acid phosphatase was measured in the supernatant obtained from centrifuging the contents of the dialysis bag at 20 000 *g* for 20 min at 4° (Symons & others, 1969).

The subcellular fraction concentrated the steroids from the surrounding medium and the amount of steroid taken up was proportional to the steroid concentration of the medium (Fig. 1). Also, it seems that our previous assumption was correct in that a high concentration of steroid within the lysosome led to a loss of stability of its membrane and higher levels of free acid phosphatase. At maximum values the amount of steroid taken up by the fraction was equivalent to 0·1 mg/g fresh liver.

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A comparison of the β-adrenoceptor stimulant properties of salbutamol, orciprenaline and soterenol with those of isoprenaline

The discovery of different structure-activity relations for the actions of catechol-ethanolamines at β-adrenoceptors in different tissues led Lands and co-workers (Lands, Arnold & others, 1967; Lands, Luduena & Buzzo, 1967) to suggest the existence of two types of β-receptors, namely β_1 and β_2. In the present work we have examined the activities of four β-adrenoceptor agonists on tissues thought to contain β_1 or β_2 adrenoceptors. The drugs used were isoprenaline, orciprenaline, salbutamol and soterenol.

Brief details of experimental methods are given in Table 1. Full dose-response curves were obtained for isoprenaline and for one of the other drugs on each preparation. The activities of the drugs were compared at 50% of the maximum effect or, if this was not possible, at suitable equi-effective dose-levels.

The dose-ratios for the β-adrenoceptor agonists compared with isoprenaline are given in Table 1. As shown in the Table the activities of the four drugs on guinea-pig ileum and colon have not been included because it was found that a major proportion of the response to isoprenaline is mediated through stimulation of α-receptors (Farmer & Levy, 1970b).

Salbutamol was selective in its actions, being much more active at β–2 than at β–1 receptors. The difference in the mean dose-ratios for activity in the β–1 and β–2 groups of adrenoceptors was highly significant ($P = 0{\cdot}002$). Some separation of effects in the β–1 group is indicated in the relatively greater action of salbutamol on rate than on force in isolated rat atria. The mean of the activities for orciprenaline at β–2 receptors was seven times greater than at β–1 receptors and this difference was just significant ($P = 0{\cdot}015$). Soterenol had high activity at β–2 adrenoceptors

Table 1. *Dose ratios for salbutamol: isoprenaline, orciprenaline: isoprenaline and soterenol: isoprenaline at β-1 and β-2 adrenoceptor sites.* (Each value is the mean of at least five determinations).

Preparation	Ref.	Receptor class	Salbutamol	Orciprenaline	Soterenol
Guinea-pig					
Right atria—rate	(a)	β-1	500	125	130
Left atria—force	(b)	β-1	2500	63	>10 000
Rat					
Right atria—rate	(a)	β-1	54	24	>10 000
Left atria—force	(b)	β-1	8000	38	>10 000
Guinea-pig					
Ileum	(c)	Excluded due to α-receptor involvement in response to β-stimulant drugs (Farmer & Levy, 1970)			
Colon	(d)				
Rabbit					
Intestine (+ 1 μg/ml phentolamine)	(e)	β-1	800	500	30
Guinea-pig					
Trachea	(f)	β-2	6	14	6
Vas deferens	(g)	β-2	1	20	2
Rat					
Diaphragm	(h)	β-2	5	40	3
Uterus	(i)	β-2	3	4	1
Dog					
Hind limb blood flow	(j)	β-2	5	33	3
Chick					
Colon	(d)	β-2	9	10	5

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and except for high positive chronotropic activity on guinea-pig isolated right atria, low activity at β-1 adrenoceptors. The significance of this exception is not clear.

Lands and others have already shown that structural modifications of the ethanolamine side chain of cateholamines can produce selectivity of action. The present work shows that replacement of the 3-hydroxy group by a hydroxymethyl or methanesulphonamide-group produces an even greater selectivity of action. On the other hand, replacement of a catechol by a resorcinol function, as in orciprenaline, produces much less separation of effects.

The results, in general, support the concept of two main types of β-adrenoceptor. The question whether the receptors within the groups are identical or differ slightly from one another remains to be finally answered. Evidence obtained by the use of β-adrenoceptor blocking agents indicates that a simple two-receptor hypothesis would be inadequate (Farmer & Levy, 1970a). However, the concept that different tissues have different β-adrenoceptor populations is substantiated by these results.

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Amphetamine toxicity and endogenous noradrenaline concentrations in isolated and aggregated mice

There is indirect evidence for the participation of noradrenaline in the mechanism by which aggregation augments amphetamine toxicity in mice. Experiments with amphetamine toxicity in mice pretreated with a variety of pharmacological agents known to modify the metabolism, storage, or action of noradrenaline indicate that noradrenaline released from endogenous stores plays a role in the aggregation effect (Sethy & Sheth, 1968). Although reduced tissue concentrations of noradrenaline in mice treated with amphetamine are consistent with this view (Moore, 1963, 1964; Beauvallet & Solier, 1964; Lal & Chessick, 1964; Menon & Dandiya, 1967), the relation between the enhanced toxicity of amphetamine in aggregated mice and noradrenaline depletion is not clear. We aimed to examine this relation by determining whether amphetamine-induced lethality, in either isolated or aggregated mice, is correlated with the degree of noradrenaline depletion in tissues. This approach differs from that used earlier (Moore, 1963, 1964; Lal & Chessick, 1964) in one important aspect. A distinction has been made between the degree of noradrenaline depletion in mice that died as a consequence of amphetamine treatment and in mice that survived the treatment.

Novice, male, albino mice of a random-bred Swiss strain (Maxfield; Cincinnati, Ohio), 9 to 12 weeks, 25 to 35 g, were housed 15 per cage (45 × 24 × 12 cm) for not less than 30 days; Purina laboratory chow and water were freely available. After an intraperitoneal injection of saline or (+)-amphetamine sulphate (30 or 100 mg/kg) in aqueous solution (1 ml/100 g), mice were either isolated or aggregated (3 per cage) in metal cages (7 × 7 × 7·5 cm) with a wire mesh side for observation. The rationale for using these doses of (+)-amphetamine has been previously discussed (George & Wolf, 1966, 1967). Ambient temperature was 24 ± 1°. After 3 h, surviving animals (survivors) were killed by cervical dislocation; their brains and hearts were removed and frozen in liquid nitrogen within 1 min after death. Mice that died within 3 h (non-survivors) had their tissues removed and frozen immediately after death. The degree of aggregation was maintained constant by replacing non-survivors with untreated mice. Noradrenaline in pooled samples of 4 hearts or 3 whole brains was assayed fluorometrically by the trihydroxyindole method of Anton & Sayre (1962).

Aggregation itself did not deplete brain or heart stores of noradrenaline (Table 1). The levels were not significantly different ($P>0{\cdot}05$) in saline-treated isolated versus aggregated mice. Furthermore, aggregation did not enhance the noradrenaline-depleting effect of (+)-amphetamine. Brain and heart noradrenaline levels in mice treated with 30 or 100 mg/kg of (+)-amphetamine were not significantly different in isolated versus aggregated animals. This finding differs from that of Lal & Chessick (1964) who found lower brain levels of noradrenaline in aggregated than in isolated mice 30 min after (+)-amphetamine (25 mg/kg). It is possible that high doses of (+)-amphetamine or long treatment times, such as we used, obscured any influence aggregation might have had on the noradrenaline-depleting effect of (+)-amphetamine. Moore (1963), too, reported that aggregation in mice enhanced noradrenaline deple-