EFFECT OF TETANUS TOXIN ON DESCENDING FACILITATORY AND INHIBITORY EFFECTS FROM THE MEDULLA ON MONOSYNAPTIC REFLEXES

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Experiments on cats under superficial nembutal-chloralose anesthesia have shown that during development of tetanus toxicosis stimulation of the facilitatory structures of the medulla is accompanied by increased facilitation of monosynaptic reflexes and by the appearance of polysynaptic activity. Inhibition of monosynaptic reflexes, characteristic under normal circumstances of stimulation of inhibitory structures, is replaced in tetanus by facilitation, which appears during the first few tens of milliseconds after stimulation, when normally inhibition is strongest. Under these same conditions the inhibition persists on the side opposite to that of injection of the toxin.

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Previous investigations [3, 6] showed that during the development of tetanus toxicosis facilitatory effects produced by stimulation of medullary structures are strengthened while the corresponding inhibitory effects are weakened. At the height of development of local tetanus, during stimulation of the facilitatory nuclei of the medulla direct excitatory effects arise, while during stimulation of the inhibitory structures not only does the inhibition disappear, but considerable electrical activity also appears in the muscles of the limb affected by tetanus.

The object of the present investigation was to examine the supersegmental facilitation and inhibition of monosynaptic reflexes during disturbance of inhibitory mechanisms in local tetanus.

EXPERIMENTAL METHOD

Experiments were carried out on cats under superficial nembutal-chloralose anesthesia (10 and 12 mg/kg, respectively). Extensor (stimulation of the nerves to the gastrocnemius and quadriceps muscles) and flexor/stimulation of nerves to the posterior biceps and semitendinosus muscles (PBST) and the deep peroneal nerve (PP)/reflexes were recorded by the usual method from the ventral roots of L5-S1 on the side of injection of tetanus toxin and on the opposite side. To study the temporal course of suprasegmental inhibition of the reflexes, the medullary structures were stimulated by a group of five square pulses. The amplitudes of the monosynaptic reflexes were tested throughout the experiment before every conditioning stimulus for all intervals tested. The results of the experiments were analyzed by statistical methods. Nuclei of the medulla were stimulated with both unipolar and bipolar electrodes: in the latter case the interelectrode distance was 100 μ. The electrodes (tip diameter 5-10 μ) were inserted through the cerebellum in accordance with the coordinates of Szentagothai's atlas. The localization of the nuclei of the raphe (n. raphe pallidus, n. raphe obscurus) was determined from the data of Taber et al. [9]. The position of the electrodes in the brain structures was verified macroscopically after removal of the cerebellum, and microscopically by treatment of the sections by Nissl's method. Local tetanus was produced by injection of tetanus toxin (1/200-1/50 MLD for cats) at several points into the left gastrocnemius muscle and into the posterior group of muscles of the left thigh. This method of injection of the toxin enabled it to reach the regional neural pathways in the region of the anterior horns of the spinal cord of the corresponding segments more uniformly [1].

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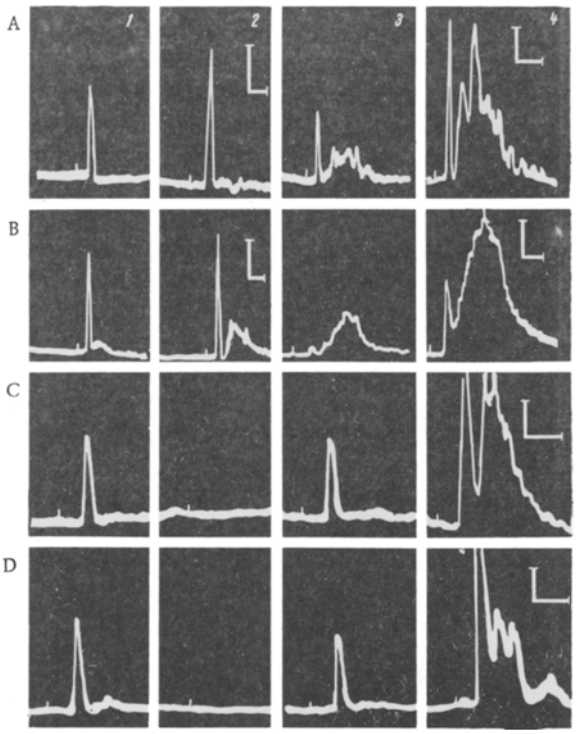

Fig. 1. Effect of stimulation of medullary structures on reflex responses in local tetanus. 1, 2) Reflex responses on side opposite to injection of tetanus toxin; 3, 4) responses on side of injection; 1, 3) before and 2, 4) during stimulation of medullary structures; A and B) effects of stimulation of parvocellular nucleus. A) Extensor monosynaptic reflexes are produced by stimulation of nerves to gastrocnemius muscle. Electrodes located in parvocellular nucleus (P8, L4, H-3), 3rd day after injection of tetanus toxin, 1/200 MLD (for cat). Note differences in strengthening of 1,2 and 3, 4. Calibration 200 μV, time 5 msec. B) Flexor monosynaptic reflexes to stimulation of nerve to PBST muscle. Electrodes located in parvocellular nucleus (P8, D4, H-2), 4th day after injection of tetanus toxin, 1/200 MLD (for cat). Case shown where initial monosynaptic reflexes depress on side of injection of tetanus toxin. Calibration 500 μV, time 5 msec. C and D) Effects of stimulation of gigantocellular and nuclei of raphe (n. raphe pallidus, n. raphe obscurus) in the same animal. Extensor monosynaptic reflexes produced by stimulation of nerve to gastrocnemius muscle. Electrodes located in gigantocellular nucleus (P7, D1, H-2) and in nuclei of raphe (P9, DL0, H-3), 6th day after injection of tetanus toxin, 1/100 MLD. Calibration 250 μV, time 5 msec. In all experiments reflexes tested after stimulation of nuclei for 25 sec; frequency of stimulation 80/sec.

EXPERIMENTAL RESULTS

The experiments showed that stimulation of structures of the parvocellular nucleus and nucleus ambiguus, stimulation of which under normal conditions with the electrodes located in the same place produced facilitatory effects – an increase in amplitude of monosynaptic reflexes on the average by 50% (150±14.6%, results of 35 observations) – in the case of local tetanus produced still greater facilitation on the side of injection of the toxin. Monosynaptic reflexes were increased by 2-3 times or more (on the average by 285.1±42.8%; $P < 0.001$) (Fig. 1A, B). The fact will be noted that an increase in amplitude of the monosynaptic reflexes also took place when their initial amplitude on the side of injection of the toxin was reduced (Fig. 1B, 3, 4). Often stimulation of facilitatory structures by stimuli which normally were subthreshold in magnitude produced excitatory effects, manifested as high polysynaptic activity. Similar responses were observed by the writers previously when recording electrical activity in muscles [3].

Stimulation of the gigantocellular nucleus and nucleui of the raphe (n. raphe pallidus and n. raphe obscurus) in healthy animals produced bilateral inhibition of reflex responses (which in some cases was complete) on the average by 57.6±26.3% (results of 25 observations). No significant difference was found in the degree of inhibition of flexor and extensor reflexes: both were reduced if stimulation of these nuclei was sufficiently strong. Monosynaptic reflexes were inhibited more easily during stimulation of nuclei of the raphe than during stimulation of the gigantocellular nucleus. The same inhibitory effects as in healthy animals (a decrease in amplitude of monosynaptic reflexes by 62.2±22.8%; results of 45 observations) were obtained in animals with local tetanus, on the side opposite to injection of the toxin (Fig. 1B, D, 1, 2).

By contrast, on the side of injection of toxin in the same animals inhibition of monosynaptic reflexes was disturbed: it was weakened on the 1st-2nd day of tetanus and disappeared completely on the 4th-7th day, being followed by facilitation of monosynaptic responses (Fig. 1C, D, 3, 4). Frequently, especially in the later stages of the toxicosis, during stimulation of the gigantocellular nucleus facilitation was accompanied by the appearance of polysynaptic activity: a high-amplitude polysynaptic discharge appeared, and this effect was observed also in cases when, with special selection of the strength of nerve stimulation, only a monosynaptic component was recorded in the initial reflex response

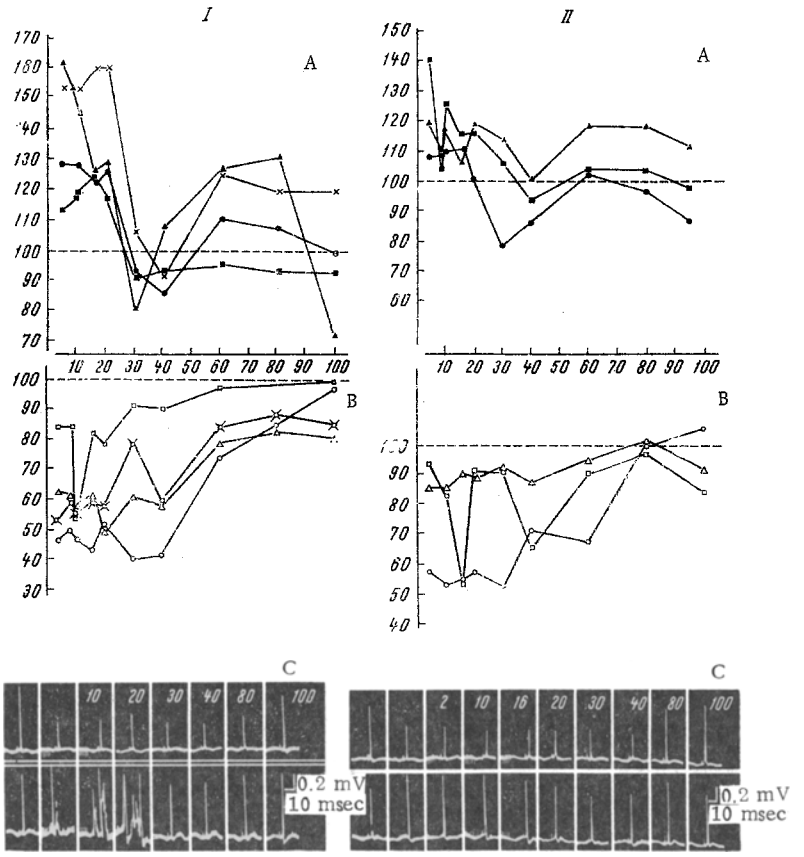

Fig. 2. Temporal course of suprasegmental inhibition of monosynaptic reflexes during stimulation of gigantocellular nucleus. A) Reflexes on side of injection of toxin; B) reflexes on opposite side. I) Extensor monosynaptic reflexes (stimulation of nerves to gastrocnemius and quadriceps muscles); II) flexor monosynaptic reflexes (stimulation of nerves to PBST muscles and PP). Graphs: ordinate, amplitudes of monosynaptic reflexes (in percent of initial); abscissa, time (in msec) between last stimulus of facilitatory stimulation of gigantocellular nucleus and test stimulus. Each curve represents averaged data of experiment on one animal (3-5 tests at the same point). B) Responses recorded in one test of an experiment (averaged curves of all tests of this experiment shown on graphs by circles): bottom row – on side of injection of toxin, top row – on opposite side. First frame shows initial amplitude of monosynaptic reflex, second frame shows monosynaptic reflex during simultaneous application of conditioning stimulus, subsequent frames show amplitudes of monosynaptic responses at different time intervals (in msec, indicated by numbers) after end of facilitatory stimulation of gigantocellular nucleus; 3rd day after injection of 1/100 MLD toxin. Electrodes located in gigantocellular nucleus (P7, L1, H-3).

(Fig. 1C, 3, 4). It is also an interesting fact that sometimes marked polysynaptic components appeared during stimulation of the nuclei of the raphe (Fig. 1D, 3, 4), stimulation of which in healthy animals had a comparatively smaller inhibitory effect on polysynaptic reflexes than stimulation of the gigantocellular nucleus.

A study of the temporal course of inhibition of monosynaptic reflexes showed that normally depression of extensor and flexor monosynaptic responses can be traced up to 100 msec, and it reaches a maximum during the first 20-40 msec. Similar relationships were also observed on the side opposite to injection

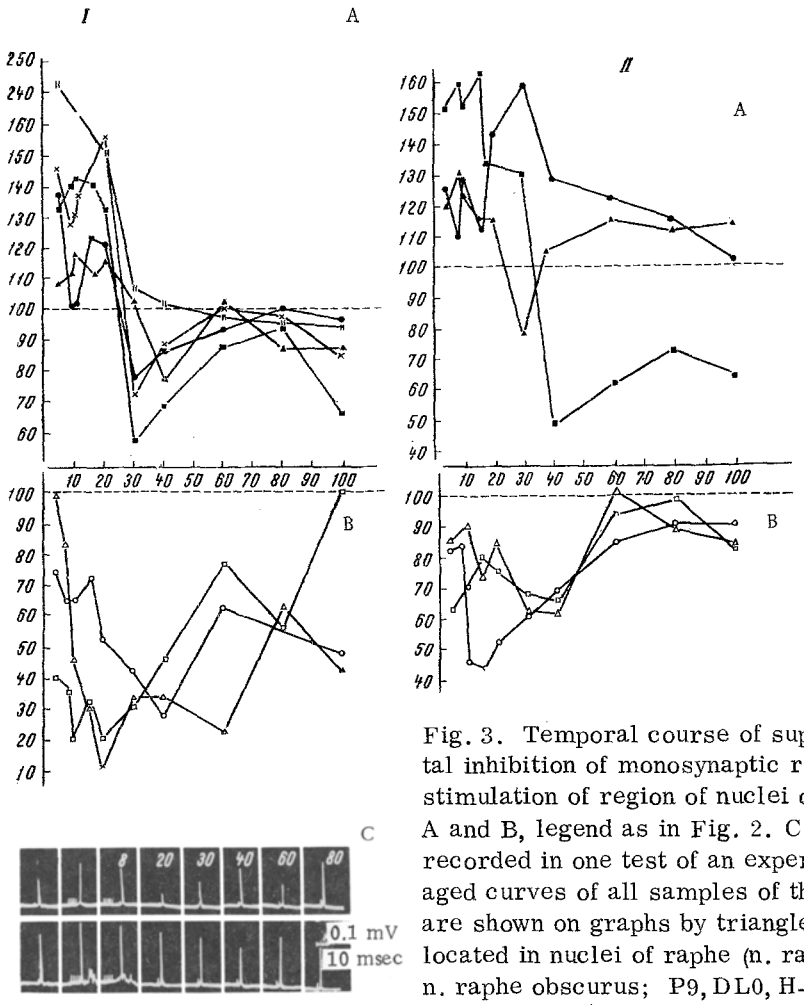

Fig. 3. Temporal course of suprasegmental inhibition of monosynaptic reflex during stimulation of region of nuclei of raphe. In A and B, legend as in Fig. 2. C) Responses recorded in one test of an experiment (averaged curves of all samples of this experiment are shown on graphs by triangles). Electrodes located in nuclei of raphe (n. raphe pallidus, n. raphe obscurus; P9, DL0, H-3), 4th day after injection of 1/200 MLD toxin.

of tetanus toxin (Figs. 2B, Fig. 3B). In tetanus, on the side of injection of the toxin inhibition of monosynaptic responses was considerably weakened or even abolished altogether, being replaced by facilitation of monosynaptic responses (Figs. 2A, 3A), facilitation reaching a maximum in the initial periods (20-40 msec) after conditioning stimulation, i.e., in the period of most marked inhibition under normal conditions. In this period, facilitation of monosynaptic reflexes could be accompanied by the appearance of high polysynaptic activity (Fig. 2C, bottom row).

These effects were found during stimulation of structures both of the gigantocellular nucleus and of the nuclei of the raphe (n. raphe pallidus, n. raphe obscurus), and they were manifested in relation to both extensor (Figs. 2A, I and 3A, I), and flexor (Figs. 2A, II and 3A, II) monosynaptic responses. Meanwhile, on the side opposite to injection of the toxin, in the same animal, preservation of the inhibitory effects was observed during stimulation of these same structures (Figs. 2B, I, II and 3B, I, II).

These investigations thus show that in tetanus toxicosis the types of short-term descending inhibition of monosynaptic reflexes, developing during or immediately after stimulation of the specified medullary structures and lasting for several tens of milliseconds, studied in these experiments are abolished. The fact that during stimulation of the same point in the medulla inhibition is abolished only on the side affected by tetanus, and is maintained on the opposite side, indicates that the abolition of inhibition is effected at the spinal level.

Special attention must be paid to the fact that in tetanus toxicosis not only are descending inhibitory responses abolished, but they are replaced by facilitatory. Similar relationships in tetanus have been found during the investigation of segmental reactions [2, 4, 5, 7] and of descending pyramidal influences [8]. This rule can be explained on the basis of suggestions [3, 4] that during stimulation of structures, stimulation of

which under normal conditions gives an inhibitory effect, the motoneurons are subjected to a mixed influence – both inhibitory and excitatory, and this mixed final message to the motoneurons is assembled at the spinal level, as a result of the arrival of stimulation from the inhibitory nuclei, at both excitatory and inhibitory interneurons. Normally the inhibitory component in the final integrative response is stronger than the excitatory; under conditions when the inhibitory component is abolished, the excitatory is revealed; it then appears considerably strengthened because of the involvement of many interneurons in the response.

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