PHARMACOLOGICAL AND BIOCHEMICAL EFFECTS OF SOME RESERPINE DERIVATIVES

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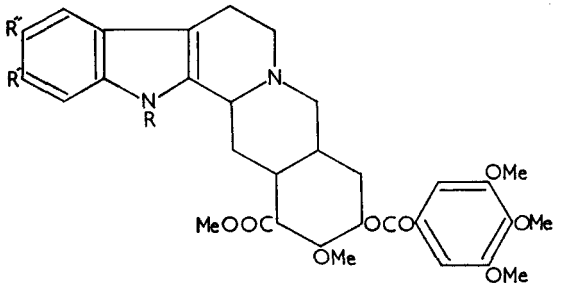
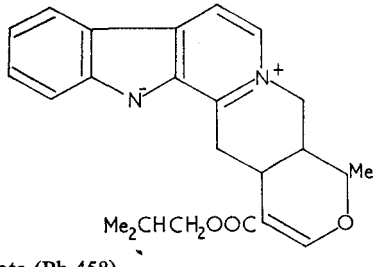
A relation exists between the hypotensive properties of three reserpine derivatives (β-diethylaminoethyl reserpine, isobutylserpentinate, 10-methoxydeserpidine) and their ability to release cardiac catecholamines. These derivatives do not show sedative properties and do not release 5-hydroxytryptamine or catecholamines from the brain.

AT the same time as the discovery of carbethoxy-siringoylmethyl reserpate, a reserpine derivative (Plummer, Barrett, Maxwell, Finocchio, Lucas and Earl, 1959) with specific hypotensive effect (Darvill, 1958; Garattini, Mortari, Valsecchi, Valzelli, 1959; Hughes Orlans, Finger and Brodie, 1960; Plummer and others, 1959) was reported, some other compounds,



I. Reserpine, R = H; R′ = OMe; R″ = H.
II. 10-Methoxydeserpidine, R = H; R′ = H; R″ = OMe.
III. DL152, R = $CH_2CH_2N(Et)_2$; R′ = OMe; R″ = H.



IV. Isobutylserpentinate (Ph.458).

with similar properties, were described. Among them the most promising appeared to be β-diethylaminoethyl reserpine (DL 152) (Buzas and Régnier, 1960); isobutyl serpentinate (IBS) (van Proosdij Hartsema, Akkerman and de Jongh, 1959) and 10-methoxydeserpidine (10 MD) (Merlene and Gérard, 1960; Peterfalvi and Jequier, 1960; Velluz, Peterfalvi and Jequier, 1958) (see formulae) since their effect on blood pressure is not

accompanied by depressant activity in the central nervous system. The aim of this investigation was to compare the effect of reserpine and these three new compounds in a number of pharmacological tests and on the 5-hydroxytryptamine and catecholamine content of various organs. It was thought that these experiments might contribute to the elucidation of the mechanism of action of reserpine, particularly to the relation between its pharmacological effects and the release of biogenic amines.

Material and Methods

Sprague-Dawley rats fed a balanced diet have been used throughout. Compounds were dissolved in a mixture of benzyl alcohol (2 g.), propyleneglycol "300" (10 g.), citric acid (0·25 g.) and distilled water up to 100 ml. and injected intravenously or intraperitoneally. Control rats received only the solvent.

For blood pressure experiments, animals were anaesthetised with ethylcarbamate (1 g./kg., s.c.). The carotid artery was isolated and cannulated for recording pressure by means of an electromanometer connected to a No. 150 Sanborn recorder. Iproniazid (100 mg./kg.) was given intraperitoneally 6 hr. before the intravenous injection of reserpine or its derivatives. Tyramine and noradrenaline were injected intravenously at the concentrations shown in Table I.

5-Hydroxytryptamine (5-HT) in extracts of the spleen and intestine was assayed by the method of Bogdanski, Pletscher, Brodie and Udenfriend (1956); noradrenaline assayed in extracts of heart by the method of Shore and Olin (1958), and 5-HT and noradrenaline assayed in extracts of brain by the method of Shore (1959) using a spectrofluorimetric technique.

Isobutylserpentinate was obtained from Prof. D. K. de Jongh (Pharmaco-Therapeutisch Instituut Nedchem, Amsterdam), β-diethylaminoethylreserpine (DL 152) from Dr. A. Buzas (Société Dautreville et Lebas, Paris), 10-methoxydeserpidine from Dr. P. Poirier (Roussel-Uclaf, Paris) and reserpine (Serpasil) from Dr. V. Ghetti (Ciba, S.p.A., Milano).

Results

Reserpine Derivatives on Blood Pressure

Table I summarises the effects of reserpine and its derivatives on the blood pressure of anaesthetised rats. Reserpine is more hypotensive than the other compounds; for example, 4 hr. after intraperitoneal administration, 1·25 mg./kg. of reserpine produces a fall in blood pressure which is comparable with that produced by 2·5 mg./kg. of DL 152 or 5 mg./kg. of IBS or 10 MD.

The doses producing hypotensive effects also prevent the tyramine hypertension. Furthermore, reserpine and its derivatives slightly increase the pressor effect of noradrenaline. In animals pretreated with a monoamine oxidase inhibitor (for example, iproniazid), reserpine elicits a hypertension which may be related to a release of peripheral catecholamines (Garrattini, Fresia, Mortari and Palma, 1960). Unlike reserpine, IBS does not induce hypertension in iproniazid-treated rats, while DL 152 and 10 DM are only slightly active. The increased effect of tyramine in

TABLE I

EFFECT OF RESERPINE AND THREE OF ITS DERIVATIVES ON BLOOD PRESSURE OF RATS IN VARIOUS EXPERIMENTAL CONDITIONS

Compound	Dose (mg./kg.)	Blood pressure after 4 hr.		Pressor effect of compounds in iproniazid-treated rats	Pressor effect of tyramine injected 4 hr. after the compounds				Pressor effect of noradrenaline (10 μg./kg.) injected 4 hr. after the compounds
					Intact rats			Iproniazid-treated rats	
		Max.	Min.		250 μg./kg.	500 μg./kg.	1000 μg./kg.	250 μg./kg.	
Solvent ..	—	155 ± 15	90 ± 13 (10)	0 (10)	+20 ± 5 (10)	+40 (5)	+70 (5)	+70 ± 12 (10)	+50 ± 4 (10)
Reserpine ..	0·63	—	—		±20 ± 5 (5)				
	1·25	120 ± 2	90 ± 9 (5)		0 (5)				
	2·5	95 ± 2	55 ± 6 (10)	+60 ± 15 (10)	+5 ± 4 (10)	0 (5)	0 (5)	+45 ± 14 (10)	+70 ± 6 (5)
IBS ..	1·25	145 ± 19	108 ± 10 (5)		+18 ± 3 (5)				
	2·5	150 ± 9	105 ± 9 (10)	0 (5)					
	5	135 ± 7	87 ± 9 (10)	0 (5)				+60 ± 5 (5)	
	10	127 ± 4	90 ± 9 (10)	0 (10)	*(10)	+30 (5)	+60 (5)	+70 ± 8 (5)	+60 ± 6 (5)
DL 152 ..	1·25	145 ± 9	103 ± 7 (5)		+20 ± 3 (5)				
	2·5	115 ± 7	90 ± 6 (10)	+18 ± 3 (5)					
	5	100 ± 5	60 ± 5 (10)	+17 ± 3 (5)	+2 ± 2 (5)			+65 ± 5 (5)	
	10	100 ± 3	58 ± 4 (10)	+17 ± 2 (5)	+1 ± 2 (5)	+25 (5)	+60 (5)	+70 ± 8 (5)	+75 ± 9 (5)
10 MD ..	1·25	155 ± 12	102 ± 11 (5)		+15 ± 5 (5)				
	2·5	135 ± 11	90 ± 10 (10)	+15 ± 5 (5)	+5 ± 3 (5)				
	5	130 ± 11	86 ± 10 (10)	+20 ± 4 (5)	0 (5)			+60 ± 5 (5)	
	10	115 ± 10	75 ± 9 (10)	+18 ± 4 (5)	0 (5)			+60 ± 5 (5)	+65 ± 8 (5)

* In 4 rats pressor effect was antagonised; in 6 rats pressor effect was comparable to controls.

iproniazid-treated animals is partially prevented by reserpine, but not by the other derivatives.

Other Pharmacological Effects of Reserpine Derivatives

Table II shows the effect of reserpine and its analogues on body temperature, sedation, ptosis, barbiturate potentiation and intestinal function. Except for reserpine, the other compounds in doses up to 10 mg./kg. are almost inactive in these tests.

Reserpine Derivatives and Tissue Amines

Table III summarises biochemical findings concerning the effect of reserpine and its derivatives on tissue amines. Both intestine and spleen 5-HT are decreased by reserpine. However, only spleen 5-HT is decreased by a high concentration (20 mg./kg.) of DL 152 or IBS. The values for intestine and spleen after 10 MD treatment are higher than the control values, due possibly to the fact that 10 MD interferes with the fluorescence

TABLE II

PHARMACOLOGICAL ACTIONS OF RESERPINE AND THREE OF ITS DERIVATIVES IN RATS. EACH VALUE REPRESENTS THE MEAN OF AT LEAST 6 DETERMINATIONS

Compound	Intra-peritoneal dose (mg./kg.)	Body temperature (°C ± S.E.)		Sedation	Ptosis	Diarrhoea	Sleeping time pentobarbitone (20 mg/kg) i.p., given 4 hr. after the compounds
		Controls	After 4 hr.				
Solvent	—	36·9 ± 0·2	36·8 ± 0·2	—	—	—	46 ± 10
Reserpine	1·25	37·1 ± 0·2	35·3 ± 0·2	±	±	±	69 ± 9
	2·5	37·1 ± 0·2	34·0 ± 0·2	+	+	+	173 ± 8
IBS ..	10	37·0 ± 0·2	36·1 ± 0·4	—	—	—	53 ± 6
DL 152 ..	10	37·1 ± 0·1	36·6 ± 0·2	—	—	—	79 ± 4
10 MD ..	10	36·9 ± 0·1	36·7 ± 0·2	—	—	—	71 ± 5

of the 5-HT. Brain 5-HT and noradrenaline are not released by these three reserpine derivatives even when analysis is made 4 or 12 hr. after intraperitoneal doses of 10 and 20 mg./kg.

DL 152, 10 DM and IBS, in decreasing order, deplete heart noradrenaline, but this depletion is less marked than that obtained with reserpine.

DISCUSSION

It is interesting to compare the pharmacological effects exerted by the three reserpine derivatives with their action in releasing tissue amines. DL 152, IBS and 10 MD, which are devoid of sedative effects, do not decrease brain 5-HT and noradrenaline even in doses of 10 mg./kg., while reserpine is active at a dose 8 times lower. On the other hand, the three derivatives, like reserpine itself, possess a hypotensive action. Thus, the hypotensive action may occur without the release of brain amines.

The three reserpine derivatives also fail to decrease the peripheral stores of 5-HT and this finding may be linked with the fact that 10 MD, unlike reserpine, does not increase the urinary excretion of 5-hydroxyindoleacetic acid (Burn and Rand, 1958).

A parallelism exists between the hypotensive properties of these compounds and their activity to decrease cardiac catecholamines. Reserpine

TABLE III

EFFECT OF RESERPINE AND THREE OF ITS DERIVATIVES ON TISSUE 5-HT AND NORADRENALINE IN RATS

Compound	Intraperitoneal dose (mg./kg.)	Time after administration (hr.)	5-HT μg./g. ± S.E.			Noradrenaline μg./g. ± S.E.	
			Intestine	Spleen	Brain	Brain	Heart
Solvent	—	4	2·67 ± 0·52 (8)	2·34 ± 0·26 (8)	0·35 ± 0·06 (16)	0·24 ± 0·02 (8)	0·57 ± 0·07 (16)
Reserpine	1·25	4			0·27 ± 0·02 (4)	0·05 ± 0·01 (12)	0·17 ± 0·01 (8)
	2·5	4	2·95 ± 0·24 (8)	1·09 ± 0·24 (8)	0·17 ± 0·05 (12)	0·03 ± 0·01 (12)	0·12 ± 0·01 (8)
	2·5	12	2·16 ± 0·15 (4)	1·04 ± 0·20 (4)	0·21 ± 0·02 (4)	0·07 ± 0·01 (4)	0·12 ± 0·01 (8)
	5·0	4	2·16 ± 0·25 (4)	0·60 ± 0·06 (4)	0·10 ± 0·01 (4)		
IBS	2·5	4		2·15 ± 0·16 (4)	0·32 ± 0·01 (4)	0·24 ± 0·01 (4)	
	10	4	2·84 ± 0·26 (4)	2·88 ± 0·56 (4)	0·30 ± 0·04 (4)	0·24 ± 0·01 (4)	0·50 ± 0·02 (8)
	10	12			0·38 ± 0·01 (4)	0·25 ± 0·03 (4)	
	20	4	3·50 ± 0·28 (4)	1·92 ± 0·21 (8)	0·32 ± 0·02 (4)	0·22 ± 0·02 (4)	0·62 ± 0·08 (4)
	20	12					0·37 ± 0·09 (4)
DL 152	2·5	4		2·47 ± 0·21 (4)	0·30 ± 0·01 (4)	0·24 ± 0·01 (4)	
	10	4	2·16 ± 0·16 (4)	1·98 ± 0·19 (4)	0·26 ± 0·02 (4)	0·25 ± 0·01 (4)	0·32 ± 0·01 (12)
	10	12			0·38 ± 0·08 (4)	0·29 ± 0·01 (4)	0·21 ± 0·01 (4)
	20	4	2·53 ± 0·26 (8)	1·25 ± 0·06 (8)	0·30 ± 0·02 (4)	0·21 ± 0·01 (4)	0·20 ± 0·01 (4)
	20	12					0·18 ± 0·01 (4)
10 MD	5	4	3·58 ± 0·56 (4)	3·20 ± 0·70 (4)	0·32 ± 0·04 (4)	0·25 ± 0·03 (4)	0·35 ± 0·04 (4)
	10	4			0·35 ± 0·06 (4)	0·23 ± 0·02 (4)	0·35 ± 0·01 (4)
	10	12					0·22 ± 0·01 (4)

Number of determinations shown in brackets.

is, from both parameters, the most active compound, followed in order by DL 152, 10 MD and IBS. Unlike reserpine, DL 152 and 10 MD elicit only weak pressor responses in rats in which monoamine oxidase is inhibited. This result may be explained by the fact that these compounds release only some of the noradrenaline and that they are probably slow acting. In fact, IBS does not release noradrenaline from the heart and is inactive as a pressor agent in iproniazid-treated animals. The inhibition of the pressor effect of tyramine is common to all the compounds although reserpine is much more active than its derivatives. According to the current views (LaBarre and Hans, 1960), tyramine induces pressor effects through a release of noradrenaline and for this reason tyramine is inactive when reserpine has depleted the noradrenaline stores. However, reduction of the tyramine pressor response may not always be related to a depletion of the noradrenaline stores.

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