"Indirect" sympathomimetic effects of L(+)-isomers and desoxy derivatives

SIR,—Molecules which are structurally similar provide a valuable tool for the analysis of drug action. In 1933, Easson & Stedman postulated that for D(−)-isomers of sympathomimetic amines there is a three point interaction with receptors: the phenyl group, the β-hydroxyl group and the amino-group. For L(+)-isomers only a two point interaction is suggested because the β-hydroxyl group is orientated differently and the molecules react essentially as if the β-hydroxyl group were absent. Thus L(+)-isomers are expected to act like desoxy derivatives (Beckett, 1959). This hypothesis has been tested (Patil, La Pidus & Tye, 1967a; Patil, La Pidus, Campbell & Tye, 1967b) and the results indicate that it holds true for indirectly-acting amines in catecholamine-depleted tissue, but not in normal tissues, in which the activity of L(+)-isomers and their corresponding desoxy derivatives appears to be related to differing affinities for the catecholamine uptake sites (Burgen & Iversen, 1965). We now concern ourselves with the effects of various experimental conditions on the relative activities of L(+)-isomers and desoxy derivatives.

The vas deferens of the rat was isolated and suspended in a 10 ml tissue bath containing Tyrode solution at 37·5°, and drug-induced contractions recorded on a smoked drum. Four different procedures were used.

(i) The normal tissue was washed four times, and 15 min after the last wash, drug-induced contractions were recorded.

(ii) The conditions were the same as in (i), but tissue was exposed to 10^{-4} or 3×10^{-4}M noradrenaline, thoroughly washed, and drug-induced contractions recorded. These results were obtained from our previous experiments (Patil & others, 1967a).

(iii) Drug-induced contractions were recorded on reserpine pretreated tissue (5 mg/kg, i.p., 16–24 hr).

(iv) Catecholamine-depleted tissue obtained as above was exposed to 10^{-4}M noradrenaline for 5 min, then washed thoroughly and drug-induced effects recorded. The final bath concentrations of desoxy derivatives and L(+)-isomers were 10^{-4}M and 3×10^{-4}M, respectively. At these concentrations the agents produce maximal effects on this tissue (Patil & others, 1967a). Only

TABLE 1. RESPONSE OF VAS DEFERENS OF RAT TO SYMPATHOMIMETIC AMINES

Drugs	Mean contraction of vas deferens (mm) with s.e. of the mean							
	Normal	N[1]	Normal[2] tissue exposed to noradrenaline	N[1]	Reserpine-pretreated	N[1]	Reserpine-pretreated tissue exposed to noradrenaline	N[1]
α-Methyldopamine, 10^{-4}M (deoxy Cobefrin)	101±8	10	125±9	8	27±5	10	93±10	10
L(+)-Cobefrin, 3×10^{-4}M	35±2	10	52±4	8	14±7	10	61±9	10
Ratio, desoxy/L(+)-isomer	2·8		2·4		1·9		1·51	
P value	<0·001		<0·001		>0·05		<0·05	
m-Tyramine, 10^{-4}M (desoxy norphenylephrine)	116±6	12	136±9	8	17±3	10	99±17	10
L(+)-Phenylephrine, 3×10^{-4}M	73±6	12	86±10	8	28±5	10	68±9	10
Ratio, desoxy/L(+)-isomer	1·5		1·5		0·6		1·4	
P value	<0·001		<0·01		>0·05		>0·05	
Tyramine, 10^{-4}M (desoxy octopamine)	121±11	12	131±5	8	14±5	8	96±16	8
L(+)-Octopamine, 3×10^{-4}M	89±9	12	112±10	8	20±11	9	99±15	9
Ratio, desoxy/L(+)-isomer	1·35		1·16		0·7		0·9	
P value	<0·05		>0·05		>0·05		>0·05	

[1] Number of observations.
[2] Data from Patil & others (1967a).

one drug was tested on any given tissue. Eight to twelve observations were made of each procedure. The significance between the two means was calculated by Student's *t*-test.

In normal, or normal tissue exposed to noradrenaline, the effects of desoxy derivatives were far greater than those of L(+)-isomers. The activity ratio (desoxy derivatives/L(+)-isomer) both in the normal tissue and tissue exposed to noradrenaline remained the same. In the reserpine-pretreated tissue all L(+)-isomers and their desoxy derivatives produced little effect, indicating that these molecules have little or no "intrinsic" activity at sites of direct action, and that their action is possibly due to the release of catecholamine from storage sites. The activity ratios appear to be decreased, possibly due to a large "indirect" component of action in the desoxy derivative compared with that of the L(+)-isomer. In reserpine-pretreated tissue previously exposed to noradrenaline the effectiveness of all compounds was regained by refilling the catecholamine stores. The results are summarized in Table 1.

Kaufman & Friedman (1965) suggested that the action of desoxy derivatives of certain sympathomimetic amines could be attributed to their β-hydroxylated derivatives, which could be easily formed *in vivo* by enzyme dopamine β-hydroxylase. If this were so, it would explain why desoxy derivatives appear more active than L(+)-isomers in normal tissue. However, these possibilities are unlikely because of the following reasons. β-Hydroxylation of desoxy derivatives is a relatively slow process, yet with the present experimental design it took only 30 to 60 sec to obtain the maximal effects of desoxy derivatives. Reserpine abolishes the effects of desoxy derivatives but not of D(−)-isomers (Patil & others, 1967b), which indicates that the action of desoxy derivatives is due to the parent molecule itself rather than to its corresponding D(−)-isomer. In other words, small amounts of D(−)-isomers may be formed but they probably do not contribute significantly to the pharmacological activities of desoxy derivatives in the described conditions.

Our results further substantiate our previous findings that the Easson-Stedman hypothesis (1933) probably holds true at the sites of direct effect, but not when the overall activity of the amines is measured.

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