COMPARATIVE STUDY OF CHRONOTROPIC EFFECTS OF CATECHOLAMINES AND TYRAMINE ON THE SA NODE OF THE DOG HEART IN SITU

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Abstract—The relative potency of chronotropic effects of dopamine (DA), l-norepinephrine (NE), l-epinephrine (EP), l-isoproterenol (ISO) and tyramine (TY) was determined by direct application into the sinus node artery of the in situ canine heart (n=10), which was treated with atropine. The order of potencies of chronotropic effect of these amines was ISO > NE = EP > DA ≥ TY. The ratio of doses required to produce 50% increase in sinus rate was ISO: NE: EP: DA: TY = 1/30: 1: 1: 10: 10. At any dose level for inducing the same grade of sinus acceleration, the duration of action of NE and EP was almost equal and short, while that of ISO was usually longer. The duration of action of DA was short at a lower dose level but became equal to ISO with a medium dose. At a larger dose to produce a maximum response, DA acted longer than ISO, which indicates direct effect of DA with induction of indirect effect of catecholamine release. This suggests a tyramine-like action of DA.

Concerning the potency of catecholamines to produce a positive chronotropic response, there are a considerable number of elaborate studies already published. These were reviewed by Trautwein (1) in 1963 and briefly summarized as follows: l-Norepinephrine was slightly more potent than l-epinephrine for an acceleration of the sinus pacemaker (2–6), and isoproterenol was found from 10 to 15 times more potent than l-norepinephrine and l-epinephrine (7). More important developments followed. James and Nadeau (8) established an unique technique for perfusion of the SA node artery of in situ heart of the dog and compared doses of epinephrine, norepinephrine and isoproterenol to produce a maximum tachycardia by direct application into the sinus node artery (9). In 1965, Angelakos (10) investigated the regional distribution of catecholamines in the dog heart and found that dopamine was particularly concentrated in the SA node region. Then Rolett and Black (11) in 1966 compared potencies of dopamine and norepinephrine by infusing both catecholamines into the sinus node artery of the canine heart and found that both amines produced a similar dose-dependent increase of sinus rate with dopamine being approx. one-tenth as potent as norepinephrine. Later similar results were obtained by James et al. (12).

Previously Hashimoto et al. (13–16) found that the administration of dopamine, norepinephrine and epinephrine from 0.1 to 10 μg often induced a deceleration of the sinus rate, while isoproterenol never induced such a deceleration response. The participation of a cholinergic mechanism in this deceleration response was confirmed by blocking it
with atropine while enhancing it with physostigmine. On the other hand, concerning the mechanism of sympathomimetic effect of dopamine, much evidence as "mixed amine" (17) has accumulated, that there is partly induced tyramine-like effect by releasing norepinephrine from sympathetic storage sites.

The present study was designed to investigate chronotropic effects of dopamine, L-norepinephrine, L-epinephrine and L-isoproterenol and compare them with tyramine in equimolar base on the SA node after atropine treatment, using a direct perfusion technique under constant pressure of 100 mmHg. The effect was investigated not only on the maximum response but also on the duration.

METHODS

Ten mongrel dogs of both sexes, weighing 8 to 12 kg, were anesthetized with sodium pentobarbital, 30 mg/kg, i.v. A tracheal tube was inserted and artificial respiration was performed by use of a Harvard respirator. The chest was opened at the right 4th or 5th intercostal space. The pericardium was incised and a pericardial cradle was arranged to keep the heart in the proper position. All experiments were performed under constant perfusion pressure at 100 mmHg as the SA node is responsive to pressure changes (18). Arterial blood pressure in the carotid or femoral artery was measured continuously with an electromanometer (Nihon Kohden RP-2). The ECG (lead II) was recorded on an electrocardiograph (Nihon Kohden ME-20-TR), and the heart rate was continuously registered by use of a cardiotachograph (Nihon Kohden RT-2), which was triggered by the R wave of lead II. Blood flow to the sinus node artery was measured by an electromagnetic flowmeter (Nihon Kohden MF-2). The average flow rate of the sinus node artery was 2.2 ± 0.3 ml/min (mean ± S.E.) at 100 mmHg of perfusion pressure (n = 9).

Sodium heparin, 500 to 1000 U/kg, was given at the beginning of the perfusion and 200 U/kg was added at one-hr intervals. Both vagi were cut at the middle of the neck. The injected volume was 0.01 to 0.03 ml given in a period of 4 sec by use of a microsyringe. This volume of saline did not cause any chronotropic effect. The details of preparation have been described in the previous papers (18, 19).

The compounds used were dopamine hydrochloride (Kyowa Hakko), L-norepinephrine (Fluka AG), L-epinephrine (Merck AG), L-isoproterenol hydrochloride (Nikken Kagaku), tyramine hydrochloride (Wako) and atropine sulfate.

RESULTS

1) Comparison of the positive chronotropic effects of dopamine, L-norepinephrine, L-epinephrine, L-isoproterenol and tyramine

As Hashimoto et al. (16) reported, naturally occurring catecholamines injected into the sinus node artery often induced a deceleration of sinus rate which was blocked by treatment with atropine. Thus, 10 µg of atropine was given into the sinus node artery before administration of test compounds. The threshold doses of dopamine and tyramine, norepinephrine and epinephrine, and isoproterenol were 0.1 nmol, 0.01 nmol, and 0.0001
nmol, respectively. Each catecholamine and tyramine induced dose-dependent sinus acceleration and a maximum rate of sinus acceleration was 240 to 250 beats/min as shown in Figs. 1 and 2. The ratios of ED 50 of tyramine, dopamine, l-norepinephrine, l-epinephrine and l-isoproterenol are 10: 10: 1: 1: 1/30, respectively. Summarized data are shown in Table 1.

Fig. 1. Effects of increasing doses of catecholamines and tyramine injected into the sinus node artery. SBP, systemic blood pressure; HR, heart rate.

Fig. 2. Effects of l-isoproterenol (ISO), l-norepinephrine (NE), l-epinephrine (EP), dopamine (DA) and tyramine (TY) on the SA nodal pacemaker. Numbers in parentheses indicate the number of animals. The values are the mean± standard errors.
### TABLE 1. Chronotropic responses to catecholamines and tyramine.

<table>
<thead>
<tr>
<th>Compounds</th>
<th>No. of dogs</th>
<th>Control HR (beats/min)</th>
<th>Maximum HR (beats/min)</th>
<th>Percent increase in HR (%)</th>
<th>Duration of PCA (sec)</th>
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<td><strong>Dopamine</strong></td>
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<tr>
<td>0.1</td>
<td>4</td>
<td>159 ± 3*</td>
<td>171 ± 8</td>
<td>9 ± 4</td>
<td>13 ± 5</td>
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<td>0.3</td>
<td>6</td>
<td>148 ± 8</td>
<td>178 ± 8</td>
<td>19 ± 3</td>
<td>43 ± 4</td>
</tr>
<tr>
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<td>6</td>
<td>150 ± 6</td>
<td>211 ± 12</td>
<td>41 ± 7</td>
<td>115 ± 19</td>
</tr>
<tr>
<td>3</td>
<td>6</td>
<td>151 ± 5</td>
<td>227 ± 11</td>
<td>51 ± 8</td>
<td>188 ± 37</td>
</tr>
<tr>
<td>10</td>
<td>6</td>
<td>151 ± 7</td>
<td>241 ± 7</td>
<td>62 ± 11</td>
<td>597 ± 53</td>
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<tr>
<td><strong>Norepinephrine</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.01</td>
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<td>149 ± 7</td>
<td>166 ± 7</td>
<td>12 ± 2</td>
<td>15 ± 3</td>
</tr>
<tr>
<td>0.03</td>
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<td>151 ± 5</td>
<td>188 ± 7</td>
<td>24 ± 6</td>
<td>28 ± 5</td>
</tr>
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<td>214 ± 10</td>
<td>44 ± 7</td>
<td>53 ± 20</td>
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<td>246 ± 11</td>
<td>61 ± 9</td>
<td>70 ± 23</td>
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<td>250 ± 5</td>
<td>75 ± 8</td>
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<td>31 ± 8</td>
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<tr>
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<td>49 ± 8</td>
<td>58 ± 19</td>
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<td>130 ± 31</td>
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<td>228 ± 13</td>
<td>47 ± 9</td>
<td>192 ± 24</td>
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<td>168 ± 6</td>
<td>12 ± 3</td>
<td>52 ± 25</td>
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<tr>
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<td>192 ± 8</td>
<td>23 ± 5</td>
<td>130 ± 74</td>
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<td>4</td>
<td>158 ± 5</td>
<td>221 ± 14</td>
<td>42 ± 9</td>
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<td>245 ± 9</td>
<td>62 ± 10</td>
<td>670 ± 173</td>
</tr>
</tbody>
</table>

* Mean ± standard error of the mean. HR, heart rate. PCA, positive chronotropic action.

2) **Comparison of the duration of actions of catecholamines and tyramine**

At a smaller dose level for inducing the same grade of sinus acceleration from 160 to 170 beats/min, the duration of action of dopamine (0.1–0.3 nmol), L-norepinephrine (0.01–0.03 nmol) and L-epinephrine (0.01–0.03 nmol) was almost identical, while that of L-isoproterenol (0.0001–0.0003 nmol) and tyramine (0.3–1.0 mmol) was significantly longer acting than others. At a medium dose level for inducing the same medium grade of sinus acceleration, approx. 200 beats/min, the duration of action of L-norepinephrine (0.1–0.3 nmol) and L-epinephrine (0.1–0.3 nmol) was almost identical. L-Isoproterenol
(0.001–0.01 nmol) and dopamine (1–3 nmol) showed almost the same duration of action which was significantly longer than 1-norepinephrine and 1-epinephrine. At a larger dose level for inducing a maximum response from 240 to 250 beats/min, 1-norepinephrine (1 nmol) and 1-epinephrine (1 nmol) were shorter acting than 1-isoproterenol (0.03 nmol), while dopamine (10 nmol) showed even longer duration of action than 1-isoproterenol. Tyramine usually produced longer acting acceleration than the other three catecholamines.

DISCUSSION

It is generally accepted that the SA node responds to catecholamines with an acceleration, however, Katoh (20) observed a deceleration of sinus rate by topical application of naturally occurring catecholamines on the dog SA node. Later, Hashimoto et al. (13–16, 21) observed frequent occurrence of such deceleration response when either naturally occurring catecholamine or phenylephrine was injected into the sinus node artery. This paradoxical response was elucidated to be caused by adrenergic-cholinergic interaction at the peripheral site which was readily blocked by treatment with atropine and tetrodotoxin. After atropine treatment an acceleration response occurred as usual. In the present experiments, the SA node was pretreated with atropine in these experiments. Although it is reported that 1-norepinephrine is more potent than 1-epinephrine in accelerating the pacemaker, a difference between 1-norepinephrine and 1-epinephrine was not observed in this study, but 1-isoproterenol exhibited the most potent chronotropic effects among these catecholamines, i.e., 30 times more potent than 1-norepinephrine and 1-epinephrine. James and Nadeau (9) reported that isoproterenol is approx. ten times potent than epinephrine and norepinephrine, and that duration of action was similar between three catecholamines. Later, James and Nadeau (22) studied the effect of tyramine injected into the sinus node artery. The duration of sinus tachycardia is much longer than that induced by norepinephrine. In 1966, Rollett and Black (11) reported that dopamine was approx. one-tenth as potent as norepinephrine by use of an original technique developed by James and Nadeau (8). In the present study, similar results were obtained. Dopamine induced dose-dependently sinus acceleration but it was one-tenth as potent as 1-norepinephrine and 1-epinephrine, however, the response pattern changed from the short acting to the long acting one when doses were increased from the threshold to a dose for inducing a maximum response. At a maximum response, the duration of action of dopamine became even longer than that of 1-isoproterenol which produced in general, a long-lasting effect. This is probably due to indirect response of released catecholamine caused by tyramine-like action of dopamine. Many investigators have demonstrated that responses to dopamine are reduced but never potentiated by pretreatment with cocaine (23, 24), desmethylinimipramine (25) and reserpine (23, 26, 27). This shows dopamine exerts cardiac effects partly by acting directly on beta-adrenergic receptors and partly by releasing norepinephrine from sympathetic storage sites and should be classified as "mixed amine" (17). Tuttle (25) compared the chronotropic and inotropic effects of dopamine in the in situ dog heart and concluded that the chronotropic response to dopa-
mine should be ascribed to the direct effect, while the inotropic response is predominantly
due to released catecholamine. The present study suggests, however, that the indirect tyra-
mine-like component becomes predominant even in the chronotropic response when a
larger dose of dopamine is administered directly into the sinus node artery.

In 1965, Angelakos (10) observed that dopamine was present in higher concentrations
in the atria than in the ventricles and was particularly concentrated in the SA node re-
gion. Since Carlsson (28) discovered a higher concentration of dopamine in the central
nervous system and a physiological role was suggested, characteristic effects of dopamine
in the peripheral circulation (29-34) and also in the pancreas secretion (35) were succes-
sively discovered, all of which are blocked by haloperidol. Thus it is that the special
dopamine receptor has come to be accepted in general. The previous report (12), how-
ever, showed that the acceleration response to dopamine can be blocked by adrenergic
beta-blocker. Thus, we suggest that the concentrated dopamine into the SA node area
plays a physiological role as the precursor of norepinephrine.

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