Role of the Prostaglandins in Norepinephrine Release during Augmented Renal Sympathetic Nerve Activity in the Dog

JUAN A. OLIVER, ROBERT R. SCIACCA, JOHN PINTO, AND PAUL J. CANNON

SUMMARY To determine the role of the prostaglandins on renal norepinephrine release, the effect of inhibition of prostaglandin synthesis was examined in anesthetized dogs during reflex activation of the renal adrenergic nerves. Hypotension increased the renal vein plasma concentrations of norepinephrine from 380 ± 59 to 608 ± 106 pg/ml (mean ± SEM; P < 0.01) and of PGE$_2$ from 55 ± 7 to 81 ± 14 pg/ml (P < 0.05). Subsequent administration of indomethacin or meclofenamate lowered the renal venous concentration of PGE$_2$ to 26 ± 3 pg/ml (P < 0.01), but had no significant effect on the norepinephrine concentration (620 ± 89 pg/ml). Administration of indomethacin or meclofenamate to dogs with sodium depletion lowered renal venin plasma concentration of PGE$_2$ from 108 ± 40 to 20 ± 3 pg/ml (0.05 < P < 0.1) but had no effect on the renal venous norepinephrine concentration (475 ± 50 vs. 397 ± 46 pg/ml). During the three experimental conditions examined, renal blood flow was lowered by inhibition of prostaglandin synthesis. These results in the dog suggest that the attenuating effect that prostaglandins exert on the renal vascular action of the adrenergic nerves is not due to inhibition of norepinephrine release.

Recent evidence indicates that the prostaglandins attenuate the vasoconstrictor action of the adrenergic nervous system. The vasoconstriction produced by stimulation of adrenergic nerves in a variety of organs, including the spleen (Ferreira and Moncada, 1971) and the kidney (Malik and McGiff, 1975; Henrich et al., 1978), is enhanced following inhibition of prostaglandin synthesis. In a series of experiments carried out with isolated organs, Hedqvist and his coworkers showed that the overflow of norepinephrine evoked by direct electrical nerve stimulation into the splenic, coronary, and renal venous effluents was decreased by infusion of exogenous PGE$_2$ (Hedqvist, 1969; Hedqvist et al., 1970; Frame and Hedqvist, 1975) and increased by pharmacological inhibition of prostaglandin synthesis (Hedqvist et al., 1971; Samuelsson and Wennmalm, 1971; Chanh et al., 1972; Frame and Hedqvist, 1975). Since other studies had indicated that electrical nerve stimulation released prostaglandin-like substances into the splenic and renal venous effluents (Ferreira and Vane, 1967; Dunham and Zimmerman, 1970), Hedqvist (1977) postulated a negative feedback loop in which norepinephrine released from axon terminals enhances the synthesis of prostaglandins which, by inhibiting norepinephrine release, blunt the vascular effects of nerve stimulation.

Uncertainties about this hypothesis still remain. Other investigators have found that inhibition of prostaglandin synthesis failed to potentiate the vasoconstriction caused by direct electrical nerve stimulation in the spleen (Dubocovich and Langer, 1975) and the kidney (Needelman et al., 1974). More importantly, the venous overflow of norepinephrine evoked by direct electrical nerve stimulation was found to be unaffected by the inhibition of prostaglandin synthesis in the spleen of the cat (Dubocovich and Langer, 1975).

No ready explanation is available for these conflicting results. In some of these studies, methods were used that were not sufficiently sensitive to distinguish between norepinephrine and its metabolites (Hedqvist et al., 1970; Chanh et al., 1972). In others, direct electrical stimulation of nerves was used; it is not clear that the response of the axon terminals to direct electrical stimulation is similar to that evoked by action potentials which propagate spontaneously down the sympathetic nerves. Last, most of these studies were performed under in vitro conditions, and the effect of organ isolation and in vitro perfusion on the activity of the sympathetic nervous system or on prostaglandin synthesis is unknown.
The present study was designed to test the hypothesis that the prostaglandins modulate the release of norepinephrine from the renal adrenergic axon terminals. We reasoned that, if prostaglandins exert a negative feedback to inhibit norepinephrine release during stimulation of the adrenergic nerves, subsequent inhibition of prostaglandin synthesis should enhance the overflow of norepinephrine from the kidney. Accordingly, the renal venous overflow of endogenous norepinephrine was measured before and after pharmacological inhibition of prostaglandin synthesis in anesthetized dogs during maneuvers that increase the overflow of norepinephrine from the kidney. The overflow of norepinephrine was estimated by measuring the endogenous norepinephrine concentration in renal venous and arterial plasma with a sensitive radioenzymatic technique while renal blood flow was monitored continuously with an electromagnetic flowmeter.

Methods

Experimental Design

Since prostaglandin synthesis inhibition has been shown to have no effect on the basal norepinephrine overflow (Hedqvist et al., 1971), activation of the adrenergic renal nerves was carried out by three different maneuvers prior to prostaglandin inhibition.

Acute Arterial Hypotension

Fluctuations in systemic arterial blood pressure are followed by opposite changes in the frequency of sympathetic nerve discharges (Koizumi and Suda, 1963) which include the renal nerves (Kirschheim and Gross, 1978). Accordingly, to stimulate the renal sympathetic nerves, acute hypotension was induced in a group of dogs with moderate sodium depletion by inhibition of converting enzyme with teprotide.

Chronic Sodium Depletion

Since, in two previous studies, inhibition of prostaglandin synthesis failed to enhance norepinephrine overflow at high frequencies of electrical nerve stimulation (Stjarne, 1973; Junstad and Wennmalm, 1973), a maneuver was sought in which low level adrenergic stimulation of the kidney was present. Studies from this laboratory have shown that the overflow of norepinephrine into renal venous blood is increased moderately in dogs with chronic sodium depletion (Oliver et al., 1980a). Accordingly, norepinephrine overflow from the kidney was measured before and after prostaglandin synthesis inhibition in dogs with chronic sodium depletion.

Acute Decrease in Cardiac Output

In a third group of dogs fed a normal sodium diet, reflex stimulation of the renal sympathetic nerves was carried out by acutely decreasing the cardiac output by inflating a balloon catheter in the thoracic inferior vena cava.

Experimental Preparation

The experiments were performed in 31 mongrel dogs of either sex weighing an average of 24.3 ± 0.9 kg (mean ± SEM) that were maintained in two different states of sodium balance. (1) Eight animals were fed a normal chow diet which provided a sodium intake of 80–100 mEq/day. (2) Dietary sodium depletion was induced in 23 dogs by the administration of 10 mg of furosemide intramuscularly 9 and 10 days before study, followed by the administration of a diet that provided about 10 mEq of sodium/day. This resulted in a weight loss from 24.7 ± 1.1 to 23.7 ± 1.1 kg (mean ± SEM, P < 0.001); the mean urinary sodium concentration in these animals was 12 ± 3 mEq/liter on the day of study.

Anesthesia was induced with sodium pentobarbital (30 mg/kg, iv) and maintained with periodic additional administration. After cannulation of the trachea, the dogs were ventilated mechanically. Through a femoral artery, a catheter was introduced into the aorta for both blood pressure measurements (P23Db Statham transducer and Grass polygraph recorder) and arterial blood collections. Via the right jugular vein, a catheter was introduced into the right atrium for the administration of indocyanine dye solution. Cardiac output was determined in triplicate as previously described (Oliver and Cannon, 1978). Through a left flank incision and retroperitoneal dissection, the left renal artery was dissected and fitted with a flow probe of appropriate diameter (Carolina Medical Electronics). Renal blood flow was measured with an electromagnetic flowmeter as previously described (Oliver et al., 1979). Total peripheral and renal vascular resistances were calculated as the ratio of mean arterial pressure (mm Hg) to cardiac output (liters/min) or renal blood flow (ml/min), respectively, and expressed in arbitrary resistance units. Through the left ovarian/testicular vein, a small catheter was introduced in retrograde fashion into the left renal vein for blood collection. In all instances, aortic and renal vein blood samples were obtained simultaneously. An interval of at least 1 hour was allowed after surgery for equilibration of the preparation before control determinations were made.

Specific Protocols

Inhibition of Prostaglandins Synthesis during Arterial Hypotension Induced by Teprotide

Twelve sodium depleted dogs were used for this protocol. After equilibration of the preparation, control measurements of cardiac output, arterial blood pressure, and renal blood flow were made and arterial and renal venous blood samples were collected. Each animal then was given a bolus intra-
venous injection of teprotide, 0.4 mg/kg, dissolved in 10 ml of 0.9% saline followed by a continuous infusion of 0.4 μg/kg per min for the remainder of the experiment. After stabilization of the blood pressure and renal blood flow recordings (20-30 minutes later), hemodynamic measurements and blood collections were repeated. To inhibit prostaglandin synthesis, indomethacin, 5 mg/kg (n = 6), prepared as previously described (Oliver and Cannon, 1978), or meclofenamate, 4 mg/kg (n = 6), dissolved in 20 ml of 0.9% saline, was administered as a bolus injection. Final hemodynamic measurements and blood collections were performed 20-30 minutes later. Since the results for all quantities measured were similar with both prostaglandin inhibitors, the data were pooled for statistical evaluation.

Inhibition of Prostaglandin Synthesis during Chronic Sodium Depletion

Eleven dogs with dietary sodium depletion were used in this experiment. After baseline hemodynamic determinations and blood collections, indomethacin, 5 mg/kg (n = 5), or meclofenamate, 4 mg/kg (n = 6), was administered intravenously as a bolus injection. Twenty to 30 minutes later, hemodynamic measurements and blood collections were repeated.

Inhibition of Prostaglandin Synthesis during Acute Reduction of Cardiac Output by Thoracic Cavai Occlusion

Eight dogs fed a normal sodium diet were used in this experiment. After they had been anesthetized, each received an infusion of Ringer’s lactate equal to 4% of the body weight, followed by a sustaining infusion of 0.2 ml/kg per min for the remainder of the experiment. A catheter with an inflatable balloon at its distal end (Fogarty dilation catheter, Edwards Laboratories) was introduced into the inferior vena cava via the femoral vein. The catheter was advanced under fluoroscopy and the balloon was positioned in the thoracic portion of the inferior vena cava. Through the other femoral vein, a catheter was introduced into the lower portion of the abdominal vena cava to record venous pressure (P23Db Statham transducer and Grass polygraph recorder) caudal to the balloon.

After 1 hour of equilibration, control hemodynamic measurements and blood collections were carried out. Subsequently, the balloon placed in the thoracic inferior vena cava was inflated until a slight reduction of the mean arterial blood pressure was observed. Fifty to 60 minutes later, hemodynamic determinations and blood collections were repeated. Indomethacin (5 mg/kg body weight) then was administered intravenously as described above. Twenty to 30 minutes later, final hemodynamic measurements and blood collections were carried out.

Measurements

The urinary sodium concentration was measured by flame photometry. Plasma renin activity (PRA) in arterial and renal venous blood was measured as previously (Oliver et al., 1980b). Plasma norepinephrine was measured by the procedure described by Peuler and Johnson (1977) and as was previously described (Oliver et al., 1980b). Prostaglandin E2 was measured by radioimmunoassay as previously described (Oliver et al., 1980a).

The rates of renal secretion of PGE2 and of norepinephrine were calculated as the product of renal plasma flow and the corresponding difference in concentration between renal venous and arterial plasma.

Statistical Analysis

All values are expressed as mean ± SEM. Data were analyzed by analysis of variance or by paired t-test (Winer, 1971). Differences were termed significant if the F or t value exceeded the 5% level.

Results

Effect of Inhibitors of Prostaglandin Synthesis on Renal Norepinephrine Overflow during Arterial Hypotension

Table 1 depicts the systemic and renal hemodynamic responses to the administration of teprotide and the subsequent inhibition of prostaglandin synthesis in 12 sodium-depleted dogs. Inhibition of the converting enzyme with teprotide resulted in significant decreases in mean arterial blood pressure and total peripheral vascular resistance. The cardiac output increased slightly but not significantly during hypotension. Blockade of the generation of angiotensin II also resulted in a marked increase in renal blood flow and a decrease in renal vascular resistance. Mean arterial pressure, cardiac output, and total peripheral vascular resistance were unchanged after inhibition of prostaglandin synthesis. However, mean renal blood flow declined significantly. Although renal vascular resistance increased slightly after inhibition of prostaglandin synthesis, the change was not statistically significant, probably because of the variability of this derived term.

Table 1 also shows the changes in arterial and renal venous PRA in this group of animals. Hypotension with teprotide significantly increased the PRA in both arterial and renal venous blood; the subsequent inhibition of the prostaglandin synthesis had no significant effect on the arterial and renal vein plasma renin activities.

During the control period in this group of dogs, the renal venous plasma concentration of PGE2 was significantly greater than the arterial PGE2 concentration (Table 2A). Teprotide-induced hypotension significantly increased the renal vein concentration of PGE2; the calculated renal secretion rate was not significantly higher than control. Subsequent ad-
ministration of the inhibitors of prostaglandin synthesis significantly decreased the renal vein concentration of PGE2 and the rate of renal secretion of PGE2.

Table 2B shows that the mean concentration of norepinephrine in the renal venous plasma was significantly greater than that of the arterial plasma during control conditions. The concentration of norepinephrine in the arterial plasma increased significantly during teprotide-induced hypotension. Despite the renal vasodilation which followed administration of teprotide, the mean concentration of norepinephrine in the renal venous plasma was significantly increased by hypotension and remained significantly higher than the concentration of arterial norepinephrine. The calculated mean renal overflow of norepinephrine also increased significantly during hypotension. Inhibition of prostaglandin synthesis during teprotide-induced hypotension had no significant effect upon the concentrations of norepinephrine in arterial and renal venous plasmas (Table 2B); the calculated rate of norepinephrine overflow from the kidney fell slightly but insignificantly.

**Effect of Inhibitors of Prostaglandin Synthesis on Renal Norepinephrine Overflow during Sodium Depletion**

Table 3 depicts the changes in systemic and renal hemodynamics and arterial and renal venous PRA induced by pharmacological inhibition of prostaglandin synthesis in this group of 11 dogs with chronic sodium depletion. Mean arterial pressure, cardiac output, and total peripheral vascular resistance were not significantly changed during inhibition of prostaglandin synthesis. Renal blood flow, however, was significantly reduced; renal vascular resistance increased slightly but not significantly. Table 3 also shows that prostaglandin synthesis inhibition did not significantly change the mean arterial or renal vein PRA.

Table 4A shows the effects of the inhibitors of
prostaglandin synthesis upon arterial and renal venous plasma concentrations of PGE₂ and the rate of renal secretion of PGE₂ in these sodium depleted dogs. During control conditions, the mean concentration of PGE₂ in the renal vein plasma was significantly greater than that in the arterial plasma; this difference disappeared after administration of inhibitors of prostaglandin synthesis. Accordingly, the renal secretion rate of PGE₂ was markedly lowered by indomethacin or meclofenemate.

Table 4B shows that, in this group of sodium-depleted dogs, the mean norepinephrine concentration in renal vein plasma was significantly greater than the arterial concentration during control conditions as well as after the administration of indomethacin or meclofenemate. Inhibition of prostaglandin synthesis had no statistically significant effect upon the concentrations of norepinephrine in renal vein or arterial plasma. Further, the rate of norepinephrine overflow from the kidney did not change significantly after inhibition of prostaglandin synthesis.

Effect of Inhibitors of Prostaglandin Synthesis on Renal Norepinephrine Overflow during an Acute Decrease in the Cardiac Output

Table 5 depicts the systemic and renal hemodynamic responses during the period of the acute decrease in the cardiac output induced by inflation of the balloon placed in the thoracic inferior vena cava and during the subsequent period when prostaglandin synthesis inhibitors were administered. Inflation of the balloon increased the mean pressure in the abdominal vena cava and resulted in a marked fall in cardiac output. Despite a significant increase of the total peripheral vascular resistance, arterial pressure fell slightly but significantly. After inflation of the balloon, renal blood flow increased slightly but significantly and renal vascular resistance fell

After administration of indomethacin during inflation of the thoracic caval balloon, there was no significant change in the pressure in the abdominal vena cava or in cardiac output or mean arterial blood pressure (Table 5). However, renal blood flow was significantly reduced; renal vascular resistance increased slightly but not significantly.

During control conditions, PRA’s in arterial and renal venous bloods were not significantly different from each other, indicating that little renin was being secreted by the kidney (Table 5). Inflation of the thoracic caval balloon increased PRA in both arterial and renal venous blood. Inhibition of prostaglandin synthesis during inflation of the balloon resulted in no significant change in the PRA of arterial and renal venous blood.

During the control period in these sodium-replete dogs, the arterial concentration of PGE₂ was not different from that of renal venous plasma, indicat-
TABLE 5  Effect of Obstruction of the Thoracic Inferior Vena Cava and Subsequent Prostaglandin Synthesis Inhibition on Systemic and Renal Hemodynamics and Plasma Renin Activity in Arterial and Renal Venous Blood

<table>
<thead>
<tr>
<th></th>
<th>IVCP (mm Hg)</th>
<th>MAP (mm Hg)</th>
<th>CO (liters/min)</th>
<th>TPVR (RU)</th>
<th>RBF (ml/min)</th>
<th>RVR (ru)</th>
<th>PRA (ng/ml per hr)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Control</td>
<td>3.1 ± 0.9</td>
<td>135 ± 7</td>
<td>3.63 ± 0.28</td>
<td>39.0 ± 4.2</td>
<td>219 ± 24</td>
<td>0.68 ± 0.10</td>
<td>1.9 ± 0.5</td>
</tr>
<tr>
<td>2. TIVCO</td>
<td>11.5 ± 1.1</td>
<td>126 ± 5</td>
<td>2.46 ± 0.27</td>
<td>55.0 ± 6.0</td>
<td>252 ± 27</td>
<td>0.53 ± 0.06</td>
<td>19.7 ± 5.9</td>
</tr>
<tr>
<td>3. PGSI</td>
<td>10.8 ± 1.1</td>
<td>124 ± 6</td>
<td>2.78 ± 0.37</td>
<td>49.5 ± 6.7</td>
<td>214 ± 25</td>
<td>0.64 ± 0.09</td>
<td>21.2 ± 6.9</td>
</tr>
</tbody>
</table>

Values are mean ± SEM; n = 8. IVCP, inferior vena cava pressure; TIVCO, thoracic inferior vena cava obstruction. For other abbreviations, see Table 1.

TABLE 6  Effect of Obstruction of the Thoracic Inferior Vena Cava and Subsequent Prostaglandin Synthesis Inhibition on Arterial and Renal Venous Plasma Concentrations of PGE2 and Norepinephrine and Their Renal Secretion Rates

<table>
<thead>
<tr>
<th></th>
<th>Arterial (pg/ml)</th>
<th>Renal vein (pg/ml)</th>
<th>ΔSEM</th>
<th>P</th>
<th>RSR (pg/min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A. Prostaglandin E2 (n = 6)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1. Control</td>
<td>26 ± 4</td>
<td>28 ± 5</td>
<td>2 ± 2</td>
<td>NS</td>
<td>135 ± 284</td>
</tr>
<tr>
<td>2. TIVCO</td>
<td>28 ± 4</td>
<td>96 ± 17</td>
<td>68 ± 15</td>
<td>&lt;0.01</td>
<td>1072 ± 2368</td>
</tr>
<tr>
<td>3. PGSI</td>
<td>23 ± 4</td>
<td>16 ± 5</td>
<td>-7 ± 6</td>
<td>NS</td>
<td>-1016 ± 697</td>
</tr>
<tr>
<td>P (2 vs. 1)</td>
<td>NS</td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
<td>NS</td>
<td>80</td>
</tr>
<tr>
<td>P (3 vs. 2)</td>
<td>NS</td>
<td>&lt;0.01</td>
<td>&lt;0.01</td>
<td>NS</td>
<td>80</td>
</tr>
</tbody>
</table>

| B. Norepinephrine (n = 8) |
| 1. Control | 30 ± 21 | 212 ± 60 | 123 ± 41 | <0.05 | 16106 ± 5690 |
| 2. TIVCO | 233 ± 59 | 496 ± 112 | 244 ± 66 | <0.001 | 37856 ± 9965 |
| 3. PGSI | 367 ± 114 | 548 ± 91 | 181 ± 62 | <0.005 | 21868 ± 6639 |
| P (2 vs. 1) | <0.05 | <0.01 | NS | NS | 80 |
| P (3 vs. 2) | NS | NS | NS | NS | 80 |

Values are mean ± SEM. For abbreviations, see Tables 1 and 2.

Discussion

The purpose of the present study was to determine whether endogenous prostaglandins inhibit the release of norepinephrine from the renal sympathetic nerves. The experimental protocols were designed to study animals in which sympathetic nerves to the kidneys were activated reflexly. The overflow of norepinephrine from the kidney into the renal venous blood was measured before and during inhibition of prostaglandin synthesis. In the three experimental conditions examined in the present study, prostaglandin synthesis inhibition failed to increase norepinephrine overflow from the kidney. These findings argue strongly against the hypothesis of Hedqvist and his coworkers (Frame and Hedqvist, 1975; Hedqvist, 1977) that prostaglandins synthesized in response to nerve stimulation inhibit norepinephrine release from renal sympathetic nerve terminals.
In the present study, the overflow of norepinephrine into the renal venous blood was used as an index of norepinephrine release from sympathetic nerves. While differences in the overflow fraction could result from differences in norepinephrine reuptake and/or metabolism, three lines of evidence suggest that changes in overflow reflect changes in norepinephrine release from the axon terminals. First, reflex activation of the sympathetic nervous system by several maneuvers, such as upright posture (Cryer, 1976), hemorrhage (Watts and Westfall, 1964), and exercise (Vendsalu, 1960), is associated with increased plasma norepinephrine concentration. Second, direct electrical stimulation of sympathetic nerves increases the overflow of norepinephrine in the venous effluent from the kidney (Oliver et al., 1980b), spleen (Hertting and Axelrod, 1961), and heart (Siegel et al., 1961), and the overflow varies with the frequency of stimulation (Oliver et al., 1980b). Finally, during norepinephrine release, vesicular enzymes such as dopamine-β-hydroxylase also are secreted, and the overflow of norepinephrine has been found to parallel the overflow of this enzyme in a variety of conditions (Weinshilboum et al., 1971a, b; Frewin et al., 1973; Noth and Mulrow, 1976).

Prior to administration of the inhibitors of prostaglandin synthesis, the sympathetic nervous system was activated by lowering the arterial blood pressure with teprotide, by chronic sodium depletion, and by an acute reduction of cardiac output produced by partial occlusion of the thoracic inferior vena cava. Figure 1 summarizes the data concerning plasma norepinephrine concentration in the renal vein in these three situations. During both hypotension and acute decrease in cardiac output, renal venous norepinephrine increased significantly above control values. Studies from this laboratory have shown that the concentration of norepinephrine in the renal venous plasma and the rate of norepinephrine overflow from the kidney in sodium-depleted dogs are significantly higher than those found in sodium-replete dogs (Oliver et al., 1980a). We have previously shown that renal venous norepinephrine concentration varies directly with the frequency of electrical stimulation of the renal nerves (Oliver et al., 1980b). Accordingly, it can be argued that the elevated renal venous norepinephrine concentrations observed in the three experimental conditions indicate that the discharge frequency of renal adrenergic nerves and the release of norepinephrine from renal axon terminals were enhanced at the time when prostaglandin synthesis was inhibited.

In all three experimental conditions, there was significant secretion of PGE₂ into the renal vein prior to administration of indomethacin or meclofenamate. As summarized in Figure 2, the concentration of PGE₂ in the renal venous plasma increased significantly from control values in response to both acute hypotension and acute reduction of cardiac output. Similarly, we have shown previously that the concentration of PGE₂ in renal venous plasma and the renal secretion rate of PGE₂ during chronic sodium depletion are higher than those in sodium-replete animals (Oliver et al., 1980a). Since prostaglandins are not stored in cells (Piper and Vane, 1971), these data show that, coincident with the enhanced renal overflow of norepinephrine, there was an increased synthesis of PGE₂ in the kidney.

The concentration of PGE₂ in renal venous plasma and the calculated rate of renal PGE₂ secretion during hypotension, sodium depletion and acute decrease in cardiac output fell markedly after administration of indomethacin or meclofenamate (Fig. 2; Tables 2A, 4A, and 6A). This is indicative of effective inhibition of renal prostaglandin synthesis. Yet, neither the concentration of norepinephrine in the renal venous plasma (Fig. 1) nor the overflow of neurotransmitter increased after prostaglandin inhibition (Tables 2B, 4B, and 6B). The data, therefore, provide evidence that, under in vivo conditions in the dog, endogenous prostaglandins do not inhibit the release of norepinephrine from renal sympathetic nerves.

![Figure 1](http://circres.ahajournals.org/)

**Figure 1** Mean concentration of norepinephrine in renal vein plasma in the three experimental conditions studied. See text for details. BP, hypotension; CO, acute decrease in cardiac output; PGSI, prostaglandin synthesis inhibition.

![Figure 2](http://circres.ahajournals.org/)

**Figure 2** Mean concentration of PGE₂ in renal vein plasma in the three experimental conditions studied. See text for details. Abbreviations as in Figure 1.
The results of the present study conflict with some, but not all, studies that have explored the role of prostaglandins in relation to norepinephrine release in isolated perfused organs. Inhibition of the prostaglandin synthesis enhanced the stimulation-evoked norepinephrine overflow in the isolated spleen (Hedqvist et al., 1971), heart (Samuelson and Wennmalm, 1971; Chanh et al., 1972), and kidney (Frame and Hedqvist, 1975). However, Dubocovich and Langer (1975), in agreement with the present study, failed to observe a measurable increase in the stimulation-evoked overflow of neurotransmitter after inhibition of prostaglandin synthesis in the isolated spleen. The reasons for these conflicting results for perfused organs remain unclear.

The results of the present study also conflict with the interpretation given to three studies carried out under in vivo conditions. Junstad and Wennmalm (1972) reported increased urinary excretion of norepinephrine in conscious rats given indomethacin by subcutaneous injection; Stjarne (1971) found similar results in rats given large doses of oral indomethacin and exposed to cold. However, in the first of these studies, subcutaneous injection of phosphate buffer alone also increased the urinary excretion of norepinephrine and, in the second, the animals showed clear signs of toxicity with reductions in urine volume and spontaneous movements and a 25% mortality rate. In a preliminary study, Hedqvist et al. (1980) have reported that prostaglandin synthesis inhibition enhanced the renal venous norepinephrine overflow evoked by electrical stimulation of the renal nerves in the in situ perfused rabbit kidney. Apart from the difference in animal species used and the fact that norepinephrine overflow was enhanced by direct electrical nerve stimulation rather than by spontaneous activation of the sympathetic nerves, no reason is clearly apparent for the discrepancy between the results of the present study and those of Hedqvist et al. (1980). Our results are consistent with the interpretation of data obtained in two studies carried out in humans. Indomethacin given to normal volunteers for 3 days (Rubin and Blaschke, 1979) and 7 days (Gullner et al., 1979) was found to produce no effect or to decrease slightly the peripheral plasma norepinephrine concentration during control conditions and during sympathetic stimulation by orthostatism. However, the significance of these results is uncertain because chronic indomethacin administration induces salt and water retention (Gullner et al., 1979), and sodium balance is known to be related inversely to plasma norepinephrine concentration (Luft et al., 1979).

In two of the experimental maneuvers in the present study (hypotension, reduction of cardiac output), norepinephrine overflow from the kidney was very large prior to the administration of the inhibitors of prostaglandin synthesis. In previous studies, inhibition of prostaglandin synthesis failed to enhance norepinephrine overflow at high frequencies of electrical nerve stimulation (Justad and Wennmalm, 1973; Stjarne, 1973). It might be argued, therefore, that failure to observe an augmentation of norepinephrine overflow after inhibition of prostaglandin synthesis during hypotension or reduction of the cardiac output was because the frequency of nerve discharges was very high. In this regard, the data for the dogs with sodium depletion that had a lower basal overflow of norepinephrine than those in which hypotension was induced with teprotide (23,334 ± 3,985 pg/min vs. 56,006 ± 11,623 pg/min; \( P < 0.05 \)) and thus, probably had a lower frequency of nerve impulses, are of interest. In those dogs, administration of indomethacin or meclofenamate again produced a marked decrease in the renal secretion rate of PGE2. Nevertheless, neither the renal vein and arterial concentrations of norepinephrine nor its rate of renal secretion was enhanced following inhibition of renal prostaglandin synthesis.

Although prostaglandin synthesis inhibition failed to increase significantly the renal venous overflow of norepinephrine, it significantly decreased renal blood flow under the three experimental conditions examined. Since prostaglandin inhibition was induced during activation of the renal sympathetic nervous system (during the three experimental conditions examined) and the renin-angiotensin system (during sodium depletion and acute decrease in cardiac output), the results are in agreement with studies indicating that the renal vasoconstriction caused by angiotensin II or norepinephrine are potentiated after inhibition of prostaglandin synthesis (Henrich et al., 1978). The data of the present study thus support the concept that renal prostaglandins attenuate the action of vasoconstrictor mechanisms in the renal circulation (McGiff et al., 1970). The present study provides strong evidence in the dog that the attenuating effect that prostaglandins exert on the renal vasoconstriction elicited by adrenergic nerves is not due to an inhibition of norepinephrine release.

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