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DIFFERENCES IN THE ACTION OF TUBOCURARINE AND STRYCHNINE ON THE SPINAL REFLEX EXCITABILITY OF THE CAT

BY

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In the previous paper dealing with the effect of *d*-tubocurarine on the monosynaptic extensor reflex (Bernhard and Taverner, 1951) the literature on earlier investigations on the central action of curare was reviewed. Among authors who claim that curare has an excitatory central effect Tillie (1890), Santesson (1920–1), von Euler and Wahlund (1941), and Eccles (1946) characterize it as "strychnine-like," whereas McGuigan (1916) was of the opinion that the convulsions produced were not like those caused by strychnine. Cohnberg (1946) believed that curare, in contrast to strychnine, exerted its influence at central nervous levels higher than the spinal cord, and Salama and Wright (1950) came to the conclusion that tubocurarine stimulates the cells of origin of the facilitatory pathways in the brain-stem.

The action of *d*-tubocurarine on the mono- and poly-synaptic reflexes has been studied on anaesthetized cats by McCawley (1949), who found that after large doses both the monosynaptic and polysynaptic reflex responses decreased.

Recently Naess (1950) compared the effect of curare and strychnine on monosynaptic and polysynaptic reflexes and found no effect of curare on either of the two systems, whereas strychnine gave an increase of the polysynaptic response but had no effect on the monosynaptic reflex.

Naess, who worked on dial cats, concluded that "strychnine in smaller doses has no effect, and in greater doses only a depressing one, on the activity of the sensoric and motoric nerve cells which take part in the monosynaptic transmission." Similar results were obtained by Kaada (1950), who tested the effect of strychnine on decapitated cats, and concluded "that after strychnine the two-neurone arc discharge was only slightly increased or even decreased." In contradiction to Kaada and Naess, Bradley and Schlapp (1950) conclude that both "two-neurone and multineurone arc responses are increased by intravenous strychnine" on the basis of experiments on curarized decapitated cats.

Bernhard and Taverner (1951) investigated the effect of varying doses of *d*-tubocurarine on the monosynaptic extensor reflex in decerebrated, decapitated, low spinal, and deafferented preparations as well as on cats anaesthetized with dial.

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They found that *d*-tubocurarine injected intravenously gave an increase in the amplitude of the monosynaptic reflex in the different preparations mentioned. It was also shown that dial abolished the central effect of tubocurarine and that the influence of tubocurarine on the monosynaptic reflex was independent of its action on the blood pressure. Their investigations show the necessity of using different preparations when estimating the central effect of a drug by electrophysiological methods. This will be further emphasized by the results below. In this work the effect of tubocurarine on the polysynaptic reflex has been studied and investigations have been performed on the effect of strychnine on mono- and poly-synaptic reflexes.

METHODS

Decerebrated, decapitated, low spinal, and anaesthetized cats were used. The reflex responses were recorded from the L_7 and S_1 ventral roots, which were divided peripherally and laid on the recording electrodes. The sural and the gastrocnemius nerves were stimulated in order to obtain on the one hand a polysynaptic reflex discharge and on the other hand a monosynaptic reflex discharge in the ventral roots mentioned (Lloyd, 1943). The monosynaptic ventral root responses following stimulation of muscle afferents are impulses returning to the muscle, the afferents of which are stimulated (Lloyd, 1943). Since in the experiments the stimulus was applied to the gastrocnemius nerve a monosynaptic extensor reflex was tested. All exposed nervous structures were covered with warm paraffin oil. Rectangular electrical pulses of 0.1 msec. duration were used as stimuli and were applied to the nerves through electrodes. The ventral root responses were recorded with a differential amplifier and cathode ray oscillograph.

The experimental procedure was as follows. Alternate stimuli of frequency 40–60 per minute were delivered to the gastrocnemius nerve and to the sural nerve. The constancy of the amplitude of the reflex responses was checked for some minutes, and the drug was then injected intravenously and the effect on the amplitude of the test reflexes was recorded. In the diagrams the amplitude of the monosynaptic response is given in per cent of the average pre-injection amplitude. The magnitude of the polysynaptic response was measured planimetrically because of the desynchronization in the polysynaptic reflex discharge, and the planimetrical values are given in per cent of the average pre-injection value. The drugs used were *d*-tubocurarine chloride (Abbott, 3 mg.=20 units/c.c.) and strychnine nitrate (0.9 g./100 c.c.).

RESULTS

Effect of tubocurarine on the polysynaptic reflex

As mentioned above it has been shown that the intravenous injection of *d*-tubocurarine, even in small doses, produces an increase of the monosynaptic extensor reflex response in decerebrate as well as in high and low spinal preparations. Fig. 1 shows the effect of an intravenous injection of a small dose of *d*-tubocurarine (0.18 mg. per kg. body weight) on the monosynaptic extensor reflex response evoked from the gastrocnemius nerve and on the polysynaptic reflex response after stimulation of the sural nerve in a low spinal preparation (decerebrate cat with spinal transection at $T_{12} - L_1$ level). Records A and B show the monosynaptic and polysynaptic responses respectively before the injection of tubocurarine, and records C and D were obtained five minutes after the injection. The monosynaptic response increases, while there is no obvious change in the polysynaptic response. Fig. 2 illustrates the difference between the actions of tubocurarine on the monosynaptic and poly-

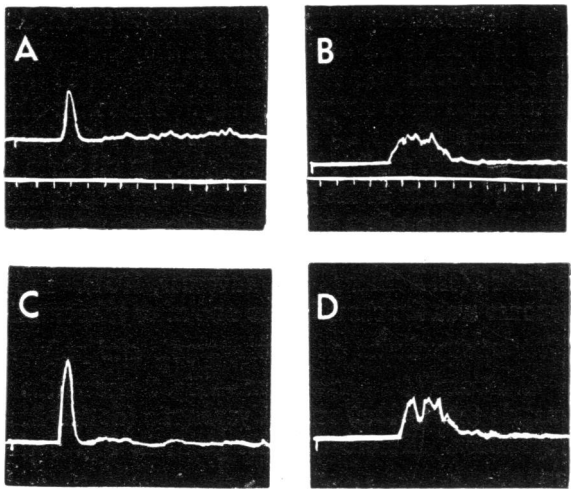

FIG. 1.—Monosynaptic extensor reflex (A and C) and polysynaptic reflex (B and D) in a low spinal preparation before (A and B) and 5 min. after (C and D) the injection of 0.18 mg. tubocurarine per kg. Time in msec.

synaptic reflex responses after repeated injections. The amplitude of the monosynaptic extensor reflex increases after the first injection to about 175 per cent of the pre-injection amplitude, and after the next injection there is a further increase to values higher than 200 per cent (heavy line). There was, however, no significant effect on the polysynaptic reflex (dotted line) even after the last injection. The areas of the polysynaptic responses varied around 100 per cent. When the amplitude of the polysynaptic responses was measured the same result was obtained.

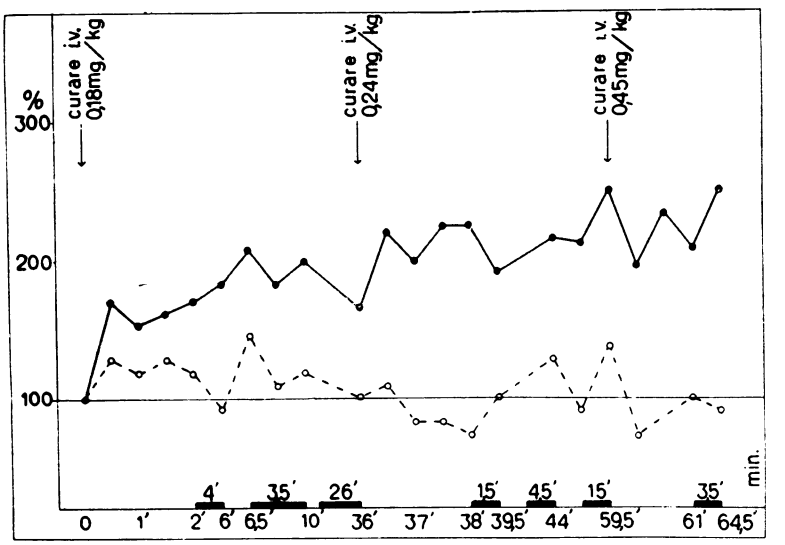

FIG. 2.—Graph showing the effect of tubocurarine on the size of the monosynaptic extensor reflex (heavy line) and the polysynaptic reflex (dotted line) in a low spinal preparation.

A series of experiments was performed on decerebrate and decapitate preparations as well as on deafferented low spinal preparations. In the latter preparation the spinal cord was transected at the level of $T_{12} - L_1$ and the $L_3 - S_3$ ventral and dorsal roots were cut peripherally. The reflex discharge in the L_7 ventral root after stimulation of the L_7 dorsal root was recorded. Varying doses were used from 0.1 to 1.8 mg. per kg. body weight. In all these different experiments tubocurarine gave an increase of the monosynaptic reflex, as described by Bernhard and Taverner (1951), but no effect whatsoever on the polysynaptic response. In anaesthetized preparations (dial) the effect of tubocurarine on the monosynaptic response was abolished (cf. Bernhard and Taverner, 1951). In these preparations the polysynaptic response was the same before and after tubocurarine.

Effect of strychnine on monosynaptic and polysynaptic reflexes

A. *Decapitate preparation.*—Fig. 3 shows the effect of strychnine on the monosynaptic (heavy line) and polysynaptic (dotted line) reflex responses in a decapitate preparation. Before the first point in Fig. 3 three doses of 0.01 mg. strychnine

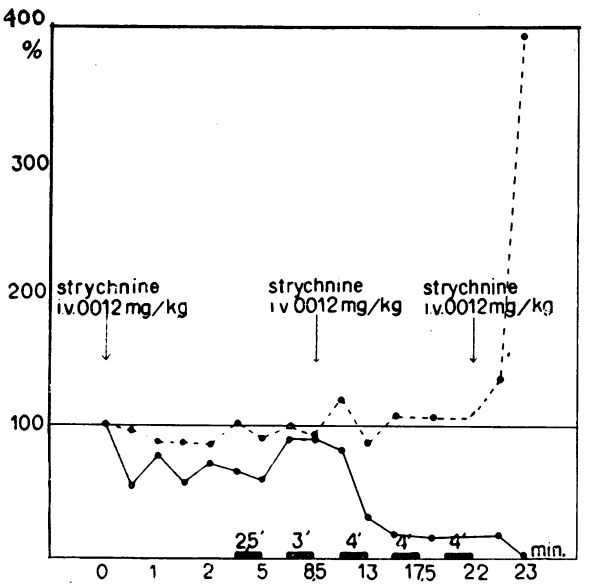

FIG. 3.—Effect of repeated small doses of strychnine on the monosynaptic extensor reflex (heavy line) and on the polysynaptic reflex (dotted line) in a decapitate preparation.

nitrate per kg. body weight were given without any effect on the reflexes. When another 0.012 mg./kg. was injected there was a significant fall in the monosynaptic reflex response, but no change in the polysynaptic reflex response. When the next injection of 0.012 mg./kg. was given the drop in the amplitude of the monosynaptic reflex response was obvious, but not until a further dose of the same size was given was there a sudden increase in the amplitude of the polysynaptic reflex response. After the last injection the experiment was interrupted because the animal developed serious convulsions. The effect obtained on spinal preparations is thus more or less the same as that previously described by Kaada (1950) and Naess (1950), although the decrease in the amplitude of the monosynaptic reflex response is more obvious in our experiments.

If, however, tubocurarine was previously injected the effect of strychnine on decapitate preparations was different. Such an experiment is illustrated graphically in Fig. 4, which shows that the polysynaptic reflex as well as the monosynaptic reflex increases after the injection of 0.08 mg. strychnine per kg. In this experiment 2.25 mg. tubocurarine per kg. was given beforehand. This effect is the same as that reported by Bradley and Schlapp (1950). Obviously the discrepancy between this and the earlier observations depends on the different types of preparation used.

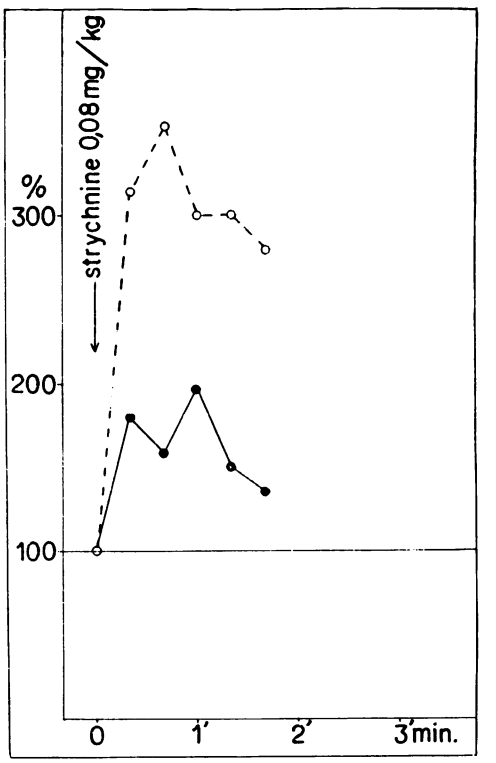

FIG. 4.—Effect of 0.08 mg. strychnine per kg. on the monosynaptic extensor reflex (heavy line) and the polysynaptic reflex (dotted line) in a decapitate cat which had previously received 2.25 mg. tubocurarine per kg.

B. *Low spinal preparation.*—If preparations were used in which the spinal cord was transected at the level of L_1, the effect of strychnine was also different from that obtained on decapitate uncurarized cats. Fig. 5 is taken from such an experiment. In order to avoid convulsions tubocurarine was given previously in a dose of 1.1 mg. per kg. body weight. After the injection of 0.07 mg. strychnine nitrate per kg., both the monosynaptic and polysynaptic reflex responses increased in parallel up to about 200 per cent. A second injection of 0.07 mg. per kg. caused a further rise of both the monosynaptic and polysynaptic reflex responses to 300–400 per cent. One and a half hours after the last injection both reflexes were still about 300 per cent of the pre-injection level. In all experiments on low spinal preparations the same results were obtained, i.e., a concomitant rise of both reflexes. The threshold dose for a significant effect was found to be about 0.06 mg. strychnine

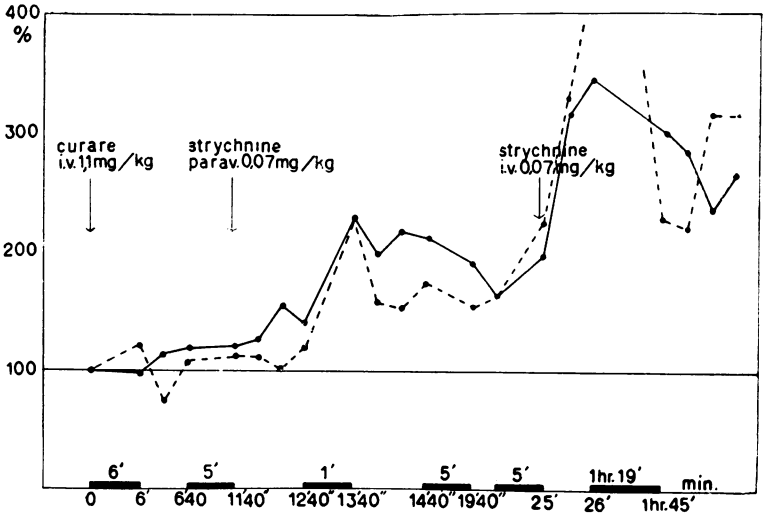

FIG. 5.—Effect of repeated injection of strychnine (0.07 mg./kg.) on the monosynaptic extensor reflex (heavy line) and on the polysynaptic reflex (dotted line) in a low spinal preparation which had previously received 1.1 mg. tubocurarine per kg.

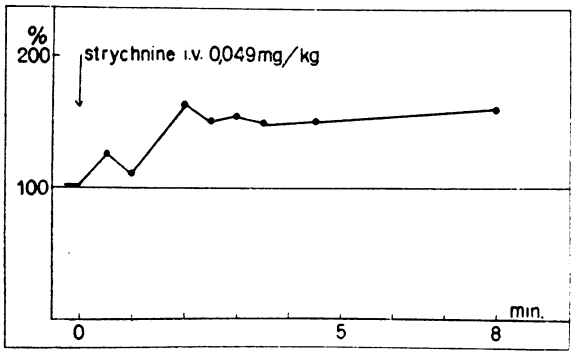

FIG. 6.—Effect of a small dose of strychnine (0.049 mg./kg.) on the monosynaptic extensor reflex in an uncurarized low spinal preparation.

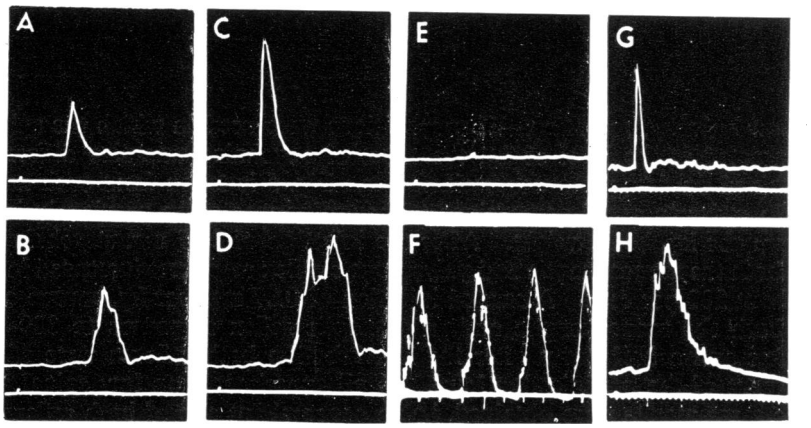

FIG. 7.—Monosynaptic extensor reflex (A, C, E, G) and polysynaptic reflex (B, D, F, H) in a curarized low spinal preparation before (A and B) and after the injection of 0.14 mg. strychnine per kg. In record F the amplification is reduced 50 per cent. Time in msecs. (in record F in 10 msec.). See text for details.

nitrate per kg. The same augmentation of the monosynaptic reflex was obtained in experiments in which tubocurarine was not injected previously (see Fig. 6).

In experiments with doses larger than double the threshold dose synchronized spike outbursts and slow potential waves of varying frequencies occurred (" onde tetaniques provoquées and onde tetaniques spontanées " ; Bremer, 1941). In that stage the polysynaptic and monosynaptic reflex responses showed large variations. The records in Fig. 7 from a low spinal preparation illustrate these different effects. Records A and B show the mono- and poly-synaptic reflex responses before the injection. C and D were obtained after the injection of 0.14 mg. strychnine nitrate per kg. and show the increase of the mono- (C) and poly-synaptic (D) reflex responses. Records E and F are from the stage in which the transient spontaneous outbursts occur (steady base line in E, large potential waves in F). As mentioned the reflexes may be abolished in this stage (record E). Records G and H, obtained about one hour after the stage illustrated in E and F, show that the polysynaptic as well as the monosynaptic reflex response is still increased.

Fig. 8 shows the same effect of strychnine on a low spinal and deafferented preparation. The monosynaptic as well as the polysynaptic part of the reflex response from the L_7 ventral root after stimulation of the L_7 dorsal root is increased after the injection of 0.04 mg. strychnine nitrate per kg. (Fig. 8 B).

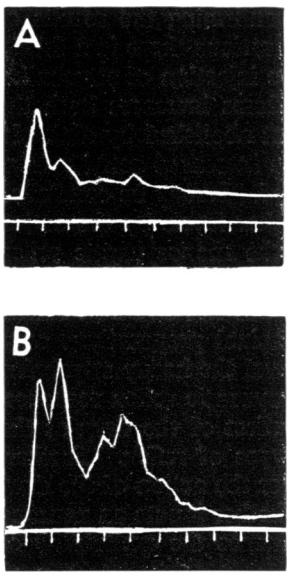

FIG. 8.—Reflex response in L_7 ventral root after stimulation of L_7 dorsal root (A) before and (B) after the injection of 0.04 mg. strychnine per kg. in a low spinal deafferented preparation. Time in msecs.

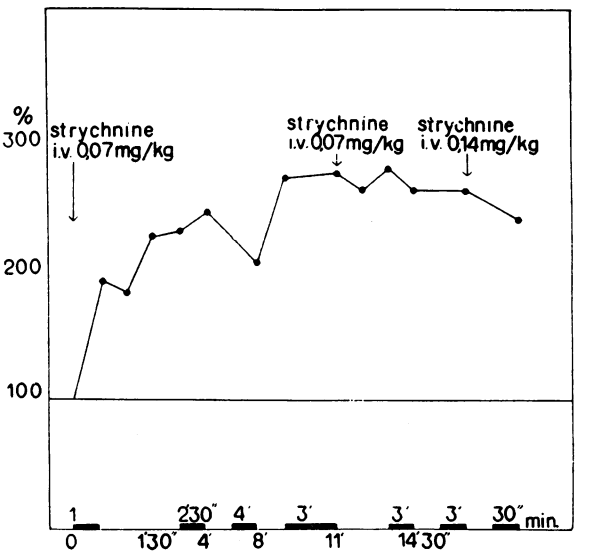

FIG. 9.—Effect of strychnine on the monosynaptic extensor reflex in a low spinal preparation which had previously received 0.45 c.c. dial per kg.

C. *Anaesthetized preparations.*—Fig. 9 shows the effect of strychnine on the monosynaptic extensor reflex response in a low spinal preparation after the previous administration of 0.45 c.c. dial per kg. body weight. There is a rise of the monosynaptic reflex response to more than 200 per cent, which is comparable to the effect in a similar preparation without dial.

Discussion

In an earlier paper (Bernhard and Taverner, 1951) it was shown that intravenously injected tubocurarine produces an increase in the amplitude of the monosynaptic extensor discharge which can be compared with the effect on the knee-jerk of the local application of tubocurarine to the spinal cord, as shown by Salama and Wright (1950). It was also demonstrated that tubocurarine, when injected intravenously, exerts its action at spinal levels as well as on supraspinal structures, and that the steady increase of the reflex is not due to changes in the blood pressure. It was further shown that dial abolishes the effect of tubocurarine.

The experiments described above show that tubocurarine does not influence the polysynaptic discharge in decerebrate, spinal, deafferented, or anaesthetized preparations. Thus, according to our experiments, *d*-tubocurarine acts differently on the monosynaptic extensor and the polysynaptic flexor reflexes.

The action of strychnine on these two reflex systems has also been tested in different preparations. In all types of preparation strychnine gives an increase in the amplitude of the polysynaptic reflex response, thus confirming the results of Kaada and of Naess. In decapitate and decerebrate preparations there is a fall in the amplitude of the monosynaptic reflex response after the injection of strychnine, but in low spinal preparations strychnine even in small doses increases both the monosynaptic and polysynaptic reflexes. This action is not abolished by dial. In decerebrate or high spinal preparations the influence of the increased activity from higher levels brought about by the drug has to be considered. It may be concluded that the augmentation of this continuous activity from higher levels by the action of strychnine is the essential factor in the inhibition of the monosynaptic reflex response in the decapitate preparations (Fig. 3). The findings explain why the enhancing effect of strychnine on the monosynaptic reflex was not noted in earlier investigations (Kaada, 1949 ; Naess, 1950) in which low spinal preparations were never used. When the cord is transected at the lumbar level the heavy strychnine activity from the upper levels no longer inhibits the monosynaptic reflex response. The change in the reflex discharge is now a sign of the effect of the drug at the segmental level, and obviously the drug augments both types of reflex. Further, it must be pointed out that it still cannot be decided whether the action of the drug is restricted to the neurones included in the reflex arcs or whether it acts on neurone structures in parallel with the afferent neurones which influence the excitability of the motorneurones.

Bernhard and Taverner (1951) pointed out that the augmentation of the monosynaptic extensor reflex after the injection of tubocurarine may depend on the drug blocking structures which normally exert an inhibitory effect on the monosynaptic extensor reflex. It is of interest (i) that strychnine gives an augmentation of the monosynaptic reflex in low spinal preparations in which structures have been

removed which have an inhibitory influence on the reflex, and (ii) that there is also an augmentation of the reflex after strychnine in curarized decapitate preparations (Fig. 4; cf. Bradley and Schlapp, 1950). These observations favour the view that tubocurarine has a blocking action on structures which inhibit the monosynaptic extensor reflex. The augmentation of the monosynaptic extensor reflex after tubocurarine should consequently be regarded as a release.

Summary

1. The effects of *d*-tubocurarine and strychnine on the monosynaptic extensor reflex and the polysynaptic reflex evoked from a cutaneous nerve have been studied in decapitate, low spinal, deafferented, and anaesthetized cats.

2. *d*-Tubocurarine, which is known to produce an increase in the amplitude of the monosynaptic extensor reflex response in unanaesthetized preparations, did not influence the polysynaptic reflex response in any of the preparations tested.

3. Strychnine was found to produce an increase in the amplitude of the polysynaptic reflex response in all types of preparation.

4. It was found that in low spinal preparations strychnine produced an increase in the amplitude of the monosynaptic reflex response parallel to its effect on the polysynaptic reflex, whether or not dial was given previously.

5. When the segment giving rise to the reflex under test is in continuity with the upper part of the spinal cord strychnine produces a decrease in the amplitude of the monosynaptic reflex response; this decrease is interpreted as being due to inhibition of the reflex by an increased continuous activity from upper segmental levels brought about by the strychnine.

6. In the same sort of preparation strychnine gives an augmentation of the monosynaptic extensor reflex if tubocurarine is given beforehand. This fact favours the view that tubocurarine blocks structures which exert an inhibitory action on the monosynaptic extensor reflex.

7. At the segmental level of the reflexes tested tubocurarine and strychnine act differently, i.e., tubocurarine produces an increase in the amplitude of the monosynaptic reflex response but has no effect on the polysynaptic reflex, whereas strychnine increases the amplitude of both reflexes in parallel.

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