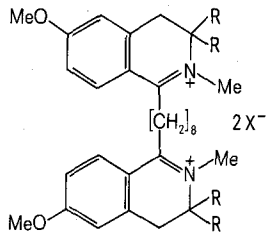
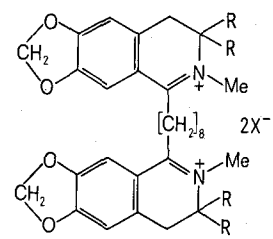
## Two New Short-Acting Non-Depolarizing Neuromuscular Blocking Agents

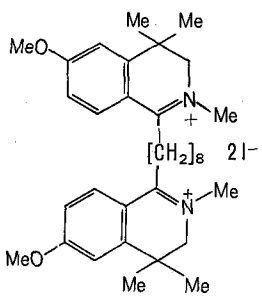
Although widely used in clinical practice as a short-acting skeletal muscle relaxant, suxamethonium is inadequate in certain respects. As an aftermath to depolarization of neuromuscular junctions it is known to cause post-operative muscle cramps[1]. Additionally, in a minority of patients, failure of the normal rapid hydrolysis of suxamethonium by plasma cholinesterase may lead to prolonged paralysis and apnoea[2,3]. In such cases neuromuscular function is usually not restored by treatment with inhibitors of acetylcholinesterase which effectively reverse paralysis caused by non-depolarizing agents such as tubocurarine and gallamine. These latter agents however have the disadvantage of a much longer duration of action than suxamethonium. Consequently there remains a need for a short-acting, non-depolarizing muscle relaxant the effect of which can be readily reversed should an atypical response make this necessary.



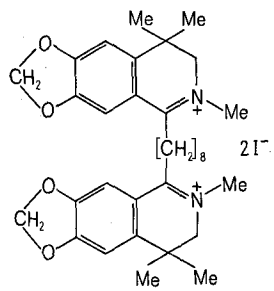
(I; B.W. 252C64; R = Me, X = Cl)
(II; B.W. 634C64; R = H, X = I)



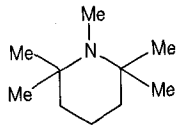
(III; B.W. 403C65; R = Me, X = $MeSO_4$)
(IV; B.W. 122C66; R = H, X = I)



(V; B.W. 331C68)



(VI; B.W. 624C66)



(VII)

As an approach to such an agent we studied a series of α.ω-*bis*-(3,3-dialkyl-3,4-dihydroisoquinolinium) alkanes[4,5], here exemplified by compounds (I) and (III) and also the related compounds (II), (IV), (V) and (VI). It is well established that certain bases in which the nitrogen is sterically hindered possess ganglionic blocking activity[6] and pempidine (VII), one of the most active of these bases, contains 2 gem dimethyl groups adjacent to the nitrogen atom. Since acetyl-choline is the chemical transmitter at the neuromuscular junction as well as in autonomic ganglia, it seemed possible that the introduction of such groups into a *bis*quaternary ammonium molecule might enhance intrinsic ability to depress neuromuscular transmission. The compounds (I)–(VI) allowed investigation of this possibility.

The paralyzing effects of the compounds were determined on isolated rat phrenic nerve diaphragm preparations[7] and on the responses of the gastrocnemius muscle to single shocks applied to the sciatic nerve following i.v. injection in cats anaesthetized with chloralose (Table).

Comparisons of (I) with (II) and of (III) with (IV) indicate that, for these 2 pairs of compounds, distinctly higher activity is present in the 3,3-dimethyl derivatives, i.e., (I) and (III); other examples are available within the entire series[4,5]. The 4,4-dimethyl derivative (VI) was less active than (III) but the relevant potencies of (I) and its 4,4-dimethyl analogue (V) depended on the test situation. On balance these results support the concept that, like ganglion blocking activity, neuromuscular blocking activity is influenced by the degree of steric hindrance around the nitrogen atoms.

All new compounds in the Table were shorter-acting than tubocurarine or gallamine in cats whilst (I) and (III) were the most active. At paralysing doses, (II) markedly lowered blood pressure, (IV) suppressed cardiac slowing in response to vagal nerve stimulation and (V) and (VI) produced salivation and bronchial secretion suggestive of anticholinesterase activity.

Absence of depolarization of the tibialis muscles of cats during paralysis by (I) and (III) was demonstrated using the method of BURNS and PATON[8] and in keeping with this both compounds paralyzed chick biventer cervicis preparations[9] without causing a contracture. Whereas non-depolarizing agents are expected to be antagonized by anticholinesterase agents, antagonism of paralysis caused by (I) and (III) was dependent upon the test situation. In cats, paralysis by (III) was reliably antagonized by neostigmine and edrophonium whereas paralysis by (I) was usually slightly deepened. Both were


[1] H. C. CHURCHILL-DAVIDSON, Br. med. J. *1*, 746 (1954).
[2] F. T. EVANS, P. W. S. GRAY, H. LEHMANN and E. SILK, Lancet *1*, 1229 (1952).
[3] F. T. EVANS, P. W. S. GRAY, H. LEHMANN and E. SILK, Br. med. J. *1*, 136 (1953).
[4] F. C. COPP, Brit. Patent 1, 125, 619 (1968).
[5] G. G. COKER and F. C. COPP, unpublished (1968).
[6] R. WIEN, in *Progress in Medicinal Chemistry*, (Eds. G. P. ELLIS and G. B. WEST; Butterworth, London 1961), Vol. 1, p. 34.
[7] E. BÜLBRING, Br. J. Pharmac. Chemother. *1*, 38 (1946).
[8] B. D. BURNS and W. D. M. PATON, J. Physiol., Lond. *115*, 41 (1951).
[9] B. L. GINSBORG and J. WARRINER, Br. J. Pharmac. Chemother. *15*, 410 (1960).

Cationic concentrations causing 50% paralysis at 5 min in isolated rat phrenic nerve diaphragms (mean for 2 diaphragms); i.v. doses reducing by 50% the responses of the gastrocnemius muscles to stimulation of the sciatic nerves in cats, together with the times for onset of peak effect and subsequent full recovery

| | Rat diaphragm | | Cat | | | |
|---|---|---|---|---|---|---|
| | Concentration | | $n$ | Dose of cations (mg/kg) | Peak time (min) | Recovery time (min) |
| | $\mu M$ | ($\mu$g/ml) | | | | |
| Tubocurarine | 1.6 | 1.0 | 8 | 0.15 (0.10–0.21) | 3.8 (1.8– 8.1) | 19.5 (15.2–25.0) |
| Gallamine | 400 | 100 | 7 | 0.65 (0.42–1.02) | 3.7 (1.7– 8.3) | 12.3 (9.4–16.1) |
| I B.W. 252C64 | 29 | 15 | 3 | 0.22 (0.11–0.41) | 2.5 (0.7– 8.5) | 6.4 (4.3–9.6) |
| II B.W. 634C64 | 100 | 47 | 2 | 2.14 (0.99–4.60) | 2.6 (0.6–11.6) | 3.8 (2.3–6.3) |
| III B.W. 403C65 | 16 | 9 | 9 | 0.13 (0.09–0.19) | 1.8 (0.9– 3.7) | 5.5 (4.3–6.9) |
| IV B.W. 122C66 | 57 | 28 | 2 | 0.98 (0.45–2.11) | 3.5 (0.8–15.0) | 4.6 (2.8–7.5) |
| V B.W. 331C68 | 25 | 13 | 3 | 0.33 (0.15–0.62) | 2.9 (0.8– 9.8) | 6.1 (4.0–9.1) |
| VI B.W. 624C66 | 53 | 29 | 2 | 0.64 (0.30–1.38) | 4.2 (0.8–16.8) | 5.7 (3.5–9.4) |

95% fiducial limits of the estimates are in brackets. $n$ = number of tests.

antagonized by physostigmine consistently in chick biventer cervicis preparations but rarely in isolated rat diaphragms. There observations may reflect differing inhibitory effects of the compounds themselves on the muscle cholinesterases of the two species.

Anticholinesterase activities were determined using structure bound acetylcholinesterase[10,11] prepared from homogenates of mixed back and hind limb muscle; acetylcholine at 0.5 m$M$ was used as substrate. In these conditions the respective concentrations causing 50% inhibition of rat and chick enzymes were 0.16 and 1.6 $\mu M$ of (I), 2.5 and 25 $\mu M$ of (III) and, for comparison, 0.025 and 0.063 $\mu M$ of physostigmine. For rat enzyme these concentrations of (I) and (III) are so much less than those required to paralyze diaphragm preparations (Table) that it seems likely that paralysis would be accompanied by strong inhibition of acetylcholinesterase. If this were the case then addition of a second anticholinesterase would not be expected to reverse paralysis. By contrast in chick preparations the much lower neuromuscular paralyzing concentrations of (I) (1.9 $\mu M$) and of (III) (0.92 $\mu M$) would probably, almost certainly in the case of (III), leave much acetylcholinesterase activity uninhibited. The relatively high anticholinesterase activity of (I) could similarly account for failure to reverse paralysis in the cat with anticholinesterase agents and also for the occurrence of salivation after doses higher than those needed to induce paralysis.

Neither (I) nor (III) affected contractions of the nictitating membrane caused by stimulation of the preganglionic sympathetic nerves but at paralyzing doses both reduced the cardiac slowing caused by vagal nerve stimulation. This effect is attributed to competition with acetylcholine at muscarinic sites since the effects of injected acetyl $\beta$-methylcholine were also reduced in cats and the spasmogenic effect of acetylcholine at submaximal concentrations in isolated guinea-pig ileum was suppressed by 1 $\mu$g/ml of either compound. Vagal blockade was less with (I) than with (III), again perhaps partly in consequence of differing anticholinesterase properties.

Both compounds have been studied in man by Professor T.C. GRAY, Dr. R.S. AHEARN, Dr. I.C. GEDDES and Dr. J.E UTTING in the Department of Anaesthesia of the University of Liverpool. They observed neuromuscular paralysis but the required dosage and the persistence of effect found were greater than in cats. At the larger dosages required for paralysis (I) not unexpectedly caused excessive salivation and bronchosecretion indicative of its anticholinesterase effect. Like other muscle relaxants that block the cardiac vagus[12], (III) caused a greater degree of tachycardia and hypertension in man than suggested by animal studies.

*Résumé.* Le chlorure de 1,1′-octaméthylène et le méthylsulfate de 5,5′-octaméthylènebis ont chez le chat un effet de blocage neuro-musculaire non dépolarisant, de courte durée. Les comparaisons faites avec des analogues voisins suggèrent que l'activité de ces composés est influencée par le degré d'empêchement stérique autour des atomes d'azote quaternaire. Les composés inhibent les acétylcholinestérases et s'opposent à des degrés divers aux effets muscariniques de l'acétylcholine.

F.C. COPP, G.G. COKER, A.F. GREEN, R. HUGHES and R.H. NIMMO-SMITH

*The Wellcome Research Laboratories, Beckenham (Kent, BR3 3BS, England), 12 July 1971.*

[10] M. ULBRECHT, Biochim. biophys. Acta *57*, 438 (1962).
[11] G. ULBRECHT and P. KRUCKENBURG, Nature, Lond. *206*, 305 (1965).
[12] R. HUGHES, Br. J. Anaesth. *42*, 928 (1970).