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THE ACTION OF NICOTINE ON THE MOTOR ENDPLATE IN THE CAT

BY

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Although nicotine gives its name to one of the principal actions of acetylcholine—the "nicotinic" action on skeletal muscle—the effects of nicotine on mammalian muscle (as distinct from amphibian or avian muscle) have been studied relatively rarely. The most recent analysis was that by Bacq & Brown (1937) who showed that arterial injections of nicotine into the gastrocnemius of the cat produced a contraction of the muscle followed by a prolonged depression of the response to nerve stimulation. They also noted that repetitive stimulation of the nerve relieved the depression of the twitch. There is no information, however, which allows a confident comparison between the effects produced by nicotine and those produced by, for instance, decamethonium or *d*-tubocurarine. The experiments described here analyse, on cat muscle, some of the characteristics of the action of nicotine in the light of recent knowledge.

METHODS

Cats anaesthetized with chloralose after induction by ethyl chloride and ether were used in all experiments. A tracheal cannula was inserted, and a venous cannula in either the femoral or jugular vein. For injecting into the iliac artery of one leg, the opposite iliac artery and the descending pelvic artery were ligated and a Gordh needle was inserted into the ligated iliac artery pointing centrally so that injections were carried into the open artery on the other side. Some minutes after each injection, an injection of saline 0.5 ml. was made to wash out the needle and arterial deadspace. The contractions of tibialis, soleus or gastrocnemius were recorded using a flat spring myograph writing with a light lever on a smoked drum. The sciatic nerve in the thigh was tied close to the hip; shielded platinum stimulating electrodes were placed on it during its course through the hamstrings. For stimuli, square wave maximal shocks of 0.5 msec duration were used. For close arterial injections into the tibialis muscle, the technique of Brown (1938) was followed.

Depolarization of the muscle was recorded in one of two ways. In the first, an electrode was placed on a small swab of cotton wool moistened with saline, and placed on the belly of the muscle through a small incision in the skin; by this means a diffuse lead from the surface of the muscle was obtained, insensitive to the precise position of the electrode. The other electrode was inserted under the skin of a toe. Silver-silver chloride electrodes in 1% agar saline carried in a glass tube, tapering to a point through which a saline wick protruded, picked up the potentials; these were recorded after amplification by an adapted Pye pH meter and on an ink recorder. Instrumental fluctuations ± 0.5 mV occur. Second, a determination of the spatial distribution of depolarization produced by nicotine was determined on the gracilis muscle, in a manner similar in principle to that used by Burns & Paton (1951). One electrode was arranged so that it could be carried on a rack and pinion device along a length of the gracilis muscle, the movement of the electrode being transmitted directly by a Bowden

cable system to the paper drive of the ink recorder. The potential developed between this electrode and the reference electrode was applied to the galvanometer of the ink recorder, so that the spatial distribution could be obtained with a single sweep of the electrode along the gracilis.

RESULTS

Experiments with distant arterial injection into tibialis, soleus and gastrocnemius muscles

The injection of 50 μg of nicotine into the iliac artery usually has little effect itself on these muscles, or on their response to motor nerve stimulation, although it may potentiate the twitch slightly, and increase the effect of succinylcholine (50 μg) given afterwards. To produce appreciable neuromuscular block, doses of nicotine between 250 μg and 2 mg need to be given (Figs. 1 and 2). The block seems to affect tibialis preferentially and gastrocnemius and soleus are relatively spared. This differential effect on the muscles is greater than is seen with succinylcholine. The block is quick in onset, and tends to be very prolonged ; it may be an hour before full recovery takes place.

If, during a partial neuromuscular block of the tibialis with nicotine, a tetanus at the rate of 41/sec is interposed, the tetanus is well sustained, and reaches a maintained tension higher than that achieved by single twitches. The tension of single twitches after the tetanus was for a short period slightly increased.

Neostigmine, in doses of 20–50 μg intra-arterially had, for the most part, little obvious effect on block caused by nicotine, whether given before nicotine or during its action. Sometimes a small transitory deepening of the block was produced which seemed to be followed by an acceleration of recovery (Figs. 5 and 7). That the doses of neostigmine used were large enough to produce their characteristic action is shown by their effect on unblocked muscle (for example, the gastrocnemius in Fig. 7c).

When the interaction with other blocking agents was studied, it was found that succinylcholine was not antagonized by a previous dose of nicotine. Further, in the experiment of Fig. 1 a comparison was made between the effects of nicotine 250 μg, of succinylcholine 50 μg, and of a mixture made up of half the dose of each of these (nicotine 125 μg and succinylcholine 25 μg) given in a single injection. Such a test allows an estimation of whether one drug antagonizes, sums with, or potentiates the other, according as the response to the mixture is smaller than, equal to, or greater than the control responses to each drug alone, the doses of the latter being equi-active. The test is not very satisfactory with substances acting for different times ; but it showed that the mixture had an effect on tibialis and gastrocnemius not far from the mean between the control responses, indicating a simple summation, and possibly a mutual potentiation, between succinylcholine and nicotine in their neuromuscular effects.

Such a result suggests that administration of a competitive blocking agent such as gallamine or *d*-tubocurarine in appropriate dose would antagonize the action of nicotine, as they do that of decamethonium and succinylcholine. This was found to be the case ; Fig. 2 shows the way in which gallamine in small doses accelerates the recovery of tibialis from nicotine block. Similar results were obtained with *d*-tubocurarine. Figure 2 also shows how, on the soleus muscle, the first effect of gallamine was to deepen the block slightly.

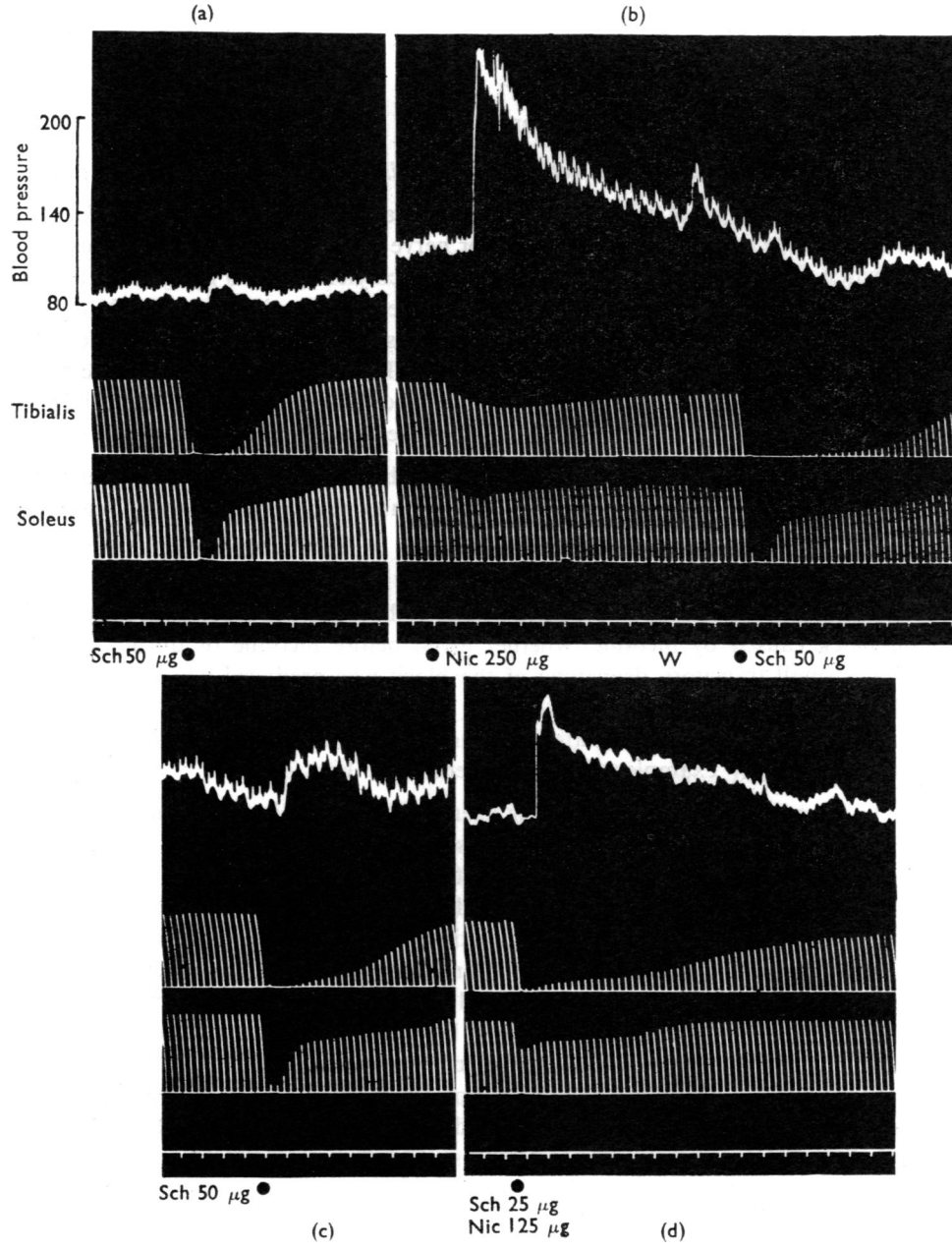


Fig. 1. Cat, 3.7 kg, chloralose. Records of blood pressure and of responses of tibialis and soleus to single maximal shocks to sciatic nerve, 3/min. Injections into iliac artery. Time in minutes. (a) and (c), Succinylcholine (Sch) 50 μg. (b), Nicotine (Nic) 250 μg followed by succinylcholine 50 μg. (d), Succinylcholine 25 μg with nicotine 125 μg. The blood pressure tracings in this and Fig. 2 are 1.5 min to the right of the muscle records.

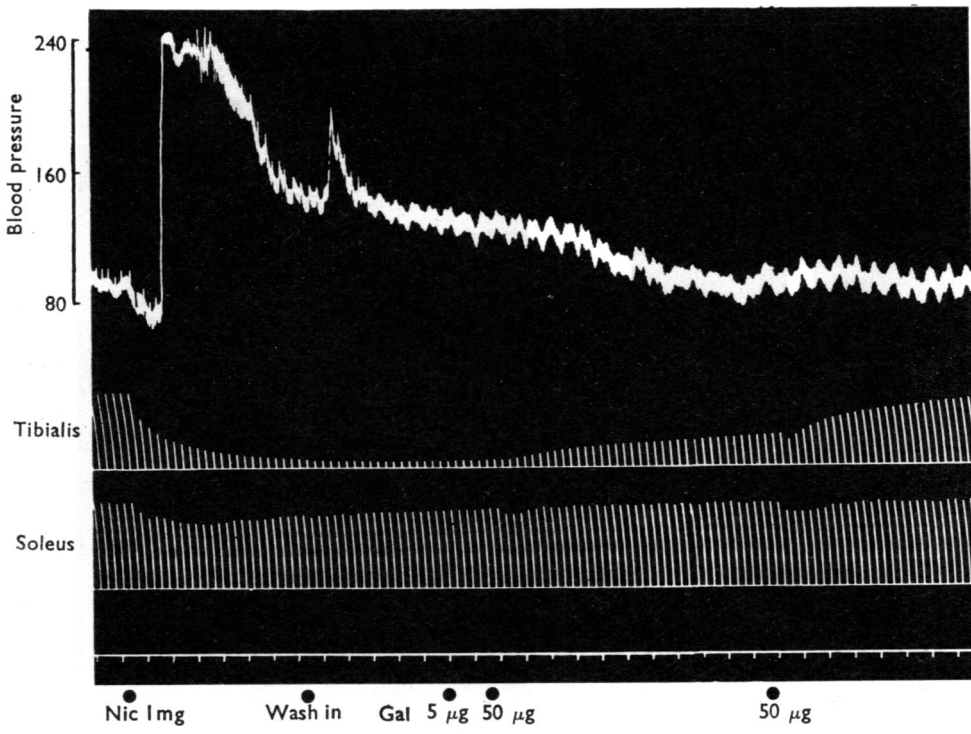
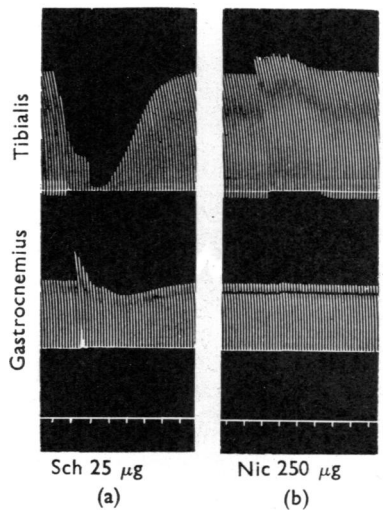


Fig. 2. Same experiment as Fig. 1. Nicotine (Nic) 1 mg produces almost complete block in tibialis, with moderate effect on soleus. Gallamine (Gal) 5 μg is ineffective but two successive doses of 50 μg accelerate recovery of tibialis and produce a transient depression of soleus.

In a few experiments the response of tibialis to nicotine was compared with that of gastrocnemius and Fig. 3 illustrates such a test. The gastrocnemius seems, if anything, to be even less sensitive than soleus; in the experiment of Fig. 3 a dose of nicotine sufficient to produce a prolonged block of tibialis did no more than evoke fasciculations in the gastrocnemius, although the response of the gastrocnemius to succinylcholine 25 μg included a phase of partial block.

There was no obvious sign in experiments of this kind that nicotine changed its pattern of action with repeated administration, and the selective action on tibialis and the initial stimulant action re-appeared with each dose. In one experiment in which four doses of intra-arterial nicotine totalling 2.75 mg had been given, a final dose of 4 mg intra-arterially produced total neuromuscular block lasting for 32 min; 11 min later, a dose of *d*-tubocurarine (50 μg) arterially was still able to accelerate recovery.

The question also arises, since nicotine is not quaternary and could readily penetrate nervous tissue, whether its neuromuscular blocking action is comparable with that produced by hemicholinium or triethylcholine, and associated with an interference with acetylcholine output by the motor nerve endings. A useful test for such an action is the increase in block of transmission as the frequency of indirect stimulation is raised (Reitzel & Long, 1959; Bowman & Rand, 1961). In Fig. 4, the action of nicotine 2.5 mg



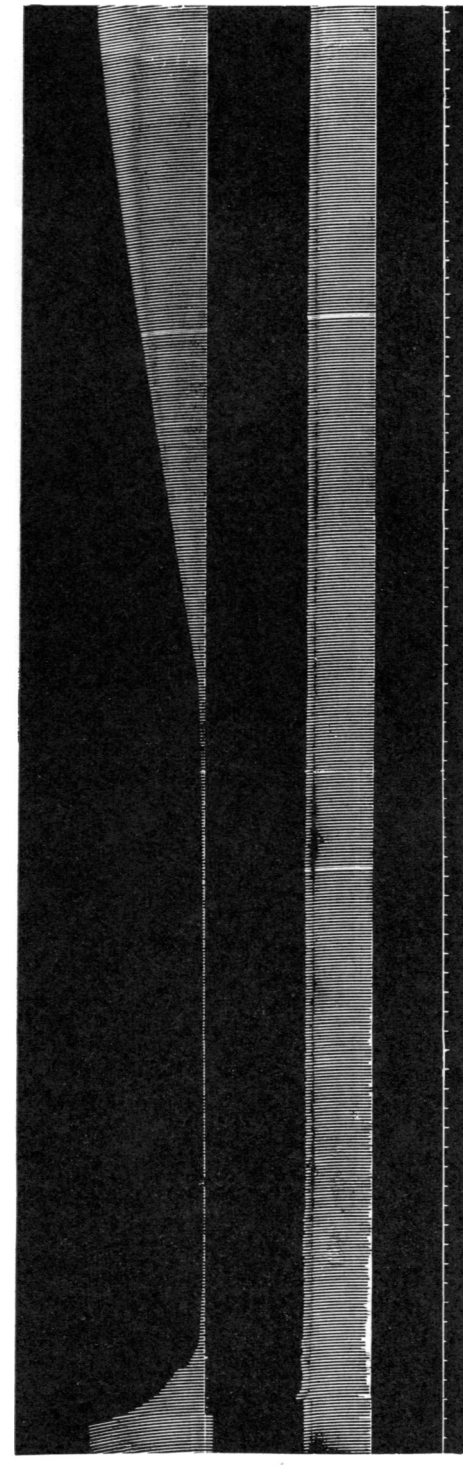


Fig. 3. Cat, chloralose. Tibialis and gastrocnemius responses to maximal shocks to sciatic nerve 6/min. Iliac arterial injection. (a), Succinylcholine (Sch) 25 μg. (b), Nicotine (Nic) 250 μg. (c), Nicotine 1 mg.

on tibialis was tested, stimulating the nerve only occasionally to sample the state of transmission. The intensity and time course of the block which developed did not differ significantly from the normal pattern recorded with excitation at a rate of 6/min ; and partial block was already present when the first test shock was given after the nicotine injection.

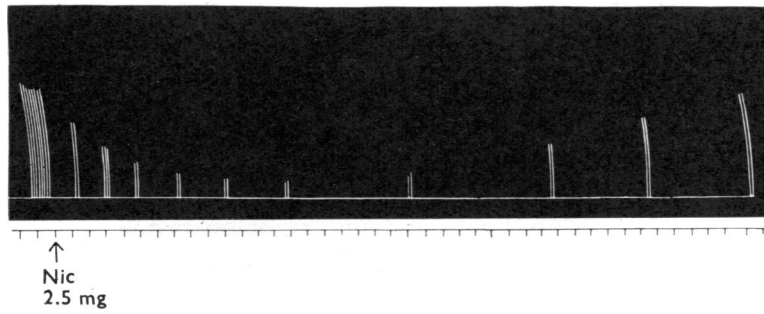


Fig. 4. Cat, chloralose. Record of tibialis twitch to occasional shocks to sciatic nerve. After injection of nicotine (Nic) 2.5 mg, block is already present at first test shock and develops in normal way.

Pressor response to intra-arterial nicotine

During these experiments, the blood pressure was recorded as a check on the systemic actions of the injected nicotine. Substantial rises in blood pressure occurred ; but instead of appearing at a time compatible with the passage of the drug through the muscle vessels to the venous system, heart and lungs, and thence to the general systemic circulation, the pressor response began within 1 or 2 sec. Because only small volumes were injected, it did not seem possible that any arterial reflux of nicotine could have occurred, such as might reach the aortic chemoreceptors. The response was graded with dose ; a rise of 60 mm mercury was produced by nicotine 50 μg, and 1 mg caused a rise of about 180 mm mercury, a response which is indeed much larger than it would usually produce if given intravenously. Repeated responses of this kind could be obtained in one animal. After the largest responses, a period of hypotension sometimes followed. From the magnitude of the response, it seems unlikely that it could result from a vasoconstrictor action of nicotine on the blood vessels of the leg, if only because complete ligation of leg vessels produces a far smaller pressor effect. If, then, it is of reflex origin, from excitation of chemoreceptive nerve endings in the limb, the afferent nerves concerned are unlikely to be carried in the sciatic nerve, because this was always ligated at the sciatic notch firmly enough to prevent reflex movement in response to electrical stimulation below the point of ligature. Succinylcholine did not share this prompt pressor action ; the response is therefore unlikely to originate in muscle spindles, the afferents of which are excited by both drugs (Albuquerque & Smith, 1965). The vigour of the response suggests the excitation of pain fibres in the walls of the arteries themslves, which are exposed to the highest concentration of nicotine ; but further analysis of the phenomenon was deferred.

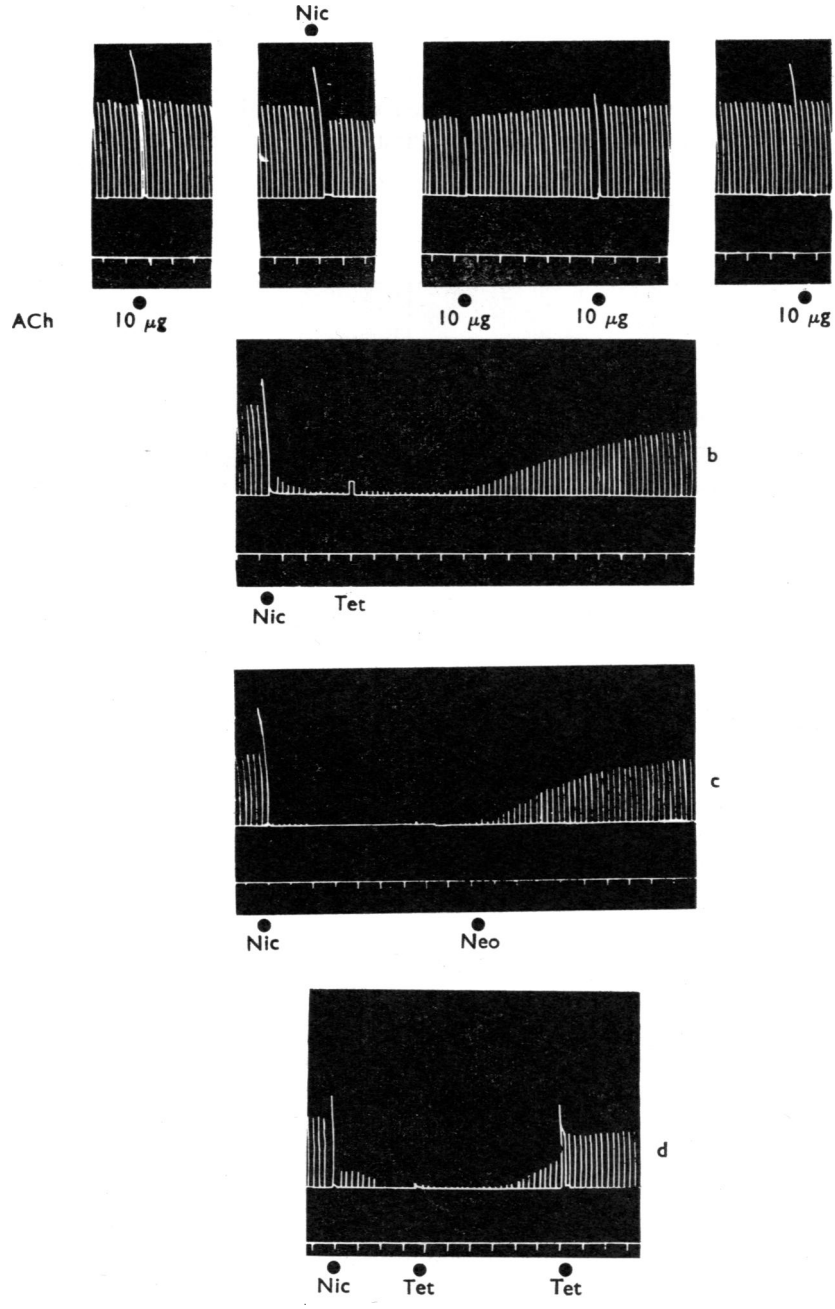
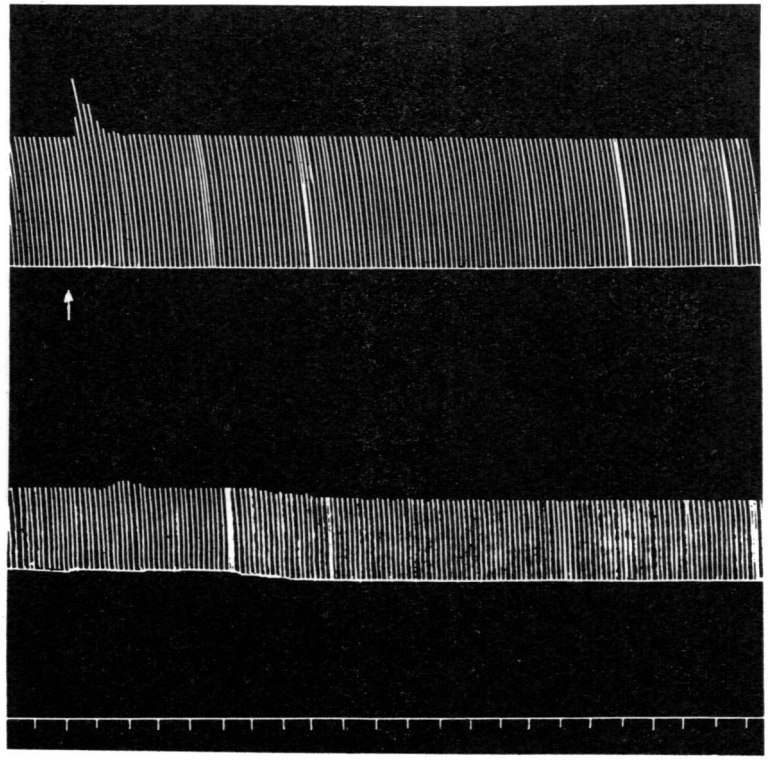


Fig. 5. Cat, 2.5 kg, chloralose. Tibialis prepared for close arterial injection. Shocks to sciatic 4/min. (a) Responses to acetylcholine (Ach) 10 μg before and after nicotine (Nic) 0.2 mg. (b) Response to nicotine 1 mg; during the block, the nerve was excited at 41/sec for 15 sec; the tetanus is sustained, and there is little post-tetanic relief of the block. (c) Response to nicotine 1 mg; as the block began to pass off, neostigmine (Neo) 20 μg was injected intra-arterially. (d) Response to nicotine 1 mg; nerve stimulation at 41/sec for 15 sec applied when block complete and during recovery.

Response of tibialis to close arterial injections of nicotine

Nicotine injected close-arterially can produce a vigorous twitch, greater than that from a single shock to a nerve ; a dose of 200 μg produces an effect roughly comparable with that caused by 10 μg or less of acetylcholine. After the twitch neuromuscular block may develop if the dose has been large enough, although it does not necessarily do so. If neuromuscular block is produced, the response to acetylcholine is depressed, proportionately more than the nerve-excited twitch. It was also possible to show under these conditions that tetanic stimulation gave rise to a maintained contraction, and was followed by only slight post-tetanic unblocking ; that neostigmine 2 μg had no effect on recovery ; and that neostigmine 20 μg even deepened the block briefly before recovery was resumed (Fig. 5).

Figure 6 (a)



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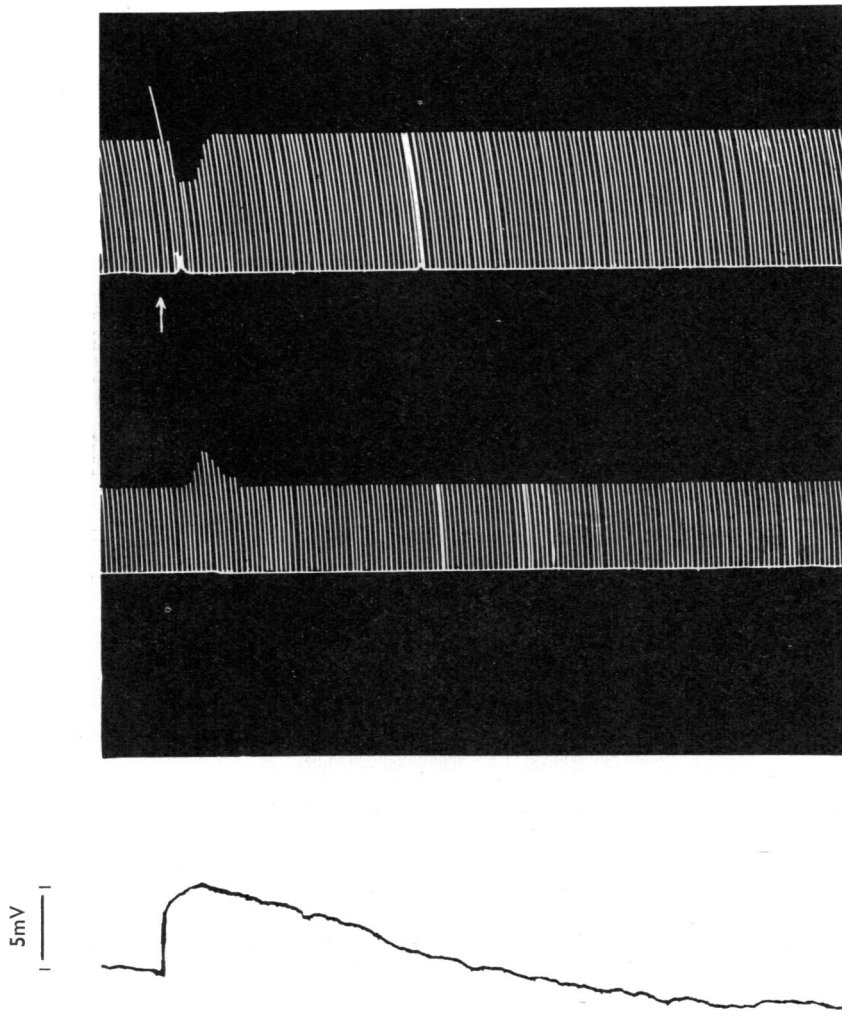

Figure 6 (c)

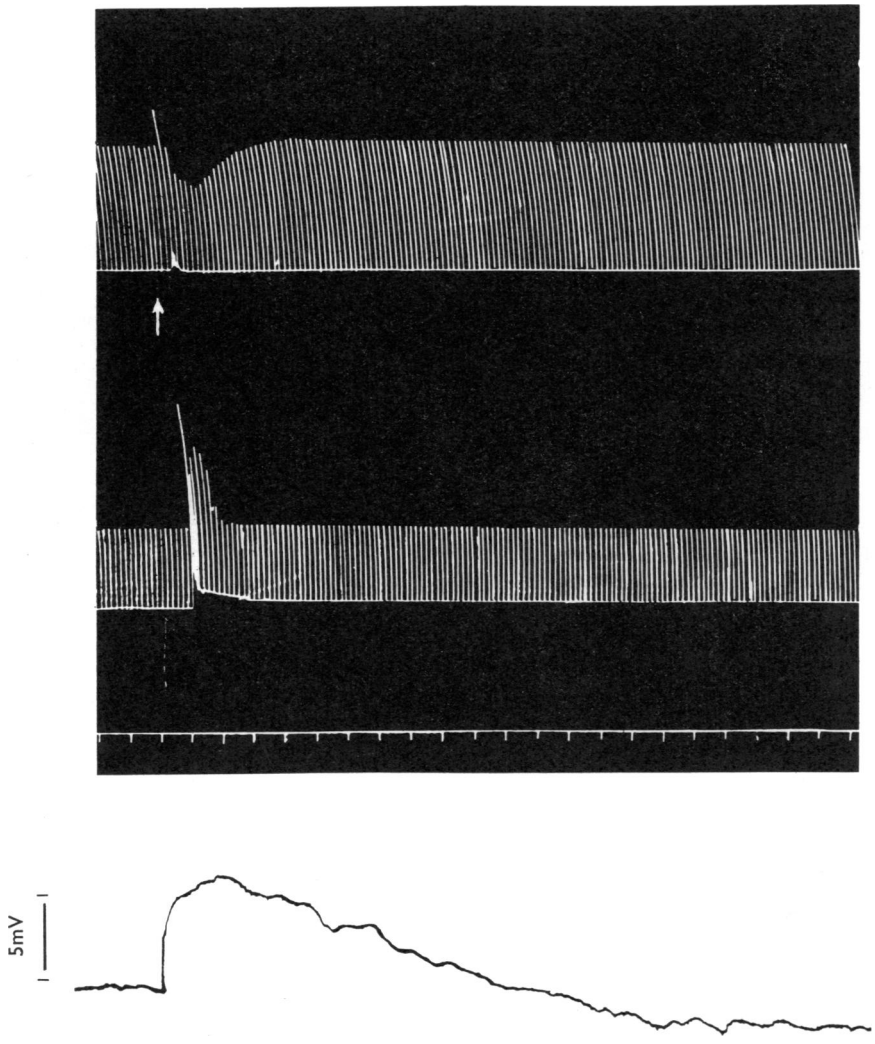

Fig. 6. Cat, 2.9 kg, chloralose. Records of tibialis (above) and gastrocnemius (below) twitch to shocks to sciatic nerve 6/min, and of potential between belly of tibialis and reference electrode on a toe. Iliac injection of succinylcholine: (a) 5 μg; (b) 10 μg; (c) 25 μg. Time in minutes.

Depolarization of tibialis gastrocnemius and gracilis muscles by nicotine

The use of a diffuse electrode on the belly of tibialis is a convenient method of recording the intensity and time course of an endplate depolarization, when it is wished to obtain simultaneously a record of the contractions of the whole muscle. Doses of drugs were spaced so that depolarization had passed off completely before a further dose was given. With succinylcholine, intra-iliac doses from 5 μg upwards will produce a brief neuro-

muscular potentiation or block but a relatively much more prolonged depolarization (Fig. 6). With nicotine a strong depolarization was observed, of slower onset than with succinylcholine and far longer lasting (Fig. 7).

Figure 7 (a)

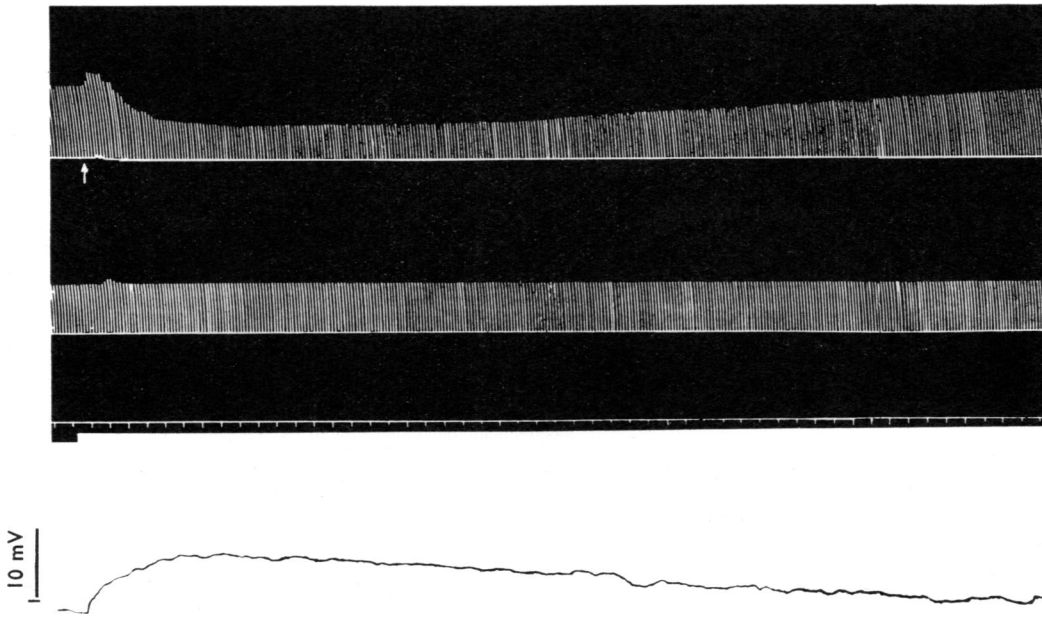

Figure 7 (b)

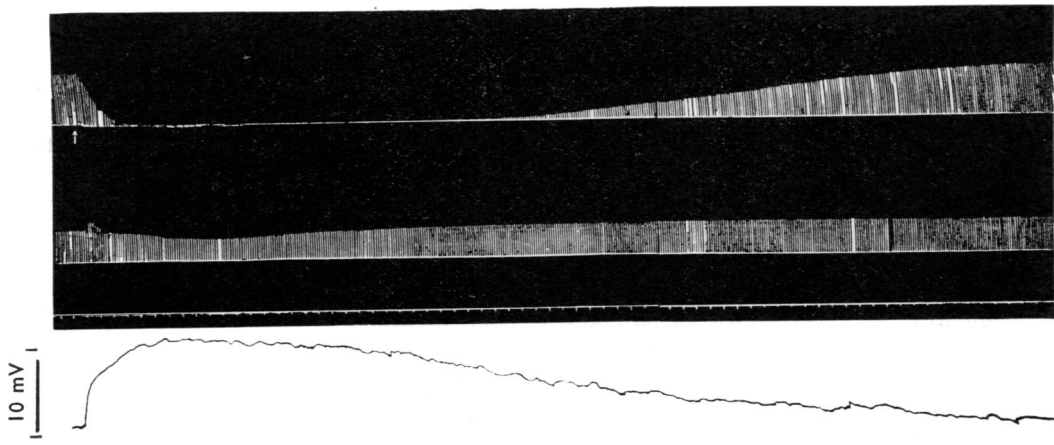

Figure 7 (c)

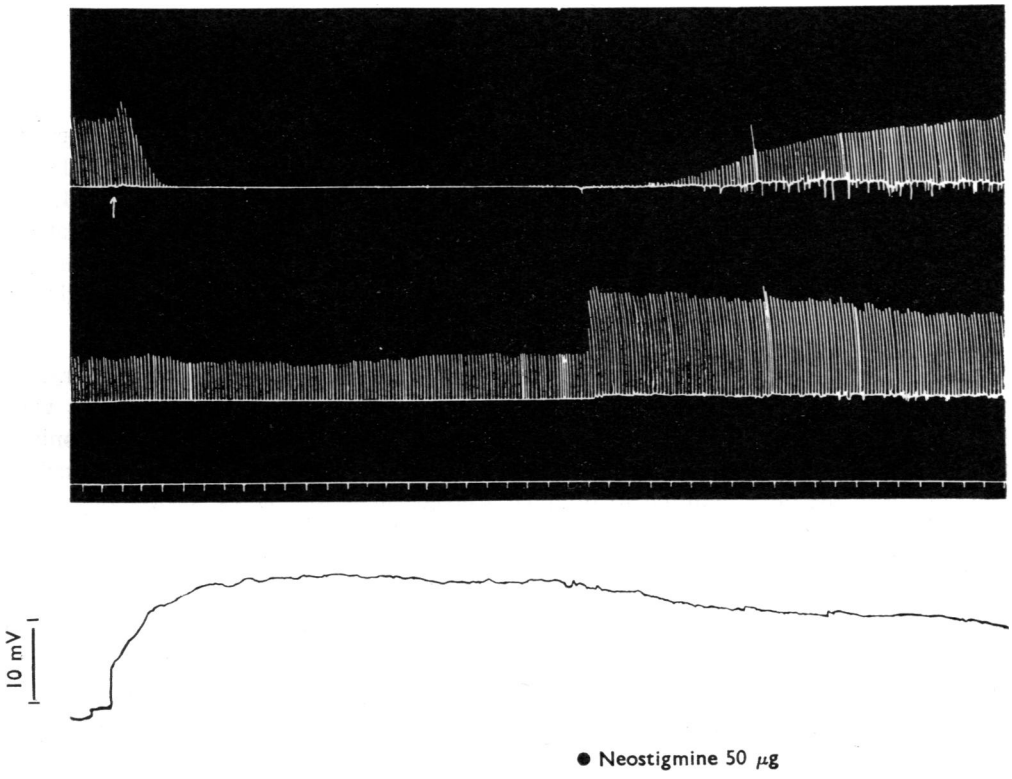

Fig. 7. As Fig. 6. Iliac injections of nicotine: (a) 500 μg; (b) 1 mg; (c) 1 mg given for the fourth time, followed by neostigmine 50 μg.

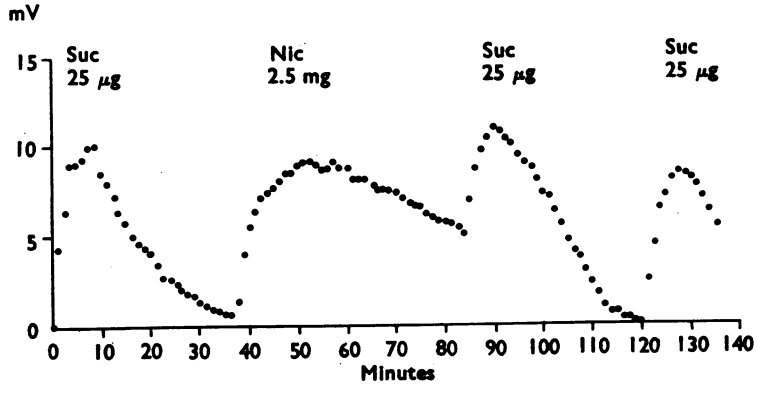

Fig. 8. Graph of changes of potential recorded between electrode on the belly of gastrocnemius and reference electrode under skin of ankle, in cat under chloralose. Injections into iliac artery of succinylcholine (Suc) 25 μg and nicotine (Nic) 2.5 mg.

In a single experiment recording the potential between a diffuse electrode on the belly of gastrocnemius and a reference electrode on the ankle, an intra-iliac injection of nicotine 2.5 mg produced a maximal depolarization of 8 mV; succinylcholine 25 μg produced a similar effect, but nicotine had (as on tibialis) a much slower and more prolonged action.

At no time was nicotine found to produce block without at the same time giving rise to a substantial depolarization. If repeated doses of nicotine were administered, the depolarization produced remained the same within ordinary limits of variation. If the action of succinylcholine was compared before and after a nicotine depolarization, the effect of succinylcholine was never diminished and often increased. If succinylcholine was administered during the decline of a nicotine depolarization, it summed with it (Fig. 8).

In experiments analysing the spatial distribution of the depolarization in the gracilis muscle, nicotine produced a pattern of depolarization identical to that obtained with succinylcholine (Fig. 9), and to that obtained earlier with acetylcholine, decamethonium and anticholinesterases (Burns & Paton, 1951; Douglas & Paton, 1954). The differences were simply in the dose required to produce a given effect, and in the slower onset and disappearance of the depolarization produced by nicotine. As with the other drugs, the depolarization of the endplate region spreads a little with lapse of time. In one experiment (Fig. 10) a dose of tubocurarine was administered after the effect of nicotine had become established; this led to a more rapid repolarization of the endplates and then to a brief period of relative hyperpolarization such as has also been found with decamethonium and with the depolarization produced by anticholinesterases.

Analysis of the relationship between depolarization and block

All the observations described point to the general conclusion that the primary action of nicotine is to produce a depolarization block. But there are reasons for believing that an element of non-depolarizing block also intervenes. It has already been noted that a modest post-tetanic unblocking action can be detected, and that, particularly on soleus muscle, the first effect of gallamine or tubocurarine given during a block produced by nicotine was to deepen the block slightly. Further evidence emerged when the time courses of depolarization and block were analysed (see Fig. 11). With succinylcholine, depolarization considerably outlasts the block it produces; but with nicotine, whether in small (250 μg) or large (1 mg) dose, the block slightly outlasts the depolarization. Particularly interesting, however, was the fact that if neostigmine (50 μg) was injected at the peak of the nicotine block, the time course of the depolarization was hardly affected, but the recovery from neuromuscular block was evidently considerably accelerated. This phenomenon can be displayed still more strikingly if block is related directly to depolarization. With succinylcholine, block is greater during onset of depolarization than during offset, so that the path of the developing and dwindling paralysis traces a clockwise loop with these co-ordinates. After nicotine, however, the loop is reversed, and block is considerably greater for a given depolarization during offset. The effect of neostigmine now shows itself as abolishing the loop, so that the relationship of block to depolarization during onset of block was indistinguishable from that during offset.

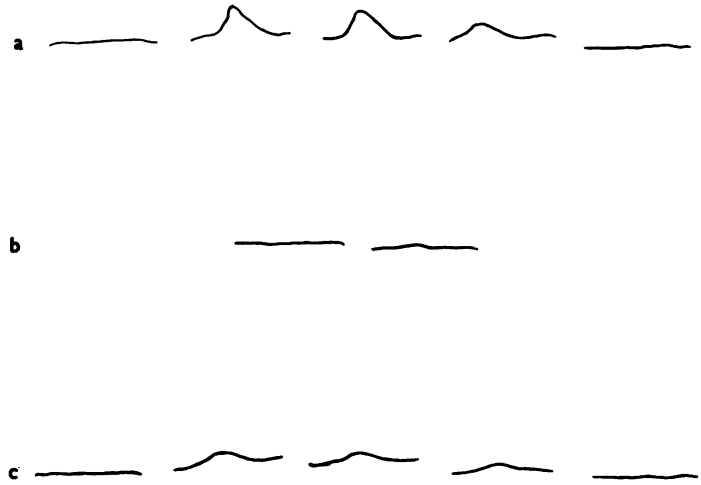

Fig. 9. Cat, 3.3 kg. Gracilis muscle. Iliac injections. Record of spatial distribution of potential round an endplate region. (a) Baseline followed by response to succinylcholine 25 μg at 30 sec, 2 min, 5 min and 20 min. (b) Baseline followed by response to nicotine 250 μg at 5 min. (c) Baseline followed by response to nicotine 1 mg at 5 sec, 10 min, 20 min and 30 min.

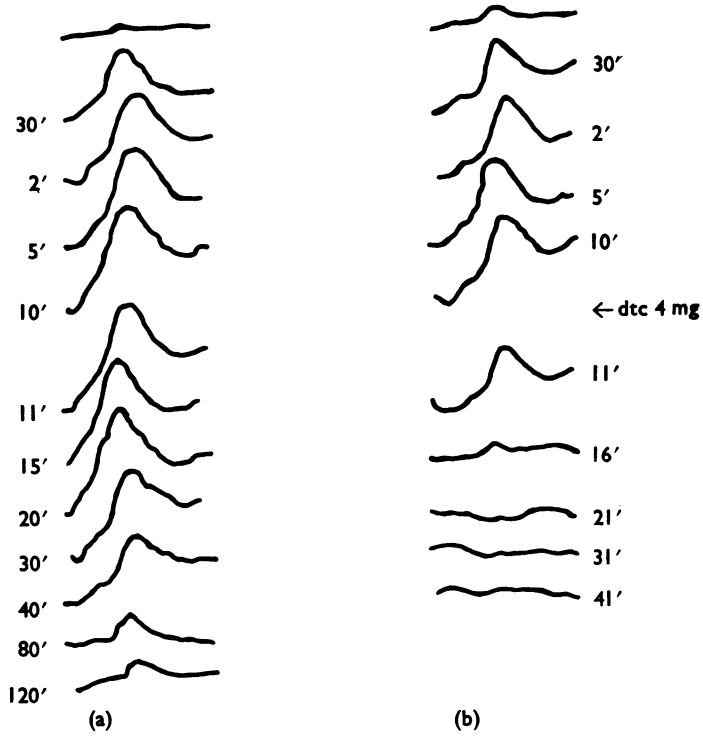

Fig. 10. Same preparation as Fig. 7. (a) Baseline followed by response to nicotine 4 mg at times indicated opposite successive traces. (b) Similar tracing, with *d*-tubocurarine (dtc) 4 mg injected at 10.5 min, showing rapid repolarization of endplate region, more rapid at centre than periphery.

Figure 11 (a)

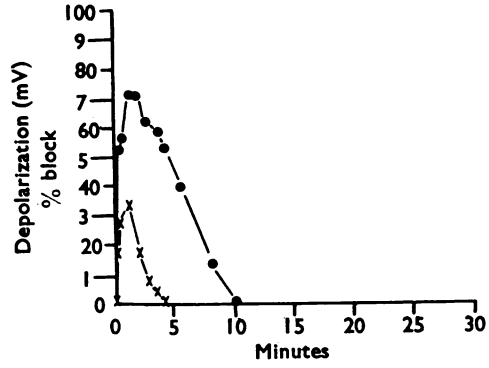

Figure 11 (b)

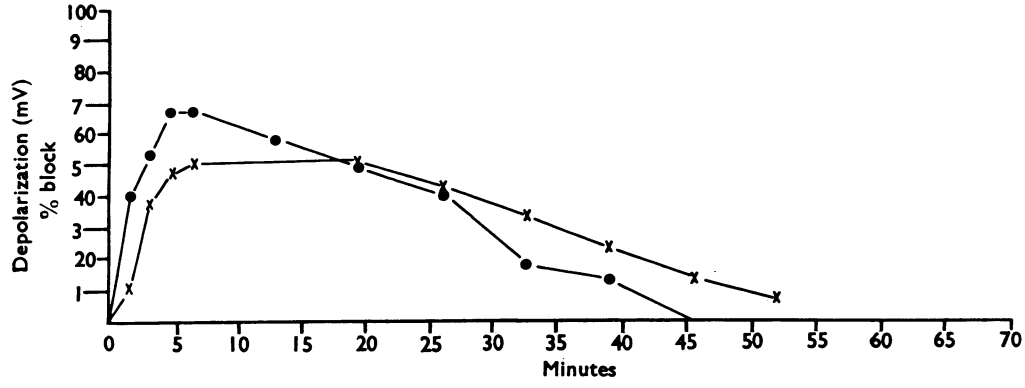

Figure 11 (c)

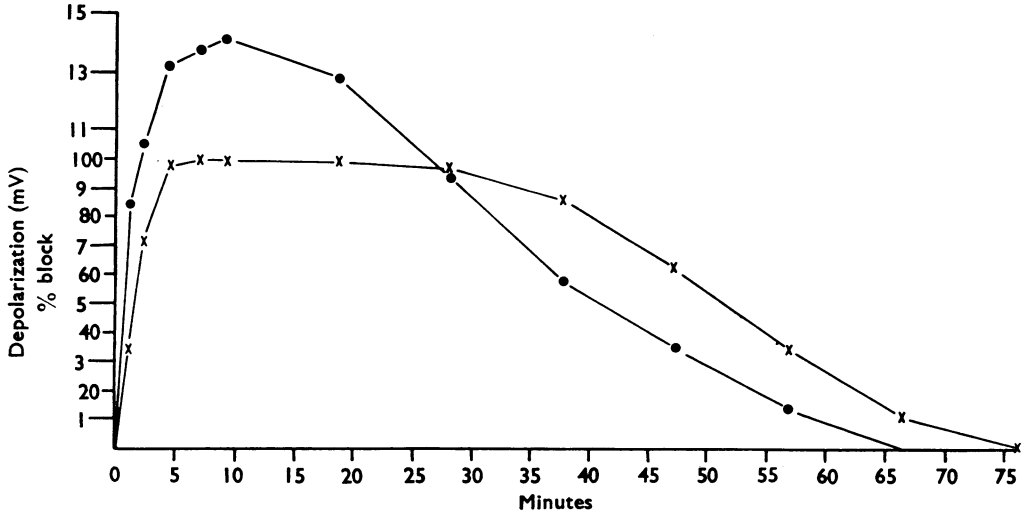

Figure 11 (d)

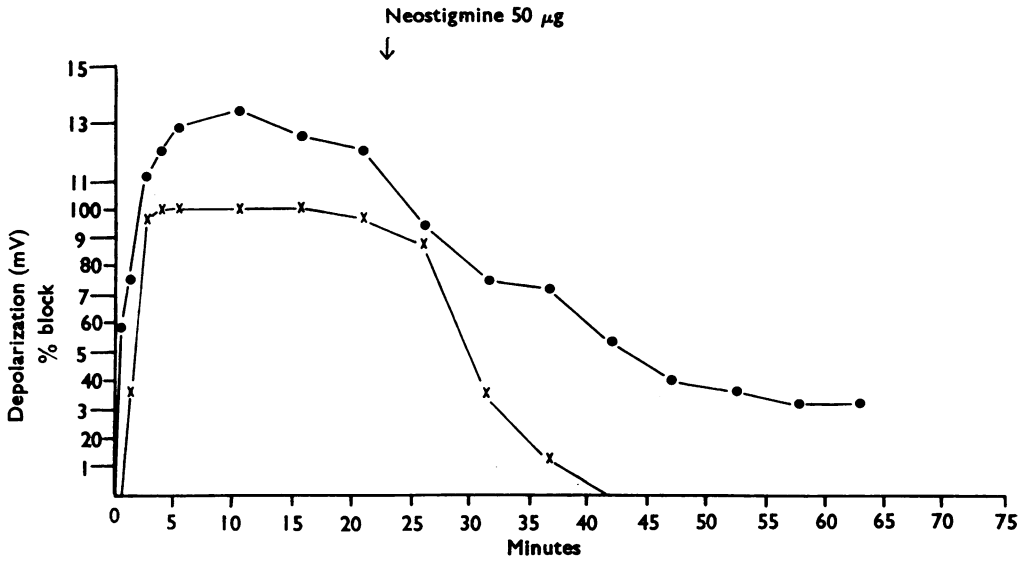

Fig. 11. Graph of time course of depolarization (●——●) and block (×——×). (a) From Fig. 6 (c), and (b), (c) and (d) from Fig. 7 (a), (b) and (c).

DISCUSSION

Although nicotine has lent its name to characterizing the action of actylcholine at the mammalian neuromuscular junction, the analysis of its action at this site has been very limited. Langley (1905) went so far as to say " nicotine does not stimulate all the muscles of the bird, and it does not, so far as we know, stimulate any in the mammal ". Thomas & Franke (1928) showed, in dogs under ether anaesthesia, that nicotine in a dose of 15 mg/kg produced peripheral neuromuscular block of the diaphragm before paralysing the respiratory centre. The observations of Bacq & Brown (1937), that nicotine given arterially could evoke a contraction of tibialis, followed by a prolonged block which could be stopped by tetanization of the nerve, suggest a complex action. Cahen (1953) showed, in the rabbit, that nicotine acted peripherally as well as centrally in causing tremor and twitching of the gastrocnemius muscle, and Ling (1959) showed that nicotine caused a contraction of the denervated gastrocnemius in the cat. None of these observations, however, allows an adequate comparison of the action of nicotine at the mammalian neuromuscular junction with that on the autonomic ganglion, where the characteristic initial excitation gives way to block without ganglionic depolarization (Paton & Perry, 1951). In addition, the fact that all the compounds hitherto found to depolarize the mammalian endplate are quaternary, whereas nicotine is not, made it desirable to characterize the action of nicotine more closely, particularly because it is possible that a non-quaternary compound can penetrate the nerve endings and interfere with acetylcholine release.

Our experiments showed, however, that the action of nicotine on cat tibialis, gastrocnemius and gracilis can for the most part be simply characterized as that of an endplate-depolarizing drug, of prolonged action and rather low potency, between 1/20 and 1/100

that of succinylcholine. Nicotine potentiates the indirectly excited twitch, and preferentially blocks tibialis ; the partly blocked muscle yields a sustained tetanus when the nerve is repeatedly stimulated ; there is only slight post-tetanic relief of block (except when recovery is well advanced) ; the block largely resists neostigmine, although recovery may be hastened ; the block is reversible by *d*-tubocurarine and gallamine ; and if nicotine is given together with succinylcholine there is no mutual antagonism but rather a potentiation. To these general features can be added the facts that the blocking action of nicotine is accompanied by a well developed endplate depolarization differing from that produced by succinylcholine only in time course of onset and offset, and that the decay of the depolarization can be considerably accelerated by tubocurarine.

Non-depolarizing component of neuromuscular block caused by nicotine

Closer examination showed that, although block by nicotine was always associated with depolarization, the block lasted slightly longer than the depolarization. This component of what will be termed (non-committally) non-depolarizing block is probably even larger than this observation alone indicates ; for example, the modest post-tetanic reduction of nicotine block, and the transient augmentation of it by gallamine, occur at a time when depolarization is still likely to be present. It is notoriously difficult to disentangle this situation (see Bowman, 1962, for discussion) ; we cannot give a quantitative description of the neuromuscular events concerned, which vary with animal, muscle and experimental conditions ; even comparisons between tests on a particular muscle in a single animal are subject to variation with time in physiological parameters such as blood flow, which can modify drug distribution and elimination. But the procedure of relating block to depolarization, and of then comparing the patterns so obtained with different drugs, may go some way to clarify the position. The analysis of Fig. 12 shows quantitatively that the block produced by nicotine in its later stages involves to a considerable extent some process not invoked in the test with succinylcholine. The fact that neostigmine can remove this component of block reinforces the conclusion.

Figure 12 (a)

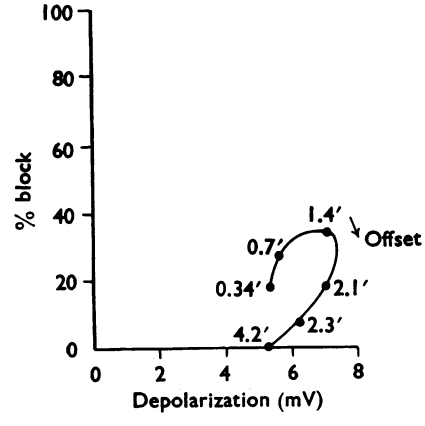

Figure 12 (b)

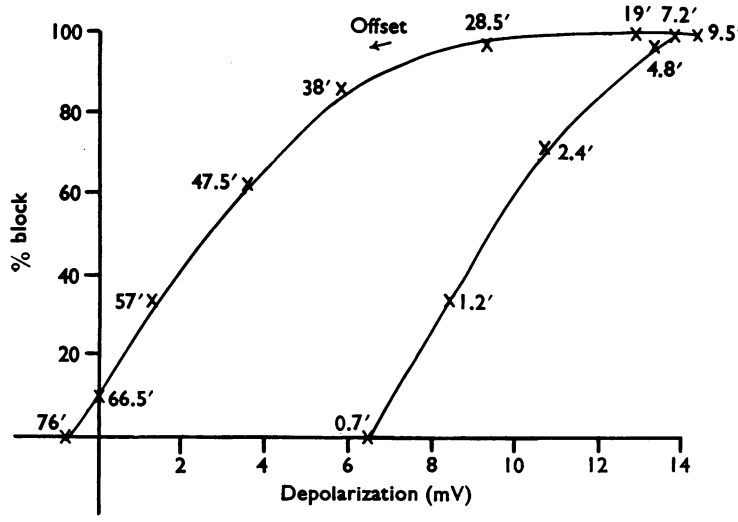

Figure 12 (c)

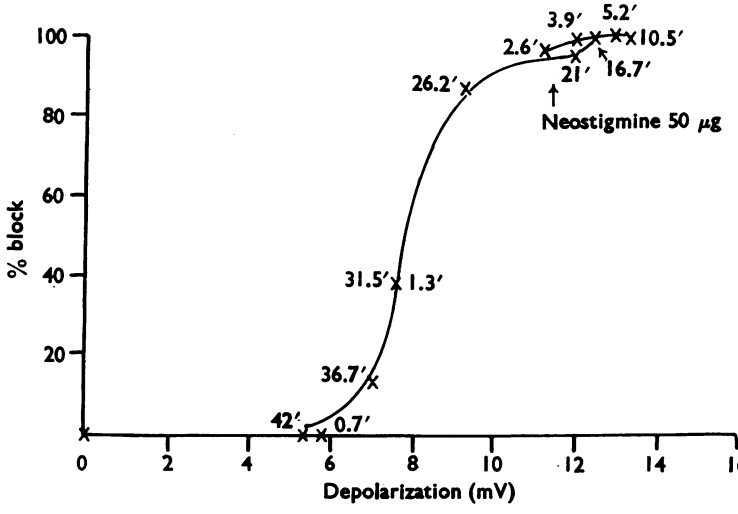

Fig. 12. Graph of relationship between depolarization and block. (a) From Fig. 11 a, (b) from Fig. 11 c, and (c) from Fig. 11 d.

This result in itself does not reveal the mechanism of the non-depolarizing block ; the effect of neostigmine merely means that augmentation of transmitter action will overcome it. A number of mechanisms can be suggested. Interference by nicotine with acetylcholine release has already been discussed and rejected because the block in insensitive to rate of stimulation. It may be noted also that in the ganglion Feldberg & Vartiainen (1934) showed that nicotine did not reduce acetylcholine output. A second process might be the development with time of an advancing occupancy of the receptors by nicotine,

leading to a competitive block both of itself and of other agonists and partial agonists. A third mechanism might be the desensitization of the motor endplate by depolarizing drugs, first brought into prominence by Thesleff (1955a). But both these explanations are hard to reconcile with the observation that nicotine does not in fact seem to render the endplate particularly insensitive to other depolarizing drugs, and certainly does not antagonize them by the factor of 4 or 5 necessary, as was found by Paton & Waud (1967), to produce at least threshold block (compare Figs. 1 and 8). A final possibility arises from the observation that persistent endplate depolarization leads to an electrical inexcitability of the endplate region, and that this inexcitability can outlast the depolarization (Burns & Paton, 1951 ; Paton, 1955). Such a development of electrical inexcitability could leave the specific chemoreceptive response of the endplate relatively unimpaired, but would raise the propagation threshold from endplate potential to muscle action potential.

This analysis suggests that it should be possible, operationally, to distinguish four types of postsynaptic block: (1) depolarization block characterized by a variety of stimulant phenomena accompanying depolarization, and by a stability to procedures which alter curare block ; this stability may arise from the fact that the receptors at the endplate are operating near the top of their dose-response curve and are thus relatively insensitive to changes in transmitter effectiveness (Paton, 1951) ; (2) desensitization block which, as Thesleff (1955b) has remarked, will also be stable, and is associated with a fading or absent depolarization and with an insensitivity both to anticholinesterases and to further depolarization ; (3) curare-like block in the strict sense, resulting from occupancy of specific receptors by drug molecules, characterized by sensitivity to anticholinesterases but antagonism to depolarization ; (4) block caused by residual electrical inexcitability, characterized by sensitivity to anticholinesterases together with relatively unimpaired sensitivity to depolarization. Our experiments provide some evidence that the action of nicotine involves not only the first of these, but also the last, possibly because of the prolonged action exerted by nicotine.

Comparison between motor endplate and ganglion

The action of nicotine at the neuromuscular junction is thus best interpreted as predominantly stimulant ; and it seems to lack the intense and prolonged phase of self-antagonizing block seen at the autonomic ganglion. It provides another example of the manner in which the type of action as well as the potency of a drug may change when it is tested in different cholinergic synapses, and of the tendency for excitation to be favoured at the neuromuscular junction, and for block to be favoured in the autonomic ganglion. Thus *d*-tubocurarine has been claimed to excite denervated muscle, but only blocks the ganglion ; tetraethylammonium is primarily a ganglion-blocking agent free from initial ganglionic excitation, but possesses a distinct endplate-depolarizing action ; decamethonium is predominantly a depolarizing agent at the motor endplate, but a competitive blocking agent on the ganglion (McIntyre & King, 1943 ; Jarcho, Berman, Eyzaguirre & Lilienthal, 1951 ; Paton & Perry, 1953 ; Paton & Waud, 1962). In terms of the rate theory of drug action, such observations suggest that if a given drug molecule is active at both ganglionic and neuromuscular synapses it will be a little more firmly bound at the former, and dissociate more readily at the latter.

Effect of nicotine intra-arterially

The rapid pressor effect of nicotine injected into the iliac artery is a striking phenomenon. Hilton (1954) has observed a similar response to arterial injection into the gastrocnemius muscle, as well as a vasodilation of the muscle. It seems probable that the response arises as a result of stimulating pain fibres, possibly in the vessel walls themselves, evoking the " pseudo-affective " reflex described by Woodworth & Sherrington (1904). It is a curious fact that, although the pressor response to nicotine is classically attributable to excitation of ganglia and suprarenal medulla, it is far more active as a pressor agent injected arterially into a vascular area beyond both the suprarenal glands and the sympathetic chains.

SUMMARY

1. The action of nicotine on the neuromuscular junction, given by distant and by close arterial injection, has been investigated on the tibialis, soleus, gastrocnemius and gracilis muscles of the cat under chloralose.

2. Nicotine has a neuromuscular blocking potency between 1 and 5% of that of succinylcholine, and a prolonged action. The block is characterized by initial potentiation of the twitch, ability to sustain a tetanus, and a preferential effect on tibialis with sparing of soleus and gastrocnemius. Following a tetanus, block is little reduced, except when recovery is well advanced. Neostigmine initially deepens block, but may hasten recovery. Gallamine or tubocurarine antagonize the block but may slightly deepen it at first. The actions of nicotine and succinylcholine summate, without any sign of mutual antagonism. Block by nicotine does not require repeated indirect excitation for its appearance.

3. Nicotine produces a localized depolarization of the motor endplate region, similar to that produced by succinylcholine, but much longer lasting. The passing off of the depolarization is much accelerated by tubocurarine, and a period of relative endplate positivity appears. The depolarization produced by succinylcholine summates with that produced by nicotine.

4. The relationship between the depolarization and the block produced by nicotine has been analysed. During the later stages of nicotine action the intensity of block, for a given depolarization, increases. The analysis reveals a component of the blocking action which can be overcome by neostigmine but is not caused by a reduced sensitivity of the endplate to depolarizing drugs. It is suggested that this is caused by an electrical inexcitability of the endplate region, hindering action potential propagation, as a result of the prolonged depolarization.

5. Intra-iliac injection of nicotine in low dosage causes a sharp rise of blood pressure within a few seconds, possibly by evoking the " pseudo-affective " reflex.

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