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## Relative susceptibility of peripheral sympathetic nerves to adrenergic neurone blockade by bethanidine

J. M. ARMSTRONG and A. L. A. BOURA*, *Pharmacology Laboratory, The Wellcome Research Laboratories, Langley Court, Beckenham, Kent*

The intravenous doses of bethanidine sulphate necessary in cats for complete abolition of responses of the heart to sympathetic postganglionic nerve stimulation (0·4–0·8 mg/kg) were much lower than those required for inhibiting contractions of the nictitating membrane caused by indirect stimulation (3·2 mg/kg) (Boura & Green, 1963; Armstrong & Boura, 1970). The ability of bethanidine to block the responses of the heart and nictitating membranes to stimulation of their respective sympathetic postganglionic nerves has now been studied further and related to any concomitant change in effector tissue sensitivity.

Cats were anaesthetized with chloralose, and autonomic reflexes were blocked by bilateral vagotomy, sympathetic cardiac nerve section or ganglion blockade (pentacynium 2 mg/kg intravenously). Low intravenous doses of bethanidine (0·4–0·8 mg/kg) slightly reduced the magnitude of the nictitating membrane contractions caused by nerve stimulation (0·3–30·0 Hz), and potentiated responses to intra-arterial injection of 0·25–16·0 $\mu$g (−)-noradrenaline bitartrate (2·6 fold), 0·1–4·0 $\mu$g (−)-adrenaline bitartrate (2·1 fold) and 2·5–40·0 $\mu$g acetylcholine bromide (approximately 1·2 fold). Higher dose levels (1·6 mg/kg) caused a 5·5, 3·3 and a 2·8 fold increase in sensitivity to noradrenaline, adrenaline and acetylcholine respectively.

The most likely explanation for the hypersensitivity of the nictitating membranes is that bethanidine resembles other adrenergic neurone blocking agents by blocking uptake of catecholamines into tissue stores (Boura & Green, 1965), thereby permitting increased concentrations to reach effector sites. This conclusion is supported by the finding that noradrenaline was potentiated more than adrenaline, the former amine being taken up more readily from the circulation than the latter (Iversen, 1967). The potentiation of the effects of acetylcholine could be due to elevated background levels of noradrenaline as caused by cocaine (Trendelenburg, 1962).

Little potentiation of the positive chronotropic effects on the heart of noradrenaline (0·5–8 $\mu$g) and adrenaline (0·5–8 $\mu$g) given intravenously could be detected after the above doses of bethanidine. This contrast between the magnitude of the myocardial and nictitating membrane sensitivity changes to catecholamines is again analogous to that which occurs after cocaine (Innes & Kosterlitz, 1950; Fleming & Trendelenburg, 1961).

It is concluded that the apparently high resistance of the postganglionic cervical sympathetic nerve to the blocking action of bethanidine can be accounted for by the relatively greater hypersensitivity developed by the nictitating membrane to catecholamines. Any reduction in the output of transmitter, resulting from administration

of low doses of bethanidine, would be compensated for by the concomitant increase in sensitivity of the nictitating membrane to its action.

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## Extrinsic and intrinsic acetylcholine and barbiturate effects on frog skeletal muscle

P. R. ADAMS*, H. C. CASH and J. P. QUILLIAM, *Department of Pharmacology, St. Bartholomew's Hospital Medical College, Charterhouse Square, London E.C.*1

Del Castillo & Katz (1957) demonstrated that tubocurarine simultaneously reduced the depolarization response of frog skeletal muscle to both neurogenically released (intrinsic) and electrophoretically applied (extrinsic) acetylcholine (ACh). Quilliam (1955) reported that barbiturate drugs decreased the ACh contracture of the frog isolated ileo-fibularis muscle without a concomitant block of neuromuscular transmission. He also noted (unpublished observations) that the depolarizing response to ACh added to the fluid bathing the frog isolated toe muscle was profoundly decreased by low concentrations of barbiturate compounds which left the muscle action potential undiminished. Thesleff (1956) suggested that pentobarbitone might be more effective in reducing the response to extrinsic than to intrinsic acetylcholine. Since these findings seem to imply some difference in the action of intrinsic and extrinsic ACh, we have investigated this phenomenon further on the frog isolated motor nerve-sartorius muscle preparation.

Transmission was blocked with 6–10 mM Mg for the observation of end-plate potentials. An intracellular microelectrode recorded the end-plate potentials (epps) and miniature end-plate potentials (mepps), and also the electrophoretic potentials produced by pulses of current through an acetylcholine-filled micropipette placed on the end-plate region. These potentials were evoked concurrently, photographed, and also monitored on a chart-recorder.

Concentrations of amylobarbitone (2 to $8 \times 10^{-5}$ g/ml) which were without neuromuscular blocking action produced a large depression of the electrophoretic potentials, while there was little or no effect on the mepps or epps. Essentially similar results were obtained using thiopentone (2 to $8 \times 10^{-5}$ g/ml).

Two possibilities exist. First, the observed differences were due to different rates of ACh release, neurally and electrophoretically. The effect of amylobarbitone on electrophoretic potentials of different time course but of comparable amplitude was examined and found to be the same. Second, barbiturate drugs might be acting like