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THE ACTION OF THIOPENTONE SODIUM ON SKELETAL MUSCLE

BY

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Of those barbiturates which have a short duration of action as central nervous system depressants, thiopentone sodium was the most active in augmenting the muscle twitch elicited by single maximal electrical stimuli applied to the motor nerves of frog or rat nerve-muscle preparations (Quilliam, 1955). Analysis of the pharmacological data obtained from these isolated tissue studies left unsolved the precise mechanism of this barbiturate effect. It seemed that an examination of the changes in the muscle action potential of the rat anterior gracilis muscle, excited through its motor nerve before and after the intra-arterial injection of thiopentone sodium, might shed some light upon the mode of action of barbiturates.

The experiments reported in this paper show that, after the intra-arterial injection of thiopentone sodium, the voltage of the muscle action potential was reduced, its latency was increased and its duration was prolonged—these changes being consistent with a decreased rate of propagation of the potential over the surface of the muscle fibre. The significance of these changes and their relation to the augmentation of the muscle twitch are discussed. Some of the results were communicated to the British Pharmacological Society in July, 1952.

METHODS

Rat Sciatic Nerve Gastrocnemius-Soleus Preparation.—In adult rats, lightly anaesthetized with 1.2 g./kg. of urethane intraperitoneally, the tendo Achillis was carefully freed from the surrounding tissues and attached to an isometric spring steel myograph. The sciatic nerve was exposed in the thigh and severed. Its cut peripheral stump was stimulated maximally by suitable condenser discharges applied through platinum electrodes at the rate of one/sec. and the muscle twitches recorded upon a smoked drum.

Rat Obturator Nerve Anterior Gracilis Preparation.—The anterior gracilis muscle was exposed by a minimum of dissection in the thigh of a rat lightly anaesthetized with urethane administered intraperitoneally. The obturator nerve supplying it was prepared for stimulation with platinum electrodes (Jarcho, Eyzaguirre, Berman, and Lilienthal, 1952). Skin flaps were raised so that a pool of liquid paraffin maintained at 39° C. covered the area exposed. Under these conditions, single maximal stimuli (condenser discharges) applied to the nerve at the rate of one/sec. evoked muscle action potentials, all of which appeared to have the same characteristics for long periods of time. A branch of the abdominal aorta, or the contralateral femoral artery, was prepared so that thiopentone sodium could be injected into the blood supplying the muscle with the minimum of disturbance to the preparation. Throughout each experiment single maximal stimuli were applied to the obturator nerve at the rate of one/sec.

Platinum electrodes, placed upon the superficial surface of the muscle, led off the muscle action potentials to the recording equipment. The amplifiers used were A.C. coupled and had a frequency response to a sine wave-form which was practically flat from 2 cycles/sec. up to 10 kcycles/sec. Thus the amplification system did not introduce appreciable frequency distortion of the muscle action potentials. The amplified potentials were displayed on and photographed from oscilloscopes.

Intra-arterial injections of 0.9% sodium chloride solution at body temperature altered neither the height of the muscle twitch nor the form of the muscle action potentials.

RESULTS

Rat Sciatic Nerve Gastrocnemius-Soleus Preparation.—The intra-arterial injection of thiopentone caused a marked augmentation of the muscle twitches in response to single maximal stimuli applied to the sciatic nerve stump. In the experiment illustrated in Fig. 1, the injection of 2 mg. thiopentone sodium was followed by a rapid augmentation of the muscle twitch which became maximum within about 1 min. and, thereafter, the twitch height declined. In this experiment the respiration failed after about 7.5 min. However, if not more than 1 mg. was injected intra-arterially, the rats survived (see Figs. 3 and 5), but, with 1 to 2 mg. of the drug intravenously, respiratory failure usually followed within one minute.

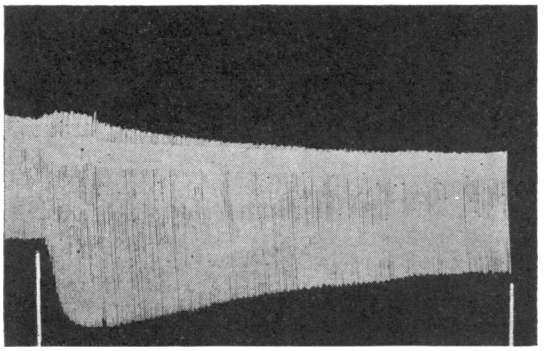

FIG. 1.—Rat sciatic nerve gastrocnemius-soleus muscle. Isometric records of the muscle twitch in response to single maximal stimuli applied to the sciatic nerve at 1/sec.; 2 mg. thiopentone sodium was injected arterially at first mark. Respiration failed at second mark, 7.5 min. later.

As the anatomical arrangement of the fibres in the soleus and gastrocnemius muscles was complex, the anterior gracilis muscle was selected for electrical studies.

Rat Obturator Nerve Anterior Gracilis Preparation.—The amplitude of the muscle twitch elicited by single maximal stimuli applied to the obturator nerve was augmented by intra-arterial thiopentone in a manner similar to that observed with the soleus-gastrocnemius muscle group.

The anterior gracilis muscle is about 35 mm. long, 5 mm. wide, and about 1 mm. thick, running as a parallel-sided strap across the thigh and with a thin flat tendon at each end (Fig. 2). Some muscle fibres appear to run the whole length of the muscle, while others do not, but all the fibres are arranged parallel one to another. The end-plates are gathered in two bands each about 1 mm. wide in the region of the junction of each one-third of the muscle. These are indicated by the letters E–E in Fig. 2. Thus, the muscle action potential recorded in any region may contain certain components arising from both end-plate concentrations, and may vary considerably from point to point along the muscle. When the amplifier leads were upon region C of the muscle (Fig. 2), the record contained, as a major diphasic component, the potential arising from the adjacent end-plate focus, followed by a minor component from the distant end-plate focus. Such a record is illustrated in Fig. 3 at time 0 min. in which a small late deflection was all that was seen of the muscle action potential arising from the distant end-plate focus. Similarly, the muscle action potentials recorded from region A were dominated by a large diphasic component from the local end-plate focus with a minor contribution from the distant focus.

In region B (Fig. 2), the contributions from each end-plate focus were nearly equal in value and the form of the muscle action potential recorded was quadriphasic as in Fig. 5. These results were similar to those of Jarcho *et al.* (1952).

In none of the electrical records made after intra-arterial thiopentone was there any evidence of repetitive muscle action potentials in response to maximal stimuli applied to the obturator nerve. Thus it seemed probable that some mechanism of action of the drug should be sought other than one involving an anticholinesterase action.

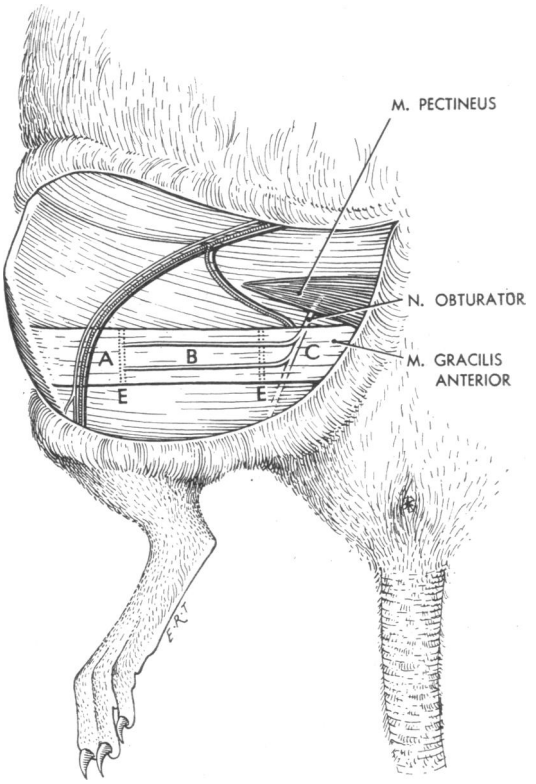

FIG. 2.—Diagram of the rat obturator nerve anterior gracilis muscle preparation. The end-plate regions are localized in two bands about 1 mm. wide denoted by E–E.

In Fig. 3, the muscle action potential was recorded at intervals from the same area in region C of Fig. 2 during a control period of 23 min. of maximal single stimuli applied to the obturator nerve at a frequency of 1/sec. In all, nearly 1,400 stimuli were given, but the form and voltage (amplitude) of the various components and the latencies (i.e., the intervals between the stimulus artifact and the various peaks) of the muscle action potential records remained constant throughout.

FIG. 3.—Action potentials from the rat anterior gracilis muscle in region C (Fig. 2); the obturator nerve was stimulated with maximal shocks at 1/sec. At 25 min. 1 mg. thiopentone sodium was injected intra-arterially and at 39 min. 3 mg. was injected. Oscillograms touched up.

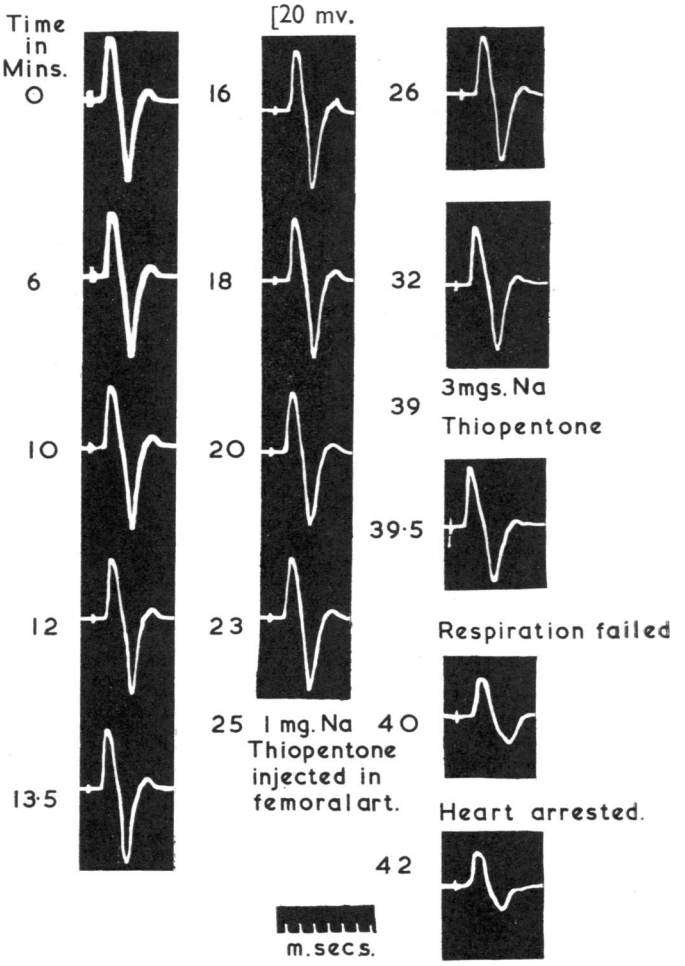

FIG. 3

After 1 mg. thiopentone sodium intra-arterially, examination of the peaks of the potentials recorded one and seven minutes later showed a slight decrease in voltage and an increase in latency. A larger dose (3 mg.), injected at 39 min., caused a considerable reduction in voltage and further increase in latencies of the peaks of the muscle action potential; this large dose caused a failure of respiration followed by cardiac arrest.

The graphs in Fig. 4 have been constructed from measurement of the potentials given in Fig. 3. The solid circles joined by solid lines indicate the increase in latency of peaks *a* and *b*, while the open circles and broken lines demonstrate the fall in voltage of peaks *a* and *b* after the two injections of thiopentone. It can be seen that neither the voltage nor the latency returned to the original values during the short interval between the two injections. At least one hour elapsed before the voltage and the latency of the potentials returned near to normal (Fig. 6).

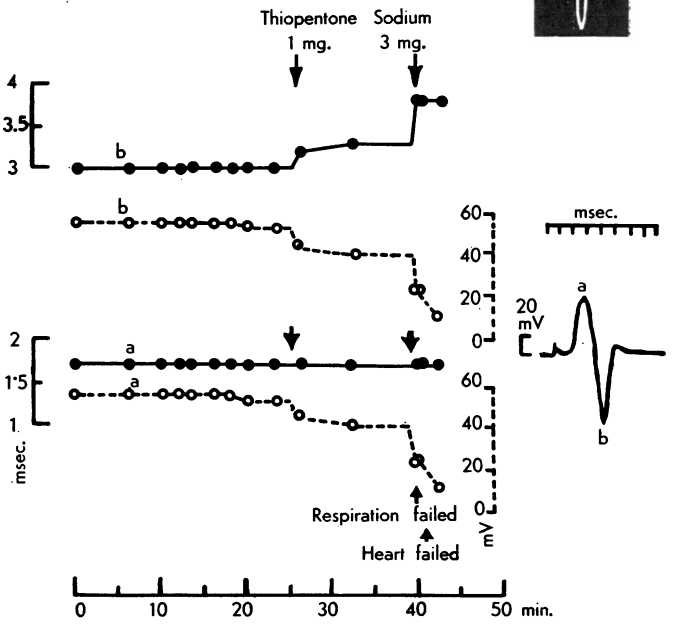

FIG. 4

In another experiment, the muscle action potential, recorded from a point upon the surface of region B (Fig. 2) of the anterior gracilis muscle, presented four peaks for analysis. The injection of 1 mg. thiopentone sodium into the abdominal aorta at 3 min.

FIG. 4.—Summary of experiment depicted in Fig. 3. The open circles represent the voltages of peaks *a* and *b* of the muscle action potential and the solid circles represent the latencies of peaks *a* and *b*.

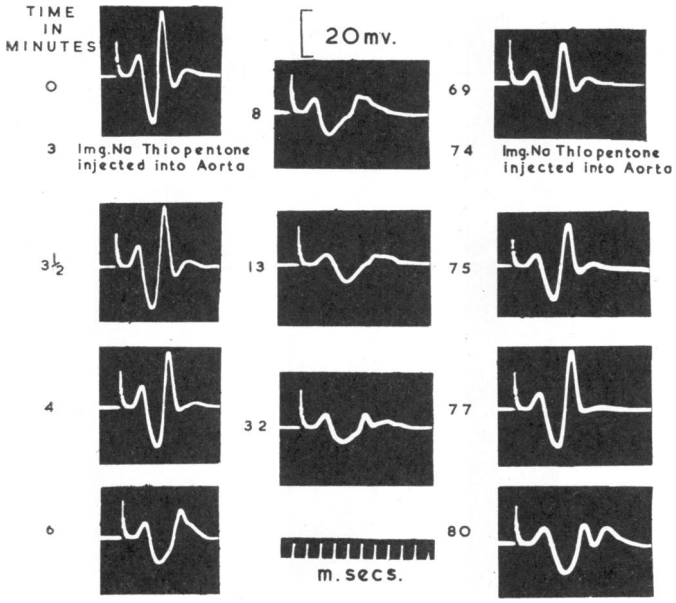

FIG. 5.—Rat obturator nerve anterior gracilis muscle preparation. The muscle action potentials were recorded from a point in region B (Fig. 2). The obturator nerve was stimulated 1/sec.; at 3 min. and at 74 min. 1 mg. thiopentone sodium was injected into the abdominal aorta. Oscillograms touched up.

produced an increase in latency and a decrease of the voltage of all four peaks (Fig. 5); these changes were maximal at about 10 min. after the injection. Recovery of the potentials began after 30 min., and even 66 min. after the injection was not quite complete.

A second injection of 1 mg. thiopentone sodium produced a similar sequence of events. At 77 min. the voltage of the third peak was greater than it was immediately after this injection—a change not seen after the first injection; the last peak of the complex, moreover, was diminished.

The increase in latency and the fall in voltage of each peak of the action potential, as well as its total duration, are expressed graphically in Fig. 6.

DISCUSSION

The results reported in this paper show that intra-arterial injection of thiopentone sodium into rat skeletal muscle augments the muscle twitch elicited by maximal stimuli applied to the motor nerve. The mechanism underlying this effect might be thought to be an anticholinesterase action of the barbiturate promoting the accumulation of the transmitter substance. Such an accumulation might be expected to result in repetitive firing of the muscle action potential after a single nerve impulse. No such repetitive firing was seen. Barbiturates are believed to depress tissue cholinesterase activity only when administered over prolonged periods (see Augustinsson, 1948). Thus it seems unlikely that the effects which thiopentone can exert upon rat skeletal muscle operate by way of an anticholinesterase mechanism.

The present work shows that thiopentone decreases the voltage of the muscle action potential, increases its latency and prolongs its duration. These effects could occur if the rate of propagation of the potential was slowed, or if the wave of depolarization was wider, thereby taking longer to pass across the point from which the electrical record was made. Within the limits of the technique used here it cannot be decided which of these two possibilities operates.

However, there is evidence to show that agencies which slow the rate of propagation of the wave of depolarization along the muscle fibre membrane are associated with an augmentation of the twitch tension. For example, this is so at subnormal temperatures, and during the post-tetanic augmentation of the muscle twitch (Brown and von Euler, 1938). These authors found that the injection of potassium chloride could augment the twitch tension, prolong the muscle action potential and, in addition, depress the peak voltages. Similar changes have been found with quinine by Harvey (1939). More recently, the action of adrenaline in augmenting twitch tension in the isolated rat phrenic nerve-diaphragm preparation has been shown to be associated with similar electrical changes (Brown, Bülbring and Burns, 1948). These authors thought that, while temporal dispersion of the contractions of the individual fibres might play a part in producing the augmented tension with adrenaline, its contribution was not large.

Increase in the twitch tension, such as that illustrated in Fig. 1 with thiopentone, is consistent with a slower propagation of the muscle action potential associated with a slower and more prolonged contraction. In this connexion it may be remembered that in a simple muscle twitch there is insufficient time, before relaxation has set in, for the full tension of the muscle to be developed (Hill, 1949).

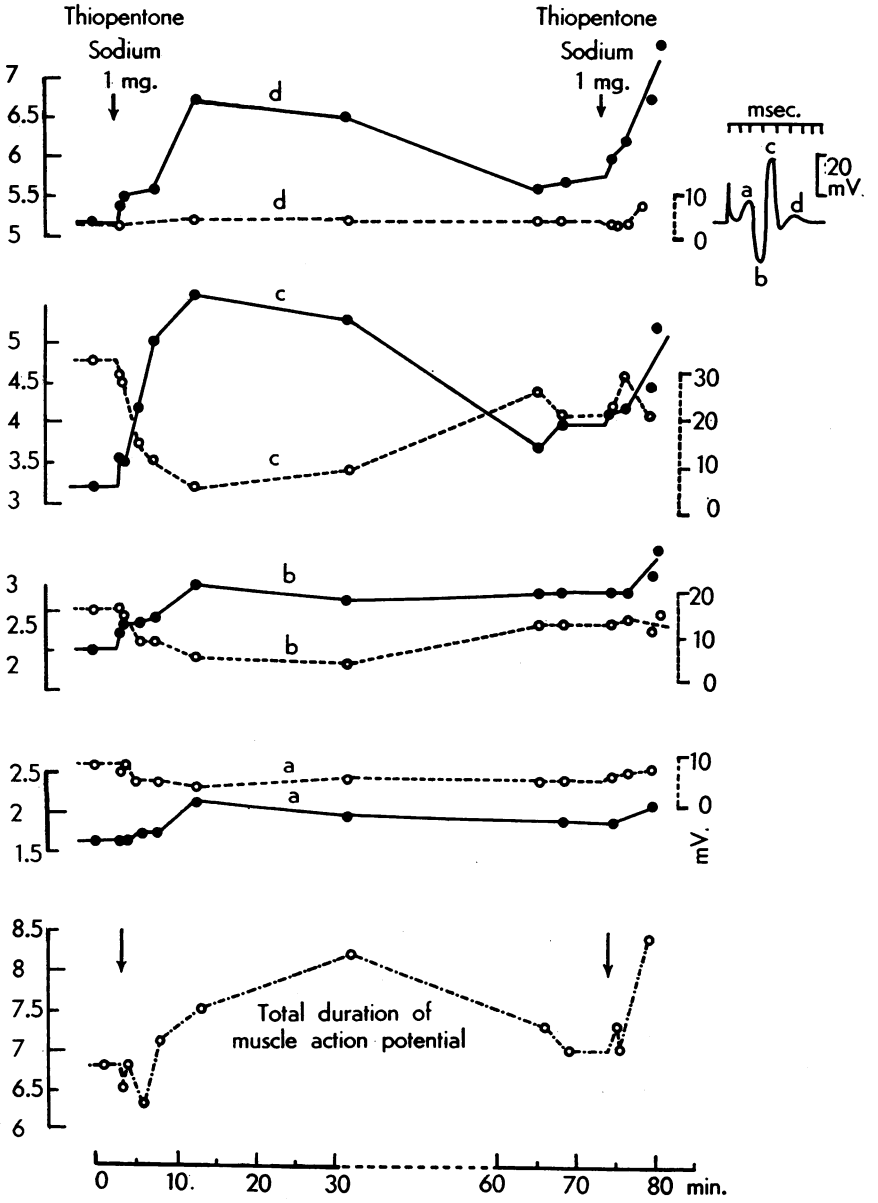

FIG. 6.—Graphical record of the experiment illustrated in Fig. 5. The open circles represent the voltage and the solid circles the latencies of the four peaks *a*, *b*, *c*, and *d* of the action potential record. Right-hand ordinates, amplitude in mV. Left-hand ordinates, latency in msec.

Little is known about the precise mode of action of barbiturates upon tissues. In two instances, however, respiratory enzyme systems are implicated —in the depression of respiration of rat brain slices *in vitro* (Quastel, 1943; Ghosh and Quastel, 1954), and in the depression of the oxygen consumption, associated with repetitive activity, in the rabbit's isolated superior cervical ganglion (Larrabee, Ramos, and Bülbring, 1950). But whether thiopentone sodium acts on skeletal muscle by affecting a barbiturate-sensitive stage of a respiratory enzyme remains to be determined.

SUMMARY

1. Thiopentone sodium, injected intra-arterially into the gastrocnemius-soleus muscle of a rat anaesthetized with urethane, augmented the twitch of the muscle when stimulated indirectly. This effect was similar to that seen in the rat obturator nerve anterior gracilis muscle preparation and in isolated frog or rat nerve-muscle preparations.

2. The intra-arterial injection of thiopentone sodium was associated with a decrease in voltage, an increase in latency and a prolongation of duration of the muscle action potential recorded from the surface of the anterior gracilis muscle of the anaesthetized rat.

3. The significance of these electrical changes in relation to the mechanical response of the muscle is discussed.

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