α-Adrenoceptor binding in guinea-pig lung using [^{3}H]-prazosin

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β-Adrenoceptors in lung membranes have been characterised by radioligand studies using (−)-[^{3}H]-dihydroalprenolol ([^{3}H]-DHA) (Rugg, Barnett & Nahorski, 1978), but there has been no previous study of α-adrenoceptor binding in pulmonary tissue. We have used [^{3}H]-prazosin, a new radioligand of high specific radioactivity (33 Ci/mmol) to study α-adrenoceptors in guinea-pig lung membranes. There is considerable evidence that α-adrenoceptors in peripheral tissues may be classified as postjunctional (α_1) or prejunctional (α_2) and α-adrenoceptor agonists and antagonists vary widely in their relative pre- and postsynaptic potencies. Prazosin is an α-adrenoceptor antagonist which has a high selectivity for peripheral α_1 receptors (Cambridge, Davey & Massingham, 1977) and [^{3}H]-prazosin has recently been shown to bind to rat brain α-adrenoceptors with a high degree of specificity (Greengrass & Bremner, 1979).

Guinea-pig lungs, dissected free of major bronchi, were homogenised, centrifuged at 50,000 *g* and the final pellet resuspended at a concentration of approximately 1 mg protein/ml. Aliquots of the homogenate were incubated at 25°C for 15 min with various concentrations of [^{3}H]-prazosin from 0.05 nM to 4.0 nM in a final volume of 1.0 ml. Bound radioactivity was isolated on GF/B filters followed by 2 × 6 ml washes with incubation buffer and quantified by scintillation counting. Specific binding, defined as that displaced by phentolamine (1 μM), comprised 70–80% of the total binding at ligand concentrations of 0.05–1.0 nM.

Specific [^{3}H]-prazosin binding was rapid ($T_{\frac{1}{2}}$ association = 2 min) at a ligand concentration of 0.26 nM, remained at a steady state for more than 30 min, and was reversible ($T_{\frac{1}{2}}$ dissociation = 1 min). Specific binding was saturable reaching a plateau between 1 and 2 nM [^{3}H]-prazosin and Scatchard analysis ($n = 5$) revealed a single receptor population with K_D of 0.24 ± 0.05 nM (mean ± s.e. mean) with a maximum binding capacity (Bmax) of 54 ± 7 fmol/mg protein. A Hill plot gave a slope of 1.06 indicating absence of cooperativity.

For comparison, binding of the β-adrenoceptor antagonist [^{3}H]-DHA was measured under identical conditions and specific binding was defined as that displaced by propranolol (10 μM). This gave a K_D of 0.93 ± 0.1 nM and a B_{max} of 870 ± 112 fmol/mg protein. Thus in guinea-pig lung membranes the ratio of beta:α adrenoceptor binding sites is approximately 16:1.

Adrenoceptor agonists inhibited [^{3}H]-prazosin binding in the order: (−)adrenaline > (−)noradrenaline ≫ (−)phenylephrine > (−)isoprenaline; (+)noradrenaline was 100 × less potent than (−)noradrenaline. α-Adrenoceptor antagonists competed for binding in the order: prazosin > phentolamine ≫ piperoxan > yohimbine, indicating that [^{3}H]-prazosin binding is probably to α_1 adrenoceptors as it is inhibited by potent α_1 adrenoceptor antagonists but not by drugs more potent at α_2 sites. Propranolol, methysergide and sulpiride inhibited binding only at concentrations greater than 10 μM.

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Interaction between prazosin and benzodioxan antihypertensives (R 28935 and R 29814); a competition for central α_1-adrenoceptors

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Previous studies have established that the erythro-isomer of 1-{1-(2-(1,4-benzodioxan-2-yl)-2-hydroxyethyl)-4-piperidyl}-2-benzimidazolinone (R 28935) lowers arterial pressure via an action within the central nervous system. Although appreciably less active than R 28935, the threo-isomer (R 29814) also displays a central hypotensive effect. However, the exact mechanism is still unknown. Central α-adrenoceptors have been excluded to play a substantial role, since central α-adrenoceptor antagonism accomplished by yohimbine, piperoxan, tolazoline and phentolamine did not affect the responses (Finch, 1975; Wellens *et al.*, 1975; van Zwieten, 1975; Taylor & Antonaccio, 1978). The present study reports on the impairment of