A COMPARISON OF THE EFFECTS OF TYRAMINE AND EPHEDRINE ON ATRIAL CONTRACTIONS IN RABBITS AND GUINEA-PIGS

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Bertler, Carlsson and Rosengren (1) found that the pressor effect of tyramine is decreased in animals that have previously been reserpinized, and Burn and Rand (2) have reported that in such animals the change in blood pressure, response of vessel wall, or nictitating membrane to tyramine recovers after the treatment of noradrenaline. It has also been reported that the catecholamine content of various organs decreases after the administration of reserpine (3–5). The generally accepted explanation of these findings has been that the decreased response to tyramine in reserpinized animals may be due to the decrease in endogenous catecholamine available to tyramine after reserpine administration.

On the other hand, tyramine and ephedrine show similar pharmacological effects both in vivo and in vitro. The action mechanism of ephedrine also remains to be clarified, despite the many investigations that have been carried out since Chen and Schmidt (6) reported on the pharmacological effects of this drug. Ephedrine is also effective as a bronchodilator and, in some cases, as a pressor drug. Clinically, however, a decrease in response to ephedrine after repeated administration has often been encountered, especially in blood pressure response, but tyramine yields to tachyphylaxis scarcely. It is generally held that mechanism of this so-called “tachyphylaxis” phenomenon is due to receptor occupation by ephedrine (7).

We have previously carried out a series of experiments on rabbit and guinea-pig atria with a number of drugs assumed to have the ability to mobilize and activate endogenous catecholamine (8, 9). We have used the similar experimental procedure in our attempt to clarify the action mechanisms of tyramine and ephedrine, and to explain the mechanism of tachyphylaxis phenomenon that displays following repeated administration of ephedrine. In the present experiments also, reserpine and nicotine were used as experimental means, reserpine for its reduction of endogenous catecholamine, and nicotine for its indirect activation.

METHODS

Rabbits weighing 1.5 to 2.5 kg and guinea-pigs weighing 250 to 350 g were used. They were sacrificed without anesthesia by severing both common carotid arteries.
heart of each animal was immediately isolated and immersed in a Locke's solution saturated with oxygen at 30° to 31°C. This solution comprised 9.0 g sodium chloride, 0.42 g potassium chloride, 0.24 g calcium chloride, 0.2 g sodium bicarbonate and 1.0 g dextrose per litre of water. The atria were then prepared from the heart and their contractions recorded on a smoked drum by means of a spring lever. These immersed atria continued to contract rhythmically for 15 hours or more.

In some experiments Locke's solution was modified to contain 2.5 times higher concentration of potassium as follows: 8.5 g sodium chloride, 1.05 g potassium chloride, 0.24 g calcium chloride, 0.2 g sodium bicarbonate and 1.0 g dextrose per litre of water. This "high K Locke's solution" was used to examine the drug effects in abnormal inorganic ion milieu.

The experiment was begun one hour after the atrial contractions spontaneously became rhythmically and constant. Changes in contraction amplitude and rate were recorded on the addition of drugs. The concentrations of drugs were expressed in terms of g/ml. The drugs used were the following: adrenaline hydrochloride, cocaine hydrochloride (Takeda), ephedrine hydrochloride, nicotine (Merck), reserpine (Serpasil, CIBA), and tyramine hydrochloride.

RESULTS

1) The effect of tyramine or ephedrine on atrial contractions

Tyramine $10^{-6}$ gradually increased contractions 20 to 80% in amplitude and 3 to 40% in rate. Ephedrine $10^{-6}$ also showed a similar effect. In an increasing concentration up to $10^{-4}$, the effect of tyramine markedly increased in both amplitude and rate, whereas the effect of ephedrine remained unchanged. Tyramine $10^{-4}$ augmented contractions more and more, but ephedrine in this concentration suppressed them after a transient and slight augmentation for one to two minutes.

In the presence of ephedrine $10^{-5}$ or $10^{-4}$, tyramine did not show the above described augmentation effect in any concentration, and the effect of adrenaline $10^{-7}$ also was suppressed (Fig. 1). On atrial contractions of rabbits and guinea-pigs, tyramine and ephedrine in any concentration could scarcely potentiate the augmentation effect of adrenaline $10^{-5}$ to $10^{-3}$.
2) The effect of nicotine on the addition of tyramine or ephedrine

Nicotine $10^{-5}$ to $2 \times 10^{-5}$ began by decreasing contractions in amplitude and rate for 10 to 30 seconds, but then increased them to a degree surpassing the originals. When tyramine or ephedrine $10^{-6}$ to $10^{-4}$ has been added to the bath one to two minutes before nicotine $10^{-5}$ to $2 \times 10^{-5}$, however, the latter stimulant action of nicotine was suppressed (Figs. 2 and 3).

**FIG. 2.** Contractions of isolated guinea-pig atria: response to nicotine $10^{-5}$ was suppressed by preceding addition of tyramine (Tyr. $10^{-4}$). Numerals above the record give atrial frequencies (beats/min). At (W) the bath fluid was changed. The concentration of each drug is shown in g/ml. Time intervals are one minute.

**FIG. 3.** Contractions of isolated guinea-pig atria: response to nicotine $2 \times 10^{-5}$ was suppressed by preceding addition of ephedrine (Ephed. $10^{-4}$). Numerals above the record give atrial frequencies (beats/min). At (W) the bath fluid was changed. The concentration of each drug is shown in g/ml. Time intervals are one minute.
3) The effect of tyramine or ephedrine on atria excised from animal treated with reserpine 1 to 3 mg/kg subcutaneously 24 hours prior to experiment

On the contractions of reserpine pretreated atria, tyramine exercised an augmentation effect in higher concentration than $10^{-5}$, but the intensity of this effect was up to 10% of the effect on normal atria. On these atria ephedrine also showed an augmentation effect in higher concentration than $10^{-6}$, but the intensity of this effect was about a half of the effect on normal atria. The augmentation effect of adrenaline $10^{-8}$ to $10^{-6}$ was suppressed in the presence of ephedrine $10^{-4}$ or tyramine $5 \times 10^{-4}$ more distinctly on these atria than on normal atria (Fig. 4).

![Fig. 4](image)

**Fig. 4.** Contractions of isolated atria, excised from guinea-pig pretreated with reserpine 3 mg/kg subcutaneously 24 hours prior to experiment: the augmentation effect of adrenaline $10^{-8}$ was decreased in the presence of tyramine $5 \times 10^{-4}$. Numerals above the record give atrial frequencies (beats/min). The concentration of each drug is shown in g/ml. Time intervals are one minute.

4) The effect of tyramine or ephedrine on atria of rabbit which had been arrested by the addition of reserpine to the bath

The experiment was carried out in a manner similar to that of the experiments reported in our papers (8, 9) concerning L-methionine and strophanthin-G. When reserpine $3 \times 10^{-4}$ was added to the bath, atrial contractions were arrested within 15 to 120 minutes. Spontaneous contractions could not be restored after repeated wash out, if the atria had been kept 60 to 90 minutes after arrest in the Locke's solution containing reserpine, but it could be restored by the addition of tyramine $10^{-4}$ or ephedrine $10^{-4}$ (Figs. 5 and 6).

In some cases the intermittent mode of contraction restored by tyramine was very curious: the contractions would last for 10 to 20 seconds, remain arrested for 5 to 10 seconds, and then resume. With atria excised from rabbit subcutaneously pretreated reserpine 1 to 3 mg/kg 24 hours prior to experiment, tyramine or ephedrine could not restore the arrested contraction in any concentration, but adrenaline $10^{-6}$ could.
FIG. 5. Contractions of isolated rabbit atria were arrested by the addition of reserpine $3 \times 10^{-5}$ and thereafter the atria were kept in reserpine containing Locke's solution for 60 minutes. At (W) the bath fluid was changed, but the resumption of atrial contraction did not occur spontaneously. Thus treated atria could be resumed by tyramine (Tyr.) in higher concentration than $10^{-4}$. Numerals above the record give atrial frequencies (beats/min). The concentration of each drug is shown in g/ml. Time intervals are one minute.

FIG. 6. Contractions of isolated rabbit atria were arrested by the addition of reserpine $3 \times 10^{-5}$ and thereafter the atria were kept in reserpine containing Locke's solution for 60 minutes. At (W) the bath fluid was changed, but the resumption of atrial contraction did not occur spontaneously. Thus treated atria could be resumed by ephedrine $10^{-4}$. Numerals above the record give atrial frequencies (beats/min). The concentration of each drug is shown in g/ml. Time intervals are one minute.

5) The effect of tyramine, ephedrine, or adrenaline on the addition of cocaine

Cocaine $10^{-6}$ showed scarcely an influence on atrial contractions, but in the presence of cocaine $10^{-6}$ neither tyramine nor ephedrine exercised the augmentation effect in any concentration. And the effect of adrenaline $10^{-7}$ was scarcely influenced by previous addition of cocaine $10^{-4}$.

6) The effect of tyramine or ephedrine in a high K Locke's solution

Tyramine $10^{-4}$, ephedrine $10^{-4}$, or adrenaline $10^{-6}$ could restore the arrested contraction of atria in a high K Locke's solution. When the added tyramine or ephedrine had been washed out with the high K Locke's solution, the restored contractions usually increased in both amplitude and rate (Fig. 7). The contractions, however, restored by adrenaline decreased in amplitude after the wash out of the drug.
DISCUSSION

The fact that atria arrested by the addition of reserpine $3 \times 10^{-5}$ could be restored by the addition of tyramine or ephedrine $10^{-4}$ in experiment 4, shows that tyramine and ephedrine can activate endogenous catecholamine that has been decreased by reserpine to a degree below that at which atrial contraction can continue. When the atria had been prepared from reserpine pretreated animal, the addition of tyramine or ephedrine could not restore contraction. This result suggests that the degree of decrease was too great to be activated by tyramine or ephedrine. Experiment 2 shows that tyramine and ephedrine suppress the stimulant action of nicotine. In our experiment 5 and in a report of Tainter et al. (10), it is shown that the effects of tyramine and ephedrine are depressed in the presence of cocaine.

These results may suggest that 1) tyramine and ephedrine enter into atrial cells and reveal their effects through activation of endogenous catecholamine, 2) each drug decreases the effect of nicotine that may activate endogenous catecholamine in the granule structure of sympathetic or chromaffin cells, and 3) the effect of cocaine exerts on the granule structure through which tyramine and ephedrine enter into catecholamine storage sites and release the stored amine.

Then, if catecholamine receptors are posturated on atrial cells, the receptors may consist of two kinds of receptors: the catecholamine granule-receptor and the catecholamine membrane-receptor. The former may be an intracellular catecholamine storage structure and the latter an atrial cell membrane structure. For instance, nicotine may attack the former but not attack the latter. The latter, however, may be attacked by exogenously
added catecholamine such as adrenaline and also by released endogenous catecholamine. On the basis of these postulations, the phenomenon that the augmentation effect of nicotine was suppressed by previous addition of tyramine or ephedrine $10^{-4}$ may be explained as follows: 1) the stimulant effects of tyramine and ephedrine on the catecholamine membrane-receptor are very weak as shown in experiment 4, 2) the catecholamine granule-receptor is combined with endogenous catecholamine that will be released spontaneously or by nicotine, but also with tyramine or ephedrine, 3) endogenous catecholamine and bound tyramine and ephedrine molecules are released by nicotine from the catecholamine granule-receptor, and 4) these three kinds of molecules attack the catecholamine membrane-receptor. Therefore, the possibility of attacking of the endogenous catecholamine molecules released by nicotine on catecholamine membrane-receptor is assumed to fall in the presence of tyramine or ephedrine in each high concentration.

In respect to a catecholamine membrane-receptor, the receptor occupation effects of ephedrine and tyramine also could be scarcely deniable. This effect of ephedrine was shown in experiment 1, in which the augmentation effect of adrenaline on atrial contractions was suppressed in the presence of ephedrine in its high concentration. Experiment 6 also showed that the arresting atria in a high K Locke's solution were resumed by the addition of tyramine or ephedrine, and when each drug had been washed out with the same solution, the contraction amplitude and rate were increased to a greater degree than that in the presence of each drug. These results suggest that the catecholamine membrane-receptor stimulation by endogenous catecholamine was manifested after the removal of each drug in an abnormal inorganic ion milieu. Therefore, those drugs on atrial cell membrane are considered to have blocked the stimulant action of endogenous catecholamine.

As shown in experiment 3, on the atria excised from reserpinized animal, the augmentation effect of adrenaline was suppressed by previous addition of tyramine or ephedrine in each high concentration, though on normal atria the effect of adrenaline was not suppressed by tyramine but was suppressed by ephedrine. Therefore, the catecholamine membrane-receptor of atria excised from reserpinized animal might be denatured, and tyramine and ephedrine may be found to have a similar occupation effect on the receptor.

Clinically, the tachyphylaxis phenomenon is more often encountered with ephedrine than with tyramine. In vivo application also has shown that the reduction of pressor response by repeated administration is brought out more easily by ephedrine than by tyramine. As shown in experiment 2, the effects of these drugs on the catecholamine granule-receptor were so similar, that the difference between the degrees of tachyphylaxis by tyramine and ephedrine may be due to that between their occupation effects on catecholamine membrane-receptor.

Though we have emphasized the suppression of tyramine or ephedrine on the effect of catecholamine in this paper, the fact also has been reported that a pressor effect of adrenaline is potentiated by previous administration of tyramine or ephedrine. Experiment 1, however, showed that on the atrial preparation the augmentation effect of adrenaline
is scarcely potentiated by previous addition of tyramine or ephedrine. The difference of each experimental result between in vivo and in vitro may be attributed to that of response of each organ to tyramine and ephedrine.

SUMMARY

The effects of tyramine and ephedrine on atrial contractions of rabbits and guinea-pigs regarding catecholamine receptor and endogenous catecholamine mobilization have been investigated and discussed on the difference in pharmacological action mechanism between tyramine and ephedrine.

1. Tyramine and ephedrine showed the augmentation effect on atrial contractions, and the effect of each drug was indistinct if the atria had been prepared from reserpine pretreated animal.

2. The augmentation effect of adrenaline on the contractions was not influenced by the previous addition of tyramine or ephedrine, but was suppressed by a high concentration of ephedrine. On the atria prepared from reserpinized animal, the effect of adrenaline was suppressed in the presence of tyramine as well as ephedrine in each high concentration.

3. The augmentation effect of nicotine on the contractions was decreased in the presence of tyramine as well as ephedrine.

4. The contractions of normal atria, arrested by the addition of reserpine to the bath could be restored by tyramine or ephedrine addition after added reserpine had been washed out.

5. In the presence of cocaine neither tyramine nor ephedrine showed the augmentation effect on atrial contractions.

6. Atrial contractions arrested in modified Locke's solution which contains a high concentration of potassium were restored by the addition of tyramine or ephedrine, and the restored contractions were augmented by wash out of the drug with the same solution.

From these experimental results it was discussed that 1) tyramine and ephedrine may activate endogenous catecholamine in the granule structure, 2) ephedrine may possess the occupation effect on the catecholamine membrane-receptor of atria, 3) on normal atria, the positive inotropic and chronotropic effects of ephedrine through the activation of endogenous catecholamine surpass the catecholamine membrane-receptor occupation effect, 4) on the atria excised from reserpinized animal, tyramine may act in a similar manner to ephedrine on denatured catecholamine membrane-receptor, and 5) cocaine may act on the granule structure in atria and depress the endogenous catecholamine release caused by tyramine and ephedrine.

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