CARDIOVASCULAR RESPONSES TO PGI₂ (PROSTACYCLIN) IN THE DOG

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SUMMARY Arachidonic acid (AA) produces characteristic hemodynamic changes in the canine circulation. These responses are blocked by prostaglandin (PG) synthetase inhibitors, indicating that AA and its nonprostanoic metabolites are not vasoactive. The hemodynamic effects of the cyclic endoperoxides, thromboxanes, and PGD₂, PGE₂, and PGF₆ᵅ, differ from those produced by AA. PGI₂, a newly identified product of AA, is reported to relax arterial strips. However, its cardiac and systemic effects are unknown. In 12 open-chest, anesthetized dogs, PGI₂ (0.25–5.0 μg/kg) produced a dose-related decrease in systemic arterial pressure (BP) and myocardial contractile force (MCF). In five left ventricular bypass preparations, PGI₂ produced only a slight decrease in MCF at all doses, whereas the BP decreases were parallel to those in the intact preparation. AA, PGD₂, PGE₂, and PGI₂ were administered in random order by bolus intravenous injections in approximately equidepressor doses to intact dogs. BP fell with each agent (AA, 300 μg/kg, -25%; PGI₂, 5 μg/kg, -26%; PGE₂, 5 μg/kg, -26%; PGI₂, 0.5 μg/kg, -26%). The vasodepressor action of PGI₂ was approximately 10 times greater than that of PGD₂ and PGE₂. Pulmonary arterial pressure (PAP) rose significantly with PGD₂ and PGE₂ (AA, -1%; PGI₂, +66%; PGE₂, +20%; PGI₂, -1%). Only PGE₂ had a significant effect on MCF (AA, +7%; PGI₂, +5%; PGI₂, +20%; PGI₂, -0.3%). At this dose, PGI₂ resembles AA in that it has little effect on either PAP or MCF. Of all known AA metabolites the response to PGI₂ most closely resembles that of exogenous AA in the dog.

ARACHIDONIC ACID (AA) produces a characteristic hypotensive response in several species. The ability to block this response with prostaglandin (PG) synthetase inhibitors indicates that one or more of the vasoactive products of AA metabolism are responsible for this systemic hypotensive response, rather than AA itself. Studies of cardiac and hemodynamic responses to exogenous PGE₂, PGF₂α, PGI₂, and PGD₂ indicate that still other AA metabolites probably are involved in these responses. For example, prostacyclin (PGI₂, formerly known as PGX), a recently isolated product of AA metabolism, has been reported to relax arterial muscle strips and thus may be a depressor product of AA metabolism. Biosynthesis of PGI₂ does not produce enough PGI₂ for in vivo studies. However, a convenient synthesis has been achieved.

The objectives of this study were to establish the acute systemic effects of synthetic PGI₂ administered intravenously into the intact dog, to quantify the hemodynamic response to a range of administered doses and to compare the PGI₂ response to that produced by AA.

METHODS

Seventeen mongrel dogs of either sex weighing 14–20 kg were anesthetized with sodium pentobarbital, 30 mg/kg, iv, intubated with a cuffed endotracheal tube, and ventilated with ambient air by a positive pressure respirator. Airway pressure (AP), which is an index of airway resistance, was measured via a needle inserted into the endotracheal tube and connected to a strain gauge. The right femoral artery and vein were catheterized to record systemic arterial pressure (BP) and for intravenous administration of test substances, respectively. A thoracotomy was performed at the 4th left intercostal space. An indwelling catheter was inserted into a branch of the left pulmonary artery and attached to a strain gauge to measure PAF. In the intact dogs, a Walton-Brodie strain gauge arch was sutured to the right ventricular wall for measurement of MCF.

In the left ventricular bypass preparation, all pulmonary venous return was diverted into a reservoir from which it was pumped at a controlled flow rate into a T-tube inserted into the descending thoracic aorta. The strain gauge arch was sutured to the left ventricular wall in this preparation. Additional details regarding the left ventricular bypass have been described previously. Before the bypass was initiated, heparin (8 mg/kg) was administered intravenously.

The sodium salt of arachidonic acid (5,8,11,14-eicosatetraenoic acid, >99% pure from porcine liver; Sigma and NuChek) was prepared by dissolving in sodium carbonate (100 mM) during constant stirring under nitrogen, in the absence of light. PGE₂ and PGD₂ were generously provided by the Upjohn Co. Solutions of each prostaglandin in ethanol (1 mg/ml) were evaporated to dryness under nitrogen. The residues were dissolved in normal saline to a final concentration of 100 μg/ml. PGI₂, 9-deoxy-6,9α-epoxy-Δ²-PGF₆ᵅ, was synthesized as previously indicated. Samples were prepared in Tris buffer (pH 9) and then diluted to 100 μg/ml before use; 15-
hydroperoxy arachidonic acid was prepared by the method of Hamberg and Samuelsson.\textsuperscript{9} It was obtained as an oil which was suspended in phosphate buffer to 30 mg/ml with a sonicator. Hexamethonium (2 mg/kg), a ganglionic blocking agent, was administered to one dog.

Prostaglandins were administered in random order by intravenous bolus injection (0.35-1.0 ml). Before administration of each test substance, AP, BP, MCF, and PAP were allowed to return to control values.

**Results**

In 12 dogs with an intact circulation, the typical cardiovascular responses to PGI\textsubscript{2} (2 \(\mu\)g/kg) were characterized by a significant drop in both systolic and diastolic BP and by little or no change in either AP, PAP, or MCF (Fig. 1).

Varied doses of PGI\textsubscript{2}, (0.25-5.0 \(\mu\)g/kg) were administered. The changes in the PAP responses were variable; mean percentage change remained close to the control values at all doses employed. Mean variations in heart rate (\(-1.3\) to \(+4.0\)\%) were negligible during the maximal vasodepressor responses at all dose levels of PGI\textsubscript{2}. As shown in Figure 2, there was a dose-dependent decrease in both MCF and BP. MCF was slightly elevated at the lower doses and significantly decreased at the upper doses. Administration of Tris buffer vehicle in the same volumes produced no change in any of the recorded parameters.

In five ventricular bypass preparations, PGI\textsubscript{2} (0.5, 2.0 and 5.0 \(\mu\)g/kg) produced a slight but consistent decrease in MCF. In contrast, one dog developed an increased MCF at all dose levels; after ganglionic blockage with hexamethonium, similar reductions in MCF were noted. Although less pronounced, the changes in BP observed in the bypass preparation were parallel to the response in the intact dog (Fig. 2). There were no changes in PAP or AP in this dose range.

The effects of equidepressor doses of AA (300 \(\mu\)g/kg), PGE\textsubscript{2} (5 \(\mu\)g/kg), and PGD\textsubscript{2} (5 \(\mu\)g/kg) were compared to PGI\textsubscript{2} (0.5 \(\mu\)g/kg). The results of this comparison are presented in Table 1. PGD\textsubscript{2} produced the most significant increase in PAP, followed by PGE\textsubscript{2}, whereas AA and PGI induced little change. Only PGE\textsubscript{2} induced a significant increase in MCF. The onset of AA response was more delayed when compared to PGI\textsubscript{2} and PGE\textsubscript{2}. The durations of action of AA and PGI\textsubscript{2} were comparable. However, the duration of action of PGE\textsubscript{2} was shorter and that of PGD\textsubscript{2} was substantially longer than that of AA or PGI\textsubscript{2} (Table 1).

In three dogs, the administration of 15-hydroperoxy AA (300 \(\mu\)g/kg) had no inhibitory effect on the systemic hypotensive response to an immediate administration of AA (300 \(\mu\)g/kg).

**Discussion**

The systemic hypotensive response produced by AA was reported as early as 1959 by Jaques,\textsuperscript{10} yet the mechanisms and site of action have not been identified. Inhibition of the vascular response to AA\textsuperscript{3} by nonsteroidal antiinflammatory drugs\textsuperscript{11} shows that AA itself or the products of the nonprostanoate pathways have little cardiovascular activity. Consequently, attention has been directed to the prostanoic products of AA metabolism to explain this hypotensive response.

Endoperoxide intermediates, PGG\textsubscript{2} and PGH\textsubscript{2}, known to be contractors of vascular smooth muscle strips,\textsuperscript{12-13} produce triphasic alterations in BP when administered intravenously to the guinea pig.\textsuperscript{14} The terminal hypotensive phase has been attributed to a potent vasodepressor metabolite of these endoperoxides.\textsuperscript{15} Thromboxane A\textsubscript{2}, a major component of rabbit aorta-contracting substance and a product of endoperoxide metabolism, contracts all isolated blood vessel preparations.\textsuperscript{16} Similarly, PGF\textsubscript{2\alpha} administered intravenously produces a hypertensive response.\textsuperscript{3,17,18} The known prostanoic metabolites of AA that consistently produce a systemic hypotensive response in the dog are PGD\textsubscript{2} and PGE\textsubscript{2}.\textsuperscript{3} However, a comparison...
of the cardiovascular responses to equidepressor doses of AA, PGD₂, and PGE₂ has demonstrated significant differences in their modes of action. PGE₂, unlike AA, consistently produced a direct increase in MCF. Moreover, PGD₂ and PGE₂ increased PAP and AA produced only a variable pulmonary response.

PGE₁ is a newly discovered substance formed by the endothelial microsomal enzymes from the endoperoxide intermediate(s) that induces relaxation of arterial muscle strips and inhibition of platelet aggregation. Moreover, 6-keto PGF₁α is the primary product of PGI₂ metabolism and has been identified as the principal metabolite when coronary arteries are incubated with AA.

In the study described here, the vasodepressor action of PGI₂ was approximately 10 times greater than that of PGD₂ and PGE₂. The time to onset of action of PGI₂ was similar to that of PGE₂, and probably represents the transit time from point of injection to the site of action in the systemic arterioles, whereas the delayed onset of action of AA suggests a delay for the enzymatic conversion to a vasoactive product(s).

PGI₂ resembles AA in that it does not increase PAP, as does PGD₂ and PGE₂. Furthermore, at lower doses of PGI₂, there was no apparent effect on MCF, unlike the positive inotropic effect of PGE₂.

The decreased MCF observed at higher doses of PGI₂ could be related to alterations in preload or afterload, or to a negative inotropic effect. In the left ventricular bypass preparation, PGI₂ produced only a slight decrease in MCF that was not dose dependent. Therefore, MCF changes in the intact dog must be due at least in part to variations in preload or afterload.

Inhibitors of PGI₂ synthetase would be useful to clarify this point further: 15-hydroperoxy AA, administered in a bypass preparation, PGI₂ produced only a slight decrease in PAP, whereas the delayed onset of action of PGI₂ was similar to that of PGE₂, and probably represents the enzymatic conversion to a vasoactive product(s).

Hemodynamic Changes Produced by Equidepressor Doses of PGI₂, AA, PGE₂, and PGD₂ in Intact Dogs

<table>
<thead>
<tr>
<th>Dose</th>
<th>μg/kg</th>
<th>n</th>
<th>Time to onset (sec)</th>
<th>BP (%)</th>
<th>PAP (%)</th>
<th>MCF (%)</th>
<th>Duration of action (sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AA</td>
<td>300.0</td>
<td>13</td>
<td>15.0 ± 0.8</td>
<td>-25.0 ± 3.0</td>
<td>-0.7 ± 2.6</td>
<td>+7.5 ± 2.4</td>
<td>174.8 ± 18.3</td>
</tr>
<tr>
<td>PGE₂</td>
<td>5.0</td>
<td>5</td>
<td>7.3 ± 0.7</td>
<td>-23.4 ± 2.2</td>
<td>+20.1 ± 2.9</td>
<td>+20.1 ± 6.0</td>
<td>138.4 ± 26.9</td>
</tr>
<tr>
<td>PGD₂</td>
<td>5.0</td>
<td>6</td>
<td>12.3 ± 1.1</td>
<td>-25.6 ± 5.4</td>
<td>+66.4 ± 10.4</td>
<td>+4.6 ± 5.4</td>
<td>239.6 ± 44.8</td>
</tr>
<tr>
<td>PGI₁</td>
<td>0.5</td>
<td>11</td>
<td>8.6 ± 0.7</td>
<td>-26.2 ± 3.4</td>
<td>-1.4 ± 2.9</td>
<td>-0.3 ± 2.9</td>
<td>187.5 ± 17.7</td>
</tr>
</tbody>
</table>

Values in columns 4-8 are expressed as mean ± SE. n = number of responses; BP = systemic arterial pressure; PAP = pulmonary arterial pressure; MCF = myocardial contractile force.

References

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