(Żebrowska-Lupina, Przegaliński, Sloniec & Kleinrok, 1977; Drew, Gower & Marriott, 1977).

We have investigated the modification of the centrally mediated cardiovascular effects of clonidine by using prazosin, a relatively selective antagonist of postsynaptic α-adrenoceptors in the peripheral cardiovascular system of the rat (Cavero, Lefèvre & Roach, 1977).

In 8 urethane (1.5 g/kg, i.p.) anaesthetised normotensive Charles River rats clonidine (10.0 μg) injected into the lateral cerebral ventricles (c.v.) induced peak falls in mean arterial blood pressure (MABP) and heart rate (HR) of 23.5 ± 2.8 mmHg and 66.9 ± 10.1 bts/min which occurred respectively 20 and 15 min after its injection (initial MABP and HR levels: 94.5 ± 7.0 mmHg, 363.0 ± 11.0 bts/min). In another group of normotensive rats i.c.v. or i.v. prazosin (25 μg) lowered blood pressure to a level which was not significantly different from that achieved after i.c.v. clonidine. This problem was avoided by administering prazosin orally for 3 days (first day, 1.0 and 0.5 mg/kg; second and third days 2 × 0.5 mg/kg). The blood pressures (93.3 ± 4.5 mmHg, $n = 9$) and heart rates (354.7 ± 15.3 bts/min, $n = 9$) of these animals after inducing urethane anaesthesia 12 h after the last dose of prazosin were not significantly different from those of the untreated rats. In these prazosin pretreated animals, the hypotensive and bradycardic effects of clonidine (10.0 μg, i.c.v.) were almost entirely abolished.

In two day old chicks, clonidine (0.5, 1.0 and 2.0 mg/kg, i.m.) induced a sleep-like state, the duration of which was a function of the dose (14.8 ± 1.6, 20.8 ± 1.2 and 32.0 ± 2.9 min, respectively; $n = 14$/group). These effects were reduced (3.2 ± 1.3, 7.7 ± 0.6 and 11.9 ± 0.9 min; $n = 10$) when clonidine was given 10 min after 15.0 mg/kg, i.p. phentolamine, an antagonist of pre- and postsynaptic α-adrenoceptors. In contrast, the same dose of prazosin tended to increase the clonidine hypnosis in chicks (16.1 ± 1.2, 25.2 ± 3.0 and 41.7 ± 3.0 min; $n = 12$).

In conclusion, oral prazosin antagonized the clonidine induced hypotension and bradycardia in rats probably by inhibiting receptors which possess similar characteristics to vascular postsynaptic α-adrenoceptors. In contrast, the lack of activity of prazosin in preventing clonidine hypnosis in chicks may indicate that the receptors responsible for evoking this effect may respond in a similar way as the rat cardiac presynaptic α-adrenoceptors to agonists or antagonists.

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A simple and cheap vacuum filtration

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Filtration under reduced pressure is a technique which has been applied in studies where speed of separation and rapid washing of trapped material are desirable. Described here is a simple, cheap and effective manifold for separating subcellular particles from incubation medium which has been used in this laboratory for uptake, release and receptor binding studies.

Six filtration units as shown in Figure 1 are connected by 5 cm lengths of 22 mm copper tube. The Swinnex 25 mm filter support is presssed onto the side arm of the 'T' piece using a vice after the Luer needle attachment and stem have been drilled out with a 12 mm bit. Twenty-two mm Yorkshire 'elbows' at the corners and 22 mm copper tubing is used to complete a ring structure. Another 22 mm Yorkshire 'T' piece is placed at a convenience point in the 'ring', and this latter 'T' piece has the side arm fitted with a 22 mm to 7 mm reducer, into which is soldered a short length of 7 mm copper tubing. All other connections are made with rubber pressure tubing. The manifold is connected by a three way glass tap to a 4 litre vacuum

flask which serves as a vacuum reservoir and waste trap. The side arm of the vacuum flask is connected to a liquid nitrogen vapour trap which in turn leads to a high vacuum pump (Edwards, I.S. 15).

With six wetted Whatman GF/B filters in place, a flow rate of 300 ml/min is achieved through each filter.

Using this apparatus we have found that binding of [^{3}H]-quinuclidinyl benzilate to membrane prepared from rat corpus striatum has a $K_D = 0.43$ nM, and a Scatchard analysis reveals 0.55 pmoles of binding sites/mg protein. These values are in good agreement with those published by Yamamura & Snyder (1974). IC_{50} and RC_{50} values for the inhibition of uptake and release of [^{3}H]-dopamine by (+)-amphetamine in rat striatum synaptosomes were found to be 1×10^{-8}M and 2×10^{-7}M respectively, values in good agreement with those found using centrifugation to separate the synaptosomes (Kruk & Zarrindast, 1976).

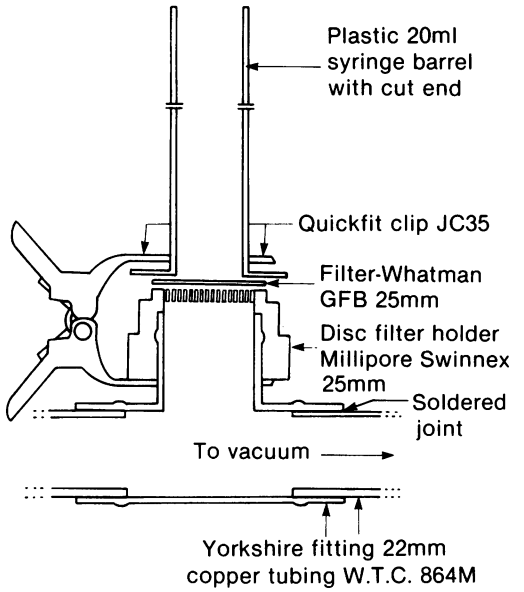

Figure 1 Transverse section of vacuum filtration unit.

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Effect of atropine on prejunctional muscarinic and α-adrenoceptors on postganglionic sympathetic neurones in an isolated blood vessel

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Atropine enhanced the overflow of tritium evoked by electrical-field stimulation of an isolated blood vessel preloaded with [^{3}H]-noradrenaline, which may be due to inhibition of prejunctional α-adrenoceptors (Nedergaard & Schrold, 1975; 1977). Atropine antagonizes the inhibitory effect of acetylcholine on stimulation-evoked tritium release from adrenergic neurones in many tissues (cf. Starke, 1977). The present investigation was undertaken in order to obtain direct evidence for the view that atropine acts on prejunctional α-adrenoceptors and muscarine receptors which both modulate the neuronal transmitter release.

Rabbit isolated pulmonary artery preloaded with [^{3}H]-noradrenaline was used. Atropine (10^{-4}M) and phentolamine (10^{-6}M) increased the stimulation-induced overflow of tritium, while clonidine (10^{-6} to 10^{-5}M) and acetylcholine (10^{-6}M) diminished it. After the overflow had been increased by either atropine (10^{-4}M) or phentolamine (10^{-6}M), clonidine (10^{-6}M) decreased the overflow below the control value. Clonidine (10^{-5}M) prevented the enhancement of tritium overflow evoked by atropine (10^{-4}M). A lower concentration of clonidine (10^{-6}) only caused a partial prevention. Atropine (10^{-7}M), in a concentration which was without any effect on the stimulation-induced tritium overflow, prevented the reduction caused by acetylcholine (10^{-6}M).

We conclude that atropine in a low concentration blocks prejunctional muscarinic receptors; at higher concentrations it blocks in addition prejunctional α-adrenoceptors.

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