Maintenance of Renal Vascular Reactivity Contributes to Acute Renal Failure during Endotoxemic Shock

Jean-Jacques Boffa and William J. Arendshorst

Department of Cell and Molecular Physiology, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina

Septic shock is characterized by hypotension and decreased systemic vascular resistance and impaired vascular reactivity. Renal vasoconstriction markedly contrasts with sepsis-induced generalized systemic vasodilation, which is strongly dependent on nitric oxide. Whether maintained renal vascular reactivity to vasoconstrictors contributes to the decrease in renal blood flow (RBF) and GFR observed during LPS-induced sepsis was tested by assessment of the acute effects of pressor agents on mean arterial pressure (MAP) and renal hemodynamics in endotoxemic and control mice. LPS-injected mice displayed lower MAP, RBF, and GFR than controls ($P < 0.001$). Despite a lower MAP, basal renal vascular resistance (RVR) was higher during endotoxemia ($P < 0.02$). Angiotensin II infusion produced a weaker MAP response in septic mice (24 versus 37%; $P < 0.005$), suggesting impaired vasoconstriction and hyporeactivity. A similar MAP increase was observed between groups during norepinephrine (NE) infusion. The MAP increase to nitric oxide synthase inhibition by N$^\text{G}$-nitro-l-arginine methyl ester (L-NAME) was much greater in LPS-treated mice (41 versus 15%, $P = 0.01$), indicating a strong influence of nitric oxide in sepsis. In contrast, the RBF and RVR responses to angiotensin II, NE, or L-NAME were similar in both groups. Moreover, vasopressin produced greater changes in MAP, RBF, and RVR in septic mice than in controls. Among the vasoconstrictor challenges, only NE ameliorated the decrease in GFR 14 h after LPS injection. The in vivo results demonstrate that the renal microvasculature displays a normal or enhanced reactivity to constrictor agents as compared with nonrenal circulatory beds. Such responsiveness may contribute to reduced RBF and GFR during endotoxemia.


The incidence of severe sepsis is increasing in the United States. At present, there are approximately 215,000 deaths per year with an estimated annual cost of $16.7 billion (1,2). Acute renal failure (ARF), clinically defined as a reduction of GFR, is a common complication of septic shock in humans. The mortality rate of ARF is 75% in septic patients, as compared with 45% in patients without sepsis (3). ARF independent of sepsis increases morbidity and mortality (4,5).

Septic shock is characterized by generalized systemic vasodilation and hypotension accompanied by vascular hyporesponsiveness leading to failure of multiple organs and death. Sepsis-induced in vivo hyporeactivity to constrictor agents has been investigated mainly by assessing the arterial BP response to vasoactive agents. There are few measurements of regional blood flow in either humans or experimental animals. An alternative approach has been to characterize vascular responsiveness in situ using isolated vessel segments or strips, largely on large-diameter conduit vessels rather than small-diameter resistance arterioles. The general finding during endotoxemia is vascular hyposensitivity and hyporesponsiveness to constrictor substances such as norepinephrine (NE) and angiotensin II (Ang II); a weak response also holds for endothelium-dependent vasodilators (6–11). Mixed results have been reported concerning vascular effects of vasopressin (AVP) and thromboxane (TxA$_2$) (9,11). Three different mechanisms have been implicated in the failure of the vascular smooth muscle cells (VSMC) to relax or constrict appropriately and the mediation of sepsis-induced generalized vasodilation, hypotension, and hyporesponsiveness (12). They are as follows: (1) K$_{ATP}$ channel activation secondary to a decrease in cellular ATP concentration hyperpolarizes the plasma membrane of VSMC and attenuates Ca$^{2+}$ entry through voltage-sensitive L-type channels normally seen during agonist-induced vasoconstriction (13). (2) Stimulated production of the vasodilator nitric oxide (NO) as a result of increased expression of inducible NO synthase (iNOS) via Toll-like receptor–induced NF-$\kappa$B activation (14). NO may reinforce its guanosine 3',5'-cyclic monophosphate–mediated vasodilation by secondary activation of the K$_{Ca}$ channel that hyperpolarizes VSMC with effects noted above (13). Whether one or both of these actions are involved, NO is thought to play a central role in vascular dysfunction during septic shock (15). (3) An inappropriate low plasma AVP concentration may also contribute to the hypotension and generalized vasodilation.

The physiopathology of septic ARF is poorly understood (16). Changes in renal hemodynamics are thought to be central in causing ARF as histologic findings in patients with sepsis show only relatively minor focal injury with early preservation of glomerular morphology (17). Similar results are found in experimental studies (18). We and other investigators have...
previously shown that an initial phase of renal vasoconstriction leads to a reduction in GFR and ARF in anesthetized mice. A marked decrease in renal blood flow (RBF) results from an increase in renal vascular resistance (RVR) in the face of hypotension after LPS injection. This early renal vasoconstriction is accompanied by a low mean arterial pressure (MAP) that persists from 1 to 14 h of observation after LPS administration (19). The constriction involves the preglomerular vasculature (20,21). Furthermore, we have demonstrated using both thromboxane (TxA2) receptor knockout mice and a TxA2 receptor antagonist that TxA2 receptors mediate the increased RVR and contribute to the reduced GFR during endotoxemic shock (19). Other studies highlight the role of endothelin and the cytokine TNF-α in sepsis-induced ARF (22,23).

Collectively, these lines of evidence indicate that the renal circulation is relatively independent of the splanchnic circulation in that it does not contribute to the classical systemic vasodilation. In this regard, renal vasoconstriction markedly contrasts with sepsis-induced dilation of the intestinal, hepatic, splenic, and nonsplanchnic vascular beds (24). The unique nature of the constriction of the renal microcirculatory response to sepsis or endotoxin injection and the underlying mechanisms are subjects of active investigation. The general vascular hyporeactivity to vasoconstrictors (Ang II, AVP, and NE) does not seem to explain the renal vasoconstriction (6). However, renal vasoconstriction may result from impaired endothelium-dependent vasodilation, a view supported by results obtained in vitro and in vivo from both humans and animals (25–27). High plasma levels of catecholamines and Ang II found in clinical and experimental studies during sepsis may contribute to the renal vasoconstriction (12,28). However, catecholamines are subject to inactivation by reactive oxygen species (ROS) (29).

We hypothesized that reactivity to agonists that activate G protein–coupled receptors (GPCR) varies among vascular beds during sepsis. We characterized the heterogeneity of systemic and renal vascular reactivity to three different receptor ligands, namely Ang II, NE, and AVP, as well as inhibition of NO production (N5-nitro-l-arginine methyl ester [L-NAME]). An aim was to determine whether the renal microcirculation maintains its normal vascular reactivity or becomes hyporesponsive to these agents. An unchanged renal vascular reactivity may be considered exaggerated relative to most nonrenal beds and thereby may contribute to the renal vasoconstriction and ARF during endotoxemic shock. Moreover, this protocol allowed us to compare the efficiency of various vasoconstrictor treatments that are used routinely in humans to restore MAP and correct for the decreases in RBF and GFR induced by LPS. Our results demonstrate a conserved or relatively enhanced renal vascular reactivity in response to vasoconstrictor agents in endotoxemic mice contributing to ARF in sepsis.

Materials and Methods
Male Balb C mice from Harlan Laboratories (Indianapolis, IN) were housed in the University of North Carolina at Chapel Hill animal facilities. All animal experiments were performed according to Institutional Animal Care and Use Committee guidelines of the University of North Carolina at Chapel Hill.

Body weight of male mice averaged 27 ± 1 g (n = 50). Mice were anesthetized with an intraperitoneal injection of pentobarbital (50 mg/kg) and placed on a servo-controlled heating table that maintained body temperature at 37°C. A tracheotomy was performed, and a tracheal catheter (PE-90) was inserted to facilitate breathing. The right femoral vein was cannulated with three pulled PE-10 catheters for continuous infusion of BSA (2.5%, 10 μl/min) and isotonic saline solution (0.9% NaCl, 0.3 ml/kg per min). This infusion rate was selected to ensure fluid resuscitation needed and recommended in septic conditions (30). Additional doses of pentobarbital were given intravenously as required. The right femoral artery was cannulated with a tapered PE-100 catheter connected to a pressure transducer (Statham P23 DB) for continuous monitoring of MAP. FITC-inulin (0.25%; Sigma, St. Louis, MO) was added to BSA infusion for determination of GFR by clearance method (31). A PE-50 catheter was inserted into the bladder to collect urine. After the mouse was placed on its right side, the left kidney was exposed through a subcostal incision. The left renal artery was gently dissected and isolated from the renal vein for determination of RBF using a noncannulating transducer connected to an ultrasonic flowmeter (Transonic system TS420; Ithaca, NY; 0.5v probe). After completion of surgery, the animals were allowed to stabilize for at least 45 min before measurements commenced. FITC fluorescence in plasma and urine was measured using a Zeiss fluorescence microscope with a ×10 lens, and an Orca-II cooled CCD camera.

Group I Studies
Either isotonic saline (10 ml/kg) or LPS (Escherichia coli serotype 026:B6, 8.5 mg/kg in 10 ml/kg; Sigma) was injected intraperitoneally 14 h before clearance and blood flow measurements of renal function. Observations were made over a 3-h period that included two 20-min control periods and two 20-min experimental periods. In the experimental periods, each mouse received two of three challenges, separated by a 30-min recovery period. After stabilization for 10 min, measurements were made during the subsequent 20 min. Ang II (Sigma) was infused intravenously for 30 min at 30 ng/kg per min; NE (Abbott Laboratories, Chicago, IL) was infused intravenously for 30 min at 6 μg/kg per min. L-NAME (Sigma), to inhibit NO synthases, was injected intravenously as a bolus (25 mg/kg). When tested, L-NAME was always given in the second experimental period. At the end of each period, 10 μl of blood was collected for FITC and hematocrit determination. At the end of an experiment, the left kidney was weighed.

Group II Studies
Different control and LPS-treated mice were challenged with AVP (Sigma), injected intravenously (100 ng/kg per min) after two 20-min control periods.

Statistical Analyses
Data are expressed as mean ± SEM. Differences between groups were compared by unpaired t test, using SigmaStat software. A paired t test was used to detect within-group differences produced by agonist infusion versus baseline. P < 0.05 was considered statistically significant.

Results
Sustained Effects of LPS Injection on Renal Hemodynamics
Endotoxemic mice 14 h after LPS injection seemed severely ill. They developed fever (38.2 ± 0.6 versus 36.4 ± 0.3°C; P < 0.001) and diarrhea and became lethargic. The mortality rate in LPS-treated mice was 30%. All mice died within 30 min after the anesthesia, 14 h after LPS injection, as a result of severe
hypotension and stress associated with anesthesia and surgery. Continuous intravenous saline infusion restored partially and stabilized the MAP. Nevertheless, relative to values in anesthetized, saline-injected control mice (Table 1), MAP was reduced by 18% to 83 ± 3 mmHg (P < 0.001) with a concomitant 18% increase in heart rate to 656 ± 23 beats/min (P < 0.001; Table 2). RBF and GFR decreased in septic mice by 46 and 64%, respectively (P < 0.001). In contrast to the vasodilation commonly reported for most nonrenal vascular beds, we observed an increase in RVR (50%; P = 0.01) in the kidneys of endotoxemic mice in the presence of a lower MAP. Urine flow was unaffected on the average by LPS. In other animals that were challenged with AVP, similar renal hemodynamic responses were noted in septic animals (Table 3).

**Acute Pressor Responses to Ang II, NE, AVP, and L-NAME**

The MAP responses to vasoconstrictors are summarized in Figure 1 and Tables 1 through 3. Ang II (30 ng/kg per min) increased MAP in both groups but to a lesser extent in septic versus control mice (24% from 83 mmHg versus 36% from 102 mmHg; P = 0.004). These results are consistent with the classical hyporeactivity previously reported during sepsis (6,10,11). In response to NE, MAP increased to a similar extent (P > 0.2), by 28 and 34% in endotoxemic and control, respectively. Global NO synthase inhibition produced a stronger MAP pressor response in LPS-injected mice than in control mice (P = 0.01), an effect suggestive of iNOS stimulation and enhanced NO-dependent vasodilation by LPS. Surprisingly, the rise in MAP elicited by AVP infusion was considerably greater in septic mice (50 ± 9 versus 18 ± 1%; P < 0.01).

**Renal Vascular Reactivity to Ang II, NE, and AVP and Inhibition of NO Production**

In contrast to the divergent effects of vasopressor agents on MAP in septic mice, RBF and RVR responses to Ang II, NE, and NOS inhibition were similar in control and septic mice (Figure 2). Ang II decreased RBF by approximately 55% in both groups, which correlated with a similar threefold rise in RVR. NE infusion had a small but similar effect on RBF and RVR during endotoxemia and control conditions, with respective changes of 1 ± 4 versus −10 ± 7% (P = 0.2) for RBF and 26 ± 7 versus 49 ± 13% (P > 0.1) for RVR. Importantly, we observed a stronger effect of nonselective NOS inhibition on MAP in septic animals. NOS inhibition produced renal vasoconstriction in both groups; RBF fell (−71 ± 3 versus −66 ± 4%; P > 0.3) and RVR rose (336 ± 52 versus 312 ± 62%; P > 0.7). GFR was reduced during NOS inhibition to a similar extent in control and septic mice (−56 ± 5 versus −46 ± 9%; P > 0.3). Thus, renal reactivity in response to Ang II, NE, and L-NAME was conserved in the main during endotoxia compared with normal conditions. In contrast, AVP increased RBF by 50% (P < 0.001) with a concurrent 19% decrease in RVR (P = 0.01) in control mice. AVP in septic mice tended to decrease RBF by 19% (P = 0.09) and increased RVR by 96% (P = 0.005).

**Effect of Vasoconstrictors on GFR and Urine Flow**

In control mice, Ang II and NOS inhibition decreased GFR by 25 and by 44%, respectively (P < 0.001). However, the pressor agents NE and AVP had no effect on GFR (7 ± 11% [P > 0.3] and 2 ± 9% [P > 0.9]) under control conditions (Tables 1 and 3). All pressor agents produced a diuresis during hypertension in control animals. In endotoxemic mice, NE increased GFR by 45% on the average (P = 0.01), whereas the other pressor agents had little effect on GFR (Tables 2 and 3). Except for NOS inhibition, urine flow increased with the pressor responses in septic mice.

**Discussion**

ARF in sepsis is a life-, cost-, and time-consuming disease, limited to supportive rather than curative treatment. Future optimal therapy would be advanced by a better understanding of the mechanisms of hemodynamic changes that would ameliorate the outcome of septic patients. The reduction in MAP and the selective increase in RVR along with reductions in RBF

| Table 1. Renal hemodynamics in control mice during basal conditions and during systemic administration of pressor agents*
<table>
<thead>
<tr>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Control Mice</strong></td>
</tr>
<tr>
<td>---------------------------------------------------------------</td>
</tr>
<tr>
<td>Mean arterial pressure (mmHg)</td>
</tr>
<tr>
<td>Renal blood flow (ml/min per g kidney weight)</td>
</tr>
<tr>
<td>Renal vascular resistance (mmHg/ml per min)</td>
</tr>
<tr>
<td>GFR (ml/min per g kidney weight)</td>
</tr>
<tr>
<td>Urine flow (ml/min)</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
</tr>
<tr>
<td>Left kidney weight (g)</td>
</tr>
<tr>
<td>Body weight (g)</td>
</tr>
<tr>
<td>No. of mice</td>
</tr>
</tbody>
</table>

*Values are Means ± SEM. Baseline versus Ang II, NE, L-NAME: *b* P < 0.05; *c* P < 0.01; *d* P < 0.001.
and enhanced pressor effect of L-NAME and AVP. In this setting, renal vasoconstriction is maintained in absolute terms and is enhanced relative to the pressor responses. Renal vasoconstriction during sepsis may contribute to a decline in GFR. In addition, our results show a marked renal and systemic sensitivity to exogenous AVP in endotoxemic shock. Moreover, the LPS-induced reduction of GFR was restored in part by NE treatment and clearly worsened during NOS inhibition.

Renal hemodynamic changes were studied in a standard experimental model of sepsis developed and described elsewhere (23). The “endotoxin model” may not completely reproduce clinical septic shock, but it has contributed significantly to our understanding of the mechanism of renal failure during sepsis (16). Although endotoxin injection and cecal ligation and puncture, an alternative sepsis model, vary by different cytokine kinetics, both models seem to induce ARF with common signaling pathways and similar sensitivity to therapeutic targets (32). We slightly adapted the initial experimental conditions by increasing the LPS dose to 8.5 mg/kg to mimic more closely septic shock in humans as described previously (19). For this reason, we focused on a relatively late phase of sepsis to simulate a short time-window of undiagnosed clinical sepsis during which one would determine whether drugs that are used routinely, such as NE, or more recently tested (NOS inhibition, AVP) in human septic shock would ameliorate LPS-induced reductions in GFR. The doses of pressor agents were chosen to elevate MAP approximately 30 mmHg during control conditions with a remnant pressor response during septic shock. The NE dose corresponds to the highest beneficial pressor dose for humans with an appreciation that high doses are likely to produce renal vasoconstriction (30). A high dose of L-NAME (25 mg/kg) inhibits NO production by iNOS as well as endothelial NOS. It is noteworthy that the renal hemodynamic changes induced by LPS in the present mouse study agree well with our recent report and those for in vitro and in vivo studies on rodents (19–21).

Comparison of vasoconstrictor effects on MAP, a systemic hemodynamic index, and on the renal microcirculation (RBF and RVR changes) between control and septic mice yields a

**Table 2. Renal hemodynamics in endotoxemic mice during basal conditions and vasoconstrictor treatments**

<table>
<thead>
<tr>
<th></th>
<th>Baseline</th>
<th>Ang II</th>
<th>NE</th>
<th>L-NAME</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean arterial pressure (mmHg)</td>
<td>83 ± 3c</td>
<td>103 ± 4d</td>
<td>109 ± 4e</td>
<td>117 ± 3e</td>
</tr>
<tr>
<td>Renal blood flow (ml/min per g kidney weight)</td>
<td>5.0 ± 0.5c</td>
<td>2.3 ± 0.3d</td>
<td>5.2 ± 0.6</td>
<td>1.8 ± 0.4e</td>
</tr>
<tr>
<td>Renal vascular resistance (mmHg/ml per min)</td>
<td>71 ± 3b</td>
<td>219 ± 35c</td>
<td>89 ± 13</td>
<td>298 ± 61e</td>
</tr>
<tr>
<td>GFR (ml/min per g kidney weight)</td>
<td>0.38 ± 0.05c</td>
<td>0.43 ± 0.03</td>
<td>0.66 ± 0.09d</td>
<td>0.25 ± 0.06</td>
</tr>
<tr>
<td>Urine flow (ml/min)</td>
<td>10.1 ± 1.0</td>
<td>20.9 ± 2.0f</td>
<td>22.2 ± 3.5d</td>
<td>8.2 ± 1.9</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>657 ± 23c</td>
<td>623 ± 30</td>
<td>615 ± 45</td>
<td>601 ± 35</td>
</tr>
<tr>
<td>Left kidney weight (g)</td>
<td></td>
<td></td>
<td>0.26 ± 0.01</td>
<td></td>
</tr>
<tr>
<td>Body weight (g)</td>
<td></td>
<td></td>
<td>26 ± 1</td>
<td></td>
</tr>
<tr>
<td>No. of mice</td>
<td>15</td>
<td>7</td>
<td>8</td>
<td>7</td>
</tr>
</tbody>
</table>

aValues are means ± SEM. Ang II, angiotensin II; NE, norepinephrine; L-NAME, N^G^-nitro-L-arginine methyl ester.

Control versus endotoxemic mice: ^p < 0.01; ^p < 0.001.

Baseline versus Ang II, NE, L-NAME: ^p < 0.01; ^p < 0.001.

Figure 1. Percentage increase in mean arterial pressure (MAP) in control (■) and endotoxemic (■) mice during intravenous infusion of angiotensin II (Ang II), norepinephrine (NE), N^G^-nitro-L-arginine methyl ester (L-NAME), or vasopressin (AVP). Means ± SEM. LPS-injected mice *versus* control mice, *p < 0.05, **p < 0.01. And GFR are characteristics of septic ARF. The fall in RBF in the face of hypotension suggests a predominance of constrictor mechanisms that may mask an apparent lack of autoregulation. To date, the reasons for the intriguing contrast of renal vasoconstriction versus systemic hypotension and generalized nonrenal vasodilation are poorly understood. In the present study, we show that the renal microcirculation behaves differently from nonrenal vascular beds, displaying maintained vascular reactivity evidenced by similar constrictor responses to Ang II, NE, and NOS inhibition in control and septic conditions. These results contrast with the more variable MAP responses observed: Attenuated response to Ang II, unchanged MAP to NE, and enhanced pressor effect of L-NAME and AVP. In this
general assessment of homogeneity of regional vascular responsiveness in vivo. The maintenance of a normal renal vascular sensitivity to Ang II, NE, and NOS inhibition and the exaggerated reactivity to AVP in septic mice account for the reduction in RBF and also in GFR. These in vivo findings extend and add relevance to previous ex vivo studies of vascular reactivity during sepsis. For example, after 24 h of continuous intravenous LPS infusion in rat, α-adrenergic receptor stimulation produced weaker-than-normal constriction of isolated aorta and mesenteric artery; reactivity of the renal artery was largely normal. A defect in receptor signaling was suggested by the observation that vascular smooth muscle cells from all vessels studied contracted normally to high [KCl] (9). However, other investigators found a more generalized defect with impaired contractile responses to high [KCl] in segments of superior mesenteric, hepatic, and coronary arteries incubated with LPS in organ culture for 20 h. However, function of the renal artery was more preserved than in other arteries (7). Moreover, the maximum contraction elicited by either NE or TxA2 was reduced in LPS-exposed superior mesenteric, hepatic, and coronary arteries. Of particular note, contraction by the renal artery was normal in the same study (7).

In situ assessment of contractile properties of preparations of isolated arteries suffers a serious limitation in that incubation for 20 h alone has the ability to attenuate reactivity and thus produce an effect similar to that of LPS, albeit less severe (11). To our knowledge, only one study has assessed the effect of vasoconstrictor agents on renal resistance arterioles from septic animals. A 24-h infusion of LPS leads to markedly reduced responsiveness to α-adrenoceptor stimulation (for 3 min) in carotid and mesenteric vascular beds in conscious rats, whereas renal vascular reactivity approximated normal (33). Thus, there is a clear pattern of heterogeneity, with vascular reactivity differing among organs and different constrictor substances. To date, there are no data comparing reactivity to Ang II among the renal and other vascular beds in septic shock.

Our understanding of mechanisms that are responsible for this regional heterogeneity in vascular reactivity is limited by a paucity of experimental data. Using a selective iNOS inhibitor, iNOS knockout mice, or global NOS inhibition, it has been

Table 3. Renal hemodynamics in control and endotoxemic mice before and during vasopressin infusion (AVP, 100 ng/kg per h intravenously)\(^a\)

<table>
<thead>
<tr>
<th></th>
<th>Control Mice Baseline AVP</th>
<th>Endotoxemic Mice Baseline AVP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean arterial pressure (mmHg)</td>
<td>110 ± 2</td>
<td>130 ± 2(^d)</td>
</tr>
<tr>
<td>Renal blood flow (ml/min per g kidney weight)</td>
<td>11.4 ± 0.5</td>
<td>16.5 ± 0.6(^d)</td>
</tr>
<tr>
<td>Renal vascular resistance</td>
<td>32 ± 1</td>
<td>26 ± 1(^c)</td>
</tr>
<tr>
<td>(mmHg/ml per min)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>GFR (ml/min per g kidney weight)</td>
<td>1.02 ± 0.05</td>
<td>1.03 ± 0.07(^d)</td>
</tr>
<tr>
<td>Urine flow (ml/min)</td>
<td>6.7 ± 0.9</td>
<td>30.4 ± 1(^d)</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>525 ± 8</td>
<td>496 ± 23</td>
</tr>
<tr>
<td>Left kidney weight (g)</td>
<td>0.29 ± 0.01</td>
<td>0.31 ± 0.01</td>
</tr>
<tr>
<td>Body weight (g)</td>
<td>29 ± 1</td>
<td>28 ± 1</td>
</tr>
<tr>
<td>No. of mice</td>
<td>6</td>
<td>6</td>
</tr>
</tbody>
</table>

\(^a\)Data are means ± SEM.

Control versus endotoxemic mice: \(^b\)P < 0.001.
Baseline versus AVP infusion: \(^c\)P < 0.01; \(^d\)P < 0.001.

Figure 2. Percentage change in renal blood flow (top) and in renal vascular resistance (bottom) in control (□) and endotoxemic (■) mice during vasoconstrictor treatments: Ang II, NE, L-NAME, or AVP. **P < 0.01, ***P < 0.001.
clearly shown that iNOS/NO production mediates most, if not all, of the weakened responsiveness to constrictor substances (7,25,34). However, measurements during endotoxemic shock reveal a relatively low to absent stimulation of iNOS and NO production in the kidney compared with other tissues (35,36). In accordance with this, we found a greater MAP response to NOS inhibition in endotoxemic mice than in controls but similar effects on RBF and RVR in both groups, suggesting a heterogeneous stimulated production of NO between renal and nonrenal vascular beds. In addition, LPS-induced iNOS/NO production may favor excessive generation of ROS and nitrogen species in liver, heart, lung, and blood. In contrast, ROS levels seem to be unchanged in rat kidney 4 h after LPS injection (37). Endothelium-dependent vasorelaxation was tested with acetylcholine in preconstricted isolated vessels. Exposure to endotoxin attenuated the maximum relaxation response to acetylcholine in coronary and in renal arteries in opposition to endotoxin attenuated the maximum relaxation response to isoprenaline-stimulated increases in cardiac ejection fraction, consistent with a role of β-arrestin1 in β-adrenergic receptor desensitization (49).

In summary, mice that are exposed to LPS develop hypotension and ARF with maintained renal vascular reactivity. The renal microcirculation exhibits normal sensitivity to vasoconstrictors such as Ang II, NE, and NOS inhibition during sepsis in contrast to dilation characteristic of nonrenal vascular beds. However, we observe that both renal and systemic reactivity to AVP is enhanced in endotoxemic mice. Collectively, the conserved or exaggerated renal reactivity to vasopressor agents may contribute to the selective renal vasoconstriction and decrease in GFR during endotoxemia. NO and ROS seem to modulate the vascular tone. Additional studies are needed to elucidate these and other mechanisms to improve renal function and overall patient outcome during septic shock.

Acknowledgments
This work was supported by National Institutes of Health Research Grant HL-02334 and the Institut National de la Santé et de la Recherche Médicale.

References


