Effect of Endogenously Produced Leukotrienes and Thromboxane on Renal Vascular Resistance in Rabbit Hydronephrosis

Christine R. Albrightson, Alex S. Evers, Anthony C. Griffin, and Philip Needleman

Ureteral obstruction in rabbits is characterized by mononuclear cell invasion of the renal cortex and proliferative fibrosis that is associated with exaggerated prostaglandin synthesis in response to vasoactive and inflammatory cell agonists. In this investigation, we studied the effects of the chemotactic peptide N-formylmethionyl-leucyl-phenylalanine (fMLP) and bradykinin (BK) on eicosanoid synthesis and renal vascular resistance in the ex vivo perfused hydropnephrotic kidney (HNK). Administration of fMLP resulted in the dose-dependent synthesis of leukotrienes, thromboxane A₁ (TXA₁), prostaglandin E₂ (PGE₂), and prostacyclin (PGI₁). Peptidoleukotriene synthesis was monitored by specific radioimmunoassay and by guinea pig ileum bioassay and it was then validated by inhibition of the ileal contractile activity with the peptidoleukotriene receptor antagonist FPL-55712. The leukotrienes produced were identified as LTB₄, LTD₄, and LTE₄ by comigration by inhibition of the ileal contractile activity with the peptidoleukotriene receptor antagonist FPL-55712 and that was completely inhibited by the thromboxane synthase inhibitor OKY-1581. Consistent with this result, exogenous administration of LTC₄, resulted in the synthesis of TXA₂, and in a renal vasoconstriction that was inhibited by either FPL-55712 or OKY-1581. These studies provide conclusive evidence that leukotrienes are produced in renal inflammation, that endogenously produced LTC₄, can stimulate the synthesis of TXA₂, and that LTC₄- induced vasoconstriction is mediated by the production of TXA₂. The vasoconstrictor effects of endogenously produced leukotrienes and thromboxane suggest a pathophysiologic role for these substances in renal inflammation. (Circulation Research 1987;61:514-522)
Leukotrienes, Thromboxane, and Vasoconstriction in HNK

Table of Abbreviations

Table

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
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<tbody>
<tr>
<td>fMLP</td>
<td>N-formylmethionyl-leucyl-phenylalanine</td>
</tr>
<tr>
<td>BK</td>
<td>bradykinin</td>
</tr>
<tr>
<td>HNK</td>
<td>hydronephrotic kidney</td>
</tr>
<tr>
<td>LT</td>
<td>leukotriene</td>
</tr>
<tr>
<td>TXA₂</td>
<td>thromboxane A₂</td>
</tr>
<tr>
<td>PG</td>
<td>prostaglandin</td>
</tr>
<tr>
<td>PGI₂</td>
<td>prostacyclin</td>
</tr>
<tr>
<td>RP-HPLC</td>
<td>reverse phase, high-performance liquid chromatography</td>
</tr>
<tr>
<td>UV</td>
<td>ultraviolet</td>
</tr>
<tr>
<td>LPS</td>
<td>lipopolysaccharide/endotoxin</td>
</tr>
<tr>
<td>HETE</td>
<td>hydroxyeicosatetraenoic acid</td>
</tr>
<tr>
<td>CLK</td>
<td>unobstructed contralateral kidney</td>
</tr>
<tr>
<td>RIA</td>
<td>radioimmunoassay</td>
</tr>
<tr>
<td>ODS</td>
<td>octadecylsilyle</td>
</tr>
<tr>
<td>PBS</td>
<td>phosphate-buffered saline solution</td>
</tr>
<tr>
<td>EDTA</td>
<td>ethylenediaminetetraacetic acid</td>
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<tr>
<td>PMN</td>
<td>polymorphonuclear leukocytes</td>
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Ureteral Obstruction and Ex Vivo Kidney Perfusion

Unilateral ureteral obstruction was carried out in male New Zealand white rabbits by a previously described procedure. Briefly, animals were anesthetized with sodium pentobarbital (30 mg/kg), the bladder was extruded through a small abdominal incision, and a silk suture was tied around the left ureter. The incision was repaired, and the animals were allowed to recover for 3 days. One hour before sacrifice, some animals received LPS (8 mg/kg i.v.). At the time of sacrifice, the animals were heparinized (500 U/animal), reanesthetized with sodium pentobarbital, and their renal arteries were cannulated with polyethylene tubing. The unobstructed CLK and the HNK or kidneys from rabbits not made hydronephrotic were excised, placed in separate warming jackets, and perfused at a constant flow of 10 ml/min with oxygenated (95% O₂-5% CO₂) Krebs-Henseleit buffer at 37° C. Renal perfusion pressure was monitored with a sidearm in the perfusion apparatus using a P-1000-A pressure transducer (Narco Bio-Systems, Inc., Houston, Tex.) and a Harvard 350 recorder (South Natick, Mass.). Basal perfusion pressure was comparable in both the HNK and CLK, consistent with the relatively low basal eicosanoid metabolite synthesis that occurs in the absence of agonist. The most apparent renal resistance changes between the HNK and CLK are noted during agonist (BK or fMLP) stimulation. The renal venous effluent was allowed to sequentially flow over 2 smooth muscle assay organs: rabbit thoracic aorta strips, to detect TXA₂-like activity, and guinea pig ileum strips, to detect peptide-leukotriene activity. The contraction of the smooth muscle strips was monitored by isotonic myographs and a chart recorder. The assay tissues were also superfused with a mixture of antagonists to render them insensitive to catecholamines, acetylcholine, serotonin, and histamine¹⁰ and superfused with indomethacin to prevent the assay tissue from synthesizing prostaglandins.¹¹ Before the experimental protocol, the kidneys were perfused ex vivo for 1 hour. At this time, the kidneys were challenged with intra-arterial bolus administration of either BK (30–1,000 ng) or fMLP (30–1,000 ng). Five-minute collections (50 ml) of renal venous effluent were made before and immediately following agonist administration. Aliquots (1 ml) of the effluent were saved (−20° C) for prostaglandin radioimmunoassay (RIA), and the remaining effluent was applied to a C-18 octadecylsilyle ODS column (as described below) for subsequent analysis for leukotrienes by RIA and reverse phase high-performance liquid chromatography (RP-HPLC). In inhibitor experiments, OKY-1581 (final concentration, 1 μg/ml) or FPL-55712 (0.4 μg/ml) was administered directly into the renal perfusion fluid.

Radioimmunoassay

Renal venous effluent was assayed for 6-keto-PGF₁α, the stable metabolite of prostacyclin (PGI₂), by a previously described method.⁷ PGE₂ and TXB₂, the stable metabolite of TXA₂, levels were measured by RIA using ¹²⁵I labelled ligand. Throughout the text and muscle, and by their immune cell chemotactic activity. A previous investigation in another model of inflammation, rabbit myocardial infarction, showed that fMLP stimulated the dose-dependent synthesis of peptidoleukotrienes and prostaglandins but that BK stimulated only the synthesis of prostaglandins.⁵ Based on this observation, we have tested the hypothesis that the HNK can produce leukotrienes. We now report that the rabbit HNK responds to fMLP with the synthesis of leukotrienes. These endogenously produced leukotrienes stimulate the synthesis of TXA₂ that, in turn, results in increased renal vascular resistance.

Materials and Methods

Chemicals

[¹⁹⁶]H6-keto-PGF₁α (160 Ci/mmol) and [¹²⁵]I NaI (14 mCi/μg) were purchased from Amersham, Arlington Heights, Ill., and [¹¹]LTC₄ (40 Ci/mmol) was purchased from New England Nuclear, Boston, Mass. LTC₄ antiserum was kindly supplied by Dr. R. Bell, Riker Laboratories, St. Paul, Minn., and 6-keto-PGF₁α antiserum was provided by Dr. R. Fertel, Ohio State University, Columbus, Ohio. Prostaglandin standards were the gift of Fisons Pharmaceuticals, Leicestershire, U.K.; indomethacin was a gift of Merck Sharp & Dohme, Rahway, N.J.; and OKY-1581 was a gift of Ono Pharmaceuticals, Osaka, Japan. LPS (Lipopolysaccharide endotoxin-lyophilized Escherichia coli 055:B5) was purchased from Difco Laboratories, Detroit, Mich.; fMLP was purchased from Sigma Chemical Co., St. Louis, Mo., and BK was purchased from Boehringer-Mannheim, Indianapolis, Ind.
Acid Metabolism

4°C, 16% polyethylene glycol in 50 mM PBS was added to the tubes. The precipitated bound tracer was containing 10,000 cpm. After overnight incubation at 4°C, 16% polyethylene glycol in 50 mM PBS was added to the tubes. The precipitated bound tracer was separated from the unbound ligand by centrifugation at 3,000 rpm for 30 minutes, and by decanting the supernatant. The radioactivity was counted in a Micro-medics Systems Apex automatic gamma counter. The cross reactivities of the 6-keto-PGF1α, PGE2, and TXB2 antisera have been previously described.7 Perfusion buffer in volume assayed did not affect the standard curve of these RIAs.

To assay for leukotrienes by RIA, the renal effluent was processed by a modification of the method of Powell.8 The effluent was acidified to pH 6.2 with HCl and applied to a 1-gram open-bed, preactivated C-18 ODS column (J.T. Baker Chemical Co., Philipsbury, N.J.). The column was washed with water, and the products were eluted with 10 ml of 100% methanol. An aliquot of the methanol was evaporated under nitrogen, and the products were resuspended in the LTC4-RIA buffer. The LTC4-antiserum cross reactivities at 50% displacement were 8% for LTD4, and 8% for LTE4.

### Results

**Effect of BK Stimulation on Rabbit Kidney Arachidonic Acid Metabolism**

Intra-arterial administration of BK to the ex vivo perfused HNK resulted in the dose-dependent synthesis of PGE2, PGI2, and TXA2. Treatment of the hydronephrotic animal with LPS 1 hour before removal of the ureter-obstructed kidney resulted in enhanced BK-stimulated prostanoic synthesis during the first several hours of perfusion (Table 1). These differences in peptide-stimulated prostaglandin synthesis were eliminated by prolonged ex vivo perfusion1 (current data not shown). The effects of LPS pretreatment were specific to the HNK because perfused unobstructed control kidneys from rabbits that received LPS pretreatment exhibited a minimal prostanoic synthetic response to BK (Table 2). Notably, the effluents from BK-stimulated HNK and CLK (±LPS) were devoid of guinea pig ileum contractile activity (bioassay for peptidoleukotrienes) and of detectable LTC4 immuno-reactivity.

**Table 1. The Effect of LPS-Pretreatment on the Maximal Eicosanoids Metabolite Responses of the HNK to Bradykinin and fMLP**

<table>
<thead>
<tr>
<th></th>
<th>PGE2</th>
<th>PGI2</th>
<th>TXA2</th>
<th>LTC4</th>
</tr>
</thead>
<tbody>
<tr>
<td>HNK + LPS</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>fMLP (300 ng) n = 7</td>
<td>545 ± 82*</td>
<td>357 ± 129*</td>
<td>252 ± 48</td>
<td>73 ± 9</td>
</tr>
<tr>
<td>BK (300 ng) n = 7</td>
<td>3,189 ± 788</td>
<td>1,235 ± 212</td>
<td>237 ± 81</td>
<td>...</td>
</tr>
<tr>
<td>HNK</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>fMLP (1 µg) n = 3</td>
<td>148 ± 85*</td>
<td>84 ± 13*</td>
<td>25 ± 10</td>
<td>9 ± 2</td>
</tr>
<tr>
<td>BK (1 µg) n = 3</td>
<td>1,343 ± 391</td>
<td>1,207 ± 193</td>
<td>49 ± 18</td>
<td>...</td>
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</table>

Hydronephrotic rabbits were either treated or not treated with LPS 1 hour before sacrifice. The HNKs were then removed and perfused ex vivo for 3-4 hours before stimulation with a maximal dose of either fMLP or BK. Maximal eicosanoid synthetic responses were obtained with 300 ng BK or fMLP in LPS-pretreated animals and with 1 µg BK or fMLP in nonpretreated animals. Values shown were determined by specific RIA and are expressed as nanograms of product measured in a 5-minute (50 ml) collection of effluent ± SEM. Ellipses indicate that the levels were not detectable. The synthesis of each metabolite elicited by BK was compared with the synthesis elicited by fMLP using a Student’s t test for unpaired data. The 2 pretreatment groups (±LPS) were analyzed separately. *p<0.05.
of the hydronephrotic animal with LPS 1 hour before kidney perfusion resulted in a leftward shift of the fMLP dose-response curves for eicosanoid synthesis (Figures 2A and 2B) and in an increase in the maximal response to fMLP (Table 1).

**Validation of Leukotriene Production in the fMLP-Stimulated HNK**

Since the LTC₄ antiserum cross reacts with LTD₃ and LTE₄, the RIA did not distinguish the species of leukotrienes released from the HNK. RP-HPLC separation of the products released from the fMLP-stimulated HNK revealed the presence of LTB₄, LTC₄, LTD₃, and LTE₄ in the renal venous effluent as detected by comigration with authentic leukotriene standards. RIAs for LTC₄, LTB₄, LTD₃, and LTE₄ in the renal venous effluent were inadequate for ultraviolet spectral analysis. Effluent subjected to RP-HPLC contained sufficient material for ultraviolet spectral analysis, and the RIA did not distinguish the species of leukotrienes in the HPLC fractions as described in "Materials and Methods" and are expressed as nanograms of product released (minus basal levels) during a 5-minute (10 ml/min) collection period ± SEM. Statistical comparisons of eicosanoid levels between HNK and CLK were performed for each agonist at each time point using a Student’s t test for unpaired data, *p<0.05. Ellipses indicate that the levels were undetectable.

**Effects of Endogenousty Produced Leukotrienes and Thromboxane on Renal Vascular Resistance in the Ex Vivo Perfused HNK**

Administration of BK and fMLP to the HNK from animals treated with LPS caused the synthesis of several vasoactive arachidonic acid metabolites. We used the leukotriene receptor antagonist FPL-55712 and the thromboxane synthase inhibitor OKY-1581 to determine the effects of endogenously produced peptidoleukotrienes and thromboxane on renal perfusion pressure. Under constant flow conditions, as performed in these experiments, increased perfusion pressure indicates increased vascular resistance. Administration of fMLP to the LPS-treated HNK resulted in an increase in perfusion pressure, coincident with the
synthesis of thromboxane and peptidoleukotrienes (Figure 4). Intrarenal infusion of the thromboxane synthase inhibitor OKY-1581 inhibited both the fMLP-stimulated TXA₂ synthesis and renal vasoconstriction. Administration of 100 ng of fMLP stimulated TXA₂ production from basal levels of 5.9 ± 1.2 to 121 ± 22 ng (n = 8), which were significantly inhibited by OKY-1581 to levels of 5.8 ± 2.7 ng (n = 5). Perfusion pressure increased with fMLP stimulation by 45 ± 10 mm Hg (n = 8) or by 3 ± 1 mm Hg (n = 5) in the presence of OKY-1581. Interestingly, the LT-receptor antagonist FPL-55712 partially inhibited the fMLP-stimulated TXA₂ synthesis (59 ± 20 ng, n = 7, p < 0.05) and the renal vasoconstriction [an increase of perfusion pressure of only 14 ± 4 mm Hg (n = 8)]. Stimulation of the LPS-treated CLK with 1,000 ng fMLP did not cause a change in perfusion pressure. BK administration to the LPS-treated HNK resulted in the production of TXA₂, PGE₂, and PGI₁ in association with a transient renal vasoconstriction. OKY-1581 inhibited BK-stimulated TXA₂ synthesis specifically. Therefore, the renal vasodilation observed after BK administration in the presence of OKY-1581 presumably resulted from the effect of PGE₂ and PGI₁. As would be expected, FPL-55712 had no effect on BK-stimulated vasoconstriction. A representative tracing, along with the corresponding TXA₂ values, from these experiments is shown in Figure 4. In this experiment BK stimulation of the HNK in the absence or presence of inhibitors (i.e., OKY-1581 or FPL-55712) produced 1,683 ± 91 ng PGE₂/5-min collection. Likewise, fMLP-stimulated PGE₂ production was unaffected by the inhibitors and ranged from 68 to 89 ng/5-min collection. BK- and fMLP-stimulated PGI₁ synthesis was also unaltered by OKY-1581 and FPL-55712.

**Effect of Exogenously Administered LTC₄ on Thromboxane Production and Renal Vascular Resistance in the Ex Vivo Perfused Rabbit Kidney**

Intra-arterial administration of LTC₄ (1-10 μg) to the LPS-treated HNK caused a dose-dependent release of TXA₂ (6.8-25.4 ng/50 ml). Only at the highest dose of LTC₄ (10 μg) was the renal arterial perfusion pressure increased above baseline (from 58 ± 9 to 74 ± 10 mm Hg; n = 4, p < 0.05). This dose of LTC₄ also stimulated the release of TXA₂ from 5.1 ± 1.3 to 41.7 ± 11.9 ng/50 ml; n = 4, p < 0.05). In the LPS-treated control CLK, LTC₄ did not stimulate the production of TXA₂ or cause a change in perfusion pressure. Administration of LTD₄ (10 μg) to the LPS-treated HNK or CLK did not stimulate TXA₂ production or change renal vascular resistance. Perfusion of the HNK with OKY-1581 or FPL-55712.

![Figure 1. Time-dependent effect of fMLP on eicosanoid synthesis in the isolated perfused HNK. The 3-day ureter-obstructed kidney (HNK) with (closed symbols; n = 7) or without (open symbols; n = 5) in vivo LPS treatment were perfused ex vivo (constant flow of 10 ml/min) for 1 hour before agonist stimulation. Samples of the renal effluent were collected for 5 minutes (50 ml) before agonist stimulation (basal) or immediately following intra-arterial bolus administration of fMLP (100 ng). Aliquots of effluent were analyzed by radioimmunoassay for TXB₂ (●, ◦) and 6-keto-PGF₁α (●, ◦), the stable metabolites of TXA₂ and PGI₁, respectively, and for PGE₂ (■, ◦) and LTC₄ (▲, △). Basal release of eicosanoids increased with time of perfusion; therefore, basal values (<10% of stimulated values) were subtracted from the fMLP-stimulated eicosanoid levels. LTD₄ antiserum cross reacted 50% with LTD₄ and 8% with LTE₄, and graphed values thus represent LTC₄-like immunoreactive material. All agonist-stimulated values were significantly increased over basal values and are expressed as nanograms of product in a 5-minute collection of effluent (minus basal values) ± SEM. Statistical comparison of eicosanoid levels between HNK and HNK pretreated with LPS were performed at each time point using Student’s t test for unpaired data: *p < 0.05](image-url)
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**Figure 2.** Dose-dependent effect of fMLP on eicosanoid synthesis in the isolated perfused HNK. Kidney preparations, RIA's, and symbols indicating specific arachidonate metabolites are as described in Figure 1. All agonist stimulations were performed between 1 and 3 hours of ex vivo perfusion. Values are expressed as nanograms of product released in a 5-minute collection of effluent ±SEM (n=3). Statistical comparisons of eicosanoid levels between HNK and HNK pretreated with LPS were performed at each dose using Student's t test for unpaired data: *p<0.05

Inhibited the LTC4-mediated TXA2 production and vasoconstriction (Figure 5). Full recovery of the TXA2 synthetic capacity and vasomotor activity of the HNK after administration of each inhibitor was monitored by the ability of fMLP to stimulate TXA2 synthesis and increase perfusion pressure.

**Discussion**

This study demonstrates several novel aspects of arachidonate metabolism in renal inflammation using the rabbit hydronephrotic kidney model. Leukotrienes were produced by the ex vivo perfused HNK in response to fMLP stimulation. This effect was greatly exaggerated by treating the hydronephrotic animal with LPS 1 hour before kidney perfusion. Peptidoleukotriene production in the HNK was initially detected in the renal effluent by bioassay. The presence of LTC4, LTD4, and LTE4 in the renal effluent was further confirmed by RP-HPLC and RIA. Both endogenous and exogenous LTC4 were shown to stimulate TXA2 synthesis in the HNK but not in the CLK. This LTC4-stimulated TXA2 synthesis resulted in renal vasoconstriction.

Although the cellular source of agonist-stimulated leukotriene synthesis in the HNK is unknown, there are several compelling lines of evidence suggesting an inflammatory cell origin. The fMLP stimulation of leukotriene and prostaglandin production in the HNK is most likely due to the infiltration of inflammatory cells since fMLP receptors have been demonstrated only on inflammatory cells such as polymorphonuclear...

**Figure 3.** RP-HPLC separation and LTC4 RIA of concentrated renal venous effluent from fMLP-stimulated HNK. The leukotrienes in the combined effluent from 4 HNKs stimulated 3 times with 100 ng fMLP were extracted and concentrated on an open-bed C18 ODS column and eluted with methanol. Sample was evaporated with nitrogen and applied to an RP-HPLC (Altex ultrasphere-ODS, 5µm) with a mobile phase of methanol: H2O: acetic acid (60:40:0.08), pH 6.5 (solvent A) at a flow rate of 1 mIl/min. Mobile phase was isocratic up to 25 minutes. Strength of mobile phase was increased by methanol (solvent B) gradient (from 0 to 25% between 25 and 40 minutes and 25 to 40% between 45 and 65 minutes). Leukotrienes were detected by their uv absorbance at 280 nm, and their retention times were compared with the indicated authentic leukotriene standards. Aliquots from HPLC fractions were assayed for peptidoleukotrienes (○) using an antisem that cross reacts with LTD4 (50%) and LTE4 (8%).

The presence of LTC4, LTD4, and LTE4 in the renal effluent was further confirmed by RP-HPLC and RIA. Both endogenous and exogenous LTC4 were shown to stimulate TXA2 synthesis in the HNK but not in the CLK. This LTC4-stimulated TXA2 synthesis resulted in renal vasoconstriction.
leukocytes and macrophages. Inflammatory cells in vitro stimulated with fMLP also produce leukotrienes. The unresponsiveness of the CLK to fMLP stimulation (little or no leukotriene production) is consistent with the low numbers of resident tissue macrophages. The hypothesis that the HNK-produced leukotrienes originate from inflammatory cells is further supported by the unmasked leukotriene production in the fMLP-stimulated HNK from rabbits treated with LPS, an inflammatory cell agonist. This novel production of leukotrienes is likely to result from LPS activation of the inflammatory cells recruited in vivo by the primary stimulus of tissue damage (in this case caused by ureter obstruction). Alternatively, LPS might stimulate inflammatory cell adherence to endothelium and migration into damaged organs. This seems less likely since fMLP stimulation of the CLK from rabbits treated with LPS causes minimal leukotriene production. The data obtained in this study suggest that fMLP and BK stimulate different cell types in the HNK and that the involved cells are present in the injured (HNK) but not the control (CLK) kidney. Three observations support this hypothesis. First, we are unaware of any evidence that demonstrates that BK directly stimulates macrophages or monocytes; we have treated cultured resident peritoneal macrophages with up to 1 μM BK and not observed the release of eicosanoids (data not shown). Others have reported similar results.

Secondly, BK (up to a 3,000-ng bolus) did not release detectable levels of leukotrienes from the HNK (± endotoxin in vivo pretreatment) even when assayed with a very sensitive immunoassay (10–30 pg LTC/D readily analyzed) following extraction of large volumes of renal effluent. Finally, the dose-response comparison of the agonist effect on the HNK demonstrates differential quantitative metabolite production, such that BK stimulation induces the release of 5–10 times more PGE, 6-keto-PGF, and TXB (maximal response is approximately 200 ng/50 ml with both agonists), and much less leukotriene (BK-undetectable versus fMLP approximately 50 ng/50 ml) than fMLP.

Leukotriene-stimulated TXA production has previously been demonstrated in guinea pig lungs and cultured bovine aortic endothelial cells. This leukotriene-stimulated TXA production is abolished by pretreatment with the leukotriene receptor antagonist FPL-55712. We demonstrated that the ex vivo perfused HNK (but not the CLK) from rabbits treated with LPS responds to fMLP stimulation with the synthesis of leukotrienes that, in turn, caused production of thromboxane. These experiments represent a unique example of an inflammatory state in which leukotrienes acquire the ability to stimulate TXA production. Likewise, this is a novel example of endogenously produced leukotrienes stimulating the production of TXA. These results indicate that inflammation confers on a previously unresponsive tissue the capacity for leukotriene synthesis and that endogenous leukotrienes in the HNK cause a response not detected in normal tissue.
The effect of leukotrienes (through TXA₂ synthesis) on renal vascular resistance was also altered by the in vivo inflammatory state. Only in the LPS-treated hydrenephrotic animal did the ex vivo perfusion pressure of the HNK increase with administration of FMLP and LTC₄. This newly acquired response to leukotrienes, mediated in part by the level and condition of inflammatory cells, may have an important impact on renal function. The renal vasoconstriction could reduce renal blood flow, cause tissue ischemia, and exacerbate tissue damage. The enhanced leukotriene production in the rabbit HNK also has the capacity to alter physiologic effects not examined in these studies. Potentially, altered leukotriene production in the HNK could enhance vascular leakage and stimulate inflammatory cell chemotaxis. These effects could, in turn, cause further inflammatory cell activation and further tissue damage.

The exaggerated PGE₂ and PGF₁α production induced by agonists in the HNK may conceivably provide a negative feedback modulation of the metabolic processes initiated by the invading inflammatory cells. PGE₂ suppresses: 1) the expression (as does PGF₁α) of the Class II histocompatibility molecule, Ia, on macrophages, a factor that we earlier demonstrated to stimulate eicosanoid metabolism in fibroblasts, endothelial cells, and vascular smooth muscle cells; and 3) macrophage cytotoxicity. Collectively, these observations suggest that regional production of PGE₂ at the site of injury may negatively modulate local macrophage metabolism and function.

In these studies, we demonstrate the production of leukotrienes in a renal model of inflammation, the HNK. The endogenously produced leukotrienes stimulated TXA₂ synthesis, resulting in renal vasoconstriction. The synthesis of leukotrienes and the ability of leukotrienes to stimulate thromboxane synthesis and subsequent renal vasoconstriction occurs only in the injured kidney, demonstrating both enhanced leukotriene production and enhanced leukotriene-mediated effects in hydrenephrosis. Collectively, these data demonstrate the synthesis of leukotrienes in hydrenephrosis and suggest their pathophysiologic importance in renal inflammation.

References

26. Williams JD, Robin JL, Lewis RA, Lee TH, Austin KF: Generation of leukotrienes by human monocytes pretreated...

**Key Words** • prostaglandins • renal inflammation • fMLP • bradykinin
Effect of endogenously produced leukotrienes and thromboxane on renal vascular resistance
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