Mechanism of Action of Carbocyclic Thromboxane A₂ and Its Interaction with Prostaglandin I₂ and Verapamil in Isolated Arteries

Noboru Toda
From the Department of Pharmacology, Shiga University of Medical Sciences, Seta, Ohtsu 520-21, Japan

SUMMARY. Carbocyclic thromboxane A₂ (10⁻⁹ to 10⁻⁷ m) produced a concentration-dependent contraction of helical strips of dog cerebral, coronary, mesenteric, renal, and femoral arteries and of monkey cerebral, coronary, and mesenteric arteries. Contractions induced by low concentrations of carbocyclic thromboxane A₂ tended to be greater in cerebral arterial strips. Even after 60 minutes of exposure to Ca⁺⁺-free media, approximately one-half of the contractile response of dog cerebral and mesenteric arteries to 10⁻⁸ m carbocyclic thromboxane A₂ was retained. The contractile response was attenuated by diphloretin phosphate, a prostaglandin antagonist, and was potentiated by aspirin. In dog cerebral arterial strips contracted with carbocyclic thromboxane, the relaxant response to prostaglandin I₂ was less than the response seen in mesenteric and coronary arteries, whereas, in contrast, the response to verapamil was greater in cerebral arteries. Concentration-relaxation curves for papaverine did differ appreciably. It may be concluded that contractions induced by carbocyclic thromboxane are associated with the release of Ca⁺⁺ from intracellular storage sites and, in addition, the increase in transmembrane Ca⁺⁺ influx. Greater susceptibility of cerebral arteries to verapamil may indicate that the Ca⁺⁺ antagonist is of use to relieve the persistent contraction of cerebroarterial smooth muscle. Prostaglandin I₂ appears to counteract effectively the action of potent vasoconstrictors, such as thromboxane A₂ and its carbocyclic analog, in various vascular beds. (Circ Res 51: 675-682, 1982)

THROMBOXANE A₂ (TXA₂) was first introduced by Hamberg et al. (1975) as an unstable intermediate, which is converted from prostaglandin (PG) endoperoxides by catalysis of an enzyme contained in the microsomal fraction of human and horse platelets. TXA₂ in minute amounts produces a contraction of isolated rabbit aortae (Needleman et al., 1976), rabbit coeliac and mesenteric arteries (Bunting et al., 1976), bovine cerebral and coronary arteries (Ellis et al., 1977), porcine coronary arteries (Svensson and Hamberg, 1976), and guinea pig coronary vasculature (Terashita et al., 1978) and also a platelet aggregation (Hamberg et al., 1975). Therefore, TXA₂ is regarded as one of the endogenous substances responsible for coronary and cerebral vasospasm. However, the rapid degradation of this substance in buffer solutions [the half life of about 30 seconds (Hamberg et al., 1975)] makes the analysis of its vascular actions difficult. Recently, Lefer et al. (1980) have synthesized a stable analog of TXA₂, carbocyclic TXA₂ (cTXA₂), which is a potent coronary vasoconstrictor but does not show a platelet-aggregating activity.

The present study was undertaken to compare the action of cTXA₂ on isolated dog cerebral, coronary, mesenteric, renal, and femoral arteries of similar size and on isolated monkey cerebral, coronary, and mesenteric arteries, to analyze the mechanism of action of cTXA₂ in the dog arteries, and to determine different effectiveness of vasodilator agents, such as PGI₂, verapamil, and papaverine, in a variety of dog arteries contracted with cTXA₂.

Methods
Mongrel dogs of either sex, weighing 8-15 kg, were anesthetized with intraperitoneal injections of sodium thiopental (50 mg/kg) and killed by bleeding from the carotid arteries. Japanese monkeys (Macaca fuscata) of either sex, weighing 6-11 kg, were anesthetized with intramuscular injections of ketamine (25-40 mg/kg) and sacrificed. The brain, heart, and kidneys were rapidly removed. Basilar and middle cerebral arteries (0.6-0.8 mm outside diameter in dogs; 0.5-0.7 mm in monkeys) were isolated from the brain, ventral interventricular, and circumflex branches of the left coronary artery (0.7-0.9 mm in dogs; 0.5-0.7 mm in monkeys) were isolated from the heart, and intrarenal, interlobar branches of the renal artery (0.6-0.8 mm in dogs) were isolated from the kidneys. Distal portions of superior mesenteric (0.6-0.9 mm in dogs; 0.4-0.7 mm in monkeys) and femoral arteries (0.6-0.9 mm in dogs) also were isolated. The arteries were helically cut into strips, approximately 20 mm long (Toda, 1981). The specimen was vertically fixed...
between hooks in the muscle bath containing the nutrient solution, which was maintained at 37 ± 0.3°C and aerated with a mixture of 95% O2 and 5% CO2. The hook anchoring the upper end of the strips was connected to the lever of a force-displacement transducer (Nihonkoden Kogyo Co., Tokyo, Japan). The resting force was adjusted to 1.5 g for dog arteries (Toda et al., 1978) and to 1.0 g for monkey arteries (Toda, 1981). Constituents of the solution were as follows (mm): Na+, 140; K+, 5.4; Ca++, 2.2; Mg++, 1.0; Cl-, 131.8; HCO3-, 20.0; and dextrose, 5.6. The pH of the solutions was 7.28–7.35. Before the start of experiments, all the arterial strips were allowed to equilibrate in bathing media for 60–90 minutes, during which time the bathing media were replaced every 10–15 minutes.

Isometric contractions and relaxations were displayed on an ink-writing oscillograph (Nihonkoden Kogyo Co.). The contractile response to 30 mM K+ was obtained first, then the preparations were repeatedly washed and equilibrated for 30–40 minutes. Responses to cTxA2 were obtained in arterial strips under resting conditions, and those to vasodilator drugs, such as PGI2, verapamil, and papaverine, were obtained in the strips previously contracted with 10−7 M cTxA2. The concentration-response curve was obtained by adding these drugs directly to the bathing media in cumulative concentrations. At the end of each series of experiments with the vasodilator drugs, papaverine in a concentration of 10−4 M was added to attain the maximum relaxation (Toda, 1974a), and relaxations relative to those induced by papaverine are presented. Preparations had been treated for 20–30 minutes with diphloretin phosphate (DPP), aspirin, phenolamine, chlorpheniramine, and cinanserin before the response to cTxA2 was obtained.

Some of dog cerebral and mesenteric arterial strips were studied for the dependence of cTxA2-induced contractions upon extracellular Ca++. After the contractile response to 10−8 M cTxA2 was obtained in normal media, the strips were repeatedly washed with drug-free solutions and exposed to Ca++-free media for 60 minutes, during which time the solutions were replaced twice every 20 minutes. cTxA2 (10−8 M) then was added. After the cTxA2-induced contraction leveled off, Ca++ (2.2 and 4.4 mM) was added. The results shown in the text, figures, and tables are expressed as mean values ± SEM. Statistical analyses were made using the Tukey's method after the one-way analysis of variance (Wallenstein et al., 1980) (Figure 1 and Tables 1, 2, and 3) or the Student's paired and unpaired t-test. Drugs used were carbocyclic thromboxane A2 (cTxA2), prostaglandin (PG) I2 sodium salt, diphloretin phosphate (DPP), aspirin, phenolamine, chlorpheniramine, and cinanserin. cTxA2 was synthesized in Ono Pharmaceutical Co. by the method reported by Ohuchida et al. (1979). This substance was dissolved in absolute alcohol to make a stock solution (10−3 M) then was added. After the cTxA2-induced contraction was obtained in normal media, the strips were repeatedly washed and exposed to Ca++-free media for 60 minutes, during which time the solutions were replaced twice every 20 minutes. cTxA2 (10−8 M) then was added. After the cTxA2-induced contraction leveled off, Ca++ (2.2 and 4.4 mM) was added.

The results shown in the text, figures, and tables are expressed as mean values ± SEM. Statistical analyses were made using the Tukey's method after the one-way analysis of variance (Wallenstein et al., 1980) (Figure 1 and Tables 1, 2, and 3) or the Student's paired and unpaired t-test. Drugs used were carbocyclic thromboxane A2 (cTxA2), prostaglandin (PG) I2 sodium salt, diphloretin phosphate (DPP), aspirin, phenolamine, chlorpheniramine, and cinanserin. cTxA2 was synthesized in Ono Pharmaceutical Co. by the method reported by Ohuchida et al. (1979). This substance was dissolved in absolute alcohol to make a stock solution (10−3 M) and diluted with distilled water before use.

**Results**

**Contractile Response to cTxA2 of a Variety of Arteries**

In helically cut strips of dog cerebral, coronary, mesenteric, renal, and femoral arteries, the addition of cTxA2 in concentrations in a range between 10−10 and 10−7 M caused a dose-related contraction (Fig. 1, left). Further increase in the concentration to 10−6 M produced only a slight or no additional contraction.
coronary arteries. Compared with dog arteries, monkey arterial strips responded with a greater contraction to $10^{-6}$ M cTxA2. Apparent ED$_{50}$ values in monkey cerebral and coronary arteries (Table 1) were significantly less than those in the dog arteries.

In dog cerebral arteries exposed for 60 minutes to Ca$^{++}$-free media, cTxA$_2$ ($10^{-8}$ M) produced a tonic contraction, the magnitude of which was 48.9 ± 7.9% ($n = 8$) of the contraction seen in normal media. When the cTxA$_2$-induced contraction stabilized, the addition of 2.2 mM Ca$^{++}$ elicited a marked, transient contraction followed by a relaxation and a persistent contraction (Fig. 3). Change in the Ca$^{++}$ concentration from 2.2 to 4.4 mM produced a relaxation. The persistent contraction stabilized during the exposure to Ca$^{++}$ for 30 to 60 minutes at a level similar to that attained by cTxA$_2$ in the normal media. Quantitative data are presented in Figure 4, upper panel. Similar results were obtained with mesenteric arteries exposed to Ca$^{++}$-free media (Fig. 4, lower), although the transient contraction was not so evident as that seen in cerebral arteries (123% vs. 172%, relative to contractions induced by $10^{-8}$ M cTxA$_2$ in normal media).

In cerebral arterial strips, the Ca$^{++}$ (2.2 mM)-induced contraction was potentiated by treatment with $2 \times 10^{-7}$ M ouabain; the transient relaxation was abolished and the contraction persisted for 20 minutes or longer (stabilized contractions of 195 ± 24.2%, $n = 7$, relative to contractions induced by cTxA$_2$ in normal media, cf. 103 ± 8.0% shown as B in Figure 4, upper panel), as was the response to Ca$^{++}$ in the arteries exposed to Ca$^{++}$-free media and depolarized by excess K$^+$ (Toda, 1974b). When the Ca$^{++}$-induced contraction was stabilized, the further addition of 2.2 mM Ca$^{++}$ produced an additional contraction in the presence of ouabain ($n = 6$). Treatment with verapamil ($10^{-7}$ and $10^{-6}$ M) abolished the phasic contraction and relaxation induced by Ca$^{++}$ and attenuated the tonic contraction in a concentration-dependent manner; mean values of the tonic contraction induced by cTxA$_2$ plus Ca$^{++}$ were 68.6 ± 8.5% ($n = 5$) and 26.8 ± 3.1% ($n = 6$), respectively (cf. control value of 103 ± 8.0%, $n = 8$). Aspirin ($5 \times 10^{-5}$ M) did not alter the triphasic response to Ca$^{++}$ ($n = 5$).

### Table 1

<table>
<thead>
<tr>
<th>Artery</th>
<th>$n$</th>
<th>ED$_{50}$ ($\times 10^{-9}$ M)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dog</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cerebral</td>
<td>17</td>
<td>5.09 ± 0.84</td>
</tr>
<tr>
<td>Coronary</td>
<td>18</td>
<td>5.15 ± 0.56</td>
</tr>
<tr>
<td>Mesenteric</td>
<td>13</td>
<td>3.92 ± 0.12</td>
</tr>
<tr>
<td>Renal</td>
<td>14</td>
<td>4.16 ± 0.22</td>
</tr>
<tr>
<td>Femoral</td>
<td>12</td>
<td>4.00 ± 0.20</td>
</tr>
<tr>
<td>Monkey</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cerebral</td>
<td>14</td>
<td>2.12 ± 0.50*</td>
</tr>
<tr>
<td>Coronary</td>
<td>8</td>
<td>1.34 ± 0.48†</td>
</tr>
<tr>
<td>Mesenteric</td>
<td>8</td>
<td>4.10 ± 0.99</td>
</tr>
</tbody>
</table>

* Significantly different from the value with dog cerebral arteries, $P < 0.01$.
† Significantly different from the value with dog coronary arteries, $P < 0.001$. F ratios obtained by the analysis of variance (6.21 for dog arteries and 3.9% for monkey arteries) are greater than the $P = 0.005$ and 0.05 critical values, respectively. However, there was no significant difference between ED$_{50}$ values in different dog arteries and those in monkey arteries.
Modification by Antagonists of the Response to cTxA2 of Dog Arteries

Contractions induced by 10^{-7} M cTxA2 were not completely reversed by repeated washing of preparations at intervals of 10-15 minutes during an observation period of 3-4 hours. However, the contractions induced at the lower concentrations (10^{-8} M or lower) were completely reversed by repeated washing, although it took 90-120 minutes to attain the stabilized level comparable to that prior to the addition of cTxA2. When the contractions were completely reversed, the response to 10^{-8} M cTxA2 were reproducible three times. Therefore, the influence of antagonists on the response to a single concentration (10^{-8} M) of cTxA2 was tested.

Contractile responses of cerebral and mesenteric arteries to cTxA2 (10^{-8} M) were attenuated by treatment for 30 min with diphloretin phosphate (DPP, 3 \times 10^{-6} and 10^{-5} M) in a dose-dependent manner (Fig. 5). Even when the response to 10^{-8} M of the drug was markedly suppressed by 10^{-5} M DPP, the preparations responded with a moderate contraction to 10^{-7} M cTxA2. The attenuations by DPP of the response of cerebral and mesenteric arteries did not differ significantly. Treatment with these concentrations of DPP did not inhibit the contractile response to serotonin in four of four cerebral arteries or the response to norepinephrine in four of four mesenteric arteries.

Treatment for 20 minutes with 5 \times 10^{-5} M aspirin significantly potentiated the contractions of cerebral and mesenteric arteries induced by 10^{-8} M cTxA2 (Fig. 6). The potentiation was reversed by repeated washing of preparations.

Contractile responses to cTxA2 were not significantly influenced by treatment with 10^{-6} M phentolamine, 10^{-6} M cinanserin, and 10^{-6} M chlorpheniramine; as compared with contractions before the treatment, percent changes averaged -0.7 \pm 6.4 (n = 3), +3.3 \pm 3.3 (n = 4), and -2.0 \pm 0.9 (n = 5), respec-
tively, in cerebral arteries, and +2.0 ± 0.9 (n = 4), +5.6 ± 3.1 (n = 5) and -0.4 ± 3.0 (n = 5), respectively, in mesenteric arteries.

Responses to Vasodilator Drugs of Dog Arteries Contracted with cTxA2

In cerebral, coronary, mesenteric, and renal arterial strips contracted with 10^{-7} M cTxA2, the effect of vasodilator drugs, including PGI2, verapamil, and papaverine, were compared. PGI2 rapidly relaxed the arterial strips (Fig. 2). The addition of PGI2 in concentrations ranging from 10^{-9} to 10^{-6} M caused a dose-dependent relaxation (Fig. 7, left). The relaxant response of cerebral arteries was less than the response of the other arteries (Table 2). Relaxations of mesenteric and coronary arteries were greater; apparent ED50 values of these arteries were significantly less than the value of cerebral arteries (Table 3).

Verapamil caused a slowly-developing relaxation, which leveled off within 10 to 30 min (Fig. 2). In contrast to PGI2, verapamil relaxed cerebral arteries to a greater extent than the other arteries (Fig. 7, middle). Although the maximum relaxation of cerebral arteries induced by 10^{-5} M verapamil was greater than that of the other arteries (Table 2), apparent ED50 values did not significantly differ in these arteries (Table 3).

Papaverine (10^{-7} to 10^{-4} M) relaxed the arterial strips in a dose-dependent manner. Increase in the concentration to 5 × 10^{-4} M did not produce an additional relaxation, as previously reported (Toda, 1974a). Therefore, the relaxation induced by 10^{-4} M papaverine was taken as 100%. Relaxant responses to (Fig. 7, right) and apparent ED50 values of papaverine (Table 3) in cerebral, coronary, mesenteric, and renal arteries did not differ significantly.

### Table 2

Mean Values of the Maximum Relaxation Induced by PGI2 and Verapamil Relative to the Papaverine (10^{-4} M)-Induced Relaxation in Dog Arterial Strips

<table>
<thead>
<tr>
<th>Artery</th>
<th>PGI2</th>
<th>P &lt;</th>
<th>Verapamil</th>
<th>P &lt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cerebral</td>
<td>55.0 ± 4.31</td>
<td>0.01</td>
<td>74.0 ± 2.72</td>
<td></td>
</tr>
<tr>
<td>Coronary</td>
<td>81.5 ± 2.14</td>
<td>0.01</td>
<td>54.8 ± 4.47</td>
<td>0.05</td>
</tr>
<tr>
<td>Mesenteric</td>
<td>80.8 ± 4.04</td>
<td>0.01</td>
<td>42.5 ± 3.59</td>
<td>0.01</td>
</tr>
<tr>
<td>Renal</td>
<td>62.4 ± 5.63</td>
<td>0.05</td>
<td>57.3 ± 2.27</td>
<td></td>
</tr>
</tbody>
</table>

* n = number of preparations used. F-ratios obtained from the analysis of variance (6.75 for PGI2 and 14.6 for verapamil) are greater than the P = 0.005 critical value. Only the values significantly different by Tukey's method are included.

### Table 3

Mean Values of the Apparent Median Effective Concentration (ED50) of PGI2, Verapamil and Papaverine in Dog Arterial Strips

<table>
<thead>
<tr>
<th>Artery</th>
<th>PGI2 (x 10^{-8} M)</th>
<th>P &lt;</th>
<th>Verapamil (x 10^{-7} M)</th>
<th>P &lt;</th>
<th>Papaverine (x 10^{-6} M)</th>
<th>P &lt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cerebral</td>
<td>8.17 ± 1.43</td>
<td>0.00</td>
<td>1.00 ± 0.13</td>
<td>0.05</td>
<td>2.86 ± 0.36</td>
<td>0.05</td>
</tr>
<tr>
<td>Coronary</td>
<td>1.91 ± 0.43</td>
<td>0.05</td>
<td>1.80 ± 0.24</td>
<td>0.05</td>
<td>4.38 ± 0.59</td>
<td>0.05</td>
</tr>
<tr>
<td>Mesenteric</td>
<td>2.27 ± 0.50</td>
<td>0.05</td>
<td>1.60 ± 0.38</td>
<td>0.05</td>
<td>3.19 ± 0.55</td>
<td>0.05</td>
</tr>
<tr>
<td>Renal</td>
<td>4.60 ± 1.50</td>
<td>0.05</td>
<td>2.20 ± 0.46</td>
<td>0.05</td>
<td>4.58 ± 0.69</td>
<td>0.05</td>
</tr>
</tbody>
</table>

* Numbers in parentheses indicate the number of preparations used. F ratios obtained by the analysis of variance (6.75 for PGI2 and 3.04 for verapamil) are greater than the P = 0.005 and 0.05 critical values, respectively. Only the values significantly different by the Tukey's method are included. The F ratio for papaverine (2.40) is less than the P = 0.05 critical value.
Discussion

The addition of cTxA2 elicited a concentration-related, persistent contraction in dog cerebral, coronary, mesenteric, renal, and femoral arteries and monkey cerebral, coronary, and mesenteric arteries; the contractions seen at low concentrations tended to be greater in cerebral arteries. Maximum contractions induced by 10^{-7} M cTxA2 were identical (dog cerebral arteries) to or greater than those induced by 30 mM K+ (other arteries used), whereas maximum contractions induced by 10^{-5} M PGF_{2\alpha} and E\alpha are reportedly less than the K+-induced contractions [approximately 60 and 70%, respectively, in dog cerebral arteries (Toda and Miyazaki, 1978)]. cTxA2 is one of the most potent vasoconstrictors in isolated dog and monkey arteries. TxA2, possibly generated by the addition of PGH2 (255 nM) to particulate fraction of human platelets, produces a marked contraction in bovine cerebral arterial strips (approximately 150% of the contraction induced by 40 mM K+), but produces only a moderate contraction in bovine coronary arteries and porcine coronary and renal arteries (30-50% of the contraction) (Ellis et al., 1977). Contractions induced by 10^{-8} M cTxA2 in dog cerebral and mesenteric arteries exposed for 60 minutes to Ca++-free media were 49 and 53%, respectively, of the contractions obtained in normal bathing media. Therefore, increase in the transmembrane influx of Ca++ as well as the release of Ca++ from intracellular storage sites is involved in the cTxA2-induced contraction. Since prolonged exposure of the arteries to Ca++-free media is considered to reduce intracellular Ca++, such an involvement of Ca++ release would be underestimated. On the other hand, contractile responses to PGF_{2\alpha} and serotonin in Ca++-free media relative to those in normal media are approximately 25 and 15%, respectively, in cerebral arteries, and 14 and 8%, respectively, in mesenteric arteries (Toda, 1982a). Thus, the ability of cTxA2 to release intracellularly stored Ca++ appears to be greater than that of PGF_{2\alpha} and serotonin.

In dog cerebral arteries exposed to Ca++-free media and stimulated by cTxA2, Ca++ caused a triphasic response, a transient contraction, relaxation, and sustained contraction. Similar responses are obtained in dog cerebral arterial strips placed under the same experimental conditions, except for the fact that PGF_{2\alpha} and K+ are used as stimulants (Toda, 1974b; Toda, 1982a). The relaxation was not influenced by aspirin, suggesting that the involvement of vasodilator PG's is excluded. Vasodilator PGI2 is reportedly formed from PGH2 in the wall of dog cerebral and extracerebral arteries (Toda, 1980) and is released from the arterial wall by activation of angiotensin II receptors (Toda and Miyazaki, 1981). Ouabain potentiated the Ca++-induced contraction and abolished the relaxation in dog cerebral arteries stimulated by cTxA2 (present study) as well as PGF_{2\alpha} (Toda, 1982a) and K+ (Toda, 1974b). On the other hand, the transient contraction and relaxation were abolished by treatment with Ca++ antagonists, including verapamil (present study) and nicardipine (Toda, unpublished data). These findings suggest that the rapid transmembrane influx of Ca++ triggers the relaxation, which may be associated with the mechanism sensitive to ouabain. Webb and Bohr (1978) have postulated that the Ca++-induced relaxa-
Vascular Action of Carbocyclic Thromboxane A₂

Toda

The relaxation induced by PGI₂ was in the order of coronary and mesenteric > renal > cerebral arteries.

Papaverine in a concentration-dependent manner.

Activity of Na⁺-, K⁺-activated ATPase, since the relaxation in isolated rat tail arteries is dependent on the level lower than that prior to the addition of Ca²⁺ (Fig. 3) is not explained only by this assumption.

Relaxations induced by increasing the concentration of Ca²⁺ from 2.2 to 4.4 mM in cerebral arterial strips exposed to Ca²⁺-free media were reversed to contractions after treatment with ouabain. Whether this reversal is due to an abolition of the electrogenic Na⁺ pump activated by Ca²⁺ (Webb and Bohr, 1978), or to a possible increase in the Ca²⁺ permeability (Toda, 1982a) caused by a membrane depolarization, remains to be determined. However, the latter alternative may be supported by the finding that, after contractions induced by the first introduction of Ca²⁺ (2.2 mM) leveled off, the second addition of Ca²⁺ produces a relaxation in dog cerebral arterial strips exposed previously to Ca²⁺-free media and depolarized by 30 mM K⁺ (Hayashi and Toda, 1977) but, in contrast, a contraction in the arteries depolarized by K⁺ in concentrations of 50 mM or higher (Toda, unpublished data).

As compared with vascular effects of other PG’s, including PGA₁, A₂, D₂, E₁, E₂, and F₂a, contractions induced by cTxA₂ persisted longer even after extensive washing of preparations. cTxA₂ may bind to receptive sites of vascular smooth muscle in vitro more tightly than those PG’s. The contractile response of cerebral and mesenteric arteries to cTxA₂ was attenuated dose dependently by DPP, an analog of a PG antagonist polyphloretin phosphate (PPP) (Sanner, 1974), in concentrations insufficient to inhibit the contractions induced by serotonin and norpinephrine (present study). Similar attenuation by DPP as well as PPP has been observed in contractions induced by PGF₂α, E₂, and D₂ (Toda and Miyazaki, 1978; Toda, 1982b; unpublished data with DPP). On the other hand, treatment with aspirin potentiated the contractile response to cTxA₂ as did the response to PGF₂α, E₂, and D₂ (Toda and Miyazaki, 1978; Toda, 1982b).

The release of vasodilator PG’s from the vascular wall may partly interfere with the vasoconstrictor action. These findings suggest that cTxA₂, PGF₂α, PGE₂, and PGD₂ share the mechanism of vascular actions. cTxA₂-induced contractions were influenced neither by phenolamine, cinanserin, and chlorpheniramine in dog arterial strips (present study) nor by phenolamine, phenoxybenzamine, and saralasin in isolated, perfused cat coronary arteries (Smith et al., 1981), suggesting that α-adrenergic, serotoninergic, histaminergic H₁, and angiotensin II-related mechanisms are not involved.

Dog arterial strips contracted with cTxA₂ responded with a relaxation to PGI₂, verapamil, and papaverine in a concentration-dependent manner. The relaxation induced by PGI₂ was in the order of coronary and mesenteric > renal > cerebral arteries. Cerebral arteries partially contracted with PGF₂α are also less sensitive to PGI₂ than the other arteries (Toda, 1980). Such a heterogeneity of the PGI₂ action in a variety of dog arteries may be associated with different population or sensitivity of receptive sites or with different rates of degradation of PGI₂ since a nonselective vasodilator, papaverine, produced a similar extent of relaxations in a variety of dog arteries. Endogenous PGI₂ may participate in attenuating smooth muscle contractions of coronary arteries, such as in the case of variant angina (Tada et al., 1981), more effectively than those of cerebral arteries.

Relaxant responses to verapamil were in the order of cerebral > coronary and renal > mesenteric arteries. Greater relaxations by nimodipine, a Ca²⁺ antagonist, of isolated rabbit basilar arteries than saphenous arteries contracted with cTxA₂ have also been demonstrated (Towart and Perzborn, 1981). Susceptibility of cerebral arteries to Ca²⁺ antagonists, such as verapamil, diltiazem, and nifedipine, is greater than that of mesenteric arteries when contracted with PGF₂α (Shimizu et al., 1980). The fact that the profound contraction induced by high concentrations of cTxA₂ was reversed by verapamil predominantly in cerebral and coronary arteries suggests that Ca²⁺ antagonists may be effective in the treatment of cerebral and coronary vasospasm. In fact, the latter spasms is known to respond well to the Ca²⁺ antagonist therapy (Braunwald, 1981); in contrast, the effectiveness on delayed cerebral vasospasm is still controversial.

I thank M. Yamamoto for his excellent technical assistance.

Carbocyclic thromboxane A₂, prostaglandin 1₃ sodium salt, and diphloretin phosphate were kindly provided by Ono Pharmaceutical Co., Osaka, Japan.

This work was supported in part by Scientific Research Fund 56480102 from the Ministry of Education, Science, and Culture of Japan, and by Japan Heart Foundation and Tokio Marine Research Grant for 1980.

Address for reprints: Dr. Noboru Toda, Department of Pharmacology, Shiga University of Medical Sciences, Seta, Otsu 520-21, Japan.

Received April 19, 1982; accepted for publication August 19, 1982.

References


Hayashi S, Toda N (1977) Inhibition by Ca²⁺, verapamil and papaverine of Ca²⁺-induced contractions in isolated cerebral and peripheral arteries of the dog. Br J Pharmacol 60: 35-43


carbocyclic thromboxane A₂, a stable analog of thromboxane A₂.

Proc Natl Acad Sci USA 77: 1706–1710


INDEX TERMS: Cerebral artery • Carbocyclic thromboxane A₂ • Vascular action of prostaglandin I₂ • Vascular action of verapamil
Mechanism of action of carbocyclic thromboxane A2 and its interaction with prostaglandin I2 and verapamil in isolated arteries.

N Toda

_Circ Res._ 1982;51:675-682
doi: 10.1161/01.RES.51.6.675

_Circulation Research_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1982 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7330. Online ISSN: 1524-4571

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circres.ahajournals.org/content/51/6/675