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EFFECT OF NORADRENALINE, BRETYLIUM AND COCAINE ON THE BLOOD PRESSURE RESPONSE TO TYRAMINE IN THE RAT

BY

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Tyramine is mainly a pressor agent in the rat under urethane, although the hypertensive effect of tyramine in some experiments is followed by a prolonged increase or depression of blood pressure. Bretylium, in doses up to 10 mg/kg, prolonged the response to tyramine, whereas larger doses depressed or blocked its effect. When the hypertensive effect of tyramine was blocked by bretylium, both noradrenaline and dihydroxyphenylalanine, when slowly infused, were found to restore it. The well-known block by cocaine of the hypertensive response to tyramine could also be reversed by intravenous infusion of noradrenaline and dihydroxyphenylalanine. It is concluded that the infusion of noradrenaline and dihydroxyphenylalanine makes available noradrenaline in the postganglionic adrenergic nerves which is necessary for the action of tyramine.

In spinal preparations made from cats previously treated with reserpine, tyramine and many other sympathomimetic amines which are not derivatives of catechol lose their pressor action. This action can be restored in the reserpine-treated animal by an infusion of noradrenaline (Burn & Rand, 1958a). These observations suggest that tyramine and similar substances normally act by releasing noradrenaline or adrenaline from the arterial walls. In fact it has been shown that under certain conditions intravenous injections of tyramine increase the concentrations of adrenaline and noradrenaline in a sample of plasma withdrawn from the lower aortae of cats under chloralose.

Tyramine is also classified as a sympathomimetic substance acting on postganglionic adrenergic nerve fibres because it does not act on denervated effector organs (Burn & Tainter, 1931 ; Fleckenstein & Burn, 1953). Bretylium is found to depress conduction of impulses in adrenergic neurones with subsequent failure of noradrenaline and adrenaline release, leaving at the same time intact the adrenergic receptors of the effector cell (Boura & Green, 1959). It was therefore of interest to investigate the effect of bretylium on the vascular response to tyramine. It was expected that the effect of tyramine would be particularly easy to block if this substance acted primarily on the postganglionic adrenergic fibres. This work was started in order to investigate the site and mechanism of action of tyramine in causing changes of blood pressure in the rat. The experiments with bretylium were compared with the results obtained with cocaine in order to show their similar modes of action.

METHODS

Rats of both sexes (145 to 300 g) were used and anaesthetized with 0.7 ml. per 100 g body weight 25% urethane solution subcutaneously. To record the blood pressure a cannula was inserted into the carotid artery and connected with a mercury manometer. Drugs were injected via a small polythene cannula, 0.5 mm in diameter, which was inserted into the jugular vein. Before the experiment was started, 1 to 1.5 mg/100 g heparin was injected. All doses of drugs were injected in a vol. of 0.1 ml. and washed in with the same vol. of 0.9% sodium chloride solution.

The slow intravenous infusion of noradrenaline and dihydroxyphenylalanine was given from a microburette which was connected with the venous cannula. The rate of infusion ranged from 0.66 to 3 μg/min for noradrenaline, and from 0.16 to 0.3 mg/min for dihydroxyphenylalanine.

The following substances were used: noradrenaline bitartrate, tyramine hydrochloride, bretylium tosylate and cocaine hydrochloride. Doses referred to in this paper are calculated as the salts.

RESULTS

The effects of tyramine, noradrenaline and dihydroxyphenylalanine. It was found that tyramine always caused a rise of blood pressure similar to that caused by noradrenaline or adrenaline. In some experiments tyramine caused a biphasic effect in which an initial short-lasting hypertension was succeeded by a prolonged

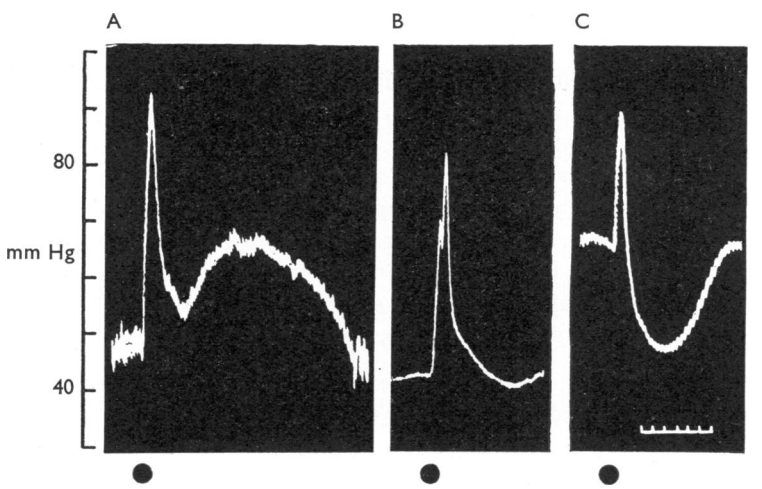

Fig. 1. Rat under urethane. Various responses of the blood pressure to the intravenous injection of 100 μg tyramine.

secondary hypertension (in 8 out of 22 experiments) or hypotension (7 out of 22). The various types of blood pressure response to tyramine in the rat are shown in Fig. 1.

The repeated injections of small doses of tyramine caused a gradual decrease in response. Fig. 2 shows an experiment in which 40 μg tyramine was injected every 10 min. After seven injections the response to tyramine had decreased from the effect shown in A to that in B. Between B and C 20 μg noradrenaline was slowly

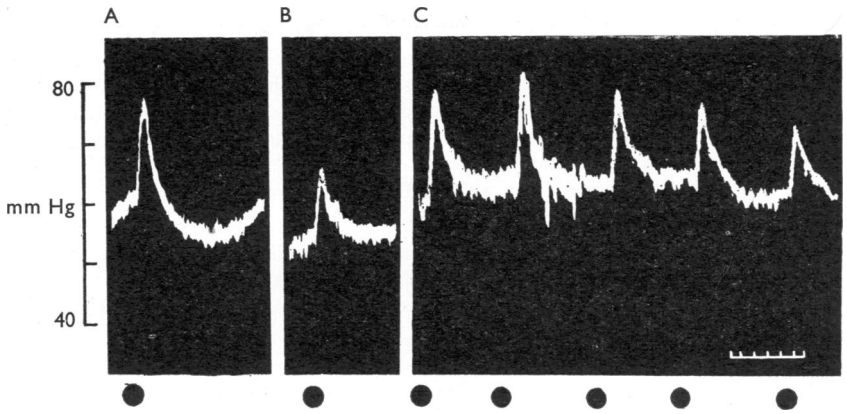

Fig. 2. Rat (190 g) under urethane. The effect of intravenous infusion of noradrenaline on the response of the blood pressure to intravenous injections of 40 μg tyramine (at dots). B, 70 min after A. Between B and C, 20 μg noradrenaline was infused over 10 min. C was taken after the infusion of noradrenaline had been stopped.

infused intravenously over 10 min. After the infusion of noradrenaline had been stopped and after the blood pressure rise caused by it had subsided, the injection of tyramine caused an increased response, as shown in C. This potentiation gradually wore off in 35 min, as also shown in C.

The infusion of dihydroxyphenylalanine produced the same effect as noradrenaline. This finding is in agreement with the finding of Burn & Rand (1960a) that the pressor action of tyramine in cats and rats treated with reserpine could be restored by intravenous infusion of various precursors of noradrenaline, for example, dihydroxyphenylalanine, dopamine, *m*-tyrosine and phenylalanine. We have now shown that even in the normal rat dihydroxyphenylalanine and noradrenaline potentiate the blood pressure response to tyramine. It was also observed that the response to tyramine after infusion of noradrenaline or dihydroxyphenylalanine was changed from a biphasic one to a pure hypertension.

The effects of tyramine, bretylium, noradrenaline and dihydroxyphenylalanine. The effect of bretylium varied according to the dose. After a dose of 10 mg/kg the response to tyramine was regularly prolonged (in 22 out of 24 experiments). Treatment of the animal by 20 mg/kg bretylium or more caused a depression or even abolition of the effect of tyramine. After such a depression the response to tyramine could be restored by an intravenous infusion of noradrenaline. A typical experiment is shown in Fig. 3. Between A and B 10 mg/kg bretylium was injected, and 15 min later the response to tyramine was prolonged, as shown in B. Between B and C 20 mg/kg bretylium was injected and 20 min later the effect of tyramine was almost abolished, as shown in C. Between C and D 30 μg noradrenaline was slowly infused intravenously over 15 min. After the infusion of noradrenaline had been stopped and after the blood pressure rise caused by it had subsided, the injection of tyramine caused an increased response, as shown in D. This type of response was observed in 6 out of 8 experiments.

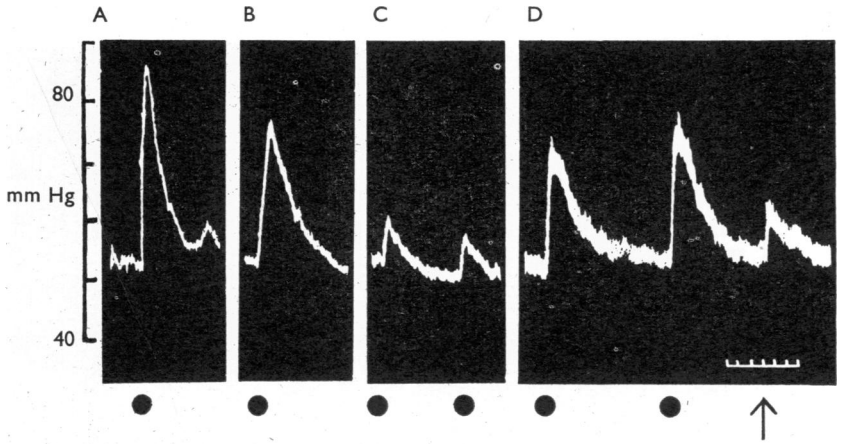

Fig. 3. Rat (155 g) under urethane. Injections into the jugular vein. The effect of bretylium and noradrenaline on the blood pressure response to tyramine. Between A and B, 10 mg/kg bretylium. Between B and C, 20 mg/kg bretylium. Between C and D, slow infusion of 30 μg noradrenaline. At dots, 50 μg tyramine; at the arrow, 0.1 ml. 0.9% sodium chloride solution.

Similar effects were obtained after infusion of dihydroxyphenylalanine. Fig. 4 shows an experiment in which 22 mg/kg bretylium abolished almost completely the effect of tyramine, as shown in B. Between B and C 3 mg dihydroxyphenylalanine was slowly infused intravenously over 15 min. After the infusion had been stopped the hypertensive response to tyramine was restored, as shown in C. This type of response was observed in 4 out of 5 experiments.

The effects of tyramine, cocaine, noradrenaline and dihydroxyphenylalanine. It is well known that cocaine can block the effect of tyramine (Burn & Tainter, 1931). We have now shown that the response to tyramine, once it was blocked by cocaine, could

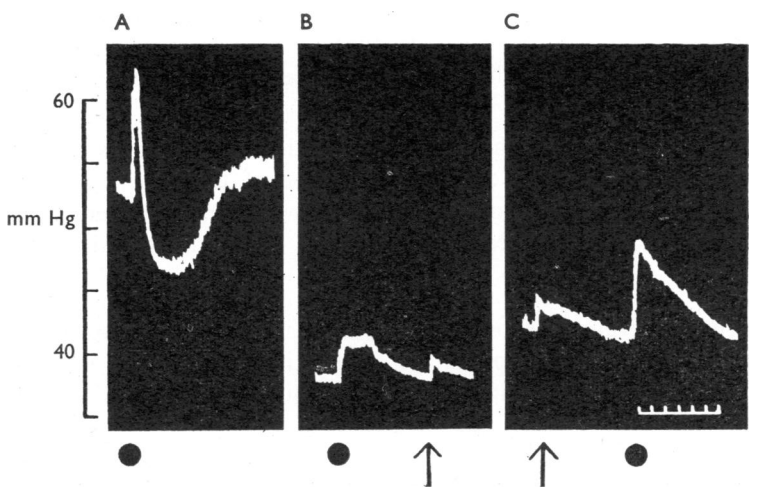

Fig. 4. Rat (170 g) under urethane. The effect of dihydroxyphenylalanine on the blood pressure response to tyramine (50 μg, at dots). Between A and B, 22 mg/kg bretylium. Between B and C, slow intravenous infusion of 3 mg dihydroxyphenylalanine. At the arrows, 0.1 ml. 0.9% sodium chloride solution.

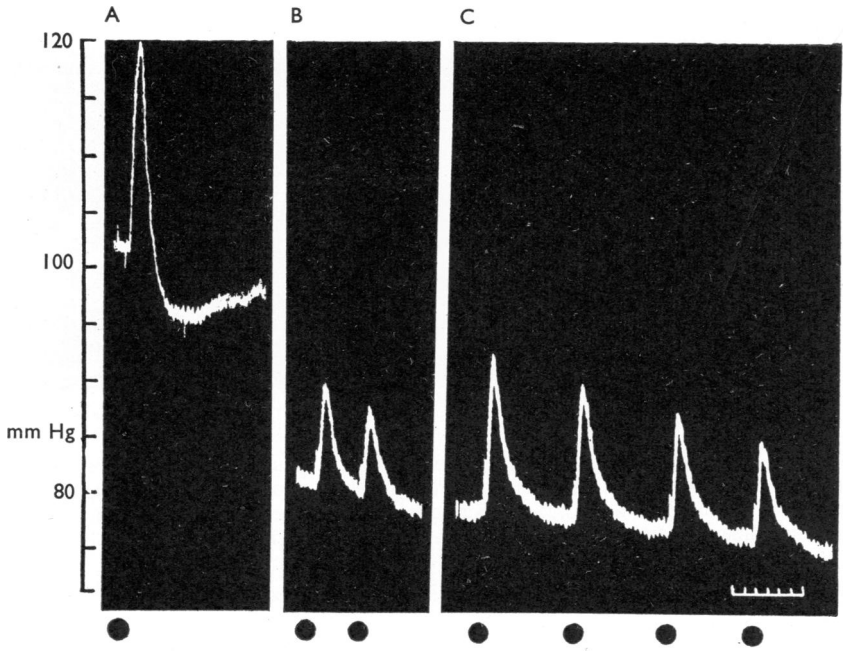

Fig. 5. Rat (210 g) under urethane. The effect of cocaine and noradrenaline on the blood pressure response to tyramine (200 μg, at dots). Between A and B, 0.5 mg cocaine. Between B and C, slow intravenous infusion of 15 μg noradrenaline.

be restored by an intravenous infusion of noradrenaline. A typical experiment is shown in Fig. 5. Between A and B 2.5 mg/kg cocaine was injected intravenously, and 20 min later the response to tyramine was very much depressed, as shown in B. Between B and C 15 μg noradrenaline was slowly infused over 15 min. After the infusion had been stopped and when the rise of blood pressure caused by it had subsided, the injection of tyramine produced a greater response than before the infusion, as shown in C. This type of response was observed in 5 out of 9 experiments.

In 2 out of 3 experiments the intravenous infusion of dihydroxyphenylalanine in animals treated with cocaine caused an increase of the hypertensive response to tyramine. This effect of dihydroxyphenylalanine was generally observed if dihydroxyphenylalanine by itself produced a small but long-lasting increase in blood pressure.

DISCUSSION

The present experiments show that tyramine is mainly a pressor agent in the rat under urethane. In some experiments this initial hypertension is followed by a prolonged increase or depression of blood pressure. The secondary fall of blood pressure caused by tyramine was similar to the blood pressure fall caused by dopamine in the guinea-pig (Hornykiewicz, 1958 ; Burn & Rand, 1958b). It was also found that the hypertensive effect of tyramine gradually diminished after repeated injections. This tachyphylaxis could be immediately reversed after infusion of noradrenaline and dihydroxyphenylalanine.

It was found in previous work that bretylium blocked the hypertensive effect of eserine in the rat, leaving at the same time intact or even potentiating the effect of noradrenaline (Lešić & Varagić, 1961). We have now found that the hypertensive effect of tyramine is usually influenced in a similar way. However, larger doses of bretylium were necessary to depress the effect of tyramine than to block the effect of eserine. The doses of bretylium up to 10 mg/kg caused a prolongation of the response to tyramine. Burn & Rand (1960b) have already observed that the effect of tyramine on the nictitating membrane of the spinal cat may be potentiated by bretylium. If the hypertensive effect of tyramine was blocked by a large dose of bretylium, both noradrenaline and dihydroxyphenylalanine, when slowly infused, were found to be able to restore that effect. Bretylium is known to impair conduction of impulses in adrenergic neurones with consequent failure of release of noradrenaline and adrenaline (Boura & Green, 1959). Tyramine, on the other hand, is known to liberate noradrenaline from adrenergic transmitter granules (von Euler & Lishajko, 1960 ; Schümann, 1960) and from the walls of the aorta in cats under chloralose after induction of lasting ganglion block with hexamethonium and after exclusion of the adrenals from the circulation (Lockett & Eakins, 1960a & b). In postganglionically denervated sympathetically innervated effector organs, as well as in animals treated with cocaine, the effect of tyramine was abolished (Burn & Tainter, 1931 ; Fleckenstein & Burn, 1953 ; Fleckenstein & Stöckle, 1955). In animals treated with reserpine the effect of tyramine was also abolished (Carlsson, Rosengren, Bertler & Nilsson, 1957). All the evidence suggested that tyramine acted through the liberation of noradrenaline (Burn & Rand, 1958a). Our experiments support the idea that the infusion of noradrenaline and dihydroxyphenylalanine in animals treated with bretylium restores the stores of noradrenaline in postganglionic adrenergic nerves. These stores are presumably necessary for the action of tyramine.

The indirect effects of sympathomimetic amines (for example, tyramine) which can be abolished by reserpine are decreased by cocaine, which presumably acts by inhibiting the liberation of noradrenaline (Holtz, Osswald & Stock, 1960). The present experiments show that the block by cocaine of the hypertensive response to tyramine could be reversed by intravenous infusion of noradrenaline and dihydroxyphenylalanine. This finding seems to suggest that " pharmacological denervation " (Fleckenstein & Stöckle, 1955) is due to impairment of noradrenaline release from postganglionic adrenergic nerves. The infusion of noradrenaline and dihydroxyphenylalanine makes available noradrenaline in the postganglionic adrenergic nerves which is necessary for the action of tyramine.

Until recently the fate of the infused noradrenaline and adrenaline was an unsolved problem. Thus Raab & Gigee (1955) observed that the heart muscle of the dog took up noradrenaline and adrenaline after intraperitoneal injection of these substances. It may be noted that the amounts of catecholamines in these experiments were very high (38 mg noradrenaline and 75 mg adrenaline for a dog of 10 kg). Von Euler (1956) infused amounts of noradrenaline between 2.5 and 7.8 μg/kg/min into cats for over 30 min and gave intraperitoneal injections of noradrenaline and adrenaline up to 2 mg/kg. In these experiments no increase of catechol amine content of the heart, spleen, liver, kidney and skeletal muscle was

observed. In experiments with tritium-labelled ^{3}H-adrenaline of high specific activity, it was found that immediately after the end of a 30 min infusion (3 μg/kg/min) the concentration of ^{3}H-adrenaline in heart, spleen, adrenal and pituitary gland exceeded that of the plasma by several-fold. Even two hours after the administration of ^{3}H-adrenaline large quantities were found in the heart and spleen, indicating that these tissues not only accumulate adrenaline but also retain it for a long period (Axelrod, Weil-Malherbe & Tomchick, 1959a & b). Our experiments suggest that the infused noradrenaline and its precursor, dihydroxyphenylalanine, are partly accumulating in the adrenergic nerve terminals. Von Euler (1956) has already expressed the view that, at least in the spleen, noradrenaline is present within the fine terminations of the nerves.

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