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A COMPARATIVE STUDY OF β-ADRENOCEPTORS IN HUMAN AND PORCINE LUNG PARENCHYMA STRIP

ROY G. GOLDIE, JAMES W. PATERSON & JANET L. WALE

Department of Pharmacology, University of Western Australia, Nedlands, Perth, 6009, Australia

1 Responses to (±)-isoprenaline (Iso), (−)-adrenaline (Adr) and (−)-noradrenaline (NA) were compared in isolated preparations of human and porcine lung parenchyma strip.

2 The order of relaxant potencies of these catecholamines in both human and porcine lung parenchyma was Iso > Adr > NA (1:0.24:0.01, human; 1:0.21:0.01, pig). These results suggest that β_2-adrenoceptors predominate in both types of lung parenchyma strip.

3 pA_2 values for the β-adrenoceptor antagonist, propranolol (non-selective), with Iso as the agonist, in human and porcine lung strips were 7.84 and 7.83 respectively and for atenolol were 6.50 and 5.35 respectively. Taken as a whole, results indicate the existence of an apparently homogeneous population of β_2-adrenoceptors in porcine parenchyma strip, while both β_1 and β_2-adrenoceptors were revealed in human lung parenchyma.

Introduction

The study of drug responses in lung tissue is important when considering therapeutic strategies for the relief of airways obstruction. It may be necessary to study drug responses at different levels of the airways as the site of airways obstruction can vary in different disease processes. Peripheral airways are the major site of obstruction in chronic bronchitis (Despas, Leroux & Macklem, 1972), while in asthma, the site of obstruction is mainly in the central airways. However, some patients have more peripheral obstruction while the site of obstruction may change in the same patient with time (Chan-Yeung, Abboud, Tsao & MacLean, 1976). β-Adrenoceptor agonists are widely used in these clinical conditions for their bronchodilator effects (Svedmyr, 1977; Paterson, Woolcock & Shenfield, 1979). In animal experiments, the response to these drugs may differ in central and peripheral airways (Lulich, Mitchell & Sparrow, 1976). Species differences are also apparent (Van der Heijden & Zaagsma, 1980).

From experiments in which the responses to sympathomimetic amines were compared in different tissues, Lands, Arnold, McAuliff, Luduena & Brown (1967) have suggested the existence of two subgroups of β-adrenoceptors. Comparison of the catecholamines noradrenaline (NA), adrenaline (Adr) and isoprenaline(Iso) shows that NA has the highest relative affinity for β_1-adrenoceptors while Adr has greater relative affinity for β_2-adrenoceptors. As a result, selective β-adrenoceptor antagonists show differential blocking potencies with the different agonists (Carlsson, Ablad, Brändström & Carlsson, 1972; O'Donnell & Wanstall, 1979). Different effector organs may also have populations of both β_1 and β_2-adrenoceptors, the relative proportions being variable for different organs in the same species (Lands *et al.*, 1967; O'Donnell & Wanstall, 1979), different substructures of the same organ (Lulich *et al.*, 1976, Carlsson, Dahlöf, Hedberg, Persson & Tangstrand, 1977), or the same organ in different species (Dickinson & Nahorski, 1981).

The present experiments were designed to characterize and compare β-adrenoceptors in lung strip preparations. The relative potencies of the three catecholamines NA, Adr and Iso were compared in human and porcine tissue and pA_2 values determined for the β-adrenoceptor antagonists atenolol (β_1-selective) (Barrett, 1977) and propranolol (non-selective), using Iso as agonist.

Methods

Longitudinal strips (approximately 25 × 4 × 2 mm) of parenchymal tissue devoid of pleural membrane were obtained from the peripheral margin of lobes of macroscopically normal specimens of human lung obtained 4–12 h post-mortem and of lung obtained from freshly killed pigs. Preparations were suspended under 500 mg tension in Krebs Henseleit solution aerated with 5% CO_2 in O_2 at 37°C. Isometric changes in tension were measured with a Grass force-displacement transducer (FTO3C) coupled to a preamplifier and Rikadenki pen recorder (Model 1328L). Preparations were left to equilibrate for 60–90 min with regular washing before any drug-induced effects were measured. Unless otherwise stated, tissues were exposed to phentolamine (15 μM)

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for the duration of experiments to inhibit α-adrenoceptors and catecholamine uptake processes (Foster, 1968; 1969; Goldie & Paterson, 1982).

Concentration-effect curves for catecholamines were constructed by cumulative administration and approximately 1 h was allowed to elapse between curves. In experiments to determine the relative relaxant potencies of Iso, Adr and NA, two consecutive concentration-effect curves to one of these amines chosen at random were constructed and a mean curve derived. Concentration-effect curves to the other two amines were then established in turn. Responses were expressed as a % of the maximum response ($E_{max} = 100\%$). Ratios of EC_{50} values for amines relative to Iso were taken as measures of relative amine potencies. In other experiments, concentration-effect curves to catecholamines were established in the absence and presence of competitive β-adrenoceptor blocking drugs and pA_2 values established by the method of Arunlakshana & Schild, (1959).

Drugs used were (−)-noradrenaline bitartrate, (−)-adrenaline bitartrate, (±)-isoprenaline hydrochloride (Sigma Chemical Co.); atenolol hydrochloride, propranolol hydrochloride (ICI); phentolamine mesylate (Ciba). All drug concentrations refer to the base. Drug solutions were freshly prepared in 0.9% w/v NaCl solution (saline). Solutions of catecholamines were stabilized with ascorbic acid (20 μg/ml).

Results

Both human and porcine lung parenchyma strip developed tone spontaneously. Iso caused complete relaxation while NA and Adr usually caused contraction. In some preparations of human lung strip, NA and Adr had little effect or caused small relaxations. In the presence of the α-adrenoceptor antagonist phentolamine (15 μM), levels of spontaneous tone were not markedly altered. However, Iso, Adr and NA each abolished tone in both human and porcine lung strips. Comparative EC_{50} values for these three catecholamines are given in Table 1.

Human lung strips

Iso was the most potent of the catecholamines in causing relaxation. Adr was approximately 4 times less potent than Iso. However, NA was more than 20 times less potent than Adr and 100 times less potent than Iso (Table 1). Thus, the relative potencies were Iso > Adr > NA (1 : 0.24 : 0.01).

Propranolol, a non-selective β-adrenoceptor antagonist was a potent inhibitor of Iso-induced relaxations of human lung strip. Schild plots indicated that the apparent pA_2 value (mean ± s.e.mean) was 7.84 ± 0.07; slope = 1.91 (Table 2). The pA_2 value for the selective β_1-adrenoceptor antagonist atenolol was 6.50 ± 0.03; slope = 1.10. The ratio of dissociation constants, K_B atenolol/K_B propranolol, indicates that propranolol was 22 times more potent than atenolol (Table 2).

Porcine lung strips

As in human lung strips, Iso was the most potent of the catecholamines causing relaxation in porcine lung parenchyma strips pretreated only with phentolamine (15 μM) (Table 1). The relative potencies for relaxation were Iso > Adr > NA (1 : 0.21 : 0.01). Thus Iso was 4–5 times more potent than Adr and 68 times more potent than NA. Iso, Adr and NA were respectively 2–3 times more potent in porcine than in human lung parenchyma strip.

Schild plots were constructed for the β-adrenoceptor antagonists using Iso as the agonist and pA_2 values determined. The pA_2 value (mean ± s.e.mean) for propranolol was 7.83 ± 0.07; slope = 0.93 and for the selective β_1-adrenoceptor antagonist atenolol was 5.35 ± 0.07; slope = 1.17

Table 1 Catecholamine EC_{50} values for relaxation of human and porcine lung parenchyma strip

	EC_{50} (M)		
	Isoprenaline	*Adrenaline*	*Noradrenaline*
Human	$2.58 \pm 0.47 \times 10^{-7}$	$1.07 \pm 0.22 \times 10^{-6}$	$2.25 \pm 0.61 \times 10^{-5}$
	(9)	(6)	(6)
Ratio	1 :	0.241 :	0.011
Pig	$0.97 \pm 0.23 \times 10^{-7}$	$4.53 \pm 0.63 \times 10^{-7}$	$6.60 \pm 0.86 \times 10^{-6}$
	(10)	(4)	(23)
Ratio	1 :	0.214 :	0.015

EC_{50} values are given as mean ± s.e.mean.
Numbers in parentheses indicate number of experiments. All experiments were carried out in the presence of phentolamine (15 μM).

Table 2 pA_2 values for propranolol and atenolol against isoprenaline-induced relaxations in human and porcine lung parenchyma strip

	Human	*Pig*
Propranolol	7.84 ± 0.07	7.83 ± 0.07
Atenolol	6.50 ± 0.03	5.35 ± 0.07
$\frac{K_B \text{ Atenolol}^*}{K_B \text{ Propranolol}}$	22	302

pA_2 values are given as mean ± s.e.mean.
*Ratio of dissociation constants (K_B) for β-adrenoceptor antagonists.
All experiments were carried out in the presence of phentolamine (15 μM).

(Table 2). The ratio of dissociation constants (K_B atenolol/K_B propranolol) indicates that propranolol was 302 times more potent than atenolol as an inhibitor of Iso relaxations in porcine parenchyma strip (Table 2). A comparison of K_B values for atenolol shows that this drug was approximately 14 times more potent in human than in porcine lung strips.

Discussion

Both NA and Adr-induced contractions of human and porcine lung parenchyma strip were mediated via α-adrenoceptors since these catecholamines caused relaxations of lung strips in the presence of phentolamine. Although NA-induced contractions of lung strips may have been due to response of alveolar contractile elements (Black, Turner & Shaw, 1981), it seems likely that contraction of peripheral blood vessels following stimulation of vascular α-adrenoceptors, contributed to these responses (Mirbahar & Eyre, 1980).

The relative relaxant potencies of the three catecholamines tested in the present study were similar (Iso > Adr > NA) in both human and porcine lung parenchyma strips (Table 1). NA and Adr are relatively selective agonists at β_1 and β_2-adrenoceptors respectively (Lands *et al.*, 1967). Since Adr was approximately 20 times and 15 times more potent than NA in human and porcine lung strips respectively, this suggests that β_2-adrenoceptors predominated in both tissues. Guinea-pig atria and trachea are known to contain relatively pure populations of β_1 and β_2-adrenoceptors respectively (O'Donnell, 1972; O'Donnell & Wanstall, 1979). Unpublished results by one of us (Wale) for pA_2 values for propranolol and atenolol against Iso were 8.30 and 7.27 respectively, in guinea-pig atria and 7.66 and 5.21 in guinea-pig trachea. Similar results have been reported by Harms (1976) in guinea-pig and human tissues. In the present study, the pA_2 value for atenolol against Iso in porcine parenchyma strip was only 5.35, supporting the suggestion that a relatively pure population of β_2-adrenoceptors exist in this tissue.

The β-adrenoceptor sub-type was less clearly defined in human lung parenchyma where atenolol was 14 times more potent than in porcine lung strip. The pA_2 value for atenolol against Iso was 6.50. This value is intermediate between that for guinea-pig atria (β_1) and trachea (β_2) indicating that human lung parenchyma has a mixed population of β_1 and β_2-adrenoceptors. Apparently homogeneous, as well as mixed populations of β_1 and β_2-adrenoceptors have been revealed in central airways of several species. In the guinea-pig, β_2-adrenoceptors predominate in the trachea (O'Donnell & Wanstall, 1979), and in the lung parenchyma strip (Siegl, Rossi & Orzechowski, 1979) while in the cat, there is a change in β-adrenoceptor sub-type from β_1 in central airways, to β_2 in lung parenchyma (Lulich *et al.*, 1976). The rabbit (Bristow, Sherrod & Green, 1970; Toda, Hayashi, Hatano, Okunishi & Miyazaki, 1978) and rat trachea (Henry, Lulich & Paterson, 1982) have mixed populations of β_1 and β_2-adrenoceptors.

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