancreatectomy [2] and in "surviving" neuromuscular preparations [3 and 4].

* At the beginning of the experiment, sub-threshold tetany was not reflected in the single contractions. With threshold stimulation which caused slight tetany, on the contrary, a decrease in the single contractions was often observed during this period, while at a later time the same tetanization caused an increase in the individual contractions and single tetanized contractions.

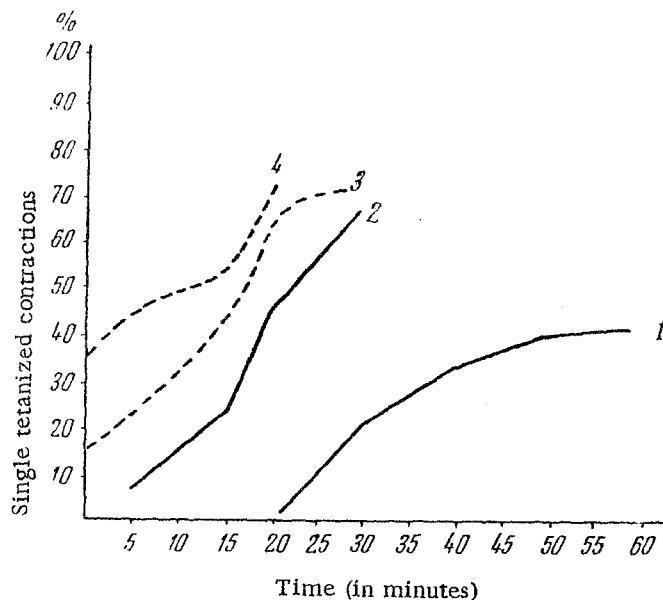


Fig. 2. Development of single tetanized contractions in unoperated frogs (1), on the 5-9th day after pancreatectomy (2), in "surviving" neuromuscular preparations which had been kept in Ringer's solution for 2-4 hours (3), and in preparations after operation and maintenance in Ringer's solution for 2-3 hours (4).

A certain analogy can be observed on examination of the results of experiments on "surviving" neuromuscular preparations and on preparations removed from frogs which had been operated previously. In both cases, the functional state is lowered, the equalization phase develops quickly and marked tetanized single contractions occur, often in the form of tetany. Thus, preliminary pancreatectomy leads to an acceleration of the development of the single tetanized contraction, to the development of a unique biological modification of the neuromuscular apparatus.

If preparations taken from operated frogs are first kept in Ringer's solution, the amount of modification increases still more; single tetanized contractions are obtained at once after 2-4 hours of preservation. In this case, the effects of the STC are especially evident and frequently represent a very prolonged tetany (Fig. 3d).^{*} When the "survival" time is longer, a fall in excitability and an absence of all contractile effects is observed. This disappearance of excitability takes place at times when the preparations, taken from unoperated frogs, give distinct results. All this indicates that there is a sharp decrease in the stability of the neuromuscular preparation after pancreatectomy, even with respect to such a weak modifying factor as Ringer's solution.

Thus, as a result of preliminary pancreatectomy, a rapid and well-evidenced development of single tetanized contractions, a decrease in the lability of the neuromuscular preparation and a decrease in its stability with respect to the action of modifying agents are observed.

On the basis of present knowledge regarding the mechanism of STC, the results described indicate that pancreatectomy leads to a substantial increase of the after-effects—a prolongation of the waves of excitation. Taking into account the data regarding the dependence of STC on the degree to which residual low-voltage potentials and the phase of exaltation are evident, it can be assumed that pancreatectomy leads to an increase of the latter.

^{*} While the size of the individual contractions is still considerable, when STC are observed during the first minutes of the experiment, single tetanized contractions are evidenced not by an increase in the strength of the individual twitches, but entirely by the acquisition of a tetanic nature by the latter.

What does the internal intimate mechanism of the observed changes consist of? The consequences of pancreatectomy are usually connected with a disturbance of the carbohydrate metabolism. However, data exist which indicate that excision of the pancreas leads to a disturbance of the fat and phospholipid metabolism [9]. Kibyakov and Uzbekov [6] showed that at certain intervals of time after pancreatectomy a disturbance of the formation of acetylcholine in the system and a disturbance of the parasympathetic innervation of the heart are observed, apparently as a result of the disturbance of the phospholipid metabolism. Zefirov and Kibyakov [5] established a decrease in lability, a decreased stability of the nerve to modification and a disappearance of tonic-like contractions in a neuromuscular frog preparation after preliminary pancreatectomy. The indicated disturbances proved to be connected with a disturbance in the formation of acetylcholine and were absent when acetylcholine was administered in compensation to the operated animals.

The above data evoke the hypothesis that in our experiments also, the change in the functional properties and development of STC after pancreatectomy is connected with a disturbance of the formation of acetylcholine. In order to check this hypothesis, acetylcholine (0.5 ml of 1:10,000 solution) was administered daily into the posterior lymphatic sacs of the operated animals, beginning on the 2-3rd day after the operation. Single tetanized contractions were absent in this case (Fig. 3e), while unique paradoxical effects were sometimes observed - decreased individual twitches associated with tetany (Fig. 3f). At the same time the pessimal

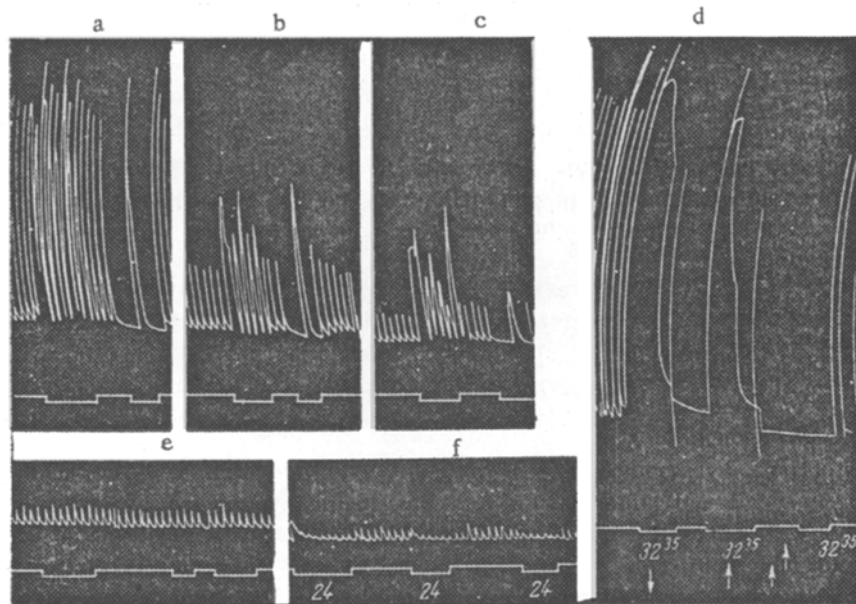


Fig. 3. Myogram of single tetanized contractions (STC). a,b,c) STC on the 8th day after pancreatectomy; the 5th, 15th and 20th minute of the experiment, in order; d) STC on the 7th day after pancreatectomy and a two-hour maintenance in Ringer's solution; e) absence of STC on the 7th day after operation with the compensatory administration of acetylcholine; f) decrease in single twittings under conditions of acetylcholine compensation with tetanization.

nature of the tetanus was observed, which indicates, apparently, that the changes in the lability of the nerve trunk and endings are contradictory. While the lability of the first is increased (as indicated by the absence of STC), the lability of the neural endings is decreased. Lower doses of acetylcholine (1:25,000 dilution) gave less evident results, but they did bring the condition of the neuromuscular preparation back to normal and slowed the development of the single tetanized contraction.

The results of the above experiments indicate that the effects of pancreatectomy are connected with a disturbance in the formation of acetylcholine and are not observed when it is administered in compensation. From this point of view, acetylcholine can be regarded as a regulator of the functional state of the neural conductor, which shortens the duration of the after-effects of excitation, increases the lability and decreases the inertness of the flowing excitation.

SUMMARY

Ablation of pancreas in frogs causes development of single tetanized contractions more rapidly and better expressed than in control experiments on the fifth and the ninth day after operation. This may be accounted for by a decrease in lability of the nervous trunk and intensification of after-excitatory processes. The effects of ablation of pancreas are apparently due to an impediment in formation of acetylcholine in the tissues.

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* In Russian.