Salbutamol: agonistic and antagonistic activity at β-adrenoceptor sites

Salbutamol is a β-adrenoceptor agonist which displays some specificity in its ability to stimulate β_2- as opposed to β_1-adrenoceptors (see Brittain, Jack & Ritchie, 1970), Farmer, Levy & Marshall, 1970, for references).

In isolated atrial preparations from the guinea-pig and rat, the maximal inotropic and chronotropic activity produced by salbutamol is less than that obtained with isoprenaline, and on this basis it has been suggested that salbutamol may be classed as a partial agonist (Farmer, Kennedy & others, 1970; Brittain, 1972; O'Donnell, 1972). However, in guinea-pig isolated tracheal preparations and in preparations from human bronchial muscle, salbutamol and isoprenaline produce a similar maximal relaxant effect (Farmer & others, 1970; Svedmyr & Thiringer, 1971; O'Donnell, 1972).

In the present experiments we have quantified the β-agonistic and antagonistic actions of salbutamol in isolated atrial preparations from the guinea-pig. In addition we have investigated agonistic activity and the possibility that salbutamol may produce β-antagonistic effects in guinea-pig tracheal preparations.

Isolated atrial and tracheal preparations from guinea-pigs were set up in Krebs-Henseleit solution maintained at 37° bubbled with 5% CO_2 in oxygen and to which ascorbic acid (20 μg ml^{-1}) was added.

Inotropic activity was assessed in left atrial preparations driven at 5 Hz and chronotropic activity in spontaneously beating whole atria. Records of rate and force were displayed on an ink-writing polygraph. Tone was induced in tracheal preparations by the addition of carbachol (0·15 μg ml^{-1}) to the bath and relaxation was recorded on a smoked drum using a lightly weighted frontal writing lever.

In all preparations, control responses to (−)-isoprenaline were first obtained, and thereafter the agonistic effects of salbutamol were determined. Both drugs were administered cumulatively until maximal responses had been attained.

As reported by previous authors, the intrinsic activities of salbutamol and (−)-isoprenaline are similar in tracheal preparations, while in atrial preparations salbutamol has a lower intrinsic activity. Table 1 shows pD_2 values and values of intrinsic activity for (−)-isoprenaline and salbutamol.

Table 1. *Agonistic and antagonistic effects of (−)-isoprenaline and salbutamol in isolated atrial and tracheal preparations from guinea-pigs expressed as* pD_2 *values, intrinsic activity* (α) *and* pA_2 *values. The slope of the relation between log(dose ratio − 1) and log(molar salbutamol concentration) is also indicated (slope). Each value represents the mean ± s.e. from 4 to 6 experiments.*

	Atria (inotropic)		Atria (chronotropic)			
	pD_2	α	pD_2	α	pA_2	Slope
(−)-Isoprenaline	8·19±0·18	1·00	8·53±0·11	1·00		
Salbutamol	5·96±0·12	0·35±0·04	5·90±0·15	0·51±0·06	5·15±0·05	0·94±0·09

	Trachea (relaxation)			
	pD_2	α	pA_2	Slope
(−)-Isoprenaline	7·45±0·09	1·00		
Salbutamol	6·53±0·08	0·98±0·01	5·64±0·10	1·22±0·34

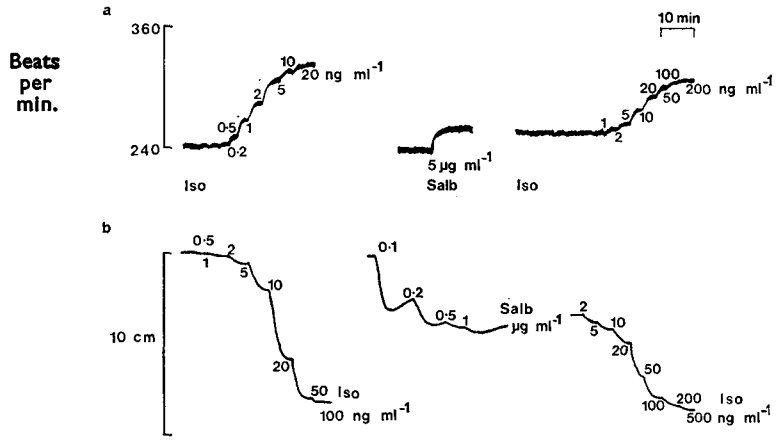

FIG.1a. Spontaneously beating atrial preparation showing rise in heart rate in response to the cumulative administration of (−)-isoprenaline (Iso) before (left) and in the presence (right) of salbutamol (Salb, 5 μg ml^{-1}, centre panel).

b. Relaxations of carbachol stimulated tracheal preparation from the guinea-pig in response to cumulative administration of (−)-isoprenaline before (left) and in the presence (right) of a total cumulative concentration of 1 μg ml^{-1} of salbutamol (centre panel).

Antagonistic activity of salbutamol was evaluated in whole atrial preparations, the effect being assessed in terms of shifts in cumulative concentration-effect curves to the positive chronotropic action of (−)-isoprenaline. Typical traces are seen in Fig. 1a. On the left is one of a series of control cumulative concentration-effect curves to (−)-isoprenaline. The centre panel shows the initial effect produced by salbutamol (5μg ml^{-1}) added to the bath and the right hand panel shows a further curve to (−)-isoprenaline superimposed after the salbutamol had been left in contact for 45 min. The chronotropic effects produced by salbutamol varied; the stimulant action was either maintained (Fig. 1a) or declined towards the resting atrial rate during the contact period.

Where antagonism was assessed, two concentrations of salbutamol (1 and 5μg ml^{-1}) were used. The superimposed (−)-isoprenaline-induced concentration-effect curves were moved to the right in a parallel fashion. At 5μg ml^{-1} of salbutamol, there was a small depression (mean $\pm$ s.e., 4·3 $\pm$ 1·6%) of the maximal response to (−)-isoprenaline. Concentration effect curves to (−)-isoprenaline were plotted, responses being expressed as a percentage of the maximal response to (−)-isoprenaline under control conditions. In each curve a point was taken from a position midway between the residual chronotropic response to salbutamol and the maximal response to (−)-isoprenaline. This point was extrapolated back to the control (−)-isoprenaline curve for the calculation of a dose-ratio. The slopes of the relations between log(dose-ratio−1) and log (molar salbutamol concentration), calculated as described by Arunlakshana & Schild (1959), have values close to unity (Table 1) suggesting that a competitive mechanism is involved in the antagonism produced.

In tracheal preparations cumulative concentration-effect curves to salbutamol show a similar maximal response to that obtained with (−)-isoprenaline (Table 1) when each dose is added at the peak response to the previous dose in the series. However, if a prolonged contact time was allowed between doses, the initial relaxation produced by the compound was followed by a return of tone in the preparation. With subsequent larger concentrations of salbutamol, relaxant effects were minimized and a concentration-effect curve to (−)-isoprenaline could be superimposed (Fig. 1b).

Antagonism was assessed from salbutamol-induced shifts in (−)-isoprenaline curves in a manner analogous to that described previously for atria. Final "blocking" concentrations of salbutamol were left in contact with the tissue for 45 min. Values for pA_2 and the slope of the relation log(dose-ratio−1) *vs* log(molar salbutamol concentration) are shown in Table 1. The slope is close to unity indicating the involvement of competitive antagonism.

In atrial and tracheal preparations cumulative concentration-effect curves to salbutamol are typical of those produced by partial agonists and full agonists respectively. Our results indicate that antagonistic actions of a competitive type are produced by salbutamol in both preparations.

The finding that antagonism of β-adrenoceptors can often be produced in tracheal preparations means that it is unlikely that the relatively selective β_2-adrenoceptor stimulant activity of salbutamol might be related to the production of self-antagonistic effects at β_1 (atrial) adrenoceptor sites.

In tracheal preparations the pD_2 values are greater than those found in atria. A similar situation also exists with respect to the pA_2 values. In each preparation pD_2 and pA_2 values differ only to a small extent; in fact, concentrations approaching those required to produce maximal responses in both trachea and atria correspond to the respective pA_2 concentrations.

There is a close structural similarity between drugs which are classified as β-receptor agonists and antagonists. The ring substituents appear to be the most important determinant in this regard. Many compounds that are classified as β-antagonists possess β-agonistic activity (Barrett & Carter, 1970; Nayler, 1972). In view of the present results it is conceivable that some agents that are classified as β-agonists may also possess antagonistic actions at β-adrenoceptors. The latter activity may play a role in the development of the tolerance and cross-tolerance displayed with such compounds in various pharmacological preparations (Atkinson & Rand, 1968; Conolly, Davies & others, 1971).

This work was supported by a grant from the National Heart Foundation of Australia.

Department of Pharmacology,
University of Melbourne,
Parkville, Vic. 3052. Australia.

C. RAPER
E. MALTA

April 26, 1973

REFERENCES

ARUNLAKSHANA, O. & SCHILD, H. O. (1959). *Br. J. Pharmac. Chemother.*, **14**, 48–58.
ATKINSON, J. M. & RAND, M. J. (1968). *J. Pharm. Pharmac.*, **20**, 916–922.
BARRETT, A. M. & CARTER, J. (1970). *Br. J. Pharmac.*, **40**, 373–381.
BRITTAIN, R. T. (1972). *Proc. R. Soc. Med.*, **65**, 759–761.
BRITTAIN, R. T., JACK, D. & RITCHIE, A. C. (1970). *Adv. Drug. Res.*, **5**, 197–253.
CONOLLY, M. E., DAVIES, D. S., DOLLERY, C. T. & GEORGE, C. F. (1971). *Br. J. Pharmac.*, **43**, 389–402.
FARMER, J. B. KENNEDY, I., LEVY, G. P. & MARSHALL, R. J. (1970). *J. Pharm. Pharmac.*, **22**, 61–63.
FARMER, J. B., LEVY, G. P. & MARSHALL, R. J. (1970). *Ibid.*, **22**, 945–947.
NAYLER, W. G. (1972). *Br. J. Pharmac.*, **45**, 382–384.
O'DONNELL, S. R. (1972). *Eur. J. Pharmac.*, **19**, 371–379.
SVEDMYR, N. & THIRINGER, G. (1971). *Postgrad. Med. J.*, **47** (Suppl.), 44–46.