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The ganglion blocking and vagolytic actions of three short-acting neuromuscular blocking drugs in the cat

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Three recently-introduced short-acting neuromuscular blocking drugs with non-depolarizing mechanisms of action, stercuronium, dacuronium and AH8165 (1,1′-azobis [3-methyl-2-phenyl-1H-imidazo (1, 2a) pyridinium] dibromide) have been tested in the anaesthetized cat on the responses of the nictitating membrane to stimulation of the cervical nerve, and of the heart to vagal stimulation. The effects of the three drugs at the superior cervical ganglion and at the cardiac neuroeffector junction have been compared with their neuromuscular blocking effects. At doses lower than those required to block neuromuscular transmission all three compounds possessed a selective atropine-like action at the cardiac vagus neuroeffector junction in that they inhibited the bradycardia produced by vagal stimulation and by acetyl-β-methylcholine, whilst the depressor action of acetyl-β-methylcholine was unaffected. The ratios of the doses of the drugs to block the responses of the preganglionically-stimulated nictitating membrane and of the tibialis anterior muscle were 16·7 for stercuronium, 8·5 for dacuronium and 3·8 for AH8165. The greater ganglion-blocking activity of AH8165 was reflected in the depressor action of the compound, whereas the weak ganglion-blocking actions of stercuronium and dacuronium were insufficient to mask the tachycardia and pressor effect caused by their blocking action on the cardiac vagus neuroeffector junction.

Of the non-depolarizing neuromuscular blocking drugs currently in clinical use, (+)-tubocurarine possesses ganglion-blocking activity (Randall, 1951) and releases histamine (Foldes, 1957), effects which result in a fall of blood pressure. Gallamine and pancuronium, although lacking ganglion blocking action, occasionally cause tachycardia in man which is thought to be a result of their atropine-like action at the cardiac vagus neuroeffector junction (Bovet, Depierre & others, 1949; Jacob & Depierre, 1950; Saxena & Bonta, 1970).

Recently, three new quaternary ammonium compounds have been tested in attempts to provide a short-acting non-depolarizing blocking drug suitable for surgical procedures of short duration. The neuromuscular pharmacology of these drugs, 1,1′-azobis-[3-methyl-2-phenyl-1H-imidazo(1,2a) pyridinium] dibromide [AH8165] (Brittain & Tyers, 1972; Bolger, Brittain & others, 1972), stercuronium (Wieriks, 1969; Derkx, Bonta & Lagendijk, 1971) and dacuronium (Buckett & Saxena, 1969; Derkx, & others, 1971), has been the subject of several separate studies. The present study constitutes a comparison of the neuromuscular and ganglion blocking activities of these drugs in the anaesthetized cat. The effect of the drugs has also been studied at the cardiac vagus neuroeffector junction.

METHODS

All experiments were carried out on cats of either sex anaesthetized with a mixture of α-chloralose (8 ml kg^{-1} of a 1% solution) and pentobarbitone sodium (2·5 mg kg^{-1}) injected intraperitoneally.

The trachea was cannulated low in the neck, and the cats were artificially ventilated (15–20 ml air kg^{-1} weight) throughout the experiments. The left cervical nerve was separated from the vagus nerve, ligated and stimulated pre-ganglionically through bipolar platinum electrodes placed peripherally to the ligation. The cervical nerve was stimulated through a stimulus isolation unit by trains of rectangular pulses of 0·5 ms duration at a frequency of 20 Hz and of sufficient strength to produce a maximal contraction of the nictitating membrane. The train duration was 10 s and the train frequency was 0·01 Hz. In some experiments the right cervical nerve was stimulated post-ganglionically after ligation of the nerve peripheral to the superior cervical ganglion, using identical stimulation parameters to those used on the contralateral nerve. Isometric tension changes in the nictitating membranes were recorded by Grass FTO3C force displacement transducers.

The right sciatic nerve was exposed in the popliteal space, ligated and crushed centrally to the ligation. The nerve was stimulated through a stimulus isolation unit by bipolar platinum electrodes, with rectangular pulses of 0·1 ms duration and of sufficient strength to produce maximal twitches of the tibialis anterior and soleus muscles, at a frequency of 0·1 Hz. The right hind leg was immobilized by drills placed through the lower end of the femur and tibia, and isometric tension changes in the tibialis anterior and soleus muscles were recorded using Grass FT10C force displacement transducers.

Blood pressure was recorded from the left femoral artery through a polythene cannula attached to a Statham P23Ac pressure transducer. Drugs were injected into the left femoral vein via a polythene cannula.

In some experiments the left vagus nerve was periodically stimulated by 10 s trains of rectangular pulses (0·5 ms duration, 20 Hz). All recordings were made on Grass Model 79 ink-writing polygraphs.

The drugs used were acetyl-β-methylcholine, atropine sulphate, adrenaline acid tartrate (British Drug Houses), (+)-tubocurarine chloride (Koch-Light), stercuronium bromide (Brocades), dacuronium bromide (Organon), AH8165 (Allen & Hanburys) and dimethylphenylpiperazinium chloride (synthesized by Dr. A. L. Green, Department of Biochemistry, University of Strathclyde).

All drug concentrations referred to in the text apply to the appropriate salts. Adrenaline acid tartrate and stercuronium bromide were dissolved in acid saline and the remaining drugs in 0·9% saline.

RESULTS

In each cat one drug only was used in any experiment. Progressively larger doses of the drugs were injected, ranging from doses that were sub-effective, or produced a small degree of neuromuscular block, to doses that produced more than a 30% reduction of the responses of the nictitating membrane. The degree of neuromuscular block produced in the soleus muscle differed from that in the tibialis anterior muscle, the degree of this difference being dependent upon the drug used (Marshall, 1973). The soleus muscle was used in these experiments as a means of assessing the duration

of the neuromuscular block. Twitches of the soleus muscle recover from non-depolarizing neuromuscular block at a much slower rate than do twitches of the tibialis anterior muscle (Marshall, in preparation), and full recovery of the soleus muscle from such neuromuscular block is probably a good indication that little cumulation of the effect of repeated doses is likely to occur in the tibialis anterior muscle. Thus, in these experiments, at least 10 min was allowed after full recovery of the responses of the soleus muscle before further doses of the test drugs were administered.

All the drugs used were selectively active on neuromuscular transmission producing depression of the twitches of the tibialis anterior and soleus muscles at lower doses than those required to block ganglia. Doses up to those required to block neuromuscular transmission completely, of stercuronium (0·125–0·75 mg kg^{-1}) and of dacuronium (0·25–0·75 mg kg^{-1}), produced a slight increase in heart rate (stercuronium 2–15; dacuronium 2–12 beats min^{-1}) and a rise in blood pressure (stercuronium 5–60 mm Hg; dacuronium 15–20 mm Hg), whilst having little or no effect on responses of the preganglionically-stimulated nictitating membrane (Figs 1, 3, 4). On the other hand, AH8165 (0·75–2 mg kg^{-1}) produced a fall in blood pressure (20–45 mm Hg) and depressed the responses of the nictitating membrane by 5–25% (Figs 1 & 5).

Further increases of the doses of all three drugs used, beyond those required to block neuromuscular transmission completely, produced a progressive depression of the responses of the preganglionically-stimulated nictitating membrane (Figs 2, 3, 4, 5). The mean ratios of the dose required to produce 30% reduction of the responses of the preganglionically-stimulated nictitating membrane, to the dose required to produce 30% reduction of the responses of the tibialis anterior muscle were 16·7 for stercuronium (Fig. 3), 8·5 for dacuronium (Fig. 4) and 3·8 for AH8165 (Fig. 5). This indicates that the latter compound is less selective than the other two compounds for the nicotinic receptors at the neuromuscular junction.

The large doses of the compounds, sufficient to produce a reduction of the responses of the preganglionically-stimulated nictitating membrane, abolished the pressor

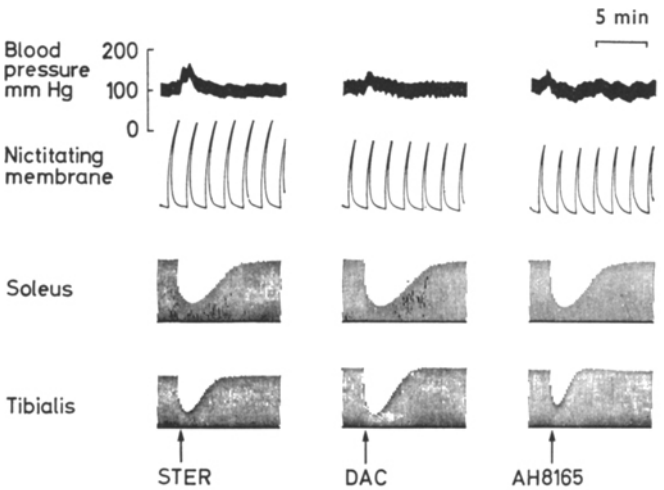

FIG. 1. Chloralose-anaesthetized cats. Effects of stercuronium (STER—0·5 mg kg^{-1}, i.v.), Dacuronium (DAC—0·75 mg kg^{-1}, i.v.) and AH 8165 (1 mg kg^{-1}, i.v.) on blood pressure, responses of the nictitating membrane to preganglionic stimulation and responses of the soleus and tibialis muscles to indirect stimulation at 0·1-Hz.

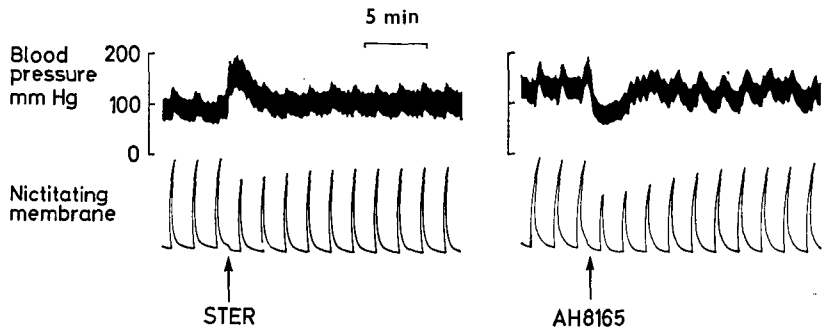

FIG. 2. Chloralose-anaesthetized cats. Effects of large doses of stercuronium (STER—4 mg kg^{-1}, i.v.) and AH 8165 (4 mg kg^{-1}, i.v.) on blood pressure and the responses of the nictitating membrane to preganglionic stimulation.

response to dimethylphenylpiperazinium (10 μg kg^{-1}), in atropine-pretreated (1 mg kg^{-1}) cats, whereas they had no effect on the pressor response to adrenaline (1 μg kg^{-1}) or on the response of the nictitating membrane to post-ganglionic stimulation of the cervical sympathetic nerve. This indicates that the site of action of the blocking drugs was the superior cervical ganglion.

All three compounds, at doses lower than those required to block neuromuscular transmission, reduced the negative chronotropic effect of stimulation of the vagus nerve (Figs 3, 4, 5), and abolished the bradycardia produced by acetyl-β-methylcholine (10 μg kg^{-1}, i.v.), whereas the depressor effect of acetyl-β-methylcholine was

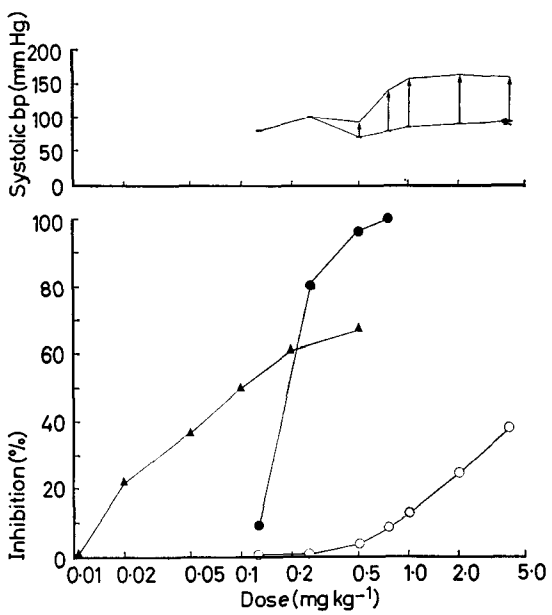

FIG. 3. Dose-response lines for stercuronium in the chloralose-anaesthetized cat. The lower part of the figure illustrates the effects of stercuronium on the bradycardia produced by vagal stimulation (-▲-), on the responses of the tibialis anterior muscle to indirect stimulation (-●-) and on the preganglionically-stimulated nictitating membrane (-○-). The upper part of the figure shows the changes in systolic blood pressure produced by stercuronium, the bases of the arrows representing the initial blood pressure, and the heads of the arrows representing the blood pressure after injection of stercuronium.

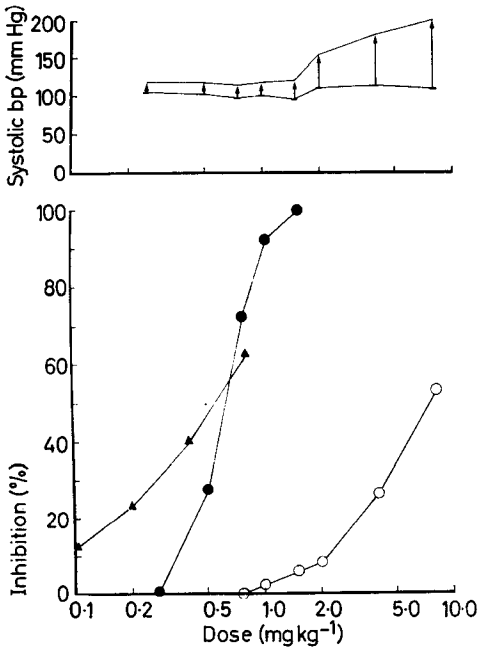

FIG. 4. Dose-response curves for dacuronium on vagal stimulation (-▲-) the tibialis anterior muscle (-●-), the nictitating membrane (-○-) and effects on systolic blood pressure in the anaesthetized cat, as in Fig. 3.

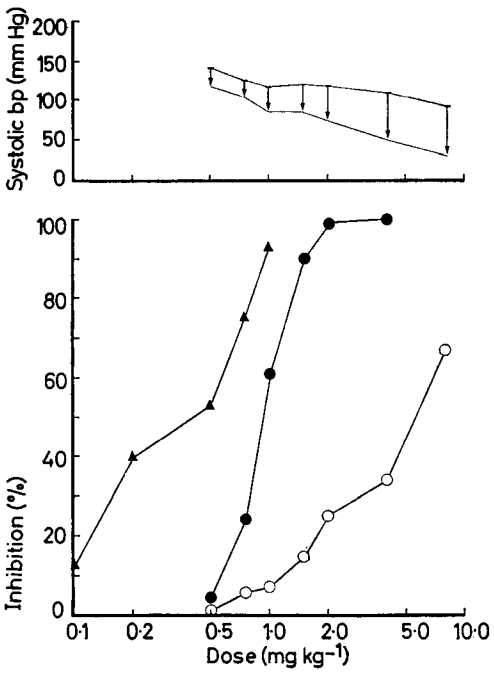

FIG. 5. Dose-response curves for AH 8165 on vagal stimulation (-▲-), the tibialis anterior muscle (-●-), the nictitating membrane (-○-), and effects on systolic blood pressure in the anaesthetized cat, as in Fig. 3.

little affected. These results indicate that, in addition to the ganglion-blocking action seen at higher doses, the compounds also possess an atropine-like action at the cardiac vagus neuroeffector junction, but not a generalized atropine-like action.

DISCUSSION

The ideal neuromuscular blocking drug should be non-depolarizing, short-acting and produce little effect on the autonomic nervous system. Effects of quaternary ammonium neuromuscular blocking agents on the cardiovascular system are generally mediated through blockade of autonomic ganglia, leading to a fall in blood pressure or through an atropine-like action on the cardiac vagus neuroeffector junction leading to tachycardia. Few neuromuscular blocking compounds show absolute specificity for the nicotinic receptors at the neuromuscular junction; e.g. (+)-tubocurarine, on a weight basis, is almost as potent as hexamethonium as a ganglion-blocking drug in the cat (Paton, 1959). Those drugs that possess large ratios between the dose required to block autonomic ganglia and that required to block neuromuscular transmission, e.g. gallamine and pancuronium (Bülbring & Depierre, 1949; Riker & Wescoe, 1951; Buckett, Marjoribanks & others, 1968; Buckett, 1972) possess additional actions on the muscarinic receptors of the heart (Bovet & others, 1949; Jacob & Depierre, 1950; Saxena & Bonta, 1970).

All the drugs tested were short-acting and of the non-depolarizing type (Marshall, in preparation). However, in addition to their primary action at the neuromuscular junction, which is evidenced by the low doses required to reduce responses of the tibialis anterior muscle, and the steepness of the dose-response curves on this muscle, all three compounds possessed actions on other cholinoceptive sites in the autonomic nervous system, resulting in effects on the cardiovascular system. All three compounds were approximately equiactive on a weight basis in blocking autonomic ganglia, but the order of potency of the compounds as neuromuscular blocking agents was stercuronium > dacuronium > AH8165. Thus, at doses that produced 90% reduction of the twitches of the tibialis anterior muscle, stercuronium and dacuronium produced less reduction (2·5 and 2% respectively) of the responses of the preganglionically stimulated nictitating membrane than did AH8165 (14·5% reduction).

At all the doses used, the possible cardiovascular effects of ganglion blockade produced by stercuronium and dacuronium were masked by their more potent action in blocking the muscarinic receptors of the cardiac vagus neuroeffector junction, leading to the production of tachycardia and a rise in blood pressure. In the case of AH8165 the cardiovascular effects of blockade of the muscarinic receptors of the heart were always masked by the effects of blockade of autonomic ganglia, resulting in a fall of blood pressure. The time course of this fall of blood pressure, paralleled the reduction of the responses of the preganglionically-stimulated nictitating membrane.

It is well documented that non-depolarizing neuromuscular blocking drugs have a greater effect as the frequency of nerve stimulation is increased (Preston & Van Maanen, 1953; Wislicki, 1958; Bowman, Hemsworth & Rand, 1962). The stimulation frequency used in the present study was chosen to avoid any possible complications due to prejunctional neuromuscular blockade, which is more likely to become evident as the stimulation frequency approaches 1–2Hz (Schueler, 1960; Bowman, Hemsworth & Rand, 1967; Bowman & Marshall, 1972). However, it is known that, at least with AH8165, the dose required to produce relaxation in man, is best predicted

from cat experiments in which the tibialis anterior muscle is stimulated at 1 Hz (Brittain —personal communication). Thus the present experiments may possibly give an overestimate of the ganglion blocking actions of the compounds in relation to their neuromuscular blocking actions.

In conlusion, although all three compounds used possess cardiovascular side-effects these effects may not prove to be a great disadvantage in a short-acting muscle relaxant, providing that the short duration of action found in animal studies can be duplicated in clinical studies. However, the tachycardia produced by both dacuronium and stercuronium in man (Norman & Katz, 1971; R. Bac—personal communication) may preclude the use of these two compounds in anaesthetic practice.

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