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THE ACTION OF RESERPINE ON THE PERIPHERAL SYMPATHETIC SYSTEM

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Reserpine causes the release of 5-hydroxytryptamine (5-HT) from all tissues in which it is normally stored. This release is shown by a fall in the tissue concentration of 5-HT and by an increase in its urinary metabolites (Pletscher, Shore & Brodie, 1955; Shore, Silver & Brodie, 1955). That reserpine may also lower the concentration of catecholamines in tissues was shown by experiments (Holzbauer & Vogt, 1956) in which the disappearance of noradrenaline from the hypothalamus was demonstrated in cats injected with small doses of reserpine (0·4 mg/kg). This depletion, however, did not occur in all organs, the denervated, in contrast to the innervated, adrenal medulla remaining unaffected by this dose of the drug.

It is tempting to try to correlate the effects on behaviour and responsiveness produced by reserpine with the loss of 5-HT and noradrenaline from the brain. The tendency has been to attribute not only the sedation, but also such signs as the fall in blood pressure, the miosis and the relaxation of the nictitating membrane to central changes alone. Although all these phenomena might be due to central causes, they could also be produced, at least partly, by disturbances in the peripheral sympathetic system. Failure of another sympathetic mechanism after reserpine was described by G. M. Everett who observed (personal communication, and Everett, Toman & Smith, 1957) that cold did not produce pilo-erection in mice injected with reserpine. This failure, too, might be due to a central or to a peripheral derangement, or to both.

The aim of the present paper is to determine the effect of reserpine on the peripheral sympathetic tissues: the investigation, abstracts of which have been published (Muscholl & Vogt, 1957*a*, *b*), deals with the loss of transmitter and the damage to function produced by reserpine in adrenergic neurones. In preliminary experiments the normal range of concentrations of adrenaline and noradrenaline in various parts of the sympathetic system was established.

METHODS

The experiments were done on adult rabbits, cats and dogs.

Operative procedures

Rabbits. The left superior cervical ganglion was removed aseptically under ether anaesthesia for subsequent estimation of its catecholamines. The wound was sewn up, and the animal recovered in a few minutes. It was then given reserpine, either intraperitoneally or into the marginal ear vein. After a varying time interval the rabbit was killed by a blow on the neck and bled out, and the right superior cervical ganglion and other tissues were removed for analysis for adrenaline and noradrenaline. When the right cervical sympathetic was stimulated, the rabbit was subjected to a brief ether anaesthesia before being killed, the cervical sympathetic chain was cut and the fibres were stimulated pre- and post-ganglionically, distal to the cut. The threshold voltage was determined which produced responses of eyelid and pupil. Stimulation of the hypogastric nerves was, at first, also done under ether anaesthesia; later it was found that the same thresholds were obtained when the animal was quickly killed by exsanguination and stimulation was carried out immediately. The best responses were seen on the vas deferens and the prostate. A Ritchie–Sneath stimulator supplied square pulses of 10 msec duration at a frequency of 33 c/s. A resistance of 0·3 MΩ was placed in series with the tissue; this reduced the current supplied to a suitable level and made the effective current in different animals less dependent on tissue resistance.

For all nervous tissue other than the superior cervical ganglia control figures were obtained on normal rabbits killed by a blow on the neck. These figures were then compared with those from animals injected with reserpine. This type of comparison was, of necessity, more statistical in character than that in which the right and left ganglia of the same animal were compared. A single superior cervical ganglion was often too small for the estimation of *adrenaline*, and in a number of experiments ganglia of several rabbits were pooled.

Cats. Essentially the same procedures were used in cats. The left superior cervical ganglion was removed in an aseptic operation under ether, and reserpine injected; on the next day the cat was given ether if electrical stimulation of the right sympathetic chain was required. After the stimulation the cat was killed by cutting the vessels in the neck. When stimulation of nerves was not intended, or when it was essential to avoid activation of the adrenal medulla by the anaesthetic, the cat was rapidly exsanguinated under chloroform.

For dissection of the splenic nerves the spleen was excised with the whole length of the splenic and coeliac arteries and a small segment of attached aorta, and the fat was congealed by keeping the tissue for at least half an hour at 4° C. Most of the mesenteric fat could then be lifted off, and the nerves dissected along the artery. Between 35 and 45 mg of tissue were usually obtained; the contamination with fat and connective tissue must have been about the same in all cats, since the concentration of amines in normal controls varied but little.

Decentralization of the left superior cervical ganglion was effected by excising about 1 cm of the preganglionic sympathetic chain, the procedure being carried out aseptically under ether.

Another preliminary operation required in some experiments was unilateral adrenal denervation. Under ether anaesthesia and with aseptic precautions, the splanchnic nerves were cut and the upper three lumbar ganglia excised on the left side. There were no complications from any of these operations.

The injections of reserpine were made intraperitoneally or intravenously. In excitable cats the intravenous injections were made under chloroform.

Dogs. The hypogastric nerves were stimulated under ether anaesthesia.

Estimation of amines

The freshly excised tissues were rapidly weighed and immersed in 1 ml. acid ethanol (0·1 ml. conc. HCl: 100 ml. ethanol) cooled in a mixture of CO_2 and acetone. The tissue was homogenized and extracted in acid ethanol, the extract purified, chromatographed on paper, and the regions

containing the individual amines were eluted separately. The amount of amines in the eluates was determined by bioassay, the blood pressure of the pithed rat (Shipley & Tilden, 1947) being used for noradrenaline and for amounts of adrenaline exceeding 5–10 ng (total), the rat's uterus stimulated by carbachol for smaller quantities of adrenaline and for isoprenaline. The latter amine was assayed simultaneously on the rat's auricles (Garb, Penna & Ganz, 1956). The threshold dose which produced an inotropic effect was usually 50–100 pg (10^{-12} g) D,L-isoprenaline added to a 6 ml. bath. The threshold on the rat's uterus was 50–100 pg in a 2 ml. bath. In view of the very small quantities of amine which exert effects on these organs, it is essential to avoid washing the region of the chromatograms carrying the control spots in the same benzene as the part of the paper to be eluted; otherwise there may be contamination of the paper with traces of isoprenaline coming off the control spots.

The ratio of the dose of isoprenaline to the equivalent dose of adrenaline varied from 0·1 to 0·25 on the uterus and from 0·02 to 0·07 on the auricles.

The pithed rat keeps its sensitivity to the smallest doses of noradrenaline much longer than the hexamethonium-treated rat previously used. It has the further advantage of a high sensitivity to adrenaline; it will often respond to as little as 1 ng. Details of the methods of extraction and assay, of necessary precautions and of recoveries of adrenaline and noradrenaline have been published (Vogt, 1952, 1953, 1954). Recoveries of isoprenaline (10 ng) were the same as those of adrenaline.

RESULTS

Noradrenaline and adrenaline: normal concentrations and effect of a single dose of reserpin

Paravertebral sympathetic system

Superior cervical ganglion

Rabbits. The noradrenaline concentration in the superior cervical ganglia of 41 normal rabbits was found to be $4·5 \pm 0·26$ μg/g (mean ± S.E. of the mean, here and throughout the paper). Table 1 shows that, in 21 of 22 experiments,

TABLE 1. Effect of injections of reserpine on the noradrenaline concentrations (μg/g fresh tissue) in the superior cervical ganglia of rabbits. (Left ganglion before, right after reserpine)

Intraperitoneal					Intravenous			
		Noradrenaline					Noradrenaline	
Rabbit no.	Dose of reserpine (mg/kg)	Left side (μg/g)	Right side (μg/g)	Loss (% of left side)	Rabbit no.	Dose of reserpine (mg/kg)	Right side (μg/g)	Loss (% of normal mean)
1	0·18	5·2	4·7	10	18	0·1	0·8	82
2	0·2	4·4	2·0	55	19	0·1	3·1	31
3	0·4	4·4	1·9	56	20	0·2	1·2	73
4	0·8	3·3	<0·6	>81	21	0·2	2·2	51
5–7	1·4	5·1	1·5	70	22	1·0	0·5	81
8–10	1·6	4·0	2·0	50	23	1·0	0·4	91
11	1·6	3·7	2·3	39	24	1·2	1·45*	68
12	1·6	4·4	1·3	70	25	1·2	1·35*	70
13	1·9	6·6	1·6	76	26–28	1·6	0·3	93
14–16	2·1	5·0	<0·4	>92	29	5·0†	0·35*	92
17	2·3	7·0	0·7	90				

The time interval between injection and removal of the right ganglion varied from 14 to 24 hr.

* Both ganglia extirpated together at the end of the experiment.

† Iproniazid, 100 mg/kg I.V., 1 hr previous to reserpine.

the noradrenaline content of cervical ganglia extirpated on the day following one injection of reserpine was much lower than normal. The only exception was rabbit 1, injected with the smallest intraperitoneal dose; even here the concentration after reserpine was 10% lower than that before. Generally speaking, the loss increased with the dose up to 0·8 mg/kg, and was greater when the same dose had been given intravenously than when it had been injected intraperitoneally. There were, however, large individual variations, as was also evident from the variable degree of sedation achieved.

In order to ascertain that the loss of noradrenaline was due to the drug and not to other factors, five rabbits were given either no injection or the reserpine vehicle only, and their left and right superior cervical ganglia were analysed; the ganglia were obtained by precisely the same operative procedures as in the experiments with reserpine. The concentration of noradrenaline was $5{\cdot}0 \pm 1{\cdot}5$ on the left side, and $5{\cdot}0 \pm 0{\cdot}6\,\mu g/g$ on the right. In another control experiment, chlorpromazine hydrochloride (10 mg/kg intravenously) was injected into four rabbits; these were killed 3·5 hr later, when they showed heavy sedation and low body temperature. The noradrenaline concentration of the cervical ganglia removed after the drug had been allowed to act was normal ($4{\cdot}9 \pm 1{\cdot}6\,\mu g/g$). Another control with morphine injections will be found in the section on cats.

Animals injected with reserpine exhibit miosis, narrowing of the palpebral fissure and relaxation of the nictitating membrane. To decide whether these signs might be caused or contributed to by the loss of transmitter from the adrenergic neurones, the peripheral end of the severed cervical sympathetic chain was stimulated electrically before and after administering reserpine in a series of rabbits; pupil and lid movements were observed (the nictitating membrane of the rabbit is unsuitable for observation), and the noradrenaline content of the superior cervical ganglia was estimated. That it was permissible to use the same ganglion for stimulation and extraction follows from previous experiments in the dog (Vogt, 1954), in which it was shown that stimulation of the cervical sympathetic trunk, even for periods of 2 hr and more, did not change the concentration of noradrenaline in the ganglia.

In eighteen rabbits, in which the noradrenaline concentration was above $1\,\mu g/g$, responses to stimulation of the cervical sympathetic were not, or only slightly, impaired by reserpine. When, however, the concentration in the right cervical ganglion had fallen below $1\,\mu g/g$, the result varied with the duration of the experiment. Of eight rabbits tested about 17 hr after the injection, four showed no response of pupil or eyelid when the pre- or post-ganglionic fibres of their right ganglia were stimulated with currents which were between 3 and 13 times stronger than those with which good responses on the left side had been obtained before reserpine. In the four other rabbits, in which the strength of the stimuli was pushed further, very small and sluggish responses

were obtained, but could not be made brisker or larger by increasing the strength of the current.

When, however, responses to stimulation of the cervical sympathetic were tested soon (between 2·5 and 4 hr) after the injection of reserpine, even severe depletion of the noradrenaline of the superior cervical ganglia did not impair the responses. Thus a fall in noradrenaline concentration to 0·3 μg/g was compatible with normal electrical thresholds (three rabbits).

That any loss of response of the smooth muscle to stimulation of the adrenergic nerves is due to the loss of transmitter can only be assumed if the excitability of the smooth muscle itself is not reduced by reserpine. To check this point, intravenous injections of noradrenaline were made into reserpinized rabbits, and the effect on the blood pressure was recorded. The pressor responses of such animals were either normal or exaggerated, never diminished. This is in good agreement with previous reports of enhanced responses to adrenaline and noradrenaline of the blood pressure and the nictitating membrane of the cat (Bein, Gross, Tripod & Meier, 1953).

TABLE 2. Effect of reserpine on the adrenaline concentration (μg/g fresh tissue) in the superior cervical ganglia of rabbits. (Left ganglion before, right after reserpine)

No. of rabbits	Dose of reserpine (mg/kg)	Interval (hr)	Adrenaline		
			Left (μg/g)	Right (μg/g)	Change (%)
1	2·3	15	0·50	0·50	0
3	2·1	16	0·13	0·20	+54
3	1·4	16	0·28	0·56	+96
1	0·8	18	0·40	0·85	+112
2	0·3	18	0·51	0·36	−29
			Mean 0·34	0·49	

The injections were made intraperitoneally. The interval refers to the time between injection and excision of the right ganglion.

In a single rabbit (no. 29, Table 1) iproniazid (100 mg/kg I.V.) was injected 1 hr before reserpine. Brodie & Shore (1957) have shown that, after the injection of this inhibitor of amine oxidase, reserpine produces a much smaller loss of 5-HT from the brain. The noradrenaline loss from the cervical ganglia, however, was severe, and the question of a possible protection of noradrenaline by iproniazid was not followed up.

Estimations of the adrenaline content of superior cervical ganglia of rabbits were difficult because of the minute quantities involved (between 1 and 7 ng/ganglion). Table 2 shows that, in five experiments on ten rabbits, there was no evidence of any fall in the adrenaline concentration after doses of reserpine which depleted the ganglia of their noradrenaline. We shall return to this point later.

Cats. Experiments on the superior cervical ganglia of cats confirmed the observations in rabbits. The results are summarized in Fig. 1. The nor-

adrenaline concentration was $3{\cdot}5 \pm 0{\cdot}3$ before, and $0{\cdot}6 \pm 0{\cdot}1\,\mu g/g$ on the day following, an injection of reserpine (1–2·5 mg/kg). In contrast to the rabbit, it made no difference whether the drug was given intravenously or intraperitoneally. The effect appeared to be as great after 1·3 as after 2·5 mg/kg.

The response of eyelid, pupil and nictitating membrane to electrical stimulation of the pre- and post-ganglionic fibres was tested in five normal and in three reserpinized cats. In one of the treated cats there was no response, and the threshold was greatly raised in the other two. Over 80% of the noradrenaline had disappeared from their ganglia. In the most severely affected animal the noradrenaline content was $0{\cdot}36\,\mu g/g$.

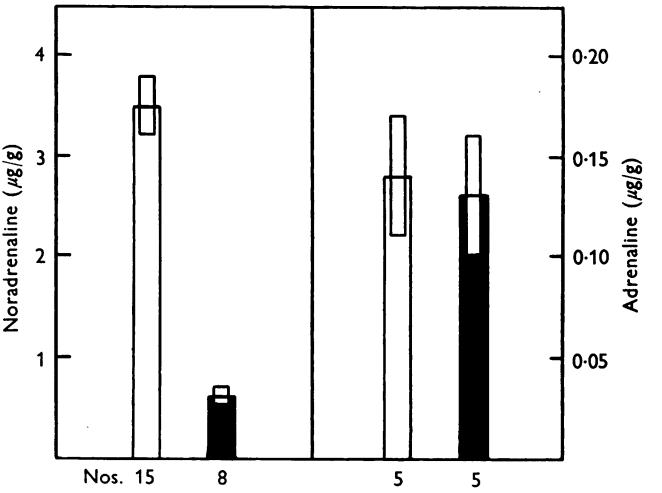

Fig. 1. Noradrenaline and adrenaline concentrations in the superior cervical ganglia of cats (mean and S.E. of the mean). Open columns, control figures. Black columns, results obtained between 13 and 20 hr after the intravenous injection of reserpine (1·0–2·5 mg/kg). Two cats were injected intraperitoneally. Number of cats at the foot of each column.

Many drugs are known to deplete the brain of its noradrenaline (Vogt, 1954); with the exception of reserpine they are all drugs which produce signs of great sympathetic activity. These drugs had not been tested on the peripheral sympathetic system before, and therefore the effect of morphine, a violent sympathetic stimulant in the cat, was compared with that of reserpine on the amine content of brain and of peripheral sympathetic tissues. Fig. 2 illustrates the results. Whereas the concentration of noradrenaline was low everywhere after reserpine, depletion after morphine only occurred in the hypothalamus and not in the cervical ganglia or splenic nerves.

In the cat, as in the rabbit, there was no evidence that reserpine produced changes in the adrenaline concentration of the superior cervical ganglion; the

mean content after reserpine was only 7% less than that of the normal cat (Fig. 1).

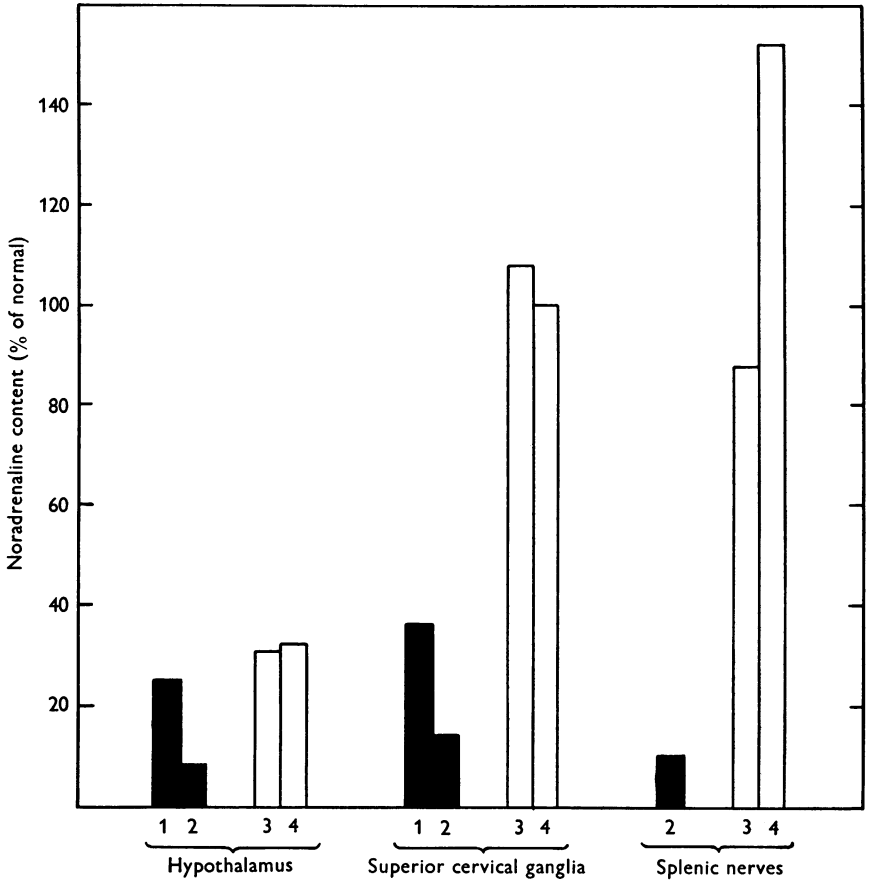

Fig. 2. Noradrenaline content of hypothalamus, superior cervical ganglia, and splenic nerves of each of four cats, as percentage of normal mean. Black columns (cats 1 and 2) after reserpine (1 and 2 mg/kg) intraperitoneally. Open columns (cats 3 and 4) after morphine HCl (40 and 35 mg/kg) subcutaneously. Number of cat at the foot of each column. Only the hypothalamus is affected by both drugs.

Stellate ganglia, thoracic and abdominal chains, splenic nerves

These parts were examined only in the cat. The stellate ganglia are anatomically strictly analogous to the superior cervical ganglia. Their normal concentration of noradrenaline was slightly, but not significantly, lower than that of the cervical ganglia, and so was that of adrenaline. As in the cervical ganglia, reserpine (1·0–2·0 mg/kg) caused an average fall of noradrenaline to 15%, whereas the concentration of adrenaline remained unchanged (Table 3).

TABLE 3. Concentration of noradrenaline and adrenaline (μg/g fresh tissue, mean $\pm$S.E. of the mean) in the stellate ganglia, the thoracic and abdominal chains, and the splenic nerves of cats. Normal animals and animals examined between 13 and 20 hr after an intravenous injection of reserpine

		Noradrenaline			Adrenaline			Percentage methylated	
Tissue	Dose range of reserpine (mg/kg)	Control (μg/g)	After reserpine (μg/g)	Amine left (%)	Control (μg/g)	After reserpine (μg/g)	Amine left (%)	Before reserpine	After reserpine
Stellate ganglia	1·0–2·0	2·9$\pm$0·4 (9)*	0·43$\pm$0·08 (7)	15·0	0·09$\pm$0·03 (7)	0·09$\pm$0·03 (5)	100	2·4	21
Thoracic chains	1·5–2·5	0·71$\pm$0·06 (4)	0·11$\pm$0·03 (5)	15·5	0·02$\pm$0·01 (3)	0·04$\pm$0·01 (5)	200	2·7	27
Abdominal chains	2·5	1·6$\pm$0·1 (4)	0·20$\pm$0·04 (4)	12·5	0·10$\pm$0·03 (2)	<0·06$\pm$0·01 (4)	<60	5·9	<23
Splenic nerves	1·0–2·5†	2·5$\pm$0·4 (9)	0·64$\pm$0·16 (9)	26·6	0·23$\pm$0·06 (6)	0·13$\pm$0·04 (7)	57	8·5	17

The last columns give the percentage of adrenaline in the mixture of the two catecholamines.

* Number of cats in parentheses.

† One cat injected intraperitoneally.

Essentially similar results were obtained for the remaining parts of the paravertebral sympathetic system, i.e. the thoracic and lumbar chains. In these, however, the ganglia were not separated from the fibres with which they form the chain. Owing to the large admixture of medullated non-adrenergic components the amine concentrations in the chains were lower than in pure ganglionic tissue (Table 3). As before, injections of reserpine (1·5–2·5 mg/kg) greatly lowered the concentration of noradrenaline without producing significant changes in the content of adrenaline.

Table 3 also contains estimations carried out on the splenic nerves. These provided a specimen of post-ganglionic sympathetic fibres containing little, if any, ganglionic tissue. The normal concentration of noradrenaline, $2{\cdot}5 \pm 0{\cdot}4\ \mu g/g$ was barely significantly lower than that in cervical ganglia; reserpine lowered it to about a quarter of normal. The adrenaline figures suggested occasional lowering by reserpine; this would seem in contrast to the data obtained on paravertebral ganglia, and bring the splenic nerves in line with the solar ganglion, from which they originate (see below); the difference, however, in the mean adrenaline concentration before and after reserpine is not significant.

TABLE 4. The effect of decentralization of the left superior cervical ganglion on the action of reserpine

	Interval (days)	Dose of reserpine (mg/kg)	Noradrenaline* Left side (μg/g)	Noradrenaline* Right side (μg/g)
Cat 1	12	0	2·7	3·1
Cat 2	19	0	3·5	3·4
Cat 3	12	1·5	0·42	0·41
Cat 4	17	1·5	0·32	0·49
Rabbit 1	3	1·2	1·5	1·4
Rabbit 2	3	1·2	1·4	1·5

The interval refers to the time between decentralization of the left ganglion and the experiment. Reserpine was given intravenously between 13 and 17 hr previous to the dissection of the ganglia.

* μg/g fresh tissue.

In view of the fact that denervation renders the adrenal medulla of the cat, and to some extent that of the rabbit, refractory to reserpine (Holzbauer & Vogt, 1956; Kroneberg & Schümann, 1957), experiments were carried out to test the effect on the action of reserpine of decentralization of the superior cervical ganglion. In four cats and two rabbits the left superior ganglion was severed from its connexions with the central nervous system by removal of a piece of the preganglionic nerve trunk. It will be seen (Table 4) that reserpine caused the same depletion in the left (decentralized) and in the right (normal) ganglia of both cats and rabbits. The average loss of noradrenaline was 88% in the cats and 68% in the rabbits. In order to ascertain that decentralization as such does not change the noradrenaline content of ganglia, cats 1 and 2 were subjected to decentralization of the left superior cervical ganglion and killed,

without being given reserpine, 12 and 19 days later. There was no significant difference between the noradrenaline content of the left and right ganglia. Thus, up to 19 days at least, the concentration of transmitter in the adrenergic neurone is not affected by a lack of impulses discharging along it, nor by the degeneration of the preganglionic fibres.

The observation that the effect of reserpine on the sympathetic ganglia persists after decentralization agrees well with the finding (Brodie, Olin, Kuntzman & Shore, 1957) that the sympathin of the heart is lost when reserpine is given to rabbits in which the cord has been divided.

Prevertebral ganglia

Rabbits. Estimations were carried out on the solar and the inferior mesenteric ganglia. The concentration of catecholamines in these ganglia is much higher than in the paravertebral sympathetic chains, and higher in the inferior mesenteric than in the solar ganglion. In contrast to what will later be shown in the cat, the range, and therefore the standard error, was large only in the inferior mesenteric ganglion. The normal concentrations and those determined after the injection of reserpine will be found in Table 5. The 'normal' rabbits actually consisted of five untreated animals and of four animals injected with chlorpromazine. It has been shown above that chlorpromazine does not alter the noradrenaline content of the superior cervical ganglia, and the same was found to hold for the prevertebral ganglia. For the purpose of Table 5 the rabbits injected with chlorpromazine were therefore classified as 'normal'. No figures for the adrenaline concentration in the inferior mesenteric ganglia are included, since the tissue available weighed only a few milligrams and estimations were rarely successful.

Row 1 contains the figures for normal rabbits; the high concentration of adrenaline in the solar ganglion is in striking contrast to the low values obtained in all paravertebral ganglia (Tables 2 and 3). When reserpine was given in a single dose of 0·1 or 0·2 mg/kg and the rabbit was killed on the following day (rows 2 and 3, Table 5), there were convincing losses of catecholamines only with the larger dose. In the following eight experiments, still larger doses (1·0–1·6 mg/kg) were injected (row 4, Table 5); the ganglia were excised not less than 14 hr after the injection. The noradrenaline concentration fell in all ganglia by approximately the same amount, and so, apparently, did the adrenaline concentration in the solar ganglion. This is in contrast to the refractoriness of the adrenaline contained in the paravertebral system. Examination of the figures, however, shows that the individual adrenaline concentrations have such a large scatter that the mean content after reserpine is not significantly different from that before reserpine. But since four of the eight values are well below the normal range, the right conclusion to draw appears to be that reserpine does lower the concentration of adrenaline, but

TABLE 5. Catecholamine concentrations (μg/g fresh tissue, mean $\pm$S.E. of the mean) in the prevertebral ganglia of rabbits. Effect of intravenous injections of reserpine

Group no.	Dose of reserpine (mg/kg)	Interval (hr)	Solar ganglion: Noradrenaline (μg/g)	Solar ganglion: Noradrenaline Lost (%)	Solar ganglion: Adrenaline (μg/g)	Solar ganglion: Adrenaline Lost (%)	Inferior mesenteric ganglion: Noradrenaline (μg/g)	Inferior mesenteric ganglion: Noradrenaline Lost (%)	Superior cervical ganglion: Noradrenaline lost (%)
1	Controls	0	6·7±0·7 (7)*	0	1·5±0·4 (9)	0	8·6±1·5 (8)	0	0
2	0·1	16	5·2 (1)	23	0·6 (1)	61	3·6 (1)	58	2
3	0·2	17	1·9 (1)	72	<0·5 (1)	67	3·3 (1)	62	59
4	1·0–1·6	14–17½	1·8±0·3 (8)	73	0·6±0·2 (8)	60	2·8±0·7 (6)	67	80
5	14 or 18 × 0·1, 8 × 0·2	—	1·3±0·1 (6)	80	0·13±0·02 (6)	91	1·4±0·3 (4)	84	80
6	1·6	¼–2½	2·4±0·4 (8)	64	1·0±0·3 (6)	33	8·2±2·1 (6)	4·7	70
7	1·6	4	0·49±0·06 (3)	93	0·29±0·08 (3)	81	6·9±3·7 (3)	20	94

* Figures in parentheses number of experiments; most figures on inferior mesenteric ganglia, and some on other ganglia, were obtained on the pooled tissue of two rabbits, and counted as one experiment.

that some ganglia have such a high initial adrenaline content that even after depletion the content may greatly exceed low normal values.

Row 5 shows the effect of repeated small doses of reserpine. A total amount of approximately 1·6 mg/kg, which depletes the ganglia when given in one injection, was given in divided doses spread over 8–18 days. The noradrenaline concentration of all ganglia was reduced to about 20% and so was the adrenaline content of the solar ganglion. Thus this treatment was at least as effective as the single dose in depleting the ganglia of their noradrenaline, and, in addition, produced a highly significant effect on their content of adrenaline.

In many rabbits electrical stimulation of the hypogastric nerves was carried out after administering reserpine. Even in animals in which the cervical sympathetic had lost so much transmitter that the response of the eyelid and pupil was absent, the vas deferens contracted on stimulation of the hypogastric nerves with minimal currents. The concentration of noradrenaline in the inferior mesenteric ganglion, from which the hypogastric fibres originate, was below 1μg/g in only one of these experiments, and perhaps there was always enough transmitter left to stimulate the effector organs. Another possibility is the presence of cholinergic fibres coursing to the vasa deferentia in the hypogastric nerves. This possibility was rendered very unlikely by the results of an experiment in which the excitability of the centrally cut hypogastric nerves of a normal anaesthetized rabbit was tested before and after the administration of eserine and of atropine. First 0·2, and later 0·4 mg/kg eserine sulphate was injected intravenously. The eserine caused defaecation and violent spasms of the colon, but responses of the bladder and vas deferens to stimulation of the hypogastric nerves were obtained at the same voltage and were not increased in size. Atropine sulphate (5·5 mg/kg intravenously) was then given. This completely relaxed the colon but was without effect on the threshold of the responses of bladder neck and vas deferens to indirect electrical stimulation.

Cats. In normal cats the catecholamine concentration in the prevertebral ganglia is much more variable than in the rabbit. This is particularly true of the inferior mesenteric ganglion, in which the figures for noradrenaline ranged from 9·6 to 46·2μg/g and those for adrenaline from 0·4 to 15·6μg/g. Large concentrations of adrenaline were found only when that of noradrenaline too was high. There is little doubt that these high and variable concentrations result from the occurrence in these ganglia of chromaffine tissue. Its development and distribution has been described by Kohn (1903). Kohn states that all sympathetic ganglia contain some chromaffine tissue, but it is obvious from his drawings that the largest chromaffine bodies invade the prevertebral ganglia on the ventral aspect of the aorta. In a few instances we have looked for chromaffine tissue in the stellate, the inferior mesenteric and the solar ganglia of cats. The tissue was fixed in a mixture of bichromate and formaldehyde, cut and stained with haematoxylin. Chromaffine bodies were found in

the inferior mesenteric and solar ganglia, but not in the stellate ganglia. This would agree with the findings of low concentrations of adrenaline in the stellate ganglion and high values in the prevertebral ganglia. Since the chromaffine tissue usually lies on the surface of the ganglia and its size and shape is very variable, the amount included in the excised ganglia is bound to vary, and this may explain the large range in amine content.

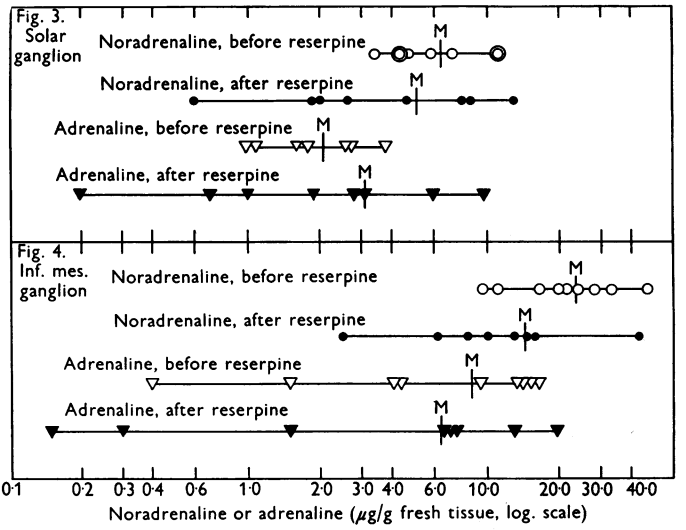

Fig. 3. Catecholamines in the solar ganglia of individual cats. Normal concentrations (open signs); and content after reserpine, 1·0–2·5 mg/kg, usually intravenously (black signs). Excision of ganglia 13–17 hr after the injection. M, mean.

Fig. 4. Catecholamines in the inferior mesenteric ganglia of individual cats. Normal concentrations (open signs); and content after reserpine, 1·0–2·5 mg/kg, normally intravenously (black signs). Excision of ganglia 13–20 hr after the injection. M, mean.

Figs. 3 and 4 illustrate the findings on normal cats and on cats given reserpine intravenously on the day preceding the extirpation of the ganglia. With doses higher than 1·0 mg/kg the results bore no relation to the size of the dose. It will be seen from the figures that the mean concentrations of catecholamines after reserpine did not differ significantly from the control means. Yet Figs. 3 and 4 show that an effect was, indeed, produced. It is manifested by the fact that, after reserpine, amine concentrations occur which are considerably below the lowest normal figures. Thus the lowest concentration of noradrenaline in the normal solar ganglia was 3·4 μg/g, but four of eight reserpine-treated cats had lower concentrations (0·6–2·6 μg/g). The effect on the mean was masked by the excessively high concentration of 12·7 μg/g found in one of the reserpinized cats.

The question may be asked why this difficulty was not encountered in the rabbit, the solar ganglion of which showed a consistent reduction in the

concentration of both catecholamines. The difference can be explained if one assumes that in both species chromaffine tissue in the ganglia behaves like adrenal medullary tissue. First, there is practically no noradrenaline in the adrenal medulla of rabbits, whereas large and variable amounts are found in the cat. In the rabbit the presence of chromaffine tissue should therefore hardly contribute to the noradrenaline content of the solar ganglion, and reserpine should deplete this tissue of its noradrenaline in the same consistent way as it depletes paravertebral ganglia. Secondly, the amines in the adrenal medulla of rabbits are much more susceptible to the action of reserpine than the amines of cat adrenals (see next section). This would explain why, in the rabbit, the chromaffine tissue loses its adrenaline as easily as the adrenergic neurones their noradrenaline. Thus, after larger and particularly after repeated doses of reserpine, noradrenaline and adrenaline disappear to about the same extent from the solar ganglia (Table 5). Only in experiments of short duration was there greater loss of noradrenaline than of adrenaline from the solar ganglia. In the cat, on the other hand, the large depletion of noradrenaline from the neurones may be masked by the localization of some of the noradrenaline in chromaffine tissue, from which it is not very readily lost.

Stimulation of the hypogastric nerves was also attempted in cats, but, in contrast to the rabbit, contractions of the vas deferens were difficult to obtain in normal animals, and contraction of the bladder was seen at a wide range of thresholds in different individuals. It was therefore not possible to decide whether the treatment with reserpine had altered the response of the organs to electrical stimulation of the nerves.

Dogs. A few experiments were carried out in the dog, in which the hypogastric nerves are thick enough for analysis and in which the inferior mesenteric ganglia are very large. The hypogastric nerves, which normally contained $2{\cdot}2 \pm 0{\cdot}4\,\mu g/g$ of noradrenaline (range of five determinations 1·7–3·6), and a very variable amount of adrenaline (range 0·09–1·23 $\mu g/g$), had lost 80% of their noradrenaline 15 hr after a single dose of reserpine (0·5 mg/kg), and 91% after four doses (one injection per day intravenously, 0·5 mg/kg on the first and last, 0·25 mg/kg on the remaining days). The adrenaline content remained within the (very wide) normal range. Electrical stimulation of the centrally cut hypogastric nerves normally caused peristaltic movements and blanching of the vasa deferentia as well as contraction of the bladder neck, but after the single dose only bladder movements occurred, and after the four doses all responses had vanished. The noradrenaline concentration had fallen to $0{\cdot}48\,\mu g/g$ in the first and to $0{\cdot}2\,\mu g/g$ in the second instance.

In the mesenteric ganglia, normally found to contain amounts of noradrenaline and adrenaline which varied but little and greatly exceeded those of cats and rabbits, reserpine caused losses which were large for noradrenaline and only slightly less severe for adrenaline. Thus the normal concentrations

were $40{\cdot}2 \pm 7{\cdot}0\,\mu g/g$ noradrenaline and $39{\cdot}0 \pm 5{\cdot}3\,\mu g/g$ adrenaline; after the single dose of reserpine the noradrenaline had fallen by 88%, the adrenaline by 83%; the repeated doses caused a loss of 91% of the noradrenaline and of 80% of the adrenaline, with corresponding tissue concentrations of $3{\cdot}6\,\mu g/g$ noradrenaline and $7{\cdot}7\,\mu g/g$ adrenaline. These values are higher than even the normal concentrations in the hypogastric nerves, and there is little doubt that they represent what was left of the catecholamines of the chromaffine tissue. The loss of response to stimulation and the low noradrenaline content ($0{\cdot}2\,\mu g/g$) of the hypogastric nerves indicate that this fraction of the catecholamines is not available as transmitter.

Adrenal medulla

Rabbits. Carlsson & Hillarp (1956) observed that a large dose of reserpine (5 mg/kg) almost completely emptied the rabbit's adrenal of its catecholamines. Kroneberg & Schümann (1957) found that 90% of the adrenaline disappeared after 2 mg/kg. They also state that denervation reduces but does not prevent the loss. We had the same experience in one rabbit, in which 24 hr after reserpine (1·6 mg/kg) the adrenaline concentration had fallen to 95% of normal on the innervated side, and to 75% on the denervated side. In this experiment, however, the more severely affected innervated adrenal contained actually twice as much noradrenaline ($1{\cdot}4\,\mu g/g$) as the denervated adrenal. The percentage of methylated amine was thus a little lower than normal (95 instead of 98%), a shift which had not been seen in experiments lasting only 4 hr. This observation was followed up in a series of experiments, in which the same total quantity of reserpine was given in divided doses over a period of many days. Table 6 contains the results. All adrenals had lost over 99% of their adrenaline, but relatively less noradrenaline, and the percentage of the methylated amine ranged from 43 to 76% instead of the normal 98%. Since the amount of noradrenaline found was always much lower than the normal concentration, these experiments might be interpreted on the assumption that adrenaline is lost more readily than noradrenaline. It is, however, more likely that the relative excess of noradrenaline indicates that resynthesis is proceeding even while the reserpine is acting, and that the process of methylation is slower than the manufacture of the noradrenaline molecule.

It was stated earlier that an amount of reserpine given in divided doses over a period of 2 weeks is as effective in depleting noradrenaline, and more effective in depleting adrenaline, from ganglia than the same quantity injected as a single dose (Table 5). Table 6 shows that the loss of medullary amines after repeated small doses of reserpine is much more complete than after a single dose, when the loss did not exceed 95%.

Cats. Previous experience had suggested that the adrenal medulla of the cat is more resistant to reserpine than that of the rabbit. With doses of

0·4 mg/kg no depletion of total amines had been found in the denervated, and an average loss of only 41% in the innervated, gland (Holzbauer & Vogt, 1956). In order to check whether this was only a question of dosage, seven cats with one denervated adrenal were given larger amounts of reserpine (1·0–2·5 mg/kg). In five of these cats (1–5, Table 7) the concentration of total

TABLE 6. Effect of repeated intravenous injections of reserpine on the medullary amines (μg/g fresh adrenal) of the rabbit

Individual dose (mg/kg)	No. of doses*	Nor-adrenaline (μg/g)	Adrenaline (μg/g)	Methylated (%)	Difference from normal: Nor-adrenaline (%)	Difference from normal: Adrenaline (%)
5 Controls	0	9·4† ±1·2	501† ±58	98	0	0
0·2	8	0·7	0·5	43	93	99·9
0·2	8	1·6	2·0	56	83	99·6
0·1	14	2·3	7·3	76	76	98·5
0·1	14	0·7	1·4	67	93	99·7
0·1	18	1·2	2·5	68	87	99·5
0·1	18	0·4	0·9	68	96	99·8

* One injection per day.

† Mean ±S.E. of the mean.

TABLE 7. Effect of a single, large intravenous dose of reserpine on the concentration of adrenaline and noradrenaline in the left (denervated) and the right (innervated) adrenal medulla of the cat

Cat no.	Reserpine (mg/kg)	Interval between denervation and expt. (days)	Left (denervated) adrenal: Catecholamines (mg/g)	Left (denervated) adrenal: Methylated (%)	Right (innervated) adrenal: Catecholamines (mg/g)	Right (innervated) adrenal: Methylated (%)	Catecholamines lost from innervated gland* (%)
1	1·0†	23	1·16	85	0·57	65	69
2	1·5	12	1·20	64	0·65	64	53
3	2·5	16	0·52	73	0·30	70	39
4	2·5	16	0·63	75	0·30	60	47
5	2·5	24	0·96	67	0·24	54	71
6	2·0	29	0·23	72	0·17	71	
7	2·5	26	0·26	57	0·18	48	

The catecholamines of the denervated adrenal of 18 cats not injected with reserpine ranged from 0·48–1·56 mg/g fresh tissue (mean 0·91 mg/g). The adrenals were removed between 15 and 19 hr after the injection of reserpine.

* Denervated gland = 100.

† Intraperitoneally.

amines on the denervated side lay within the range of 0·48–1·56 mg/g found in cats which were not injected with reserpine. It would, therefore, appear that doses up to 2·5 mg/kg do not affect the denervated adrenal. The loss on the innervated side was 54%, a little greater than with the dose of 0·4 mg/kg, and much smaller than the corresponding figure in rabbits, which is at least 90%. In two cats, however (nos. 6 and 7), obvious depletion was obtained in both adrenals, the figure for the denervated adrenals falling well below the lowest control figure. There was also very little difference in the concentration

of amine on the two sides. It seems likely that denervation was not satisfactory in these two cats. In one of them (no. 7), the lesser splanchnic nerve was not found at operation, so that incompleteness of denervation is likely to explain at least this result. Perhaps some re-innervation had taken place in the other. The ratio adrenaline to noradrenaline was not much affected by the reserpine; it fell a little in three and remained the same in two cats.

Time course of the action of reserpine

A single dose of reserpine (1·6 mg/kg) was given to rabbits, and sympathetic ganglia were examined at different intervals after the injection. The injection was intravenous when the experiments were of short duration, and intraperitoneal for the long-term experiments.

Onset of action

Figs. 5 and 6 illustrate the early phase of the depletion of noradrenaline from the different ganglia. The corresponding values for the adrenaline content of the solar ganglion are shown in rows 6 and 7, Table 5. It takes about 4 hr for the effect on the superior cervical and solar ganglia to reach its maximum, but more time is apparently required to deplete the inferior mesenteric ganglion. Fig. 6 represents all the noradrenaline estimations made of this ganglion, and shows the normal range and the individual estimations after reserpine; it illustrates the very large scatter both in the normal and in the treated rabbits. It is possible that what appears to be a greater resistance of the inferior mesenteric ganglion to reserpine is only an artifact resulting from the wide range of individual values.

Recovery

After a single injection of reserpine (1·6 mg/kg), the catecholamine content of the sympathetic system remains low for a long period of time. After 5 days (observations on four rabbits), the noradrenaline concentration of the superior cervical ganglia had recovered to just above half the normal value. Even after 8–9 days (three rabbits), the adrenals and the cervical, solar and inferior mesenteric ganglia contained reduced amounts of catecholamines, all figures but one lying below the normal range. Recovery was complete after 14 days (two rabbits) except for the adrenal medulla of one rabbit which was still partially depleted of its hormones; the ratio of adrenaline to noradrenaline was normal in the adrenals of both animals, as indeed it was in the incompletely restored glands examined after 8 and 9 days.

Other catecholamines

Dopamine (hydroxytyramine) has been demonstrated in sympathetic ganglia and splenic nerves of cattle by Schümann (1956). The quantity was of

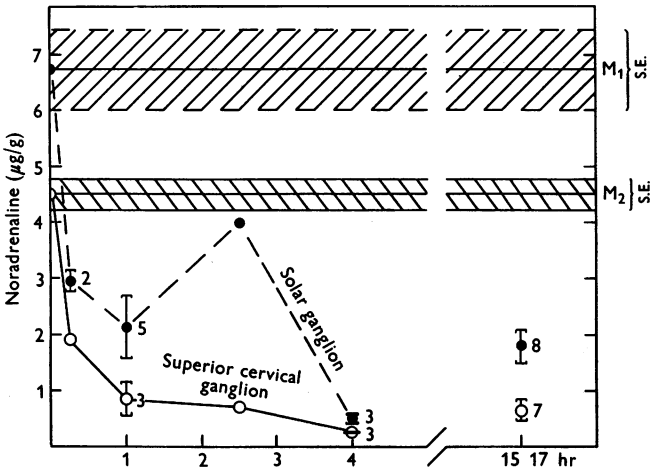

Fig. 5. Onset of effect of reserpine (1·0–1·6 mg/kg intravenously) on noradrenaline concentration in the solar and the superior cervical ganglia of the rabbit. The upper horizontal line (M_1) and hatched band represent mean and S.E. of the mean of the noradrenaline content of the solar ganglia of 7 normal rabbits, the lower horizontal line (M_2) and hatched area the corresponding figures for the superior cervical ganglia of 41 rabbits. ●– – –●, solar ganglia; ○—○, superior cervical ganglia of reserpinized rabbits. Vertical lines through points, S.E. of the mean. Number of experiments to the right of each point; when no S.E. is given, only one experiment was carried out. In most experiments, the superior cervical ganglia of two rabbits were pooled. Note that the effect of reserpine is fully developed at 4 hr.

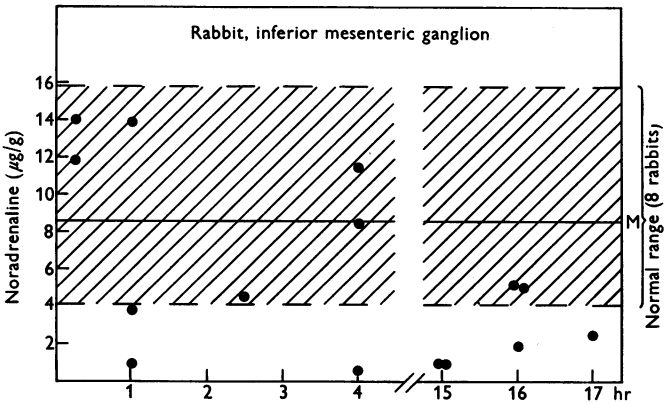

Fig. 6. Onset of effect of reserpine (1·0–1·6 mg/kg intravenously) on noradrenaline concentration in the inferior mesenteric ganglion of the rabbit. Hatched part of figure, normal range of noradrenaline concentrations (8 experiments). Continuous line M, mean normal value. Each dot represents one estimation on the ganglia of one or two reserpinized rabbits. Note that a convincing fall in noradrenaline was only obtained after 15–17 hr.

the same order of magnitude as that of noradrenaline. Since the biological activity of dopamine on the rat's uterus is several thousand times less than that of adrenaline, and on the rat's blood pressure about one hundred times less, it was obvious that it was not worth while attempting estimations of dopamine in the small amounts of tissue at our disposal. Schümann, however, expressed the view that the small quantities of adrenaline found in sympathetic ganglia might not be adrenaline but dopamine, and we carried out some control experiments to check such a possibility. Chromatograms of the pooled superior cervical ganglia of three rabbits were cut up in such a way that the strip corresponding to the adrenaline region and the strip immediately behind it, which would have contained any dopamine, were eluted separately and tested on the rat's uterus. Only the adrenaline region contained a heat-labile compound which inhibited the uterus; to obtain an inhibition due to dopamine by the eluate of the dopamine strip the quantity present would have had to be of the order of 10 μg, or 100 times the amount of noradrenaline present in the ganglia. Since Schümann found the quantities of noradrenaline and dopamine in cattle ganglia to be about equal, it is obvious that if the same held for the rabbit, amounts which could have been present in the eluates were far below the threshold of the assay.

Some experiments were done in which the isoprenaline region of chromatograms of sympathetic tissue from the cat was eluted and tested by parallel assay on the rat's uterus and the rat's auricles. In view of the work of Lockett (1957), who reported isoprenaline in the blood of cat heart–lung preparations when the thoracic chains were stimulated, the stellate ganglia and thoracic chains were repeatedly tested. The prevertebral ganglia were included in the survey owing to their greater content of catecholamines. In only one of four cats did the stellate ganglia contain a small quantity of a material which had the R_F value and the biological activities of isoprenaline. The total amount in both ganglia assayed as 1·0 ng on the uterus and 0·6 ng on the rat's auricles; the amount was nearly equal to the quantity of adrenaline in the ganglia (1·1 ng), and represented approximately 1 % of their noradrenaline. The concentration amounted to 0·035μg/g fresh tissue. In the stellate ganglia of the other three cats, in the solar ganglia of a cat and a rabbit, in the superior cervical ganglia and the whole sympathetic chain of several cats, in the splenic nerves of a cat and in the heart of a rabbit, no isoprenaline-like substance was found. It would have been detected if the concentrations had reached levels which ranged from 0·003 to 0·01 μg/g of tissue. Under these circumstances the one positive finding of a concentration of 0·035 μg/g is insufficient evidence that isoprenaline is in fact stored in sympathetic tissue of the cat.

DISCUSSION

The foregoing experiments show that cells and fibres of peripheral adrenergic neurones lose their noradrenaline after injections of reserpine. Organs with an adrenergic innervation no longer respond to electrical stimulation of preganglionic and post-ganglionic fibres when the loss of noradrenaline is severe and has persisted long enough. The finding that low noradrenaline content of the superior cervical ganglion is compatible with excitability of the sympathetic trunk in the early part of an experiment suggests that the loss of amine does not proceed evenly along the whole neurone, and that there is a period when the nerve endings have still enough transmitter to enable them to function while the ganglion cell has already reached its lowest noradrenaline concentration. Recently a disappearance of noradrenaline from heart tissue of animals injected with reserpine has been reported by several authors (Bertler, Carlsson & Rosengren, 1956; Paasonen & Krayer, 1957); this agrees well with the above findings.

These observations pose the question whether the impaired function of the peripheral sympathetic system is a factor which contributes to the clinical picture produced by reserpine in man. The answer is suggested by the experiments in which small doses were injected over a long period of time, since it is under these conditions that the animal experiments are strictly comparable with the therapeutic procedures. Daily doses of the order 0·1 mg/kg are used in psychiatry, and these doses were shown to be as effective as single large doses in depleting sympathetic ganglia of rabbits of their noradrenaline, and more effective than single doses in causing the disappearance of adrenaline from the solar ganglion and of the hormone from the adrenal medulla. It is therefore likely that an action on the peripheral sympathetic system contributes to the over-all effect of reserpine in man. Gaddum, Krivoy & Laverty (1958) have shown that patients treated for weeks with daily doses of reserpine excrete much less noradrenaline than normal; this change might be caused by the reduction of the noradrenaline stores in adrenergic neurones. The difficulty remains of deciding which role to allocate to each of the many chemical changes produced by reserpine. Is the loss of noradrenaline from the sympathetic centres more important than the loss from the peripheral sympathetic system, and is the loss of 5-HT from the hypothalamus more important than the loss of sympathin from the same region? There may also be other chemical 'lesions' produced of which we are quite ignorant but which may affect function decisively.

Except for the special case of the prevertebral ganglia of the cat, the noradrenaline was readily lost from the sympathetic neurones of all species; this loss was independent of any connexions with the central nervous system. In contrast, there was no evidence that adrenaline was lost from any tissue

in which its initial concentration was very low, such as paravertebral ganglia and post-ganglionic fibres. In the prevertebral ganglia, which contain high concentrations of adrenaline in what is probably chromaffine tissue, the loss was easily demonstrable in the dog and the rabbit, but not in the cat. The difference between the rabbit and the cat is even more pronounced with regard to the adrenal medullary amines. The adrenal medulla of the rabbit is very easily depleted by moderate doses of reserpine and denervation affords only a little protection. In the cat losses of medullary hormones from the innervated gland are very variable, and denervation affords a high degree of protection. The reason why connexion with and presumably stimulation by the central nervous system should be essential for the effect of reserpine to take place on some tissues and not on others is quite obscure, and perhaps points to different modes of binding of the amines at various sites.

Recovery of the normal concentration of amines after a single injection of reserpine took a long time, and usually more than 9 days. This is somewhat longer than the time required for the restoration of the 5-HT lost from brain tissue (Shore, Pletscher, Tomich, Carlsson, Kuntzman & Brodie, 1957). According to the same authors reserpine is not stored in tissue for more than a few hours. It is not clear how much of the time needed for restoration of the normal content of catecholamines is taken up by the period during which binding of the amines is impaired and how much simply represents the time needed for resynthesis. That the latter takes days is known from experiments with radioactive tracers (Udenfriend & Wyngaarden, 1956), and from the work on cat adrenals depleted of medullary hormone by acetylcholine (Butterworth & Mann, 1956). One interesting difference between the recovery process in the adrenal of the rabbit and that of the cat is the fact that the percentage of methylated amine is normal in the rabbit long before the total amine content is restored, whereas Butterworth & Mann found the opposite to be true of the cat, in which total amine concentration was back to normal while the percentage of adrenaline was still very low. This is a good example of the greater efficiency of the methylation process in the rabbit as compared with the cat.

Nevertheless, it was possible by repeated daily injection of reserpine to produce a rabbit adrenal containing a mixture of catecholamines in which the percentage of methylated amine was low. This finding suggests, but does not prove, that resynthesis of amines proceeds unimpeded during the presence of reserpine in the tissues, and that even in the rabbit methylation is a slow step in adrenaline synthesis.

During the early period of the action of reserpine some manifestation of the release of medullary amines and adrenergic transmitter into the circulation might be expected; and this is, indeed, seen when conditions are favourable. Thus Everett *et al.* (1957) described evanescent pilo-erection in mice or rats about half an hour after large doses of reserpine (50–100 mg/kg by mouth);

Kuschke & Frantz (1955) saw hyperglycaemia in the rabbit; tachycardia has been seen to occur in the heart–lung preparation of the dog (Plummer, Earl, Schneider, Trapold & Barrett, 1954), and many workers have observed rises in blood pressure in different species (de Jongh & van Proosdij-Hartzema, 1955; Maxwell, Ross, Plummer & Sigg, 1957; Domino & Rech, 1957). A raised level of blood adrenaline has been found in rabbits during the first hour after an intravenous dose of reserpine (Muscholl & Vogt, 1957*c*).

SUMMARY

1. Fibres and ganglia of adrenergic neurones of rabbits, cats and dogs lose a large fraction of their noradrenaline when the animals are injected with reserpine.

2. When the loss of noradrenaline is severe, and has persisted for more than four hours, it may lead to loss of response from the organs innervated by adrenergic fibres to indirect electrical stimulation.

3. The normal adrenaline content of the paravertebral sympathetic tissue is low, and shows no consistent changes after reserpine. In contrast, very high adrenaline concentrations are found in the prevertebral (solar and inferior mesenteric) ganglia of all species examined, evidently because of the presence in these ganglia of much chromaffine tissue. In the rabbit and in the dog treated with reserpine the adrenaline and the noradrenaline disappear as readily from the prevertebral ganglia as the noradrenaline disappears from the paravertebral ganglia. In the cat's prevertebral ganglia reserpine produces erratic changes in the concentration of both amines; whereas evidence of depletion is found in some, it is not seen in other individuals. This peculiar response of cat ganglia is explained on the assumption that the nervous tissue proper always loses its noradrenaline, but that the chromaffine tissue of cat ganglia is less readily depleted of its amines than the chromaffine tissue in the ganglia of rabbits, just as the adrenal medulla of the cat is more resistant to the action of reserpine than that of the rabbit. If we further assume that the percentage of methylated amine is as low in the ganglionic as it is in the medullary chromaffine tissue of cats, the resistance of the ganglia to the action of reserpine will affect all the adrenaline and that fraction of noradrenaline which is localized in the chromaffine cells.

4. The great efficacy of small daily doses of reserpine in causing a loss of catecholamines throughout the body suggests that chronic therapeutic administration of reserpine in man will also lead to a reduction in his stores of catecholamines.

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