Brit. J. Pharmacol. (1958), 13, 461.

THE ACTION OF SYMPATHOMIMETIC AMINES ON HEART RATE IN RELATION TO THE EFFECT OF RESERPINE

BY

D. BEJRABLAYA, J. H. BURN AND J. M. WALKER

From the Department of Pharmacology, University of Oxford

(RECEIVED AUGUST 1, 1958)

When the heart-lung preparation is made from a dog treated with reserpine, catechol amines such as noradrenaline and isoprenaline have a greater effect on the rate of the heart than they have in a preparation from a normal dog. Other sympathomimetic amines such as tyramine and ephedrine, on the other hand, are found to have lost their action. Since treatment with reserpine has been shown to cause the store of noradrenaline in the heart to disappear, and the infusion of noradrenaline into the preparation made from a reserpine-treated animal restores the action of tyramine, it is concluded that substances like tyramine and ephedrine normally act by liberating noradrenaline from the store, and do not act directly. Cocaine, like reserpine, increases the effect of noradrenaline and decreases the effect of tyramine on the heart rate; it appears to block the release of noradrenaline from the store in the heart.

Observations have recently been made (Burn and Rand, 1958c) on the action of sympathomimetic amines on the blood pressure of the spinal cat and on the vessels of the perfused hindleg of the dog. Experiments were performed on normal animals, and on animals treated with reserpine. In animals treated with reserpine the effect of the catechol amines such as noradrenaline, adrenaline and dopamine was greater than usual, the tissues being supersensitive to them. However, the effect of other amines such as tyramine and ephedrine was almost abolished. Since it had been shown (Burn and Rand, 1957, 1958b) that reserpine causes the dispersal of the noradrenaline in the vessel wall, the conclusion was drawn that tyramine and similar amines act by releasing noradrenaline, and that they differ from the catechol amines which act directly.

We now describe a study of the action of sympathomimetic amines on the heart rate.

METHOD

Observations were made in the heart-lung preparation of the dog, at a temperature between 36.5° and 37°. One dog was bled under ether anaesthesia. The second dog was first anaesthetized with ether and chloralose was then injected intravenously. The heart-lung preparation was then made. The venous reservoir was immersed in a thermostatically controlled water bath, with an overflow from the reservoir so that the venous pressure was constant. The heart rate was measured from the electrocardiogram recorded on a Cossor electrocardiogaph model 1314. The artificial resistance was set to maintain a pressure of 110 mm. and the systemic outflow was usually about 750 ml./min. Dogs usually weighed 12 to 15 kg. Those treated with reserpine were given 5 mg. on two successive days by intraperitoneal injection and were used on the third day. The solution of reserpine was prepared in 20% (w/v) ascorbic acid.

RESULTS

Tyramine and Phenylethylamine.—Tyramine hydrochloride was tested, as were the other amines, by injecting a dose into the tube carrying blood to the superior vena cava and observing the height and duration of the rise in heart rate which

TABLE I

EFFECT OF TYRAMINE AND PHENYLETHYLAMINE ON HEART RATE IN THE DOG HEART-LUNG PREPARATION

Substance	Dose (mg.)	Untreated		Reserpine-treated	
		Increase in Rate (Beats/Min.)	Duration of Effect (Min.)	Increase in Rate (Beats/Min.)	Duration of Effect (Min.)
Tyramine ..	0·4	25	19	2	4
	0·4	21	20	—	—
	0·8	—	—	2	3
	1·0	86	36	—	—
	1·0	55	41	—	—
	1·0	82	34	—	—
	2·0	—	—	25	22
	2·0	—	—	7	13
Phenyl-ethylamine	0·4	11	2	—	—
	1·0	67	14	—	—
	1·0	76	8	—	—
	1·0	50	6	—	—
	2·0	—	—	34	11
	2·0	—	—	10	4

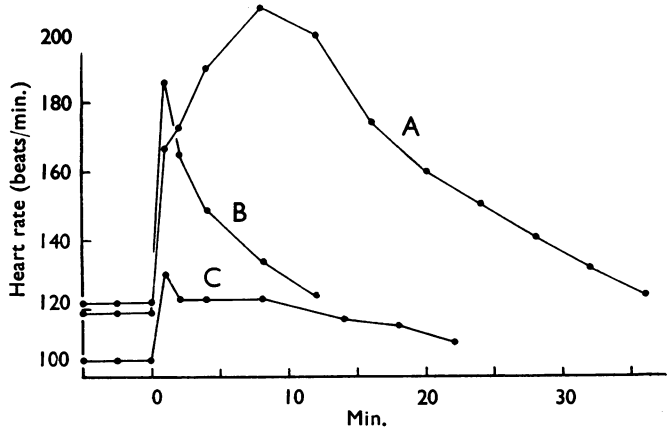

FIG. 1.—Dog heart-lung preparation. The effect upon the heart rate of injection at time 0 min. of 1 mg. of tyramine (A) and 1 mg. of phenylethylamine hydrochloride (B). The effect of 2 mg. of tyramine in a dog treated with reserpine is shown at C.

followed. As the example in Fig. 1 shows, the rise did not reach its maximum until some minutes after the injection, and it lasted for 30 to 40 min. when the dose was 1 mg. The results of observations in different experiments are shown in Table I. When the dog was treated with reserpine beforehand, the effect of tyramine was greatly reduced. The largest effect is shown in Fig. 1 **C**, and was made after the injection of 2 mg.

The rise produced by phenylethylamine differed in all observations from that produced by tyramine in reaching its maximum point in 1 min., and then rapidly declining so that the effect was finished in 10 min. The action of phenylethylamine also was much smaller in dogs treated with reserpine.

Amphetamine. — The injection of amphetamine was followed by a rise in heart rate which seemed to persist indefinitely. How much more prolonged it was in comparison with the rise due to tyramine can be seen in Fig. 2, in which an injection of 0.4 mg. of tyramine (**A**) was followed by an injection of 0.4 mg. of amphetamine (**B**). The rise caused by tyramine was over in 19 min. while the rise caused by amphetamine was still half its maximum value after 2.5 hr. In another experiment an injection of only 0.1 mg. of amphetamine caused a rise which was still half its maximum value after 1 hr. In a reserpine-treated dog, the injection of 0.4 mg. of amphetamine caused only a trace of effect on the heart rate as shown in Fig. 2 **C**.

The Phenylethanolamines. — We were able to test both optical isomers of phenylethanolamine, having samples prepared by Professor P. Pratesi of Pavia (Pratesi and Grassi, 1953). When they were compared in a normal preparation, they differed in duration of action, the effect of (−)-phenylethanolamine passing off more quickly as shown in Fig. 3. Burn and Rand (1958c) observed a similar difference on the blood pressure. In a preparation from a reserpine-treated dog, the effect of both was greatly reduced, though the injection of (−)-phenylethanolamine produced a somewhat greater effect than the other isomer, and this again recalled a similar difference on the blood pressure.

Ephedrine.—Ephedrine is known to produce a prolonged rise in heart rate (Krayer and Ourisson, 1954). Axelrod (1953) has shown that it is converted in the body into norephedrine, which is the active substance and which differs only from (−)-phenylethanolamine in having a methyl group on the α carbon atom. The relation between these two substances is therefore the same as the relation between amphetamine and phenylethylamine. The prolonged action of ephedrine and of amphetamine can therefore be ascribed to the presence of this methyl group which interferes with their destruction by amine oxidase. Fig. 4

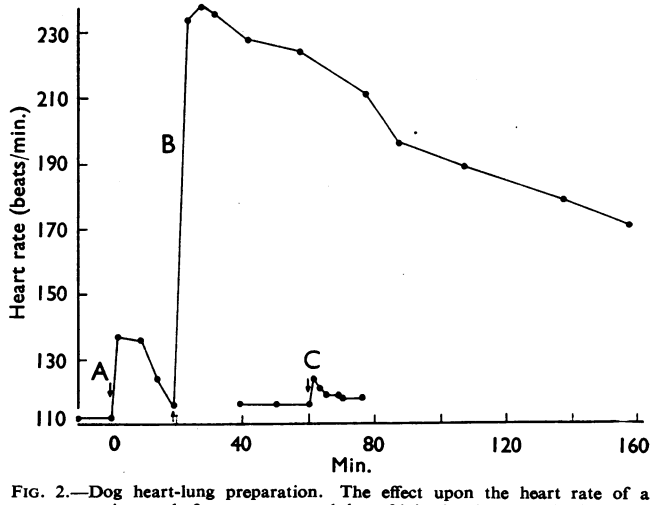

FIG. 2.—Dog heart-lung preparation. The effect upon the heart rate of a preparation made from an untreated dog of injection (at arrows) of 0.4 mg. of tyramine (A) and 0.4 mg. of amphetamine (B). The response to 0.4 mg. of amphetamine in a preparation made from a dog previously treated with reserpine is shown by curve C.

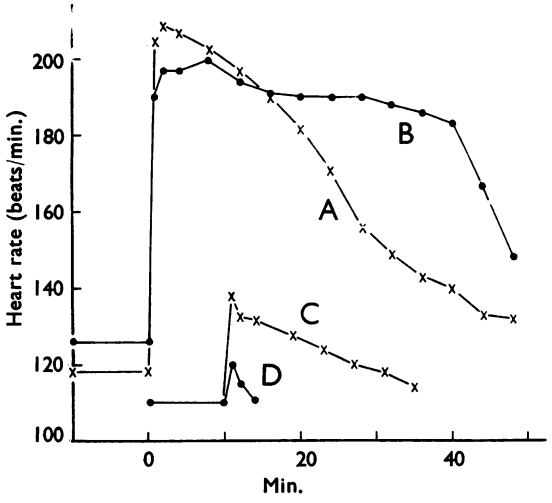

FIG. 3.—Dog heart-lung preparation. The effect on the heart rate of a preparation made from an untreated dog of 2 mg. of (−)-phenylethanolamine (A) and 2 mg. of (+)-phenylethanolamine (B). The responses of a preparation made from a dog previously treated with reserpine to 2 mg. of (−)-phenylethanolamine and 2 mg. of (+)-phenylethanolamine are shown by curves C and D, respectively.

shows a comparison between the effect on the heart rate of 0.4 mg. of (−)-phenylethanolamine and that of 0.4 mg. of ephedrine which was given immediately after. The effect of ephedrine was still half the maximal value after 2 hr. In the dog treated with reserpine, the injection of 1 mg. of ephedrine had no appreciable effect (Fig. 4 **C**).

Infusion of Noradrenaline.—When a cat was treated with reserpine, the injection of tyramine into the spinal preparation caused little or no rise of blood pressure. If, however, a slow intravenous infusion of noradrenaline was given during a period of 15 or 20 min., the total amount infused being from 0.5 to 1.0 mg., then when the infusion was stopped and the blood pressure had returned to the level at which it was before the infusion began, the injection of tyramine caused a rise of blood pressure (Burn and Rand, 1958c). We carried out experiments to see if a similar effect was to be observed in the heart-lung preparation. Having first established that tyramine had little effect on the heart rate of a preparation from a reserpine-treated dog, we infused noradrenaline slowly into the superior vena cava until a given amount had entered the blood. The infusion caused a rise in heart rate, and when the infusion was complete we waited until the heart rate returned to the rate at which it was before the infusion began. We then determined the effect of the same dose of tyramine again. In one experiment, the injection of 0.4 mg. of tyramine caused a rise of only 15 beats/min. The infusion of 2 mg. of noradrenaline was then made during 18 min. The rate returned to the pre-infusion level after 1.75 hr. The injection of 0.4 mg. of tyramine then caused a rise of 53 beats/min.

In the experiment illustrated in Fig. 5, smaller amounts of noradrenaline were infused, and the initial rise in rate was not affected. However, the duration of the effect of injecting 0.4 mg. of tyramine was increased by a first infusion of 0.25 mg. of noradrenaline (Fig. 5 curve **B**) and was still further increased by a second infusion of 0.5 mg. of noradrenaline (Fig. 5 curve **C**).

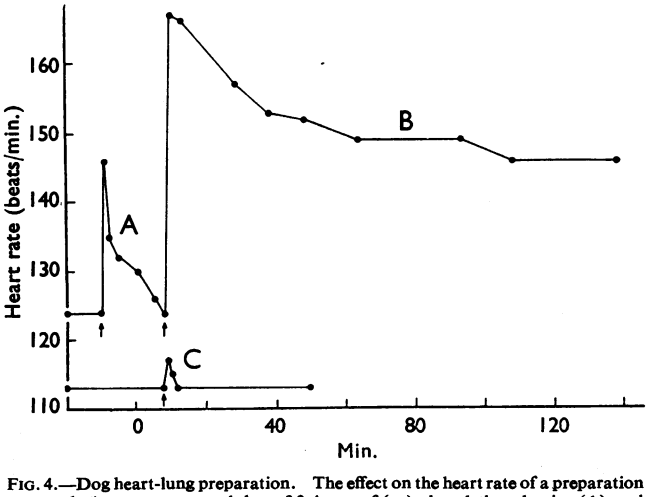

FIG. 4.—Dog heart-lung preparation. The effect on the heart rate of a preparation made from an untreated dog of 0.4 mg. of (−)-phenylethanolamine (A) and 0.4 mg. of ephedrine (B). The response in a preparation made from a dog previously treated with reserpine to 1 mg. of ephedrine is shown by curve C.

Catechol Amines.—Though the pressor effect of tyramine and related amines was very small in the reserpine-treated cat, the pressor effect of the catechol amines was very large. Comparisons were therefore made between the action of noradrenaline, dopamine and isoprenaline in the heart-lung preparation from an untreated dog and their action in the preparation from a reserpine-treated dog. The action of noradrenaline and isoprenaline was greater in the reserpine-treated preparation, and the action of dopamine was reduced. The details of all observations are given in Table II, which shows that the greater effect of noradrenaline was always seen. An example is shown on the right of Fig. 6. In observations on the blood pressure the greater effect of noradrenaline in the reserpine-treated cat was reduced after an intravenous infusion

of noradrenaline. This was also true in the heart-lung preparation. Thus the rise of 74 beats/min. caused by the injection of 4 μg. of noradrenaline in a reserpine-treated preparation was reduced after an infusion of noradrenaline to a rise of only 8 beats/min. Similarly the rise of 42 beats/min. produced in another experiment was reduced by an infusion of noradrenaline to a rise of only 18 beats/min.

TABLE II

INCREASE IN HEART RATE CAUSED BY CATECHOL AMINES IN THE DOG HEART-LUNG PREPARATION

Substance	Dose	Normal		Reserpine	
		Increase (Beats/Min.)	Mean	Increase (Beats/Min.)	Mean
Noradrenaline ..	1 μg.	0, 5, 9	4·7	11, 25, 18	18·0
,, ..	2 μg.	6, 10, 16, 14	11·5	18, 19, 29	22·0
,, ..	4 μg.	15, 31, 22	22·7	74, 42	53·0
Dopamine ..	0·1 mg.	19, 10	14·5	3, 7, 3	4·3
,, ..	0·2 mg.	57, 23	40·0	5, 18, 10	11·0
Isoprenaline ..	0·5 μg.	20		23	
,, ..	2 μg.	53		80	

While the action of dopamine on the vessels resembled that of noradrenaline, being increased in the reserpine-treated preparation, this was not true in the heart-lung preparation. The effect of dopamine was less, as shown in Table II. Moreover after an infusion of noradrenaline the action of dopamine was increased. Thus the injection of 0.1 mg. of dopamine caused a rise of only 7 beats/min. in one reserpine-treated preparation. After an infusion of noradrenaline, 0.1 mg. of dopamine caused a rise of 32 beats/min. Similarly in a second preparation the injection of 0.2 mg. of dopamine caused a rise of 10 beats/min., but after an infusion of noradrenaline it caused a rise of 22 beats/min.

The evidence concerning isoprenaline depended on only four observations. However, the duration of action was greater in the reserpine-treated animal. Thus, 0.5 μg. of isoprenaline caused a rise of 20 beats/min. in a normal preparation, which passed off in 4 min., and in a reserpine-treated preparation it caused a rise of 23 beats/min. which lasted for 20 min. Similarly, 2 μg. of isoprenaline caused a rise of 53 beats/min. which was finished in 12 min. in a normal preparation, but it caused a rise of 80 beats/min. which was not finished by 30 min. in a reserpine-treated preparation.

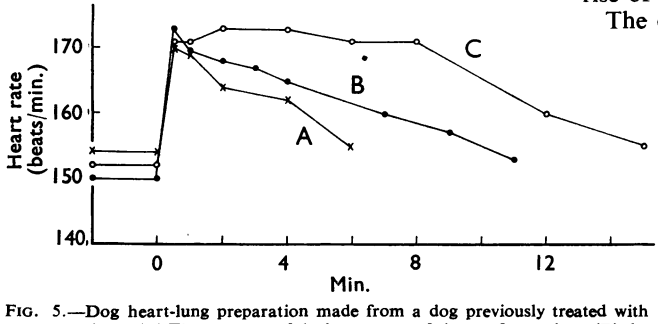

FIG. 5.—Dog heart-lung preparation made from a dog previously treated with reserpine. (A) The response of the heart rate to 0.4 mg. of tyramine; (B) the response to 0.4 mg. of tyramine after an infusion of 0.25 mg. of noradrenaline; (C) the response to 0.4 mg. of tyramine after infusion of a further 0.5 mg. of noradrenaline.

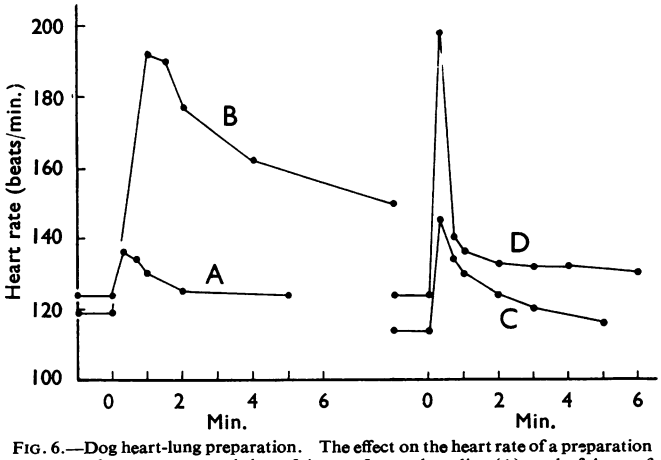

FIG. 6.—Dog heart-lung preparation. The effect on the heart rate of a preparation made from an untreated dog of 4 μg. of noradrenaline (A), and of 4 μg. of noradrenaline after the addition of 10 mg. of cocaine to the blood (B). Curve C shows the response of another preparation made from an untreated dog to 4 μg. of noradrenaline. Curve D shows the response in a preparation made from a dog previously treated with reserpine to 4 μg. of noradrenaline.

Effect of Cocaine.—When cocaine was added to the blood in the venous reservoir of the heart-lung preparation it modified the action of sympathomimetic amines in the same way as treatment with reserpine. The effect of noradrenaline and of adrenaline in doses of 4 μg. was increased and that of tyramine was diminished. An example of the effect of cocaine on the action of noradrenaline is shown in Fig. 6, in which curve **A** shows the effect before the addition of cocaine to the reservoir, and curve **B** shows the effect after the addition. The diminution of the action of tyramine is shown in Fig. 7. Curve **A** shows the effect of 1 mg. tyramine injected before the addition of cocaine, and curve **B** shows the effect after the addition.

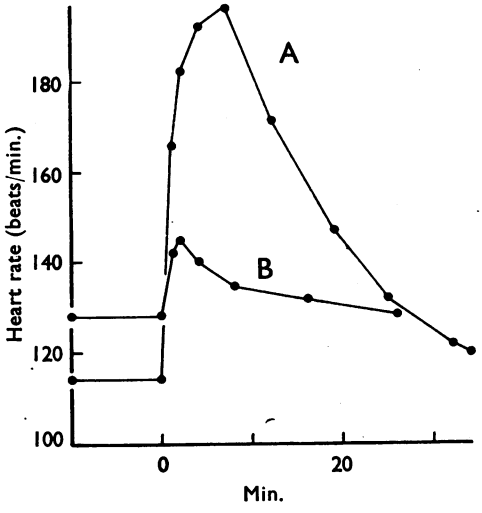

FIG. 7.—Dog heart-lung preparation made from an untreated dog. The response to 1 mg. of tyramine before A and after B the addition of 5 mg. of cocaine to the blood.

Effect of Hexamethonium.—After the injection of a large amount of hexamethonium, up to a total of 50 mg., the action of phenylethylamine, amphetamine and tyramine was similar to their action in other normal preparations. The action of noradrenaline appeared to be slightly increased.

Effect of Neosynephrine and 5-Hydroxytryptamine.—When neosynephrine was injected in amounts up to 0.2 mg. the increase in heart rate was no more than 5 beats/min. We also confirmed the finding of Paasonen and Krayer (1958) that the injection of 5-hydroxytryptamine in amounts up to 0.2 mg. did not increase the heart rate.

DISCUSSION

Bertler, Carlsson, and Rosengren (1956) showed that when rabbits were treated with reserpine, the catechol amines present in the heart disappeared, and Paasonen and Krayer (1958) have shown that this also occurs in the dog. They gave 0.5 mg./kg. of reserpine by intraperitoneal injection on two consecutive days and found that the noradrenaline was reduced to about 2% of the control value. The adrenaline content of normal dog hearts was very much lower (0.04 μg./g. as compared with 1.2 μg./g. for noradrenaline) and did not decline as a result of treatment with reserpine.

Our observations show that in the heart-lung preparation of the reserpine-treated dog, the group of sympathomimetic amines which is formed by phenylethylamine, phenylethanolamine and their derivatives (a group which includes tyramine and ephedrine) have lost their action. This action in the normal preparation seems therefore to be due to the release of noradrenaline from cardiac tissue. The action of the catechol amines, with the exception of that of dopamine, is, however, greater in the preparation from the reserpine-treated animal.

Certain differences were observed in the action of the amines on normal hearts. The action of phenylethylamine reached its peak within the first minute after injection and then declined, being finished in less than 10 min. The action of tyramine took nearly 10 min. to reach its peak and lasted up to 40 min., the time varying according to the dose. This may indicate that the —OH group in tyramine slows the rate at which it enters the cells. Both isomers of phenylethanolamine reached their maximum effect very quickly and acted longer than phenylethylamine. The action of the (+)-isomer was more prolonged than that of the (—)-isomer, as was observed on the blood pressure (Burn and Rand, 1958c). Amphetamine and ephedrine both exerted an effect so prolonged that it was impossible to determine the duration. Apart from the difference between the action of tyramine and that of phenylethylamine, the other differences in the duration of action appear to be explicable on the assumption that the duration depended on the rate of destruction by amine oxidase.

While the failure of these substances to act in the reserpine-treated preparation was clear enough, the sensitization of this preparation to noradrenaline was not so immediately obvious as when studies were made on the blood pressure. However, careful examination showed that the preparation which had been freed from noradrenaline by reserpine was more sensitive to noradrenaline. Hence the evidence from the heart supported the suggestion already made by Burn and Rand (1958c) that the store of noradrenaline in the blood vessel wall continuously discharges some noradrenaline, and that this noradrenaline occupies some of the receptors, creating some degree of tone. By so doing it reduces the effect of noradrenaline from outside. In the same way the store of noradrenaline in the heart would appear to discharge noradrenaline, increasing the rate of the pacemaker. In his observations on the effect of veratramine in abolishing the effect of adrenaline on the heart rate, Krayer (1950) observed that when full doses of veratramine were given the heart rate was sometimes much lower than it was initially, and pointed out that this might indicate the effect of

catechol amines released from within the heart in raising the pacemaker rate. Burn and Rand (1958a) found that atria from reserpine-treated rabbits beat at a mean rate of 112/min. in contrast to a rate of 146/min. for untreated controls.

It is of interest that the heart-lung preparation of the dog treated with reserpine resembles the preparation from an untreated dog after the addition of cocaine to the blood. After the addition of between 5 and 10 mg. cocaine, the effect of noradrenaline and of adrenaline is increased within a few minutes while the effect of tyramine and similar amines is greatly reduced. The effect of cocaine is not due to the dispersal of noradrenaline, at least not from the walls of the blood vessels. Burn and Rand (unpublished observations) injected large amounts of cocaine on two consecutive days into five rabbits, in doses of up to a total of 120 mg./rabbit. The amount of noradrenaline in the wall of the aorta remained normal. Burn and Rand (1958c) suggested that cocaine might stop the discharge of noradrenaline from the store in the organ. This would explain the inactivity of amines like tyramine which release noradrenaline, since cocaine would block the release. Rosenblueth and Schlossberg (1931) failed to observe that cocaine sensitizes the heart to the accelerating action of adrenaline; this may have been because the sensitization is less evident when the dose of adrenaline is large.

The addition of even so large an amount of hexamethonium as 50 mg. did not modify the action of amphetamine in causing a prolonged rise of heart rate. This result speaks against the conclusion of Reinert (1957) that amphetamine has a nicotine-like action.

We observed that the very small rise in rate which tyramine caused in the reserpine-treated heart became much greater after the slow intravenous infusion of noradrenaline. This result was comparable to the increase in the constrictor effect of tyramine in the perfused hindleg of the dog which followed a similar infusion of noradrenaline (Burn and Rand, 1958c). Both these observations indicate that the store of noradrenaline in the heart and in the vessel wall can be refilled from noradrenaline circulating in the blood. Burn (1932) observed the same effect in the blood vessels after an infusion of adrenaline. How far the refilling of the store by the amines circulating in the blood is a normal physiological phenomenon is not known. v. Euler (1956) failed to observe any increase in the catechol amine content of the heart after infusing noradrenaline into the cat at rates up to 8 μg./kg./min. for more than 30 min. Moreover, Goodall (1951) showed that the store in the heart is greatly depleted after removing the stellate ganglia: the depletion occurred after 2 weeks in animals in which the adrenal glands were present.

In conclusion, attention must be called to dopamine. Its action is diminished in the reserpine-treated heart-lung preparation, and the action is increased by an infusion of noradrenaline as is the action of tyramine. Hence, in the heart dopamine belongs to the class of drugs which release noradrenaline. In the vessels, however, dopamine, like noradrenaline, was found to act directly and to have a greater action in the reserpine-treated preparation than in the normal preparation.

We wish to express our indebtedness to the Wellcome Trust for providing the Cossor electrocardiograph (model 1314) with which the observations were made.

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