Summary

Anesthetized dogs were studied to characterize the renal hemodynamic response to acute, transient hypoxemia and to determine the role of renal prostaglandins (PGE and PGF) in that response. Acute hypoxemia of 10 minutes duration (mean arterial PO₂ 32 ± 9 torr) induced a reversible increase in renal blood flow (RBF) (measured by electromagnetic flow probe) and mean arterial pressure (MAP) with a decreased urinary flow rate (V) and no change in glomerular filtration rate (GFR) or renal vascular resistance (RVR). Urinary excretion of PGE (UPGE) and PGF (UPGF) (measured by radioimmunoassay) was not significantly changed by hypoxemia under these conditions. After inhibition of the prostaglandin system by administration of indomethacin (5 mg/kg) or meclofenamate (5 mg/kg) intravenously, a significant decrease in RBF and GFR with an increase in RVR occurred in response to hypoxemia. Furthermore, there was a significantly greater decline in UPGE and UPGF in response to hypoxemia after prostaglandin inhibition than before treatment. Control (non-drug treated) animals demonstrated no difference in response to a second episode of hypoxemia relative to the first hypoxicemic response. The significant changes in UPGE (ΔUPGE), UPGF (ΔUPGF), and GFR with hypoxemia after treatment were not secondary exclusively to a decline in RBF, as shown by the similar response of ΔUPGE and GFR to hypoxemia in dogs in which RBF was enhanced with hypoxemia after treatment by means of an adjustable aortic clamp. Inhibition of prostaglandin synthesis, therefore, unfavorably altered the ability to maintain normal renal perfusion and function during acute hypoxemia in these animals. This renal response to hypoxemia may be mediated by decreased renal prostaglandin production under these conditions.


Experimental Procedures

Adult mongrel dogs of either sex weighing 10-19 kg were studied. The animals were fed standard kennel ration and were fasted for 18-24 hours prior to surgery. After a 24-hour fast, the dogs were anesthetized with pentobarbital, intubated, and ventilated with a pressure-controlled respirator. Arterial blood gases were monitored continuously and the inspired oxygen fraction was adjusted to maintain arterial oxygen saturation at 95-100%. Blood pressure was monitored with a tail cuff and aortic blood flow was measured by electromagnetic flow probe. Renal blood flow and mean arterial pressure were recorded continuously on a polygraph. Urinary flow rate and urine volume were recorded every 10 minutes. Urinary excretion of PGE (UPGE) and PGF (UPGF) was measured by radioimmunoassay. Prostaglandin synthesis was inhibited by intravenous administration of indomethacin (5 mg/kg) or meclofenamate (5 mg/kg).
to study. Anesthesia was induced with intravenous sodium pentobarbital, 30 mg/kg body weight, and anesthesia was maintained by intermittent intravenous administration of an additional 30 mg of pentobarbital as needed. The trachea was intubated and the dogs were ventilated artificially with a Harvard respirator after neuromuscular blockade with gallamine (4 mg/kg body weight, intravenously). During normoxemia, the inhaled gas mixture was room air. During hypoxemia a 1:1 mixture of air and nitrogen with a resultant fraction of inspired oxygen \((f_O_2)\) of approximately 0.10 was administered. This low oxygen gas mixture was prepared by directing the contents of cylinders containing nitrogen and air, respectively, through separate gas flow meters into a 3-liter Douglas bag where they were mixed prior to entering the respirator. Cannulation of the right femoral vein was performed for infusion of a 0.15 M NaCl solution containing either a 5 mM concentration of inulin or 1-2 µCi of \(^{125}\)I-sodium iothalamate. This solution was infused intravenously at a rate of 0.25 ml/kg body weight per minute. The right femoral artery was cannulated to obtain samples of arterial blood for analysis. Between blood samples, the arterial cannula was used for continuous monitoring of mean arterial blood pressure (MAP) with a pressure transducer (Statham P23AA).

Through a left flank incision, the left ureter was cannulated and the catheter tip was advanced to the renal pelvis for unilateral urine collection. A non-cannulating flow probe (Carolina Medical Electronics) was placed on the left renal artery after careful blunt dissection to isolate the vessel with minimal disturbance of nerve supply. The flow probe was connected to a square wave electromagnetic flow meter (Carolina Medical Electronics) and renal blood flow (RBF) along with MAP was recorded continuously on a Beckman type R dynograph. The dogs were allowed to stabilize for at least 60 minutes after completion of the surgical procedures. Six periods (10 minutes each) were monitored and urine collected quantitatively in four groups of animals.

**Dogs Treated with Indomethacin (Group I)**

Ten animals were studied throughout two experimental sequences, each consisting of prehypoxic, hypoxic and post-hypoxic periods in succession. In the first period (pre-hypoxemia #1), baseline values during 10 minutes of ventilation with room air were obtained. A low oxygen gas mixture with an \(f_O_2\) of approximately 0.10 then was administered. Over the next 2–3 minutes, serial arterial blood \(P_O_2\) (\(P_{AO_2}\)) measurements were performed. When \(P_{AO_2}\) reached approximately 33 torr (an average of 3 minutes after initiation of ventilation with the low oxygen gas mixture), a 10-minute urine collection period (hypoxemia #1) was begun. In the third 10-minute period (post-hypoxemia #1), recovery of the dogs was monitored and urine collected during ventilation with room air. Indomethacin (5 mg/kg body weight) then was administered intravenously during an interval of 3–4 minutes. Thirty minutes after the administration of indomethacin, the sequence of 10-minute pre-hypoxic, hypoxic and post-hypoxic periods was repeated. Near the end of each period, arterial blood was sampled for analysis of hematocrit (Hct), pH (\(p_H_2\)), and gas pressures (\(P_{AO_2}\), \(P_{ACO_2}\)). In addition, arterial blood was sampled for analysis of inulin or \(^{125}\)I-iothalamate content in five animals. Aliquots from quantitative urine collections also were analyzed for inulin or \(^{125}\)I-iothalamate content in these five animals. Prostaglandin E (PGE) and prostaglandin F (PGF) content in urine was measured in five animals.

**Dogs Treated with Meclofenamic Acid (Group II)**

Seven animals were monitored during two sequences of pre-hypoxic, hypoxic and post-hypoxic periods in a fashion similar to that described for the indomethacin-treated group. Following the initial sequence, meclofenamic acid (5 mg/kg body weight) was substituted for indomethacin and was administered intravenously during an interval of 3–4 minutes. Thirty minutes after the meclofenamic acid administration, a second sequence of pre-hypoxic, hypoxic and post-hypoxic periods was monitored. Near the end of each period arterial blood was sampled for analysis of Hct, pH, gas pressures and inulin or \(^{125}\)I-iothalamate content. Aliquots of quantitative urine collections were analyzed for inulin or \(^{125}\)I-iothalamate in all animals plus PGE and PGF content in five animals.

**Control (Non-Drug-treated) Group (Group III).**

Seven animals were studied as in groups I and II except for the substitution of intravenously administered carrier solution alone for meclofenamic acid or indomethacin. Arterial blood was sampled for analysis of \(p_H_2\), gas pressures, and inulin or \(^{125}\)I-iothalamate concentrations in all animals. Aliquots of quantitative urine collections were analyzed for inulin or \(^{125}\)I-iothalamate in all animals plus PGE and PGF content in two animals.

**Dogs Treated with Aortic Constriction Distal to the Renal Artery and Meclofenamic Acid (Group IV)**

Five animals were studied as in the other groups except for the addition of a maneuver to prevent the decline in RBF during the post-treatment hypoxic period. These experiments were performed to determine whether RBF was a determinant of the difference in \(\Delta U_{PGE}\) response to hypoxemia after cyclooxygenase inhibition. In these dogs, an adjustable clamp was placed on the aorta below the left renal blood flow (RBF) along with MAP was recorded continuously on a Beckman type R dynograph. The dogs were allowed to stabilize for at least 60 minutes after completion of the surgical procedures. Six periods (10 minutes each) were monitored and urine collected quantitatively in four groups of animals.
renal artery at the time of surgical preparation. The sequences of urine collection periods were performed as in previous groups except that the adjustable clamp was tightened during the post-treatment hypoxic period to cause an increased proximal aortic pressure and prevent the fall in RBF. Aortic constriction was terminated at the onset of the recovery period by releasing the adjustable clamp.

**Analytical Procedures**

Glomerular filtration rate (GFR) was determined by the clearance of inulin (seven animals) or sodium iothalamate (17 animals). Inulin was determined by the method of Schreiner (1950), and sodium iothalamate by radioactivity counting in a Beckman γ spectrometer. Arterial blood pH, PO₂, and PCO₂ were determined with an Instrumentation Laboratory digital pH/blood gas analyzer/acid base calculator. Microhematocrit (Hct) was determined by standard methods. PGE and PGF content of urine was determined by radioimmunoassay (Van Orden and Farley, 1973; Van Orden et al., 1977). Briefly, urine specimens were collected in chilled tubes and placed on ice until the end of each experiment and then stored at −20° until extraction. Sequential extractions in petroleum ether and ethyl acetate were followed by separation of prostaglandins by silicic acid chromatography. PGE and PGF column extractions were assayed using specific PGE and PGF antisera, respectively. PGE and PGF contents were determined from standard curves. Urinary prostaglandin excretion rate (U_{PGE} and U_{PGF}) was calculated as the product of urinary flow rate and urinary prostaglandin concentration. Renal vascular resistance (RVR) was calculated as the perfusion pressure (PP) across the kidney divided by the renal blood flow (RBF). Perfusion pressure was determined as the difference between the mean arterial pressure (MAP), estimated to be equal to renal arterial pressure, and renal venous pressure, measured directly by a pressure transducer.

**Table 1**

<table>
<thead>
<tr>
<th>Group Treated with Prostaglandin Inhibitors*</th>
<th>Pre-treatment</th>
<th>Post-treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Pre-hypoxemia</td>
<td>Hypoxemia</td>
</tr>
<tr>
<td><strong>PaO₂ (torr)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>88 ± 8</td>
<td>32 ± 9†</td>
</tr>
<tr>
<td><strong>PaCO₂ (torr)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>34 ± 5</td>
<td>31 ± 6†</td>
</tr>
<tr>
<td><strong>pH</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>7.36 ± 0.04</td>
<td>7.39 ± 0.05†</td>
</tr>
<tr>
<td><strong>RBF (ml/min per g)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>3.34 ± 0.58</td>
<td>3.62 ± 1.18</td>
</tr>
<tr>
<td><strong>GFR (ml/min per g)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>0.86 ± 0.22</td>
<td>0.68 ± 0.20</td>
</tr>
<tr>
<td><strong>MAP (mm Hg)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>134 ± 12</td>
<td>155 ± 12†</td>
</tr>
<tr>
<td><strong>RVR (mm Hg/min per ml)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>1.213 ± 0.408</td>
<td>1.269 ± 0.447</td>
</tr>
<tr>
<td><strong>V (ml/min)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>0.97 ± 0.60</td>
<td>0.65 ± 0.37†</td>
</tr>
</tbody>
</table>

PaO₂ = arterial blood PO₂, PaCO₂ = arterial blood PCO₂, pH = arterial blood pH; RBF = renal blood flow; GFR = glomerular filtration rate; MAP = mean arterial pressure; RVR = renal vascular resistance; V = urinary flow rate.

* Indomethacin (5 mg/kg body weight, n = 10) or meclofenamic acid (5 mg/kg body weight, n = 7) administered intravenously after pre-treatment post-hypoxemia period. Values are mean ± sd.
† Paired comparisons of pre-hypoxemia value to hypoxemia value reveal a significant difference, P < 0.05.
‡ Paired comparisons of pre-treatement pre-hypoxemia value to post-treatment pre-hypoxemia value reveal a significant difference, P < 0.05.
§ Paired comparisons of post-treatment pre-hypoxemia to post-treatment post-hypoxemia value reveal a significant difference, P < 0.05.

**Statistical Analysis**

The data were analyzed by analysis of variance and Duncan's range test (Steel and Torrie, 1960). A P value less than 0.05 was required for a difference to be declared significant.

**Results**

Comparisons of data from group I and group II revealed no significant differences; thus, these data were combined. Administration of an oxygen-poor inhaled gas mixture induced a significant fall of PaO₂ (Table 1). Arterial pH (pH) values increased significantly above baseline values in response to hypoxemia (Table 1), but the mean difference ± sd was only 0.03 ± 0.04 unit before drug treatment and 0.04 ± 0.04 unit after drug treatment. Arterial PCO₂ values also decreased slightly but significantly in response to hypoxemia. In post-hypoxic periods, PaO₂ was not significantly different from baseline values, but PaO₂ was slightly but significantly lower. Post-hypoxemia pH in the pre-treatment sequence was slightly but significantly lower than baseline values. There were no significant differences in PaO₂ or PaCO₂ between periods before and after treatment with prostaglandin inhibitor. The pre-hypoxic value for pH was slightly but significantly lower after treatment than before treatment.

Acute systemic hypoxemia induced an increase in RBF and MAP in dogs prior to treatment with prostaglandin inhibitor (Table 1). Urinary flow rate decreased under these conditions. There was no significant change in RVR in response to hypoxemia in the pre-treatment sequences. Although GFR did not change significantly in response to hypoxemia in the pre-treatment sequences, the mean difference ± sd was 0.18 ± 0.30 ml/min per g. This difference can be accounted for almost entirely by declines in GFR during pretreatment hypoxemia in three animals. In 6 of 12 animals, GFR either increased slightly or remained unchanged. After administration of indomethacin or meclofenamic acid, however, acute hypoxemia was associated with a sig-
significant fall in RBF and GFR. Furthermore, hypoxemia induced a significant increase in calculated RVR in these animals after drug treatment. MAP also increased in response to hypoxemia, and urinary flow rate decreased, as in the pre-treatment sequence. During the post-hypoxic period after drug treatment, these values returned to their respective pre-hypoxic values except for urinary flow rate, which remained significantly depressed. No significant changes were noted in values for RBF, GFR, MAP, V, or RVR in the pre-hypoxic period after treatment with prostaglandin inhibitor in relation to their respective pre-treatment baseline values (Table 1).

The difference in renal response to hypoxemia after treatment with prostaglandin inhibitor was studied further to determine whether the response to a second exposure to hypoxemia differed in a non-specific fashion from the response to an initial exposure to hypoxemia. The results of arterial blood pH and gas analysis in these control (non-drug treated) animals (group III) are depicted in Table 2. Ventilation of these animals with a low oxygen gas mixture resulted in significant lowering of PaO₂ values below pre-hypoxic values. PaO₂ values returned to pre-hypoxic values during the post-hypoxic period. PaCO₂ values did not change significantly in response to hypoxemia although five of seven animals responded to hypoxemia with a decline in PCO₂. The lack of statistical significance of this difference apparently was due to large variance. An increase in pH values occurred during hypoxemia in sequence #1 in a fashion similar to that described for the treated group. A second sequence of pre-hypoxic, hypoxic, and post-hypoxic periods resulted in respective pHₐ, PaO₂, and PaCO₂ values that were not significantly different from the first sequence. The values for pHₐ, PaCO₂, and PaO₂ in these control animals also did not vary significantly from their values in the treated animal groups.

In these control animals, values for RBF and MAP increased significantly in response to hypoxemia in sequence #1 (Table 2), whereas GFR and RVR remained unchanged. Urinary flow rate decreased in response to hypoxemia in five of seven animals, but the change did not reach statistical significance due to large variance. The second sequence of pre-hypoxic, hypoxic, and post-hypoxic periods in these animals resulted in values not significantly different from the values of the first sequence. Therefore, measured differences in response to hypoxemia between sequences in the experimental animals treated with prostaglandin inhibitor were the result of drug treatment rather than a non-specific effect of the first hypoxic sequence on the second.

Treatment of dogs with indomethacin or meclofenamic acid was associated with a significant decrease in urinary prostaglandin excretion (Fig. 1a). Total measured urinary prostaglandin excretion (PGE and PGF) decreased by 71.7 ± 15.5% (mean ± sd) after administration of prostaglandin inhibitor. No significant difference in total measured urinary prostaglandin excretion was noted in comparisons of consecutive sequences in the few control animals in which these were measured (Fig. 1b).

Inhibition of prostaglandin synthesis also significantly altered the response of UPGF and UPGF to hypoxemia. Hypoxemia induced no significant change in UPGF (from 0.643 ± 0.224 to 0.688 ± 0.417 ng/min, mean ± sd, P > 0.1) or UPGF (from 1.56 ± 2.25 to 1.91 ± 2.20 ng/min, mean ± sd, P > 0.1) in pre-treatment sequences of the drug-treated group. However, after treatment, hypoxemia was associated with a greater decrease in urinary excretion of PGE (ΔUPGF) and PGF (ΔUPGF) relative to pre-treatment values (Fig. 2a). Comparisons of ΔUPGF and ΔUPGF with hypoxemia in control animals, depicted in Figure 2b, revealed no significant difference in comparisons of sequence #1 to sequence #2. Furthermore, comparisons of ΔUPGF during hypoxemia after treatment to ΔUPGF during hypoxemia in sequence #2 in the control group revealed a significant difference (−0.300 ± 0.164 to −0.017 ± 0.099 ng/min, mean ± sd, P < 0.05). Similar comparisons of ΔUPGF with hypoxemia revealed no significant difference. Treatment with prostaglandin inhibitor, therefore, apparently induced the difference in ΔUPGF response to hypoxemia in the treated animals.

To determine whether the difference in ΔUPGF and GFR response to hypoxemia with prostaglandin inhibition was exclusively a secondary effect of the

### Table 2: Control (non-drug-treated) Group*

<table>
<thead>
<tr>
<th></th>
<th>Pre-hypoxemia</th>
<th>Hypoxemia</th>
<th>Post-hypoxemia</th>
<th>Pre-hypoxemia</th>
<th>Hypoxemia</th>
<th>Post-hypoxemia</th>
</tr>
</thead>
<tbody>
<tr>
<td>PaO₂ (torr)</td>
<td>93 ± 11</td>
<td>35 ± 14†</td>
<td>90 ± 8</td>
<td>89 ± 9</td>
<td>35 ± 13</td>
<td>89 ± 25</td>
</tr>
<tr>
<td>PaCO₂ (torr)</td>
<td>35 ± 8</td>
<td>34 ± 7</td>
<td>35 ± 8</td>
<td>34 ± 9</td>
<td>31 ± 7</td>
<td>34 ± 10</td>
</tr>
<tr>
<td>pH</td>
<td>7.38 ± 0.05</td>
<td>7.41 ± 0.06†</td>
<td>7.38 ± 0.07</td>
<td>7.40 ± 0.06</td>
<td>7.36 ± 0.07</td>
<td></td>
</tr>
<tr>
<td>RBF (ml/min per g)</td>
<td>3.78 ± 0.97</td>
<td>4.25 ± 0.96†</td>
<td>3.54 ± 0.89</td>
<td>3.83 ± 0.74</td>
<td>3.54 ± 0.74</td>
<td>3.38 ± 0.84</td>
</tr>
<tr>
<td>GFR (ml/min per g)</td>
<td>0.61 ± 0.12</td>
<td>0.59 ± 0.15</td>
<td>0.65 ± 0.14</td>
<td>0.65 ± 0.11</td>
<td>0.62 ± 0.16</td>
<td>0.70 ± 0.14</td>
</tr>
<tr>
<td>MAP (mm Hg)</td>
<td>121 ± 16</td>
<td>138 ± 17†</td>
<td>115 ± 17</td>
<td>113 ± 17</td>
<td>129 ± 13†</td>
<td>116 ± 17</td>
</tr>
<tr>
<td>RVR (mm Hg/min per ml)</td>
<td>7.06 ± 0.35</td>
<td>7.11 ± 0.13</td>
<td>7.15 ± 0.071</td>
<td>7.02 ± 0.126</td>
<td>7.05 ± 0.115</td>
<td>7.03 ± 0.106</td>
</tr>
<tr>
<td>V (ml/min)</td>
<td>1.02 ± 0.60</td>
<td>0.78 ± 0.47</td>
<td>1.07 ± 0.73</td>
<td>1.08 ± 1.01</td>
<td>1.26 ± 1.13</td>
<td>1.34 ± 1.07</td>
</tr>
</tbody>
</table>

* Animals given drug vehicle only. Values are mean ± sd.
† Paired comparisons of pre-hypoxemia value to hypoxemia value reveal a significant difference, P < 0.05.
Treatment Group | Control Group
---|---
3.0 ± 2.5 | 2.0 ± 1.5
1.0 ± 0.5 | 0.5 ± 0.2
0.5 ± 0.2 | 0.2 ± 0.1

Pre-Post-Treatment Sequence

**FIGURE 1** Urinary prostaglandin excretion rate (Upg). Each point is mean of n determinations of average urinary PGE or PGF excretion during the three urine-collection periods of each sequence. Points connected by solid (UpgE) or dashed (UpgF) lines represent values in the same animal during two separate hypoxic sequences (see Methods). Vertical line from each point represents standard deviation. The number of dogs (n) in each group is indicated in the right upper corner of each box. Treatment group consists of dogs given indomethacin or meclofenamic acid (groups I and II). Control group consists of dogs given vehicle only (group III). *Paired comparisons of pre-treatment values to post-treatment values reveal a significant difference, $P < 0.05$. NS = difference not significant.

difference in RBF response under these circumstances, another group of dogs (group IV) was studied. In these animals, RBF was enhanced during hypoxemia after prostaglandin inhibition by means of an adjustable aortic clamp below the renal artery. Administration of an oxygen-poor inhaled gas mixture to this group of dogs induced a significant fall in Pao2 (Table 3) which was not significantly different from that seen in the other groups of animals. Values for pHa increased in two of five dogs, and in three dogs pHa remained unchanged in pre-treatment hypoxic periods. In post-treatment hypoxic periods, pHa increased in four of five animals and decreased in one. Hypoxic pHa values, however, were significantly different from their respective pre-hypoxemia values only after prostaglandin inhibition. Pao2 values likewise decreased during post-treatment hypoxic periods in three of five

**TABLE 3** Group Treated with Prostaglandin Inhibitor* Plus Aortic Clamp

<table>
<thead>
<tr>
<th></th>
<th>Pre-treatment</th>
<th>Post-treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Pre-hypoxemia</td>
<td>Hypoxemia</td>
</tr>
<tr>
<td>Pao2 (torr)</td>
<td>85 ± 8</td>
<td>35 ± 9†</td>
</tr>
<tr>
<td>PacO2 (torr)</td>
<td>31 ± 3</td>
<td>30 ± 4</td>
</tr>
<tr>
<td>pHa</td>
<td>7.37 ± 0.03</td>
<td>7.38 ± 0.04</td>
</tr>
<tr>
<td>RBF (ml/min per g)</td>
<td>3.86 ± 1.06</td>
<td>4.29 ± 1.05†</td>
</tr>
<tr>
<td>GFR (ml/min per g)</td>
<td>0.62 ± 0.22</td>
<td>0.60 ± 0.38</td>
</tr>
<tr>
<td>MAP (mm Hg)</td>
<td>134 ± 23</td>
<td>156 ± 37†</td>
</tr>
<tr>
<td>RVR (mmHg/min per ml)</td>
<td>1.012 ± 0.221</td>
<td>0.966 ± 0.335</td>
</tr>
<tr>
<td>V (ml/min)</td>
<td>1.91 ± 0.80</td>
<td>1.55 ± 0.96</td>
</tr>
</tbody>
</table>

* Meclofenamic acid (5 mg/kg body weight) administered intravenously after pre-treatment post-hypoxemia period. Values are mean ± sd.
† Paired comparisons of pre-hypoxemia value to hypoxemia value reveal a significant difference, $P < 0.05$. NS = difference not significant.

**FIGURE 2** Change in urinary prostaglandin excretion rate (ΔUpg) with hypoxemia. Each point is mean of n determinations, indicated at upper corner of each box, of ΔUpgE or ΔUpgF. Vertical line from each point represents standard deviation. Points connected by solid (ΔUpgE) or dashed (ΔUpgF) lines represent values in the same animal during two sequences of hypoxemia. Treatment group consists of dogs given indomethacin or meclofenamic acid (groups I and II). Control group consists of dogs given vehicle only (group III). *Paired comparisons of pre-treatment values to post-treatment values reveal a significant difference, $P < 0.05$. NS = difference not significant.
animals and remained unchanged in two. However \( \text{Paco}_2 \) values during hypoxemia were not significantly different from their respective pre-hypoxic values because of large variance. \( \text{Paco}_2 \) and \( \text{pH}_a \) values in this group of animals were not significantly different from values during respective periods in the other groups of treated animals.

During pre-treatment sequences in these animals, values for RBF, GFR, MAP, V, and RVR were not significantly different from values during respective periods of the pre-treatment sequence in the other groups of treated and control animals (Table 3). During post-treatment sequences in these group IV animals, however, RBF during hypoxemia was maintained at the pre-hypoxic level by increasing renal perfusion pressure with an aortic clamp. RBF increased in response to hypoxemia in three of five dogs and declined in two dogs under these circumstances. RBF was not significantly different in comparisons of post-treatment hypoxic to pre-treatment hypoxic values. RVR increased during hypoxemia in four of five animals, but the mean value was not significantly different from the pre-hypoxic value due to large variance. Despite the maintenance of RBF during the post-treatment hypoxic period, GFR fell significantly.

Urinary prostaglandin data for this group of animals are depicted in Figures 3 and 4. Administration of inhibitors of prostaglandin synthesis significantly decreased average urinary prostaglandin excretion (PGE and PGF) (Fig. 3) as in the group of treated animals. The change in \( \Delta \text{UPGE} \) with hypoxemia pre-treatment (Fig. 4) was also similar to that seen in the treated animal group. However, the \( \Delta \text{UPGE} \) response to post-treatment hypoxemia was not significantly different from the pre-treatment value, apparently due to large variance. \( \Delta \text{UPGE} \), on the other hand, was not significantly different from that seen in the treated group under these conditions. Therefore, maintenance of RBF during post-treatment hypoxemia at the pre-hypoxic value failed to prevent the fall in GFR and was associated with a similar decline in \( \text{UPGE} \) and \( \text{UPGF} \) in comparison to the treated group.

Discussion

In the pre-treatment sequences of the treated animal groups and in control animals of the present study, acute systemic hypoxemia induced a reversible increase in RBF and MAP, a decrease in urinary flow rate, and unchanged GFR and RVR. These results are comparable to those of Anderson
et al. (1978) except that MAP remained unchanged and RVR increased in response to hypoxemia in their study. The difference in response probably was a result of the more prolonged hypoxemia (up to 100 minutes) in the study by Anderson et al. (1978). Another possible influence on the difference in results may be the use of the clearance of p-aminohippurate as an estimate of renal plasma flow (RPF) in their study as opposed to the use of an electromagnetic flow probe for direct measurement of RBF in the present study. Bruns (1978) also studied the response to a prolonged period of hypoxemia (up to 90 minutes) and found a reversible increase in MAP with decreased RPF and GFR. The difference in the response of GFR to hypoxemia in the studies of Anderson et al. (1978) relative to the studies of Bruns (1978) may be influenced by the extracellular fluid volume expansion in the former study. Arterial blood pH and Pco2 were controlled in both of these studies, as in the present study, and so were not a factor in the renal hemodynamic response. Previous studies of the response to systemic hypoxemia were marred by unmeasured or uncontrolled arterial blood pH and Pco2 values (Korner, 1963; Taquini et al., 1969; Wyler, 1975; Adachi et al., 1976).

In the present study, treatment of anesthetized dogs with inhibitors of prostaglandin synthesis (indomethacin or meclofenamic acid) altered the renal hemodynamic response to hypoxemia. Under these conditions, hypoxemia induced a significant fall in RBF with increased RVR. Furthermore, GFR, which is to a large extent dependent upon RBF (Tucker and Blantz, 1977), declined significantly in response to hypoxemia after administration of inhibitor. Since control animals demonstrated no difference in renal hemodynamic response to a second episode of hypoxemia relative to the first hypoxic response, the drug treatment was responsible for the altered response in the treated animals. Furthermore, the lack of change in GFR and RVR during hypoxemia in control animals further supports the lack of significant change in these values in pre-treatment responses to hypoxemia in the treated group. Therefore, treatment with inhibitor unfavorably altered the ability to maintain normal renal perfusion and function during hypoxemia in these animals.

The observed effect of indomethacin and meclofenamic acid on the renal response to hypoxemia in the current study is not likely to be due to a non-specific effect of these drugs. Indomethacin and meclofenamic acid are quite dissimilar structurally (Flower, 1974), and they belong to a large group of chemically diverse substances, the non-steroidal anti-inflammatory agents, which have in common an inhibitor effect on the cyclooxygenase enzyme system responsible for prostaglandin synthesis (Vane, 1978). Both of these drugs also have inhibitory effects on the prostaglandin system at other points in the metabolic pathway. Meclofenamic acid (and other fenamates) also block prostaglandin receptors (Collier and Sweatman, 1968) and indomethacin also inhibits renal tubular transport of prostaglandins, prostaglandin dehydrogenase, 9-ketoreductase, and phosphodiesterase (Dunn and Hood, 1977).

The renal hemodynamic effects of prostaglandin inhibition may depend on the baseline activity of renal vasoconstrictor pathways (Dunn and Hood, 1977). This hypothesis suggests that acutely stressful circumstances, such as acute anesthesia and surgical procedures, increase catecholamine or angiotensin release which then stimulates renal vaso-dilatory prostaglandin release. Under these circumstances, prostaglandin blockade may allow this augmented vasoconstrictor influence to act unopposed (Dunn and Hood, 1977). Furthermore, administration of exogenous vasoconstrictors after prostaglandin inhibition leads to an augmented vasoconstricor response. Similarly, prostaglandin inhibition has resulted in decreased RBF in some anesthetized, acutely operated (or "stressed") animals and unchanged RBF in some conscious (or "unstressed") animals and human subjects (Dunn and Hood, 1977). The conditions of anesthesia and acute surgery under which the experiments were performed in the present study would, by this hypothesis, be expected to be associated with decreased RBF in response to prostaglandin inhibition. RBF did not decrease after prostaglandin inhibition in our experiments, perhaps because of the time interval between the surgical procedures and the actual experiment. However, the additional stress of acute hypoxemia in the present study was associated with decreased RBF and GFR after prostaglandin inhibition. Since previous studies have shown that hypoxemia is associated with increased plasma renin activity and catecholamine concentration (Liang and Gavras, 1978), the results of the present study support the previously stated hypothesis.

Urinary prostaglandin excretion rate has been shown to approximate renal prostaglandin production (Dunn and Hood, 1977). However, recent evidence has suggested that urinary flow rate may be a major determinant of the urinary excretion rate of prostaglandin E in the dog (Kirschenbaum and Serros, 1980). The decreased urinary PGE and PGF excretion rate after administration of meclofenamic acid or indomethacin in the present study occurred in the absence of a decrease in urinary flow rate. Therefore, the decreased urinary excretion of PGE and PGF reflects decreased renal production of these prostaglandins under these conditions. Furthermore, although the urinary flow rate decreased during hypoxemia in these animals, the urinary flow rate was not significantly lower after treatment with prostaglandin inhibitor relative to before treatment. Therefore, the significantly greater decrease in urinary PGE and PGF excretion
during hypoxemia after prostaglandin inhibition suggests decreased prostaglandin production under these circumstances, rather than an effect of urinary flow rate alone. Variation in urinary flow rate, however, cannot be discounted as a possible influence on the urinary prostaglandin excretion rate in these studies.

Endogenous or exogenous PGE is a potent renal vasodilator (Dunn and Hood, 1977). PGF, on the other hand, has not been shown to affect the renal vasculature, and has not been shown conclusively to be a primary product of the renal prostaglandin cyclooxygenase pathway in the dog. Rather, PGF may be a secondary product of PGE metabolism within the kidney (Dunn and Hood, 1977). The greater decrease in urinary PGE excretion rate during hypoxemia after prostaglandin inhibition in the present study, then, suggests that the increased vasoconstriction in response to hypoxemia after prostaglandin inhibition may be due to the withdrawal of vasodilatory effects of prostaglandin E.

In the present study, urinary PGE and PGF excretion rates did not increase in response to hypoxemia. Although this finding is contrary to results of some studies of ischemic or anoxic organ systems (Jaffe, 1972; Herbaczynska-Cedro and Vane, 1973; Wennmalm et al., 1974; Block et al., 1975), these results have theoretical validity in that oxygen is required for prostaglandin synthesis (Samuelsson et al., 1975; Beckman and Zehr, 1975). Therefore, increased urine prostaglandin excretion as a reflection of increased renal prostaglandin synthesis would not be expected under hypoxic conditions. The current study suggests that the combination of prostaglandin inhibitor administration and hypoxemia may accentuate any decline in PGE production which occurs with either one alone.

Recently identified vasodilatory products of the renal prostaglandin cyclooxygenase pathways, prostacyclin (PGI2) and PGD2 (Dunn and Hood, 1977) were not measured in the current experiments. Furthermore, other systemic effects of hypoxemia and prostaglandin inhibition which may influence renal function were not specifically examined in this study. The role of these variables in the renal response to hypoxemia remains to be studied.

In summary, acute systemic hypoxemia of 10 minutes duration induced an increase in RBF and MAP, a decrease in urinary flow rate, and no change in GFR, RVR, and urinary excretion of PGE and PGF in anesthetized dogs. After administration of indomethacin or meclofenamic acid, however, hypoxemia induced a decrease in RBF and GFR and an increase in RVR. Urinary PGE and PGF excretion decreased significantly in response to hypoxemia after prostaglandin inhibition. Control (non-drug-treated) animals demonstrated no such differential response to hypoxemia. Furthermore, the decline in GFR, U_PGE and U_PGF with hypoxemia post-treatment was not affected by maintenance of RBF with an adjustable aortic clamp. Indomethacin and meclofenamic acid treatment, therefore, unfavorably altered the ability to maintain renal perfusion and function during hypoxemia in these animals. This response may be mediated by decreased renal vasodilatory prostaglandin production under these circumstances.

Acknowledgments

The aid and advice of Dr. H.E. Williamson in the initiation of these studies are gratefully acknowledged. Urinary prostaglandin determinations were performed by the the Cardiovascular Core Laboratory, which is supported by Program Project Grant HL-14388. Gratitude is also expressed to James Herrig for technical assistance and Linda Siegel for typing of the manuscript.

References


The Histological Lateral Border of Acute Canine Myocardial Infarction

A Function of Microcirculation

STEPHEN M. FACTOR, ELLEN M. OKUN, AND EDWARD S. KIRK

SUMMARY  Studies from this laboratory have shown that the border of a 24-hour canine infarct is histologically sharp and is composed of numerous interdigitating peninsulas of necrotic and normal tissue. To see if this sharp boundary is spatially related to the capillary beds of occluded and non-occluded arteries, the left anterior descending artery (LAD) was ligated in five mongrel dogs. Twenty-four hours later, white silicone rubber (Microfil) was injected into the LAD distal to the ligature; simultaneously and under the same pressure, red Microfil was injected into the left main coronary artery (LMCA). In hematoxylin and eosin sections from the border of the infarct, capillaries supplied by the LAD (white) were either in areas of necrosis, in normal epicardium or, rarely, in normal tissue along the lateral boundary; those supplied by the LMCA (red) were almost always in normal regions. Quantitative evaluation of this relationship revealed that the majority of the vessels in the normal and necrotic tissue were concordant (i.e., that normal tissue was supplied by the LMCA, and necrotic tissue by the LAD). However, a small zone of vascular discordance, averaging approximately 30 μm in width, was present along the infarct boundary, possibly representing a narrow border zone of little consequence. Hence, the complex interdigitation of normal and necrotic tissue in the lateral border of an infarct is predominantly a function of the interdigitation of the capillary beds supplied by the occluded and nonoccluded arteries. Circ Res 48: 640-649, 1981

THREE-DIMENSIONAL reconstructions of fully developed acute canine myocardial infarctions have revealed an extremely irregular but sharp boundary demarcating necrotic and normal myocardium (Factor et al., 1978). Isolated islands of normal myocardium, thought by some to represent a border zone of ischemic but viable tissue (Edwards, 1957; Braunwald et al., 1974), were shown to be peninsulas of myocardium in continuity with the main mass of normal heart muscle (Factor et al., 1978). Several recent studies, employing different analytical techniques, also have confirmed that when myocardial infarctions are analyzed spatially, the boundary between necrotic and normal tissue is remarkably discrete, with no obvious border zone present (Mars et al., 1975; Barlow and Chance, 1976; Hirzel et al., 1977; Factor et al., 1978; Harken et al., 1978; Janse et al., 1979). This view of a sharply defined lateral border is at variance with the widely held concept of myocardial infarction: a central zone of necrosis surrounded by an intermediate layer of ischemic but viable tissue blending into normal myocardium (Edwards, 1957; Cox et al., 1968), simulating a "bullseye" pattern. Our histological reconstruction of acute 24-hour myocardial infarctions which employed analyses by extensive serial sections demonstrated a remarkably complex interdigitation of necrotic and normal myocardium, but did not identify a physiological mechanism for the discrete boundary between the two tissue types. One possibility is that the coronary vasculature functions as an end artery system, so that the locus of necrotic myocardium is determined by the field of the occluded vessel. Accordingly, we have extended our initial observations to include identification of the vasculature within the zones of normal and necrotic myocardium along the lateral boundary of the infarct. The following report de-
Altered renal hemodynamic and urinary prostaglandin response to acute hypoxemia after inhibition of prostaglandin synthesis in the anesthetized dog.

D N Weismann

doi: 10.1161/01.RES.48.5.632

Circulation Research is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
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Print ISSN: 0009-7330. Online ISSN: 1524-4571

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