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THE EFFECT OF DENERVATION ON THE RESPONSES OF THE CAT'S NICTITATING MEMBRANE TO SYMPATHOMIMETIC AMINES

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In previous studies the effects of graded doses of sympathomimetic amines on the heart rate, and on the systolic blood pressure, were measured in atropinized sleeping bitches. It was found that removal of the sympathetic chains, from and including the stellate ganglia, downwards into the pelvis, produced a greater increase in the sensitivity of the systolic blood pressure to noradrenaline than to adrenaline, but that this procedure resulted in a greater increase in the sensitivity of the responses of the heart rate to adrenaline than to noradrenaline. Moreover, sympathectomy increased the responses of the heart rate both to β-phenylethylamine and to tyramine, and decreased the sensitivity of the heart rate toward amphetamine. By contrast, the responses of the systolic blood pressure were increased toward β-phenylethylamine, but were decreased toward both tyramine and amphetamine (Lockett, 1950a).

The results presented did not show whether differences existed in the responses of the heart rate and the systolic blood pressure to sympathomimetic amines before sympathectomy. Later evidence has shown that this is indeed the case, for the responses of the heart rate on the one hand, and of the systolic blood pressure on the other hand, to sympathomimetic amines are differently affected by pentobarbitone in dogs with intact nervous systems (Lockett, 1950b, c).

The analysis of the changes produced by sympathectomy in these two sets of responses to sympathomimetic amines was made difficult by the fact that, whereas measurement of the effect of sympathectomy on the responses of the heart rate was a measurement of change in one particular organ, the changes produced by sympathectomy in the responses of the systolic blood pressure were but the final integration of alterations throughout the whole varied cardiovascular system. Analysis was further complicated by the fact that total removal of the sympathetic chains might be expected to produce postganglionic division of the sympathetic fibres concerned with the regulation of the heart rate and of the nerves supplying the vascular bed, except in the splanchnic area. In the latter region, removal of the sympathetic chains should produce preganglionic arterial denervation. Therefore as a step in the analysis of former observations, a direct comparison has been made of the effects of preganglionic as contrasted with postganglionic denervation on the responses of a single tissue to these amines. Rosenblueth and Bard (1932) showed that the nictitating membrane in the cat is innervated solely by the homo-

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lateral cervical sympathetic fibres, which form a synapse in the superior cervical ganglion. Hence, because of the ease and certainty with which either preganglionic or postganglionic denervation can be effected, the nictitating membrane was selected for this study.

METHODS

Operative.—Operations for the denervation of nictitating membranes were performed aseptically, under pentobarbitone anaesthesia. Postganglionic denervation was produced by the removal of the superior cervical ganglion. Preganglionic denervation was effected by division of the trunk of the cervical sympathetic approximately one inch proximal to the superior cervical ganglion.

Experimental.—The cats were anaesthetized with ether, the dura was opened at the level of the atlanto-occipital membrane, the lowest part of the hindbrain was cut across, and the brain was pithed in the usual way. The carotid arteries were occluded only for the 15–20 seconds required for the destruction of the brain. Artificial respiration was necessary. The technique described provided nictitating membranes supplied with blood at a mean arterial pressure of 62 mm. Hg with a standard deviation of a single observation of 9 mm. in 27 cats (62 mm. Hg, S.D. 8 (27)). This mean arterial pressure is greater than that normally encountered when the cord is divided at the level of the second cervical spine, as when Dale's classical method of spinalization is employed.

Two hours were allowed for the animal to recover from the anaesthetic. The suprarenal glands were removed, and the cervical sympathetic nerve was divided on the side of any normal membrane, 10 minutes before the experiment began, to prevent impulses passing from the cord to that membrane. Equally weighted levers, tension 5 g., yielding similar magnification ($\times 16$), were used throughout to record responses from the nictitating membranes. Blood pressure records were taken from a femoral artery, and injections were made by means of a cannula into the opposite femoral vein.

The amine salts used were: *l*-adrenaline (Burroughs Wellcome, Ltd.) dissolved in dilute hydrochloric acid, *dl*-noradrenaline hydrochloride (kindly supplied by Dr. M. L. Tainter), tyramine acid phosphate (Burroughs Wellcome, Ltd.), β-phenylethylamine hydrochloride (Roche Products, Ltd.), amphetamine sulphate, and ephedrine hydrochloride.

RESULTS

1. *The changes produced by preganglionic and postganglionic denervation in the sensitivity of the nictitating membrane to the intravenous injection of* l-*adrenaline and* dl-*noradrenaline.*

In a series of 23 cats, weighing from 1.9 to 2.4 kg., measurement was made of the threshold sensitivity of both nictitating membranes toward the intravenous injection of *l*-adrenaline and *dl*-noradrenaline, six to eight days after operation, by the methods described. In some animals one normal membrane was compared with the other membrane which had undergone either preganglionic or postganglionic denervation; in other animals both membranes had been subjected the one to preganglionic and the other to postganglionic denervation in a single previous operation. In all, observations were made on 15 normal nictitating membranes, 18 membranes after preganglionic and 12 membranes after postganglionic denervation (Table I).

The weight of amine salt required to produce a small but constant contraction of each type of nictitating membrane varied greatly between individual animals.

TABLE I

INTRAVENOUS DOSE IN μG. REQUIRED TO PRODUCE A THRESHOLD RESPONSE IN NORMAL AND DENERVATED NICTITATING MEMBRANES OF SPINAL CATS, 6–8 DAYS AFTER NERVE DIVISION

Normal membranes		Denervated membranes			
		Preganglionic		Postganglionic	
μg. *l*-adren. HCl	μg. *dl*-noradren. HCl	μg. *l*-adren. HCl	μg. *dl*-noradren. HCl	μg. *l*-adren. HCl	μg. *dl*-noradren. HCl
4.0	10.0			1.0	1.2
1.0	10.0			0.2	0.4
5.0	25.0			0.2	0.5
2.0	20.0			0.2	0.3
7.0	25.0	1.0	3.0		
3.0	20.0	1.0	3.0		
3.0	15.0	0.8	4.0		
3.0	16.0	1.0	3.0		
3.0	12.0	0.3	2.0		
4.0	18.0	1.0	4.0		
5.0	20.0	0.5	4.0		
4.0	15.0	0.2	0.7		
2.0	10.0	0.5	0.8		
2.0	12.0	0.2	1.0		
1.0	8.0	0.5	1.0		
		0.3	1.3	0.5	0.7
		0.5	3.0	0.5	0.7
		0.7	2.0	0.5	0.5
		0.08	2.0	0.1	0.25
		1.0	6.0	0.5	1.0
		0.3	3.0	0.5	0.8
		0.2	1.3	0.5	0.5
—	—			0.3	0.6

The mean weight of *l*-adrenaline hydrochloride which produced a minimal contraction of the normal nictitating membrane was 3.3 μg., S.D. 1.6 (15). The corresponding figures for *dl*-noradrenaline were 15.7 μg., S.D. 5.5 (15).

Preganglionic denervation produced an approximately sevenfold increase in the threshold sensitivity of nictitating membranes to both *l*-adrenaline and *dl*-noradrenaline by the sixth to eighth post-operative days. Both normal membranes and membranes after preganglionic denervation were therefore about four times as sensitive to *l*-adrenaline as to *dl*-noradrenaline.

After postganglionic denervation these membranes showed no greater sensitivity to *l*-adrenaline than did membranes after preganglionic denervation. The threshold doses were 0.43 μg., S.D. 0.23 (12) and 0.56 μg., S.D. 0.33 (18) respectively; $t=1.2$, $P=0.3$. Postganglionic denervation, however, rendered nictitating membrane more sensitive to *dl*-noradrenaline than did preganglionic denervation. The mean threshold doses were 2.51 μg., S.D. 1.4 (18) and 0.62 μg., S.D. 0.28 (12) respectively; $t=4.5$, $P=0.001$. If the activity of *dl*-noradrenaline be considered due solely to its content of the laevorotatory isomer, then there was no significant difference in the sensitivity of nictitating membrane after postganglionic denervation toward *l*-adrenaline on the one hand and *dl*-noradrenaline on the other: $t=1.5$, $P=0.2$.

The wide variations in the sensitivity of the nictitating membranes of individual animals toward adrenaline and noradrenaline made results in this small series of cats inaccurate. If, however, only those seven cats are considered in which preganglionic denervation of the one and postganglionic denervation of the other nictitating membranes were performed in one operation, and in which the responses of these two membranes were simultaneously compared six to eight days later, the foregoing observations are confirmed. First, there is no difference in the sensitivity of these two types of membrane to *l*-adrenaline, the threshold doses being 0.44 μg., S.D. 0.3 (7) after postganglionic, and 0.44 μg., S.D. 0.2 (7) after preganglionic denervation. Secondly, postganglionic denervation has again produced a membrane four times as sensitive to *dl*-noradrenaline as has preganglionic denervation; the threshold doses were 0.64 μg., S.D. 0.2 (7) and 2.66 μg., S.D. 1.0 (7) respectively; $t=5.3$, $P=0.001$.

2. *The changes produced by preganglionic and postganglionic denervation in the sensitivity of nictitating membranes toward tyramine, β-phenylethylamine, amphetamine, and ephedrine.*

Preganglionic denervation resulted in membranes more sensitive, and postganglionic denervation in membranes less sensitive, than normal membranes toward tyramine, β-phenylethylamine, amphetamine, and ephedrine.

Tyramine and β-phenylethylamine.—Fifteen of the previous series of 23 cats were used to determine the changes in sensitivity of nictitating membrane to tyramine acid phosphate and β-phenylethylamine hydrochloride that resulted from denervation. The threshold dose of tyramine acid phosphate for normal membranes was 1.94 mg., S.D. 0.86 (8). Six to eight days after preganglionic denervation the corresponding figure was 0.8 mg., S.D. 0.12 (11). Preganglionic denervation had therefore produced membranes more sensitive to tyramine than normal membranes ($t=3.6$, $P=0.01$). Similarly the threshold doses of β-phenylethylamine hydrochloride were 3.0 mg. S.D. 1.4 (8) for normal membranes, and 1.13 mg. S.D. 0.6 (11) for membranes after preganglionic denervation ($t=12.3$, $P=0.001$). In addition three normal membranes failed to respond to salts either of tyramine or of β-phenylethylamine in doses up to 6 mg., and are excluded from these calculations.

The weights of amine salts required to produce threshold responses in paired membranes, one denervated by preganglionic and the other by postganglionic section of the cervival sympathetic, were compared in seven cats. The results are collected in Table II. Absence of response, delayed or biphasic responses to tyramine were obtained in membranes after postganglionic denervation as described by Bülbring and Burn (1938). Similar types of response to β-phenylethylamine were recorded from these membranes, but the responses of single membranes after postganglionic denervation to these two amines were frequently dissimilar.

Amphetamine and ephedrine.—The threshold doses of amphetamine sulphate and ephedrine hydrochloride were determined for normal and denervated membranes. Such tests were made only after those with other amines had been completed; either amphetamine or ephedrine was used, for both were not tested in the same animal.

The mean threshold doses for membranes 6–8 days after preganglionic denervation were 0.53 mg. S.D. 0.12 (6) for amphetamine sulphate and 0.78 mg. S.D. 0.23

TABLE II

INTRAVENOUS DOSE IN MG. REQUIRED TO PRODUCE A THRESHOLD RESPONSE IN PAIRED NICTITATING MEMBRANES OF SPINAL CATS, 6 TO 8 DAYS AFTER PREGANGLIONIC DENERVATION OF ONE, AND POSTGANGLIONIC DENERVATION OF THE OTHER NICTITATING MEMBRANE.

Dose which produced no response in parentheses.

Cat	mg. tyramine ac. phosph.		mg. β-phenylethylamine HCl	
	Postganglionic	Preganglionic	Postganglionic	Preganglionic
1	(3)	2.0	(3.0)	2.0
2	(3)	1.5	(2.0)	2.0
3	(2)	0.4	(2.0)	0.4
4	Delayed (0.8	0.4	(3.0)	1.0
5	response 1.5	0.6	Biphasic 2.0	0.6
6	0.6	0.2	delayed 1.0	0.4
7	Biphasic 1.0	0.5	1.5	0.4

mg. Ephedrine HCl		*mg. Amphetamine sulphate*		
Postganglionic	*Preganglionic*	*Postganglionic*	*Preganglionic*	
(4.0)	0.6	Irregular 1.2	0.8	Smooth contraction
(4.0)	0.8	jerking 2.0	0.4	
(4.0)	0.6	movements 1.0	0.4	

(6) for ephedrine hydrochloride injected intravenously. In each experiment double the dose which produced a small contraction of the one membrane which had been subjected to preganglionic denervation still failed to produce contraction of the opposite normal membrane.

The responses of one membrane after preganglionic denervation were then compared with those of the other membrane after postganglionic denervation in three experiments with each of these two amines. The results are summarized in Table II. Nictitating membranes after postganglionic denervation were much less sensitive to these amines than were membranes after preganglionic denervation. In four of a total of five postganglionically denervated membranes, no response to ephedrine was obtained with intravenous doses up to 4 mg. of ephedrine hydrochloride; a single contraction was given by a fifth membrane to an initial dose of 2.0 mg., but not thereafter to this or to larger doses. In five membranes after postganglionic denervation, 1 to 3 mg. of amphetamine sulphate produced a series of irregular jerks, unaccompanied by any sustained contraction of the nictitating membrane.

3. *The slopes of the log-dose response curves for the action of intravenous sympathomimetic amines on the responses of normal nictitating membranes, membranes after preganglionic and postganglionic denervation, and of the mean arterial blood pressure.*

Whatever may have been the mechanism by which pre- and post-ganglionic denervation produced change in the responses of the nictitating membrane to sympathomimetic amines, an alteration in threshold sensitivity toward a drug could only be regarded as an approximate measure of this change. Should denervation cause not only a shift in the log-dose response curve, but also a pronounced change in the lower slope of this curve, such as has been encountered in the upper slopes of such curves (Lockett, 1950a), then comparison of threshold doses would become a very inaccurate measure of change in sensitivity. The slopes of the lower part of the log-

dose response curves for these amines were therefore determined in normal and denervated membranes.

l-*Adrenaline.*—The log-dose response curves relating size of contraction to intravenous log-dose of *l*-adrenaline hydrochloride for normal nictitating membranes at a tension of 5 g., from five spinal cats, are shown in Fig. 1A. Similar curves were obtained from one nictitating membrane after preganglionic, and the other after postganglionic denervation, and from the mean arterial pressure, in five other spinal cats: these curves are illustrated in Figs. 1B, C, and D respectively. Fig. 1 shows, first, that no gross change is produced in the lower slope of the log-dose response curves for *l*-adrenaline by denervation of the nictitating membranes; secondly, that the sensitivity of nictitating membranes to *l*-adrenaline, 6–8 days after either preganglionic or postganglionic denervation, is of the same order as that of a normally innervated cardiovascular bed.

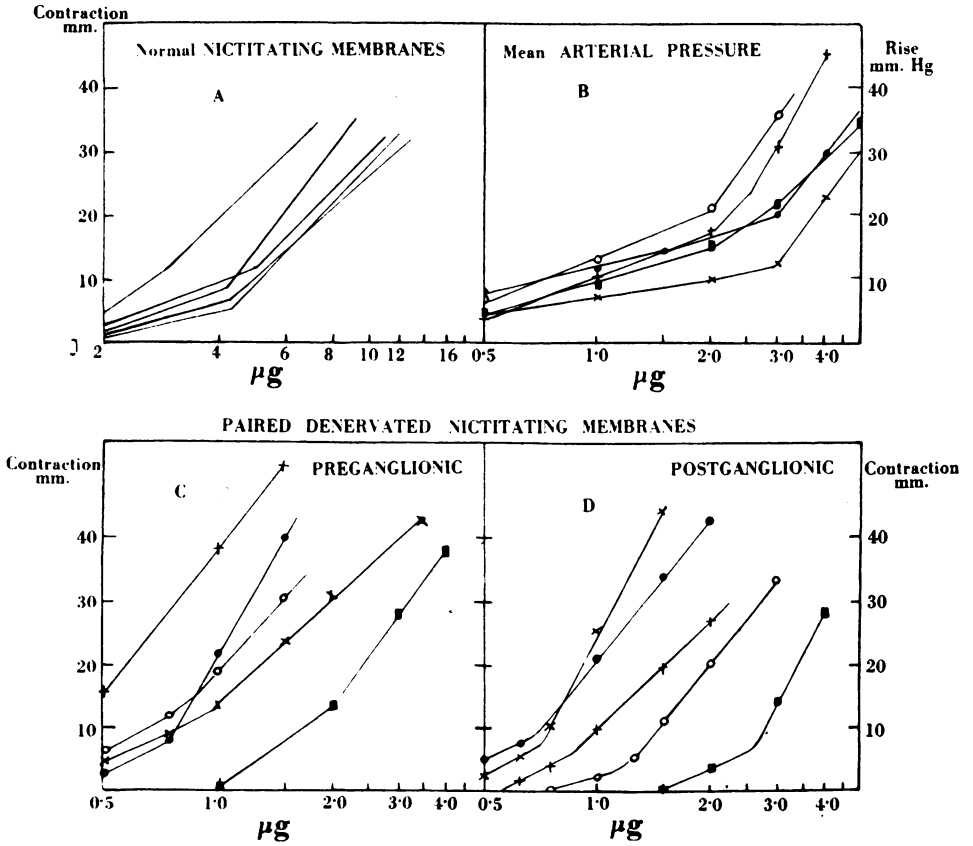

FIG. 1.—Dose response curves for the action of intravenous *l*-adrenaline hydrochloride on the mean arterial blood pressure (B), normal nictitating membrane (A), and on nictitating membranes 6–8 days after preganglionic denervation (C) and postganglionic denervation (D). For description, see text. Abscissa: μg. *l*-adrenaline hydrochlorides. Ordinates: A, C, and D, recorded contraction of nictitating membrane in mm.; B, rise of arterial pressure in mm. Hg.

dl-*Noradrenaline.*—Log-dose response curves for the action of intravenous *dl*-noradrenaline hydrochloride on normal nictitating membranes of five cats are shown in Fig. 2A. Corresponding curves from one membrane after preganglionic and another membrane after postganglionic denervation, and of the mean femoral arterial blood pressure of five other spinal cats, are shown in Figs. 2B, C, and D respectively. The animals used in these experiments were those employed for the corresponding experiments with *l*-adrenaline (Fig. 1). Fig. 2 shows first that denervation did not markedly alter the lower slopes of the log-dose response curves for the action of noradrenaline on nictitating membrane, but the extent of this slope appeared to be reduced by denervation. Secondly, only after postganglionic denervation of the nictitating membrane was the sensitivity to noradrenaline of this membrane and of the mean arterial pressure comparable in these spinal animals. It was also fre-

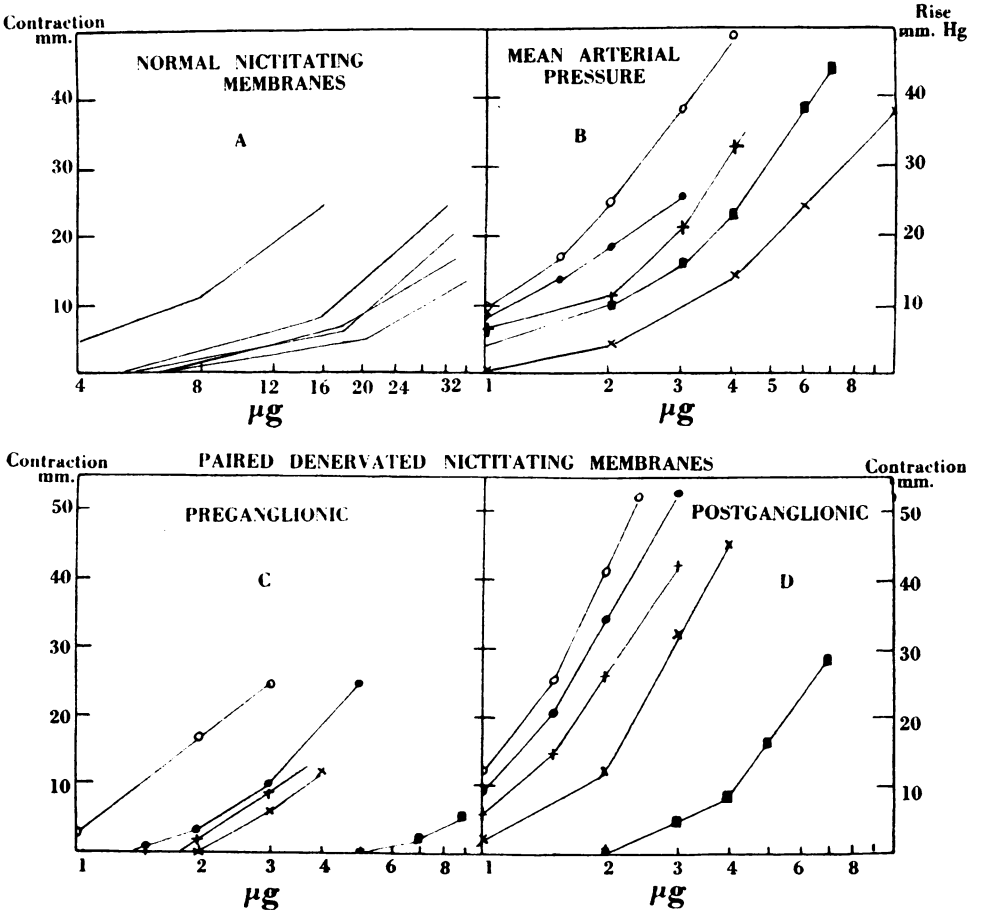

FIG. 2.—Dose response curves for the action of intravenous *dl*-noradrenaline hydrochloride on the mean arterial pressure (B), normal nictitating membrane (A), and on nictitating membranes 6–8 days after preganglionic denervation (C) and postganglionic denervation (D). For description see text. Abscissa and ordinates as in Fig. 1.

quently noted that *dl*-noradrenaline caused a reduction in the tone of nictitating membranes more especially after preganglionic denervation.

Tyramine and β-phenylethylamine.—Corresponding experiments with tyramine and β-phenylethylamine salts showed that preganglionic denervation caused a shift to the left, without marked change in the lower slopes, of the log-dose response curves for the action of these amines on nictitating membranes.

4. *Examination of the responses of (a) normal nictitating membranes, and (b) membranes after preganglionic denervation, to sympathomimetic amines, after the following procedures: emptying the eyeballs; use of atropine; excision of superior cervical ganglia; freezing of superior cervical ganglia.*

Changes produced by preganglionic denervation in the responses of nictitating membrane to sympathomimetic amines are likely to be the sum of the effects of changes in the ganglion, in the membrane, and in the smooth muscle of the orbit. Emptying of the eyeballs, as described by Cannon and Rosenblueth (1939), caused

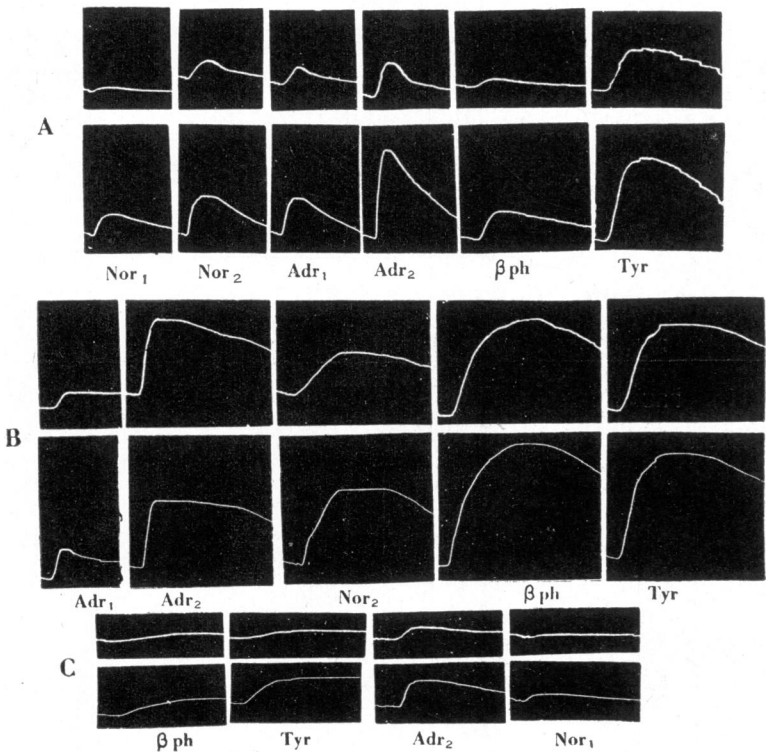

FIG. 3.—Contractions of a normal nictitating membrane above, and a membrane 3 days after preganglionic denervation below, to 10 μg and 15 μg. *dl*-noradrenaline-HCl (Nor_1 and Nor_2), 2 μg. and 4 μg. *l*-adrenaline-HCl (Adr_1 and Adr_2), 1 mg. β-phenyl-ethylamine-HCl (β-ph), and 1 mg. tyramine ac. phos. (Tyr.). Both superior cervical ganglia removed between A and B. 4 mg. atropine sulphate injected between B and C. All injections intravenous. Spinal cat, 2.6 kg.

little or no change in the responses recorded from normal or denervated membranes in these experiments. The influence of enophthalmos or exophthalmos was therefore unimportant in these experiments, as in the experiments of the previous authors.

Atropine.—Cannon and Rosenblueth (1939), working with cats under dial anaesthesia, used both atropine and curare to antagonize the effects of acetylcholine on the nictitating membrane, and noted that atropine had no greater effect on the responses to adrenaline of nictitating membrane after preganglionic denervation than on normal membrane. Their observation has been confirmed and extended. Atropine, in a dose of 1 to 2 mg./kg., reduced the responses to adrenaline, noradrenaline, tyramine, β-phenylethylamine, amphetamine, and ephedrine, in normal nictitating membranes and in membranes after both pre- and postganglionic denervation. There was, however, no apparent difference in the effect of atropine on the responses of the individual amines, neither were normal membranes or either type of denervated membrane differently affected by atropine. Fig. 3 shows that the effect of atropine was on the membrane itself, and that the responses of nictitating membranes, normal and after preganglionic denervation, were similarly reduced.

Excision and freezing of superior cervical ganglia.—Cannon and Rosenblueth (1939) showed that acute removal of the superior cervical ganglion (S. C. G.) caused an equal reduction in the responses to acetylcholine of normal nictitating membranes and nictitating membranes after preganglionic denervation. In contrast to these previous observations for acetylcholine, in each of four experiments with paired nictitating membranes one normal and the other 6–8 days after preganglionic denerva-

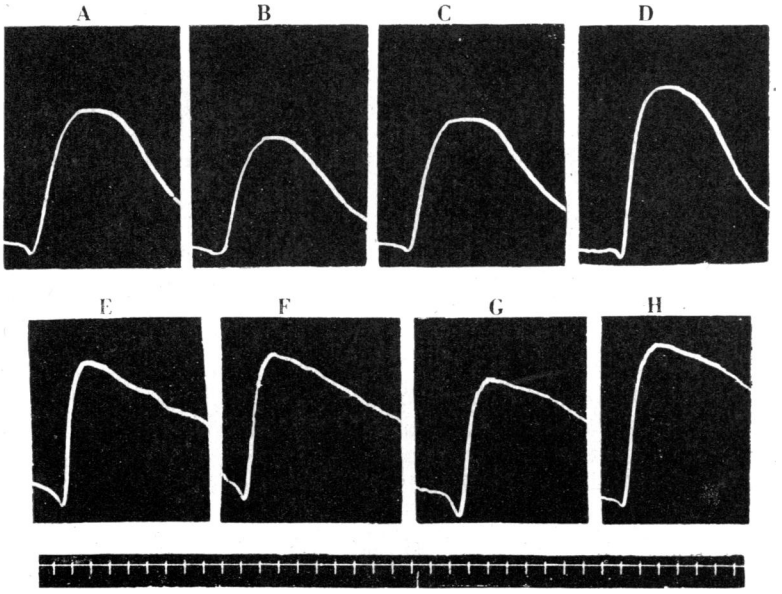

FIG. 4.—Contractions of nictitating membranes of a spinal cat. Above, normal membrane responding to 4 μg. *l*-adrenaline-HCl, intravenous. Homolateral superior cervical ganglion frozen during contraction B. Below, membrane 6 days after preganglionic denervation responding to 0.8 μg. *l*-adrenaline HCl intravenous. Homolateral superior cervical ganglion frozen during contraction F. Time marker, 30 secs.

tion, gentle excision of the S.C.G. produced approximately equal increase in the responses of these two membranes toward adrenaline, noradrenaline, tyramine, β-phenylethylamine (Fig. 3) and also to amphetamine. These changes developed within 15 minutes of excision of the S.C.G., and remained constant, or increased, during subsequent periods of observation, varying from one to four hours. The origin of this increased sensitivity to sympathomimetic amines may well have been traumatic, since in each of four experiments temporary local freezing of the homolateral S.C.G. resulted in a reversible moderate reduction in the responses to adrenaline and to noradrenaline of both normal nictitating membranes and membranes 6–8 days after preganglionic denervation (Fig. 4).

Discussion

In spinal cats, normal nictitating membranes are markedly less sensitive to intravenous *l*-adrenaline and *dl*-noradrenaline than is the mean arterial pressure. Greater accessibility of the vascular bed than of the nictitating membrane to the action of these amines might be offered in explanation. Such a hypothesis would not explain the fact that whereas in the spinal cat the ratio of the threshold sensitivity to *l*-adrenaline and *dl*-noradrenaline in the normal nictitating membrane is of the order of 1 to 4, the corresponding ratio for the arterial blood pressure is rather less than 1 to 2 (Figs. 1 and 2). The rates of diffusion of such molecules could not differ greatly. Whether sensitivity to these amines be dependent on one or many factors such as molecular configuration for selective adsorption, cell permeability, rate of destruction of amines at the site of action, etc., these different ratios suggest the probability of widely different receptor systems linked with the sympathetic nervous system. Such an interpretation might also be placed upon the dissimilar effects produced both by sympathectomy (Lockett, 1950a) and by anaesthesia (Lockett, 1950b and c), on the heart rate as contrasted with responses of the systolic blood pressure to these amines. It is certainly not possible to explain the differences produced by sympathectomy in the responses of the heart rate to sympathomimetic amines as contrasted with those of the systolic blood pressure by the theory that a postganglionic denervation was performed on the heart, and a mixed preganglionic and post-ganglionic denervation on the vascular bed, identical receptor systems throughout the periphery of the sympathetic nervous system being assumed.

Preganglionic denervation.—Increased sensitivity of nictitating membrane to adrenaline, acetylcholine, and potassium ions has long been known to follow preganglionic denervation (Cannon and Rosenblueth, 1936). In a previous note attention was drawn to the fact that preganglionic denervation increased the sensitivity of this membrane to adrenaline, noradrenaline, tyramine, β-phenylethylamine, amphetamine, and ephedrine (Lockett, 1949). Superficially, a nonspecific increase in sensitivity following chronic decentralization may most easily be explained as a release from a tonic inhibitory action of central sympathetic origin, as has been postulated by Swan (1949). The effects of preganglionic denervation may arise from changes in the ganglion or in the membrane, or at both sites. Whereas the evidence presented by Cannon and Rosenblueth, 1939, stresses the ganglionic origin of the increased sensitivity of the nictitating membrane to acetylcholine following preganglionic denervation, the experiments recorded in this paper suggest a stronger

peripheral than ganglionic effect in the increased sensitivity of this membrane to sympathomimetic amines after preganglionic denervation. Were the increase in sensitivity solely due to release from central inhibitory influences that increase should affect the responses to all amines similarly, provided that they produce contraction of the nictitating membrane through a common receptor-response system. However, 6–8 days after preganglionic denervation, a sevenfold increase in the sensitivity of nictitating membrane was found to both adrenaline and noradrenaline, but the increase in sensitivity to tyramine and β-phenylethylamine was but two- or threefold, that to amphetamine and ephedrine more than twofold. Either these amines are active through multiple receptor response systems (Lockett, 1950d), or the theory of tonic central inhibition must be discarded.

Postganglionic denervation.—Burn and Tainter (1931) showed that denervation abolished the dilator effect of tyramine on the pupil. Burn (1932) demonstrated that after removal of the stellate ganglion the constrictor effect of tyramine and ephedrine disappeared from the forearm ; and further, that removal of the superior cervical ganglion abolished the dilator action of tyramine and ephedrine on the pupil. Bülbring and Burn (1938) found that nictitating membranes varied considerably in the responses given to tyramine. Whilst postganglionic denervation usually increased the effect of small doses but decreased the effect of large doses of tyramine on the nictitating membrane, in some membranes postganglionic denervation abolished all response to tyramine. The observations recorded in this paper are compatible with and extend the findings of Bülbring and Burn. Postganglionic denervation has been found to produce membranes which did and membranes which did not respond to tyramine ; membranes after postganglionic denervation had higher threshold sensitivity to tyramine than had membranes after preganglionic denervation ; preganglionic denervation resulted in membranes more sensitive to tyramine than were normal membranes.

In 1948 Bülbring and Burn showed that postganglionic denervation of the nictitating membrane produced greater increase in sensitivity of the membrane to *dl*-noradrenaline than to adrenaline. This observation has been confirmed and extended. Burn and Hutcheon (1949) showed that postganglionic denervation of the pupil also produced greater increase in sensitivity to noradrenaline than to adrenaline. They suggested that in some tissues the presence of the sympathetic nerve supply protects the end organ against the action of noradrenaline in the blood stream, although not protecting it from the action of adrenaline.

By contrast to the nonspecific type of sensitization that follows preganglionic denervation of nictitating membrane, that following postganglionic denervation was found to be highly specific. In both normal membranes and those after preganglionic denervation, *l*-adrenaline was at least four times as active as *dl*-noradrenaline in provoking contraction. After postganglionic denervation there was no significant difference in the sensitivity of this membrane to *l*-adrenaline on the one hand and *dl*-noradrenaline on the other hand. Whereas there was no difference in the sensitivity of nictitating membrane to *l*-adrenaline after either preganglionic or postganglionic denervation, membranes after postganglionic denervation were more sensitive to noradrenaline than were those after preganglionic denervation. Sensitivity to tyramine, β-phenylethylamine, amphetamine, and ephedrine were reduced or lost after postganglionic denervation of this membrane.

Bülbring and Burn (1949) have shown that noradrenaline can be converted to adrenaline in the perfused suprarenal gland. Since the sympathetic innervation of this gland is preganglionic, it might be suggested that such a methylation is under the influence of the postganglionic sympathetic neurone, and that severance of the ganglion from the termination of the sympathetic fibres deflects an equilibrium of enzyme action and greatly increases the rate of methylation of noradrenaline to adrenaline. Such an explanation assumes some enzyme actions to be under sympathetic control.

Summary

1. The effects of preganglionic and postganglionic denervation on the threshold sensitivity and on the lower slopes of log-dose response curves for the action of intravenous salts of sympathomimetic amines on nictitating membrane have been measured in spinal cats. Normal membranes were compared with denervated membranes. The amine salts were *l*-adrenaline, *dl*-noradrenaline, β-phenylethylamine, and ephedrine hydrochlorides, tyramine acid phosphate and amphetamine sulphate.

2. Preganglionic denervation produced, after 6–8 days, a sevenfold increase in the threshold sensitivity of nictitating membrane to adrenaline and to *dl*-noradrenaline, but a two to threefold increase in sensitivity to β-phenylethylamine and to tyramine, and more than a twofold increase in sensitivity to amphetamine and ephedrine. Preganglionic denervation did not cause marked changes in the lower slopes of the log-dose response curves for the actions of these amines on nictitating membrane.

3. Postganglionic denervation resulted, after 6–8 days, in a sevenfold increase in the threshold sensitivity of nictitating membrane to *l*-adrenaline, a twentyfold increase in threshold sensitivity to *dl*-noradrenaline, and decreased sensitivity toward tyramine, β-phenylethylamine, amphetamine, and ephedrine.

4. Whereas normal membranes and membranes after preganglionic denervation were four times as sensitive (threshold dose) to *l*-adrenaline as to *dl*-noradrenaline, no significant difference was found between the threshold sensitivity of membranes 6–8 days after postganglionic denervation toward adrenaline and noradrenaline.

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