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THE MECHANISM BY WHICH NORADRENALINE RESTORES THE PRESSOR ACTION OF INDIRECTLY ACTING SYMPATHOMIMETIC AMINES IN RESERPINIZED DOGS

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There are two methods of restoring the pressor action of indirectly acting sympathomimetic amines in reserpinized animals. One is to " replete " the noradrenaline stores by an intravenous infusion of excess noradrenaline and to inject the indirectly acting amine a few minutes after stopping the infusion (Burn & Rand, 1958; Fawaz & Simaan, 1965). The other is to infuse noradrenaline in much smaller amounts and to inject the indirectly acting amine during, but not after the infusion (Fawaz & Simaan, 1964). In this paper an attempt is made to elucidate the mechanism of the restorative action of noradrenaline in each instance.

METHODS

About 110 mongrel dogs of either sex weighing 8 to 15 kg were anaesthetized with chloralose (150 mg/kg). Artificial respiration (Starling pump) was used in all experiments. The blood pressure was measured from a common carotid artery by a mercury manometer and both vagosympathetic trunks were cut. Reserpine was given as intraperitoneal injections of 0.5 mg/kg 48 and 24 hr before the experiment; such animals usually required less anaesthetic. Noradrenaline was used as the bitartrate, tyramine and cocaine as the hydrochlorides and mephentermine (Wyamine) as the sulphate, and the doses are expressed in terms of the bases. Injections into anaesthetized dogs were intravenous. Unless otherwise stated, tyramine, cocaine and mephentermine were injected in single doses of 0.2, 1 and 0.4 mg/kg respectively. Infusions were performed by the use of a regulable constant-infusion apparatus (B. Braun, Melsungen, Germany).

RESULTS

Restoration by repletion of the " small " noradrenaline store with excess noradrenaline

The " repletion " experiment performed by Burn & Rand (1958) on reserpinized spinal cats can be done on reserpinized dogs during chloralose anaesthesia by infusing noradrenaline at the rate of 4 μg/kg/min for 50 min and injecting tyramine or mephentermine 5 to 10 min after the end of the infusion when the blood pressure has reached a steady level (Fawaz & Simaan, 1965). This treatment does not completely replete the noradrenaline

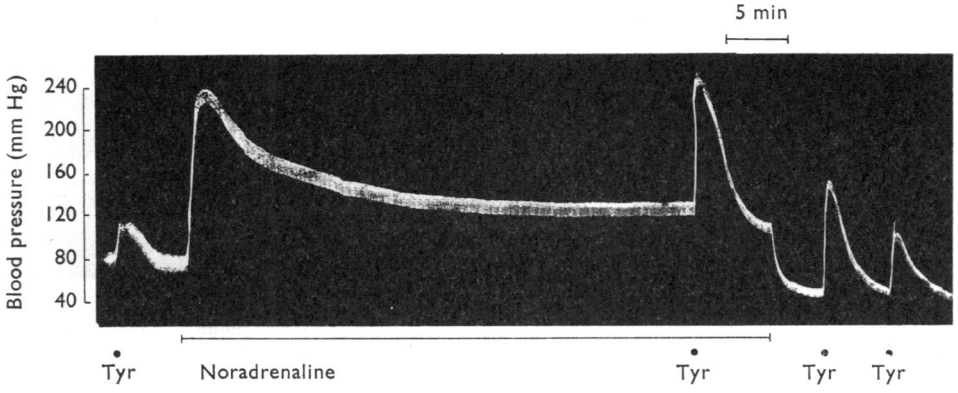

Fig. 1. Reserpinized dog, 12 kg. Record of arterial blood pressure. Tyramine (Tyr, 0.2 mg/kg intravenously) given before the end of the noradrenaline infusion (horizontal line, 4 μg/kg/min for 50 min) does not affect the postinfusion pressor response to tyramine.

store for, while in the nonreserpinized animal about six consecutive tyramine injections may be given before a reduction of the pressor response is observed, only one tyramine injection is fully effective in a " repleted " reserpinized dog, that is, it restores the pressor response to the level obtained after injecting the same dose of tyramine to nonreserpinized animals. It thus appears that in a " repleted " reserpinized dog only the " small " noradrenaline store is filled, since the pressor action of tyramine resembles that of mephentermine in a nonreserpinized dog where we postulated that mephentermine liberates noradrenaline from the small store. The so-called small noradrenaline store (Fawaz & Simaan, 1965) and the store described by Burn & Rand (1958) may thus be the same. A " repleted " store in a reserpinized dog is not indefinitely stable: if tyramine is injected 30 min instead of 5 to 10 min after the end of the infusion the pressor response is not fully restored and may sometimes be absent.

One or two tyramine injections may be given during the last 5 to 10 min of the 50-min noradrenaline infusion and a full pressor response is observed. However, this does not affect the postinfusion pressor response to tyramine indicating that the mechanisms of the pressor actions during and after the infusion are different (Fig. 1).

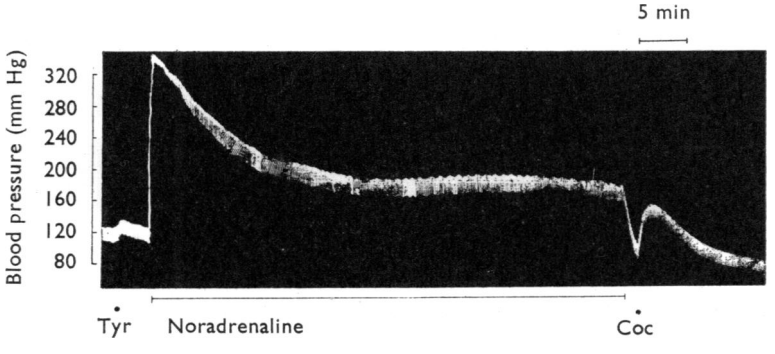

Fig. 2. Reserpinized dog, 15 kg. Record of arterial blood pressure. Cocaine (Coc, 1 mg/kg intravenously) given shortly after noradrenaline infusion (horizontal line, 4 μg/kg/min for 50 min) shows pressor effect in all experiments. Tyramine (Tyr, 0.2 mg/kg) was given intravenously.

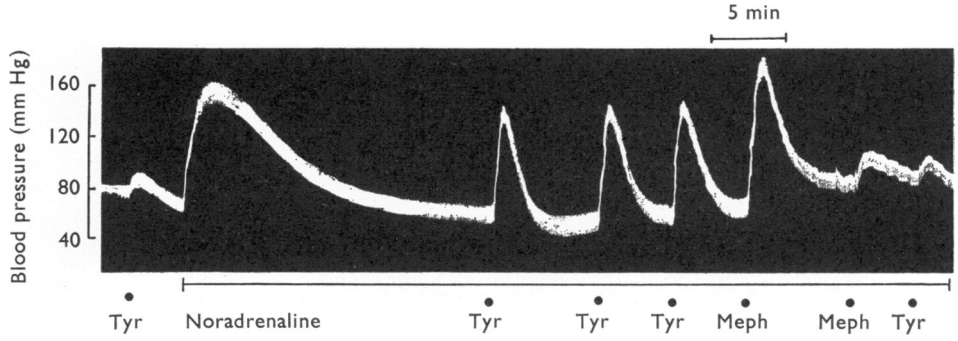

Fig. 3. Reserpinized dog, 13 kg. Record of arterial blood pressure. Noradrenaline infusion (horizontal line, 0.5 μg/kg/min) restores the pressor response of tyramine (Tyr, 0.2 mg/kg) for any number of tyramine injections if given during the infusion. Mephentermine (Meph, 0.4 mg/kg) pressor action is also restored but for one injection only. Tyramine after mephentermine is no more effective.

When, instead of tyramine or mephentermine, cocaine was injected in the repletion experiments 5 to 10 min after the end of the noradrenaline infusion a pressor response was observed in five out of ten experiments. However, if cocaine is injected immediately after stopping the noradrenaline infusion, at a time when the blood pressure just returns to the preinfusion level, a pressor response after cocaine is always observed (Fig. 2). In either instance, injection of mephentermine or cocaine before the noradrenaline infusion cancels the postinfusion pressor response due to cocaine just as it does for tyramine and mephentermine (Fawaz & Simaan, 1965). This indicates that the pressor effects of tyramine, mephentermine or cocaine in reserpinized animals after infusion of noradrenaline are due to release of noradrenaline from repleted stores and that the repletion by noradrenaline was blocked by prior treatment with mephentermine or cocaine (Fawaz & Simaan, 1965). Cocaine rarely shows a pressor effect in a normal, nonreserpinized dog, but it exhibits a pressor effect if administered immediately after a large single dose of noradrenaline (10 μg/kg) or after a noradrenaline infusion (4 μg/kg/min for 50 min) when the blood pressure just returns to the preinfusion level. Here also cocaine given before the noradrenaline nullifies the post-noradrenaline pressor response due to cocaine.

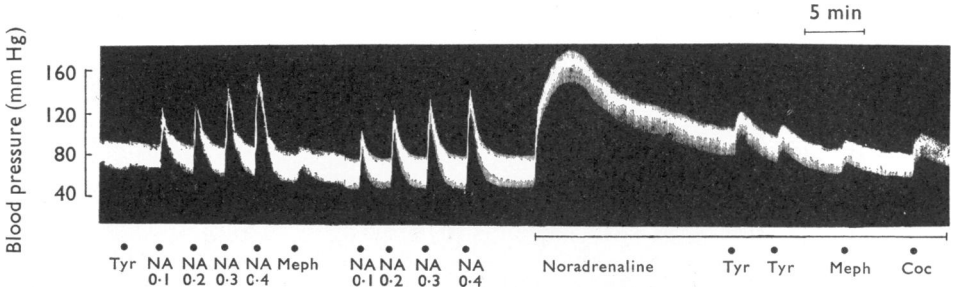

Fig. 4. Reserpinized dog, 15 kg. Record of arterial blood pressure. One injection of mephentermine (Meph) does not potentiate the pressor effects of single doses of noradrenaline (NA) but greatly reduces or nullifies the pressor action of subsequent doses of tyramine (Tyr), mephentermine or cocaine (Coc) which is otherwise always observed when any of the last three substances is injected during the noradrenaline infusion (horizontal line, 0.5 μg/kg/min). All doses in μg/kg.

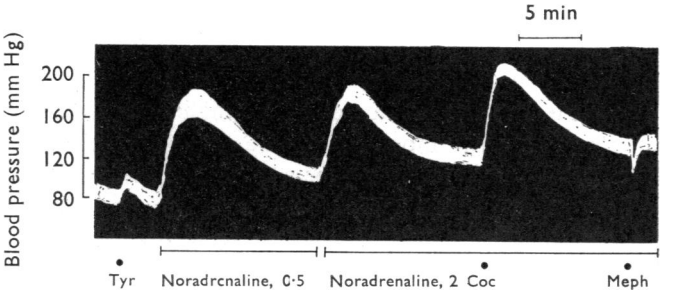

Fig. 5. Reserpinized dog, 14 kg. Record of arterial blood pressure. Noradrenaline infusion (horizontal lines, 0.5 and 2 μg/kg/min). The second peak marks the effect of suddenly quadrupling the rate of noradrenaline infusion. Tyramine (Tyr, 0.2 mg/kg), cocaine (Coc, 1 mg/kg) and mephentermine (Meph, 0.4 mg/kg) were injected where shown. Cocaine exhibits a pressor effect if given during the noradrenaline infusion; mephentermine after cocaine is ineffective.

Restoration by infusion of small amounts of noradrenaline and injection during the infusion

This procedure has been described earlier (Fawaz & Simaan, 1964). The indirectly acting sympathomimetic amine has to be injected during the noradrenaline infusion (0.5 μg/kg/min) but not after. In contrast to the situation in the normal animal (Fawaz & Simaan, 1965), consecutive injections of tyramine result in no diminution of the pressor response as long as the noradrenaline infusion continues. The pressor action of mephentermine is also restored but only for the first dose, the second dose being very slightly effective. Tyramine (or cocaine) given after mephentermine shows little or no pressor effect (Fig. 3). Mephentermine given before the start of the noradrenaline infusion greatly reduces or nullifies the pressor effect of tyramine, mephentermine or cocaine injected during the infusion (Fig. 4). Cocaine behaves almost exactly like mephentermine in that it exhibits a pressor response if injected during the noradrenaline infusion, although the descending limb of the pressor response curve is less steep than that usually observed after mephentermine (Fig. 5), and nullifies the pressor effects of subsequent injections of tyramine or mephentermine if it is given during or before the noradrenaline infusion.

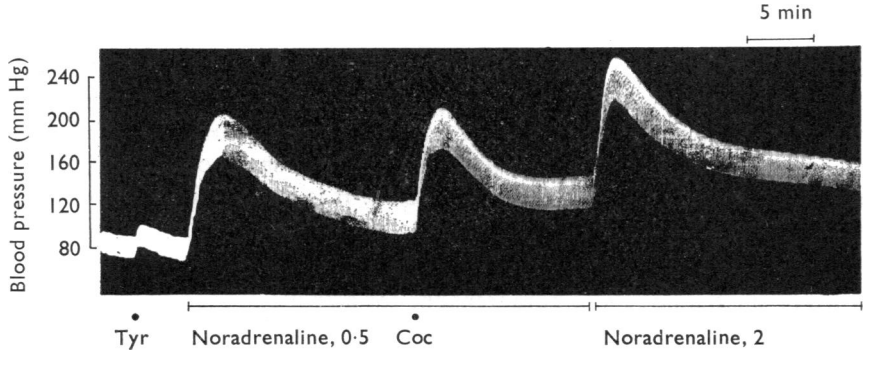

Fig. 6. Reserpinized dog, 12 kg. Record of arterial blood pressure. Noradrenaline infusions (horizontal lines, 0.5 and 2 μg/kg/min). Injection of cocaine (Coc, 1 mg/kg) is equivalent to quadrupling rate of noradrenaline infusion (compare with Fig. 5). Tyramine (Tyr, 0.2 mg/kg) was given where shown.

At first sight it would appear that the pressor responses obtained with tyramine, mephentermine and cocaine during the noradrenaline infusion in the reserpinized animal are due to potentiation of the action of infused noradrenaline, the actions of these amines being tantamount to a sudden increase in the rate of noradrenaline infusion. Tyramine, mephentermine and cocaine are known to block the uptake of noradrenaline by the stores. This would make more of the infused noradrenaline available to the receptors thus resulting in potentiation of the pressor effects. Plausible as this explanation might seem, it conflicts with the experimental findings. It can be seen from Fig. 4 that one dose of mephentermine does not potentiate the pressor effect of single doses of noradrenaline in the reserpinized dog. Actually, four such doses are needed before potentiation begins to be observed. Furthermore, one dose of mephentermine given before a noradrenaline infusion (0.5 μg/kg/min) does not alter the peak of the pressor response or subsequent course of the pressure curve. In thirty-seven reserpinized dogs, infusion of noradrenaline (0.5 μg/kg/min) gave an increase in blood pressure of 130 mm Hg at the peak of the pressor response, while in twelve dogs treated with one dose of mephentermine before the infusion the average increase was 126 mm Hg. It is thus seen that one dose of mephentermine which blocks the uptake of noradrenaline by the " small " store (Burn & Rand, 1958; Fawaz & Simaan, 1965) fails to potentiate the pressor action of noradrenaline. The situation with cocaine is only quantitatively different. Cocaine in doses of 1 mg/kg potentiates the pressor response of single doses of noradrenaline in reserpinized animals not more than twice (Fawaz, unpublished). Yet quantitative experiments show that the effect of injecting cocaine in the course of a noradrenaline infusion is equivalent to a fourfold increase in the rate of infusion of noradrenaline (Figs. 5 and 6). Thus it cannot be said that the pressor effect of cocaine during the infusion is solely due to its ability to potentiate the pressor action of noradrenaline.

DISCUSSION

Three possibilities come to mind if one attempts to elucidate the mechanism of the pressor actions of tyramine, mephentermine or cocaine when these substances are injected in the course of a noradrenaline infusion in reserpinized dogs.

(1) These substances block the uptake of infused noradrenaline by the stores, temporarily with tyramine—owing to its destruction by amine oxidase—and for a longer period with mephentermine or cocaine. This would make more noradrenaline available to the receptors and thus result in a peak in the pressure curve. This possibility has been ruled out, as indicated in Results. A dose of mephentermine which suffices to completely block the store of Burn & Rand (1958) does not potentiate the pressor effect of single doses of noradrenaline in reserpinized animals nor does it alter the shape of the pressure curve of a noradrenaline infusion if given before the infusion. It is thus difficult to see why it should potentiate noradrenaline during the infusion.

(2) Tyramine, mephentermine and cocaine have pressor actions of their own but need the presence of small amounts of noradrenaline for activity. This view has been expressed by Fawaz & Simaan (1964). However, a further assumption would have to be made, namely, that a substance other than noradrenaline and found in the stores is necessary for this action, since if these stores are blocked by one dose of cocaine or mephentermine given before or during the noradrenaline infusion the pressor effect of tyramine, mephentermine

or cocaine is nullified. A reserpinized dog treated with cocaine or mephentermine may be likened to a sympathectomized preparation: in the first case the stores are blocked, in the second they are absent. In unpublished experiments we have found that in a heart-lung preparation from dogs whose hearts had been denervated by the method of Cooper, Gilbert, Bloodwell & Crout (1961) noradrenaline or adrenaline was unable to restore the chronotropic action of mephentermine, while in preparations from reserpinized dogs adrenaline can restore the action of mephentermine (Fawaz, 1961). The difference between the two preparations is that the denervated heart, unlike the reserpinized heart, has no stores.

(3) The third possibility postulates the presence of a third noradrenaline store which may be called the "labile" store. In a previous publication (Fawaz & Simaan, 1965) we postulated the presence in nonreserpinized animals of a large store from which noradrenaline can be released by tyramine and a "small" store from which noradrenaline can be released by tyramine or mephentermine. The small store, equivalent to that described by Burn & Rand (1958), but apparently not the large store, is the one that is repleted in a reserpinized animal after the acute infusion of excess noradrenaline. The labile store is expected to be empty not only in a reserpinized but also in a normal animal and to fill up after administration of small amounts of noradrenaline. This store can also be blocked by cocaine and mephentermine and its contained noradrenaline can be released not only by tyramine and mephentermine but also by cocaine. Cocaine exhibits a sure pressor effect if injected immediately after an infusion of excess noradrenaline into a reserpinized or nonreserpinized dog at a time when the blood pressure declines to the preinfusion level, but not if mephentermine or cocaine is administered before the infusion. The postulated labile store evidently needs very small quantities of noradrenaline to be filled but its effectiveness indicates that it is conveniently located with regard to the receptors. Cocaine thus not only blocks the entry of noradrenaline into the stores but also releases noradrenaline at least from the labile stores. The noradrenaline-releasing ability of cocaine has also been postulated by Rosenthale & Dipalma (1963) and by Furchgott, Kirpekar, Rieker & Schwab (1963).

SUMMARY

1. In reserpinized dogs treated with an infusion of excess noradrenaline (0.2 mg/kg) one dose of tyramine (0.2 mg/kg), mephentermine (0.4 mg/kg)or cocaine (1 mg/kg) exhibits a strong pressor effect if given after the infusion. A second dose is very slightly effective. Cocaine is always effective if given immediately after the infusion, while tyramine and mephentermine are effective if given within 10 min after the end of the infusion and only partially effective after 30 min. In all instances, one dose of mephentermine or cocaine given before the infusion nullifies the postinfusion pressor effects.

2. In reserpinized dogs infused with small amounts of noradrenaline (0.5 μg/kg/min), tyramine, mephentermine and cocaine show pressor effects if given during, but not after the infusion. Tyramine shows no diminution of pressor response after consecutive doses—in contrast to its actions in nonreserpinized dogs—while mephentermine and cocaine injections are effective only once. Mephentermine and cocaine given before or during the infusion nullify subsequent pressor responses of tyramine, mephentermine or cocaine.

3. The mechanism of these pressor actions of mephentermine, tyramine and cocaine summarized in (2) is discussed. Potentiation of noradrenaline by blocking the stores does not explain the pressor action of mephentermine and only partly that of cocaine. A dose

of mephentermine that completely blocks the "small" noradrenaline store does not potentiate noradrenaline. The other two explanations are: that tyramine, mephentermine and cocaine have pressor actions of their own but need minimal amounts of noradrenaline and another substance liberated from the stores; that tyramine, mephentermine and cocaine liberate noradrenaline from an exceedingly labile store which can be filled by minute amounts of noradrenaline but which is suitably located with regard to the receptors. This labile store can also be blocked by mephentermine or cocaine.

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