Renal and Systemic Hemodynamic Effects of Synthetic Atrial Natriuretic Peptide in the Anesthetized Rat

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To characterize the hemodynamic events responsible for alterations in renal function during administration of atrial natriuretic peptide, we studied the systemic, renal, and glomerular circulatory effects of intravenous rANP[126-149], administered as a 4 μg/kg prime and 0.5 μg/kg per minute continuous infusion in anesthetized, euvolemic rats. With this protocol, a small decline in mean systemic arterial blood pressure occurred in the context of markedly enhanced urinary sodium excretion, hemoconcentration, and reduced left ventricular end-diastolic pressure and +dP/dt. However, despite a significant decrement in renal vascular resistance, total peripheral resistance remained constant, thereby denoting a preferential renal vasodilatory effect of this peptide in vivo. Whole kidney and single nephron GFR increased by approximately 20%, while effective renal and glomerular plasma flow rates remained stable, resulting in a substantial rise in filtration fraction. Of all the parameters potentially capable of augmenting single nephron GFR, only glomerular capillary hydraulic pressure increased significantly and therefore accounted entirely for the hyperfiltration observed during ANP infusion. This rise in glomerular capillary pressure, in turn, resulted from afferent arteriolar vasodilatation and concurrent effluent arteriolar vasoconstriction, findings that proved independent of both endogenous angiotensin II activity and ANP-induced reductions in renal perfusion pressure. These renal hemodynamic effects are unique when compared with actions of previously studied renal vasodilators.

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PEPTIDES isolated from mammalian atrial myocytes have been shown recently to function as circulating hormones that are capable of interacting at several major control loci in the regulation of body fluid homeostasis. Such actions of these atrial natriuretic peptides (ANP) include an elevation of glomerular filtration rate (GFR) and renal sodium excretion; inhibition of the secretion of renin, aldosterone, and vasopressin; relaxation of contracted vasculature in vitro; and reduction of systemic arterial blood pressure in vivo. The potential importance of these effects of ANP in modulating extracellular fluid volume and arterial blood pressure under both physiologic and pathophysiologic conditions remains to be determined.

The augmentation of GFR induced by these peptides has been observed in many animal species, as well as in man, despite only a transient or undetectable increase in renal plasma flow rate. Such an elevation of effective whole kidney filtration fraction is an unusual action for a renal vasodilator and has been taken as evidence that ANP produces an increase in postglomerular vascular resistance and/or glomerular ultrafiltration coefficient; however, direct determination of these effects on the glomerular circulation is lacking. The present studies, therefore, were undertaken to evaluate the effects of ANP on the systemic, renal, and glomerular circulations in order to delineate the hemodynamic events responsible for peptide-induced alterations in renal function.

Materials and Methods

Seven groups of male, Munich-Wistar rats (Simonson Laboratory, Gilroy, Calif.), weighing 235-340 g, were employed in these studies. All animals were fed standard rat chow ad libitum (Wayne Rodent Blox #8604; Continental Grain Company, Chicago, Ill.) and were housed at 24°C under an alternating 12-hour light/dark cycle for at least 1 week prior to study.

Micropuncture Studies (Groups 1-3)

In preparation for micropuncture study, rats were anesthetized with Inactin (thiobutabarbital, 100 mg/kg b.w., i.p.; Byk Gulden, Konstanz, F.R.G.) and positioned on a thermistor-controlled table to maintain central body temperature at 37.5°C. Following a tracheostomy, a polyethylene (PE-50) catheter was inserted into the left femoral artery for periodic sampling of blood and for monitoring arterial blood pressure via an electronic transducer (Model P23Db, Statham Instru-
ments Division, Gould, Inc., Oxnard, Calif.) coupled to a direct-writing recorder (Model 7754 A, Hewlett-Packard Co., Palo Alto, Calif.). Catheters (PE-50) were also inserted into the left femoral vein for infusion of plasma and into the right and left jugular veins for infusion of inulin/PAH and ANP (or vehicle), respectively. Since plasma volume is reduced by approximately 20% during preparation for micropuncture, all rats received an intravenous infusion of isoncotic rat plasma equivalent to 1% of body weight over 45 minutes, after which the infusion rate was reduced to 1.5 ml/kg per hour; this protocol has been shown to maintain plasma volume at pre-anesthesia levels.

All animals also received a continuous infusion of 0.9% NaCl containing 7% inulin (Taylor Pharmaceutical Co., Decatur, Ill.) and 0.8% PAH (para-aminohippurate sodium; Merck Sharp & Dohme, West Point, Pa.) at a rate of 1.2 ml/hour. Following midline and left subcostal incisions, the left kidney was suspended on a Lucite holder, its surface illuminated with a fiber-optic/quartz rod light source, and bathed with warmed 0.9% NaCl. The left ureter was cannulated with PE-10 tubing and the bladder vented by a curved 19-gauge needle.

After allowing 45-60 minutes for animal equilibration following the reduction in plasma infusion rate, micropuncture and clearance measurements were begun. Late surface convolutions of several proximal and distal tubules were located by observing the passage of 0.2% lissamine green dye injected into proximal tubule segments. Precisely timed (1.0-3.0 minutes) collections of tubule fluids were then obtained from 2-3 nephrons at late proximal tubule sites and from 2-3 separate nephrons at distal tubule sites for measurement of tubule fluid flow rate and inulin concentration. Samples of efferent arteriolar blood were taken from 2-4 surface star-vesseis for determination of efferent plasma protein concentration. Concurrently, femoral arterial blood samples were obtained for measurement of systemic arterial hematocrit and plasma concentrations of total protein, sodium, inulin, and PAH; and urine from the left kidney was collected in calibrated glass cylinders under paraffin oil at 10- to 20-minute intervals for determination of urine flow rate and concentrations of sodium, inulin, and PAH. Time-averaged hydraulic pressures were measured in surface glomerular capillaries (Poc), efferent arterioles (Pp), and proximal tubules (Pw) with a continuously recording, servo-nulling system (Model 3, Instrumentation for Physiology and Medicine, San Diego, Calif.), which employed 2-3 μm (outer tip diameter) micropipettes filled with 2.0 M NaCl. Hydraulic output from the servo system was coupled electronically to a second channel of the Hewlett-Packard recorder by means of a pressure transducer (Statham Model P23Db).

Following completion of measurements during the control period (30-40 minutes) in Group 1 animals (n = 6), synthetic ANP (Auriculin A, rANP[126-149]; Peninsula Laboratories, Inc., Belmont, Calif.) in 0.9% NaCl was infused intravenously as a 4 μg/kg priming dose over a 2-minute interval, followed by a continuous IV infusion at the rate of 0.5 μg/kg per minute (1.56 ml/hour). All micropuncture and clearance studies were repeated during the infusion of ANP (30-40 minutes); tubule fluid samples were recollected from the same tubule segments studied during the control period. To obviate dead-space artifact during the transition from low to high urine flow rates, these studies were begun 3-5 minutes after completion of the priming ANP dose, an interval that allowed maximal urine flow rates to be achieved.

Group 2 animals (n = 7) were subjected to an identical protocol except that an adjustable silk ligature was positioned proximal to the renal arteries and tightened during the control period to reduce renal perfusion pressure to the slightly lower level that occurs during infusion of ANP. The ligature then was loosened at the start of the peptide infusion period, thereby maintaining renal perfusion pressure at a constant level throughout both periods of study.

The protocol for Group 3 animals (n = 8) differed from that of Group 1 only by the addition of saralasin (Sarenin, Norwich-Eaton Pharmaceuticals, Norwich, N.Y.) to the standard inulin/PAH infusion at the outset of animal preparation so as to deliver 5 μg/kg per minute of this angiotensin II receptor antagonist throughout the experiment.

Chemical Analyses and Calculations

The volume of fluid collected from individual late proximal and distal tubules was measured in a constant-bore capillary tube of known internal diameter. The concentration of inulin in tubule fluid was determined, usually in duplicate, by a microfluorescence technique. The concentrations of inulin and PAH in plasma and urine were measured respectively by a macro-anthrone method and by a colorimetric technique. Plasma and urine sodium concentrations were analyzed by flame photometry. Total protein concentration in femoral arterial and efferent arteriolar plasma was determined, usually in duplicate, using a microfluorometric technique, and was used to derive values for the colloid osmotic pressure of plasma entering (πl) and leaving (πp) glomerular capillaries, respectively. Values for single nephron GFR (SNGFR), single nephron filtration fraction (SNFF), glomerular capillary plasma flow rate (Qc), glomerular blood flow (GBF), single afferent arteriolar resistances (Rael) and efferent arteriolar resistances, total arteriolar resistance (Rae) and the glomerular capillary ultrafiltration coefficient (Kc) were calculated using equations reported in detail previously. Since ANP has been shown to have no effect on renal PAH extraction, these equations were modified to calculate renal blood flow (RBF) and whole kidney vascular resistance (Rk) from PAH clearance measurements.

Cardiovascular Studies (Groups 4-7)

To characterize the renal effects of ANP in relation to overall circulatory dynamics, additional groups of rats were prepared in a manner similar to that for renal micropuncture, except that left kidney suspension and...
fiber-optic illumination were omitted. In Groups 4 and 5, only relatively noninvasive cardiovascular measurements were obtained both to ensure comparability with Groups 1–3 and to provide a reference point for the more invasive studies in Groups 6 and 7.

Groups 4 and 5. Following renal surgical preparation and initial plasma replacement, infusion of plasma was reduced to the maintenance rate of 1.5 ml/kg per hour and the animal was prepared for cardiovascular monitoring. A PE-50 catheter, joined to a micro-tip pressure transducer (Model PC350, Millar Instruments, Inc., Houston, Tex.) by a series of tubing connections (PE50 to PE90 to 0.04" i.d. Silastic) to a stopcock, was inserted into the right carotid artery and advanced into the left ventricle for continuous monitoring of systolic and end-diastolic pressures, as well as generation of a continuous recording of the maximum rate of rise in pressure (+dP/dt) via a derivative computer. This saline-filled, catheter-transducer system has an undamped natural frequency of about 100 Hz. Although the fluid filled catheter system overestimates +dP/dt, the values of this parameter thus obtained were used solely for the purpose of relative comparison and not as an absolute measurement. Another PE-50 catheter, connected to a Statham pressure transducer (Model P50), was inserted into the right jugular vein and positioned near the right atrium to monitor central venous pressure; an intervening stopcock permitted the continuous infusion of 0.9% NaCl, as in Groups 1–3. A 23-gauge needle, connected to a Statham P50 pressure transducer via PE-50 tubing, was inserted directly into the left renal vein for continuous monitoring of renal venous pressure.

Following the 45–60-minute equilibration period, phasic and mean ventricular, systemic, and venous pressures were recorded every 2 minutes over a stable 10-minute period and were averaged to provide baseline hemodynamic values. An intravenous priming dose and subsequent continuous infusion of ANP then were administered to Group 6 animals (n = 6) in a fashion identical to that in Groups 1–4, while Group 7 animals (n = 6) received a 0.9% NaCl vehicle infusion. Phasic and mean pressures and flows were recorded at 10, 15, 20, 25, and 30 minutes following initiation of the priming infusion of ANP and were averaged to provide hemodynamic values for the experimental period. Total peripheral resistance (TPR) was calculated from mean arterial pressure (AP), central venous pressure (CVP), and cardiac output (CO): TPR = (AP − CVP)/CO. Urine from the left kidney again was collected under paraffin oil at 10- to 20-minute intervals for determination of urine flow rate and sodium concentration during both the control and ANP (or vehicle) periods of study.

Statistical Analysis

All results are expressed as means ± SEM. Statistical analyses were performed by either paired or Student's t test, as appropriate. Statistical significance was defined as p < 0.05.

Results

Micropuncture Studies

Group 1. As summarized in Tables 1 and 2, the infusion of ANP in Group 1 animals resulted in an 18-fold increase in urinary sodium excretion (U_{Na}\text{,} V, from 0.49 ± 0.26 to 9.21 ± 0.82 μEq/minute) in the setting of a modest reduction in mean arterial pressure (AP). An attendant trend toward hemoconcentration was observed, but failed to achieve statistical significance. Whole kidney inulin clearance (C_{inu}), a measure of GFR, increased substantially, on average by 20% over control values (from 1.21 ± 0.05 to 1.45 ± 0.03 ml/minute), while effective renal plasma flow rate, estimated from PAH clearance (C_{PAH}), was largely unaffected by infusion of ANP. However, renal blood flow (RBF) increased significantly in the setting of a reduction in whole kidney vascular resistance (R_{k}).

At the single nephron level in surface glomeruli of these animals, the percent increment in GFR was similar to that observed for the whole kidney: Single nephron GFR (SNGFR) increased on average by 19% whether measured at late proximal (from 44.5 ± 3.3 to 52.8 ± 2.5 nl/minute) or distal (from 43.2 ± 4.4 to 51.2 ± 3.1 nl/minute, p < 0.05) tubule sites. This
TABLE 1. Whole Kidney and Renal Microcirculation Studies (Group I)

<table>
<thead>
<tr>
<th>Activity</th>
<th>Control (μEq/min)</th>
<th>ANP (μEq/min)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urinary sodium excretion</td>
<td>47.5 ± 0.1</td>
<td>46.4 ± 0.6</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Glomerular ultrafiltration</td>
<td>3.69 ± 0.1</td>
<td>3.69 ± 0.0</td>
<td>0.05</td>
</tr>
<tr>
<td>Glomerular capillary hydraulic pressure (Pc)</td>
<td>53.5 ± 2.3</td>
<td>53.5 ± 1.7</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Mean glomerular filtrate fraction (Kf)</td>
<td>0.005</td>
<td>0.005</td>
<td>0.2</td>
</tr>
<tr>
<td>Mean glomerular capillary hydraulic pressure (Pc)</td>
<td>45.7 ± 0.6</td>
<td>45.7 ± 0.6</td>
<td>0.05</td>
</tr>
<tr>
<td>Mean glomerular capillary hydraulic pressure (Pc)</td>
<td>14.1 ± 0.3</td>
<td>14.1 ± 0.3</td>
<td>0.05</td>
</tr>
</tbody>
</table>

Means ± SEM; p values derived from paired t test analysis of control versus ANP periods; NS, p > 0.05.

An adjustable suprarenal aortic clamp was employed in Group 2 animals to prevent changes in renal perfusion pressure during the infusion of ANP so that the possible role of ANP-induced reductions in renal perfusion pressure per se in mediating the alterations in pre- and post-glomerular resistances observed in Group 1 animals could be examined. Figure 1 and Table 2 demonstrate the clamp’s effectiveness in maintaining constant and changes observed in Group 1 animals during ANP infusion. However, despite this lower control value, a significant decrement in Rg, again occurred during ANP infusion (from 1.00 ± 0.02 to 1.45 ± 0.03, p < 0.005) and produced a large increase in PEF (from 38.0 ± 2.4 to 63.5 ± 1.7 mm Hg, p < 0.05), which was counted only minimally by a small increment in n (from 0.080 ± 0.017 and 0.064 ± 0.012 (nl/min)/(s·mm Hg), control and ANP periods, respectively; p > 0.2) or unique (5 of 7 animals; 0.067 ± 0.017 and 0.046 ± 0.006 (nl/min)/(s·mm Hg), respectively; p > 0.1) values.

Afferent arteriolar resistance of Group 2 animals during the control period was somewhat lower than that observed in Group 1 animals, presumably reflecting an autoregulatory response to the deliberate reduction in renal perfusion pressure. However, despite this lower control value, a significant decrement in Rg, again occurred during ANP infusion (from 1.00 ± 0.02 to 1.45 ± 0.03, p < 0.005) and produced a large increase in PEF (from 38.0 ± 2.4 to 63.5 ± 1.7 mm Hg, p < 0.05), which was counted only minimally by a small increment in n (from 0.080 ± 0.017 and 0.064 ± 0.012 (nl/min)/(s·mm Hg), control and ANP periods, respectively; p > 0.2) or unique (5 of 7 animals; 0.067 ± 0.017 and 0.046 ± 0.006 (nl/min)/(s·mm Hg), respectively; p > 0.1) values.
the prevention of a fall in renal perfusion pressure during ANP infusion, the changes in whole kidney hemodynamics and in glomerular pressures, flows, and resistances were similar to those seen in Group 1 animals where the ANP-induced fall in renal perfusion pressure was allowed.

**Group 3.** To examine the possibility that endogenous angiotensin II prevented a fall in R\(_K\) during ANP infusion and thereby contributed to the ANP-induced increases in P\(_{\text{GC}}\) and SNGFR, an intravenous infusion of saralasin was begun at the outset of surgical preparation in Group 3 animals and maintained throughout the experiment. When the action of endogenous angiotensin II was suppressed in this manner, the infusion of ANP resulted in a more substantial decline in AP than that observed in Groups 1 and 2, but the ANP-induced fall in R\(_K\) was again < 0.05. As hematocrit increased from 48.2 ± 1.2 to 50.1 ± 1.2 vol%, a substantial rise in C\(_{5}\) followed, as hematocrit increased from 48.2 ± 1.2 to 50.2 ± 1.2 vol% (p < 0.001).

Hemodynamic changes at the single nephron level were remarkably similar to those observed in Groups 1 and 2. SNGFR again increased on average by 24% when measured at late proximal tubule sites (from 50.9 ± 3.7 to 63.0 ± 3.9 nl/minute, p < 0.001), and by 20% when measured at distal tubule sites (from 50.8 ± 3.6 to 60.8 ± 3.4 nl/minute, p < 0.001). This hyperfiltration response again occurred in the absence of a change in Q\(_{\text{a}}\) (224.6 ± 25.4 and 235.0 ± 21.7 nl/minute during control and ANP periods, respectively; p > 0.5) or GBF and, thus, denoted a substantial elevation (p < 0.02) of SNGFR, P\(_{\text{GC}}\) again increased dramatically (from 52.3 ± 2.1 to 66.1 ± 3.4 mm Hg, p < 0.001, resulting in a large rise in AP (from 36.2 ± 2.2 to 47.3 ± 2.4 mm Hg, p < 0.001) that was not tempered by alterations in Q\(_{\text{a}}\) (22.3 ± 0.4 and 23.4 ± 0.9 mm Hg during control and ANP periods, respectively; p > 0.2). As in Groups 1 and 2, K\(_{\text{f}}\) failed to change significantly whether calculated as minimum (0.197 ± 0.068 and 0.077 ± 0.012 nl/[s-mm Hg], control and ANP infusion periods, respectively; p > 0.1) or unique (5 of 8 animals; 0.138 ± 0.036 and 0.064 ± 0.013 nl/[s-mm Hg], respectively; p > 0.1) values.

Afferent arteriolar resistance declined substantially during ANP infusion (from 1.20 ± 0.16 to 0.64 ± 0.08 x 10\(^{-3}\) dyne·s·cm\(^{-2}\), p < 0.002) and resulted in an overall reduction in R\(_{\text{T}}\) (from 2.06 ± 0.25 to 1.57 ± 0.19 x 10\(^{-3}\) dyne·s·cm\(^{-2}\), p < 0.005). Nevertheless, despite the antagonism of endogenous angioten-
sin II action, a significant increase in $R_e$ again was observed (from $0.77 \pm 0.13$ to $0.94 \pm 0.13 \times 10^{10}$ dyne·s·cm⁻², $p < 0.05$).

**Cardiovascular Studies**

**GROUPS 4 AND 5.** Control values for $\overline{A}P$, heart rate (HR), central venous pressure (CVP), renal venous pressure (RVP), left ventricular end-diastolic pressure (LVEDP), and $+dP/dt$ in the closed-chest preparation of Group 4 [ANP] animals were similar to those in Group 5 [vehicle] ($p > 0.05$ for each parameter, Table 3). During the infusion of ANP in Group 4 animals, significant decrements in $\overline{A}P$, LVEDP, and $+dP/dt$ occurred, while HR, CVP, and RVP remained stable. The decline in $\overline{A}P$ and increase in $U_{Na}$V (from $0.29 \pm 0.07$ to $5.12 \pm 0.75 \mu$Eq/minute, $p < 0.001$) were of the same magnitude as those observed in Group 1 studies. In contrast, during the infusion of vehicle in Group 5 animals, no change occurred in any of these hemodynamic parameters or in $U_{Na}$V ($0.31 \pm 0.09$ and $0.60 \pm 0.11 \mu$Eq/minute, control and vehicle infusion periods, respectively; $p > 0.05$).

**GROUPS 6 AND 7.** Control values for $\overline{A}P$, HR, $+dP/dt$, cardiac index (CI), and total peripheral resistance index (TPRI) in Group 6 [ANP] animals were similar to those in Group 7 [vehicle] ($p > 0.5$ for each parameter, Table 4). The simultaneous measurement of cardiac output and ventricular pressures necessitated the replacement of the ventricular PE-50 catheter with PE-10 tubing prior to positioning the flow probe.

![Figure 1. Hemodynamic parameters for Group 2, in which renal perfusion pressure was maintained at a constant level throughout control and ANP infusion periods. Values are expressed as means ± SEM; *p < 0.05 versus control period by paired t test.](http://circres.ahajournals.org/)

![Figure 2. Hemodynamic parameters for Group 3, in which saralasin was administered at a constant rate throughout control and ANP infusion periods. Values are expressed as means ± SEM; *p < 0.05 versus control period by paired t test.](http://circres.ahajournals.org/)
on the ascending aorta and therefore resulted in control values for +dP/dt that were lower than those observed in Groups 4 and 5. However, prior to catheter replacement, +dP/dt determinations in Group 6 (10.590 ± 380 mm Hg/second) and Group 7 (11.611 ± 866 mm Hg/second) were similar in magnitude to values obtained in Groups 4 and 5 (p > 0.2 Group 4 versus Group 6, p > 0.5 Group 5 versus Group 7).

In vehicle-infused Group 7 animals, +dP/dt and CI declined slightly with time (by 4.4% and 5.4%, respectively), while AP and TPRI remained stable. Despite a significant reduction in AP, LVEDP, left ventricular end-diastolic pressure; +dP/dt, maximum rate of rise of left ventricular pressure.

**Table 3. Cardiovascular Studies (Groups 4 and 5)**

<table>
<thead>
<tr>
<th></th>
<th>AP (mm Hg)</th>
<th>HR (beats/min)</th>
<th>CVP (mm Hg)</th>
<th>RV (mm Hg)</th>
<th>LVEDP (mm Hg)</th>
<th>+dP/dt (mm Hg/sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Group 4</strong></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Control</td>
<td>112 ± 4</td>
<td>348 ± 15</td>
<td>0.7 ± 0.5</td>
<td>1.6 ± 0.5</td>
<td>4.5 ± 0.5</td>
<td>11.919 ± 822</td>
</tr>
<tr>
<td>ANP</td>
<td>106 ± 3</td>
<td>349 ± 7</td>
<td>0.8 ± 0.4</td>
<td>1.4 ± 0.6</td>
<td>2.7 ± 0.3</td>
<td>10.381 ± 596</td>
</tr>
<tr>
<td>p</td>
<td>&lt;0.02</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>&lt;0.02</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td><strong>Group 5</strong></td>
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</tr>
<tr>
<td>Control</td>
<td>112 ± 2</td>
<td>362 ± 13</td>
<td>-0.1 ± 0.2</td>
<td>1.6 ± 0.7</td>
<td>3.1 ± 0.4</td>
<td>12.281 ± 640</td>
</tr>
<tr>
<td>Vehicle</td>
<td>115 ± 3</td>
<td>362 ± 13</td>
<td>-0.3 ± 0.3</td>
<td>1.7 ± 0.5</td>
<td>3.5 ± 0.4</td>
<td>12.508 ± 472</td>
</tr>
<tr>
<td>p</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
</tbody>
</table>

Means ± SEM; p values derived from paired t test analysis of control versus experimental periods within each group; NS, p > 0.05.

AP, mean systemic arterial pressure; HR, heart rate; +dP/dt, maximum rate of rise of left ventricular pressure; LVEDP, left ventricular end-diastolic pressure; +dP/dt, maximum rate of rise of left ventricular pressure.

While the majority of these investigations have been performed in anesthetized animals, quantitatively similar increases in GFR have been documented in conscious dogs,5 rats,7 and humans21 given ANP, as well as in isolated, perfused rat kidneys.22-24 In contrast, the effect of ANP on renal blood flow (RBF) has been variable. During administration of ANP to dogs and rats, RBF, estimated by a variety of techniques, has been shown to decrease,26-27 remain stable,28-29 or increase.20,29-34 In isolated, perfused rat kidneys, which constitute an extremely vasoconstricted preparation, a consistent decrease in perfusion flow rate has been observed,22-23 but an equally consistent increase in perfusion flow rate was found if the preparation was vasoconstricted with angiotensin II, norepinephrine, or vasopressin prior to ANP.22-24 In concert with the latter observation, the in situ auto-perfused (and therefore auto-vasoconstricted) rat kidney preparation, in which perfusion rate is held constant, has been observed to vasodilate following intraarterial bolus doses of atrial extract.35 Therefore, alterations in RBF induced by ANP might be determined, in part, by the baseline level of renal vasoconstriction in the various animal models studied.3

Despite this variability of RBF responses to ANP, whole kidney filtration fraction has consistently in-

**Table 4. Cardiovascular Studies (Groups 6 and 7)**

<table>
<thead>
<tr>
<th></th>
<th>AP (mm Hg)</th>
<th>HR (beats/min)</th>
<th>+dP/dt (mm Hg/sec)</th>
<th>CI (ml/min per kilogram)</th>
<th>TPRI (mm Hg/ml-min/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Group 6</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>105 ± 5</td>
<td>357 ± 9</td>
<td>6.923 ± 497</td>
<td>181 ± 7</td>
<td>0.57 ± 0.03</td>
</tr>
<tr>
<td>ANP</td>
<td>95 ± 5</td>
<td>351 ± 10</td>
<td>5.967 ± 685</td>
<td>160 ± 5</td>
<td>0.58 ± 0.02</td>
</tr>
<tr>
<td>p</td>
<td>&lt;0.05</td>
<td>NS</td>
<td>&lt;0.02</td>
<td>&lt;0.02</td>
<td>NS</td>
</tr>
<tr>
<td><strong>Group 7</strong></td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>101 ± 4</td>
<td>359 ± 7</td>
<td>7.384 ± 543</td>
<td>184 ± 12</td>
<td>0.56 ± 0.06</td>
</tr>
<tr>
<td>Vehicle</td>
<td>97 ± 4</td>
<td>358 ± 8</td>
<td>7.062 ± 473</td>
<td>174 ± 11</td>
<td>0.56 ± 0.06</td>
</tr>
<tr>
<td>p</td>
<td>NS</td>
<td>NS</td>
<td>&lt;0.05</td>
<td>&lt;0.005</td>
<td>NS</td>
</tr>
</tbody>
</table>

Means ± SEM; p values derived from paired t test analysis of control versus experimental periods within each group; NS, p > 0.05.

AP, mean systemic arterial pressure; HR, heart rate; +dP/dt, maximum rate of rise of left ventricular pressure; CI, cardiac index; TPRI, total peripheral resistance index.
increased both in vivo17,18,28 and in vitro,23-22 implying elevation of post-glomerular vascular resistance. In vitro studies in isolated glomeruli perfused at constant pressure have demonstrated significant increases in SNGFR, SNFF, and $P_{oc}$ without significant changes in $R_e$ or $Q_A$, thereby also suggesting efferent vasoconstriction as an important action of ANP.26 The present study is the first to directly assess the effects of ANP on glomerular hemodynamics in vivo and demonstrate that all of the primary determinants of glomerular ultrafiltration, only $\Delta P$ increased significantly and therefore accounted entirely for the ANP-induced elevation of SNGFR. The dramatic increase in $P_{oc}$ responsible for this elevation of $\Delta P$ resulted from afferent arteriolar vasodilation and concurrent efferent arteriolar vasoconstriction.

Given the large number of studies that have demonstrated vasorelaxant properties of ANP in isolated vascular segments24,37-38 and the presence of high affinity ANP binding sites in renal vasculature,39 a decrease in $R_e$ during infusion of ANP is not surprising, especially in the context of a concomitant reduction in renal perfusion pressure. Little, if any, of this fall in resistance appears to be directly related to a reduction in renal perfusion pressure since the decrease in $R_e$ of Group 1 animals was virtually identical to that of Group 2, in which perfusion pressure was not allowed to decline. Inhibition of renal sympathetic nerve activity by ANP via the activation of cardiac vagal afferents has been suggested.40,41 Blunting of the tubuloglomerular feedback response during ANP infusion42,43 also may contribute to the reduction in $R_A$. Furthermore, a direct vascular action of ANP has been suggested by in vitro studies of precontracted ring preparations from renal arcuate and interlobular arteries of the rat.44 However, despite the finding of specific ANP receptors in isolated rat renal glomeruli and in cultures of glomerular mesangial cells,45 localization of specific receptors to afferent arterioles has not been conclusively demonstrated, and preliminary studies of isolated rabbit afferent arterioles in vitro have failed to reveal a vasodilatory effect of arterial peptides.46

No evidence has been advanced to support the notion that ANP-induced renal vasodilation results from a direct interaction of arterial peptides with other vasoactive systems. In vitro studies of precontracted rabbit aortic and renal artery strips have demonstrated that arterial extract-induced relaxation is not inhibited by the addition of propranolol, atropine, or indomethacin,47 and studies of ring preparations from the renal arcuate and interlobular arteries of rats have revealed similar findings with synthetic ANP and pretreatment with propranolol, phentolamine, atropine, or ouabain.48 In the isolated, perfused rat kidney, no change in norepinephrine or dopamine excretion was observed with the addition of arterial extract to the perfusate,25 and injection of extract into perfused, hydrenephrotic rabbit kidneys in vitro produced a decrease in renal vascular resistance without an attendant release of prostaglandins.49 Similarly, no inhibition of ANP-induced vasodilation was observed in the auto-perfused rat kidney by pretreatment with indomethacin, phenoxybenzamine, propranolol, atropine, haloperidol, or the histamine antagonists, pyrilamine, and metiamide.50

Vasodilation in vivo may alter the distribution of intrarenal blood flow. Indeed, atrial extract administered as an intravenous bolus in anesthetized rats29 has been shown to shift blood flow from outer to inner renal cortex, as measured by microsphere techniques, and to increase renal papillary plasma flow, as estimated by accumulation of 125I-albumin, when administered either as an intravenous bolus dose29 or a continuous infusion.49 Although not directly addressed in the present study, the differential effects of ANP infusion on blood flow to the whole kidney and to superficial glomeruli of animals in Groups 1 and 2 (Table 2) are consistent with such a redistribution phenomenon. In both groups of animals, a significant increase in RBF was observed even though superficial GBF remained relatively stable, suggesting a preferential augmentation of blood flow to deeper cortical and/or medullary structures.

The failure of $R_e$ and SNFF to decline during ANP infusion in the present study contrasts with the known effects of other renal vasodilatory agents. Acetylcholine, bradykinin, papaverine, or secretin, when infused into the renal artery in dogs, resulted in substantial reductions in whole kidney filtration fraction.50-53 A similar decline in SNFF, associated with a reduction in both afferent and efferent arteriolar resistances, has been reported in hydropenic rats receiving acetylcholine, bradykinin, prostaglandin E$_1$ (PGE$_1$), or histamine.5,55

In the present study the increase in $R_e$ during ANP infusion was not the result of a reduction in renal perfusion pressure or an action of endogenous AII. A direct vasoconstrictive effect of ANP on the efferent arteriole is possible but seems unlikely in view of the vasorelaxant properties of the peptide on several other vascular segments in vitro44,37-38 and its failure to constrict isolated rabbit efferent arterioles.56 The secondary release of a more slowly acting renal vasoconstrictor has been suggested on the basis of a number of indirect observations: 1) a slower time-course of atrial extract-induced vasoconstriction in isolated, perfused rat kidneys compared with the prompt vasodilatory response in pre-constricted preparations;25 2) a transient ANP-induced elevation of RBF in intact rats and dogs, followed by a substantial reduction in RBF in some studies6,17,27; and 3) an increase in renal vascular resistance above control levels following ANP infusion in intact dogs.6

That extrarenal cardiovascular effects of atrial peptides may play an important role in the non-uniformity of the renal effects observed in the various animal preparations studied is suggested by the fact that cardiac output, measured by a variety of techniques, declined substantially during the administration of atrial extract or synthetic peptide in some studies, yet remained stable in others.26,30,31,33 A decrease in cardiac output has been attributed to a reduction in central venous27,28 and left atrial pressures,27 as well as a de-
pression of myocardial contractility. In the current study, in which ANP infusion was accompanied by hemoconcentration and a fall in LV EDP, a reduction in cardiac index was associated with a decrease in both LV EDP and +dP/dt and no change in heart rate. While the observed hemoconcentration and fall in LV EDP can be explained by a loss and/or redistribution of intravascular volume, the basis for the reduction in +dP/dt is less evident. As discussed previously, in vivo studies have suggested that atrial peptides may activate inhibitory cardiac vagal afferents. Although in vitro studies of rabbit papillary muscle have failed to demonstrate a direct effect of ANP on electrically-induced contractile force, high concentrations of atrial peptides have been observed to produce modest reductions in this parameter in isolated guinea pig atria and to substantially reduce myocardial contractility in isolated, perfused heart preparations from guinea pigs, dogs, and rats, by virtue of a vasoconstrictive effect on coronary arteries. That this coronary vasoconstriction may obtain in vivo is supported by microsphere studies in conscious SHR and WKY rats receiving bolus doses of atrial extract but not by a number of other investigations with synthetic ANP, in which coronary blood flow was measured by microspheres in conscious SHR's or by Doppler flow probes in anesthetized and conscious dogs.

In keeping with the interstudy variability observed for a number of hemodynamic parameters, total peripheral vascular resistance has been found to decrease, remain stable, or increase during the administration of atrial extract or synthetic ANP and has demonstrated no consistent correlation with simultaneous alterations in cardiac output in the various animal models studied. Not surprisingly, therefore, the relation between the effects of atrial peptides on the renal and extra-renal vasculatures in vivo also has been highly variable. Some investigators have found no in vivo evidence for a selective effect of ANP on the renal vasculature and have implicated a role for neural cardiovascular reflexes in counteracting the ANP-induced decline in renal vascular tone. However, a number of studies have shown a preferential renal vasodilatation in both conscious and anesthetized dogs and rats despite the suggestion that anesthesia and very high doses of atrial peptides may obscure this finding. The probes in anesthetized and conscious dogs. In the present study the authors are indebted to Dr. Thomas Maack for his generous provision of purified ANP during early phases of these studies.

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