Diuresis and Natriuresis During Continuous Dopamine-1 Receptor Stimulation

JOSEPH M. HUGHES, N. VIRGINIA RAGSDALE, ROBIN A. FELDER, ROBERT L. CHEVALIER, BERNARD KING, AND ROBERT M. CAREY

SUMMARY Stimulation of renal dopamine-1 (DA₁) receptors for 3 hours produces an increase in renal plasma flow and sustained natriuresis. The present study was designed to assess the response of renal hemodynamic and tubular function to long-term DA₁ receptor stimulation. Fenoldopam, a selective DA₁ receptor agonist, was infused intravenously for 24 hours in 10 normal male subjects in metabolic balance at 150 mEq sodium and 60 mEq potassium intake in a single-blind, vehicle-controlled protocol. During DA₁ receptor activation, urine flow rate and fractional excretion of sodium increased for the first 5 hours, 16.9 ± 0.9 ml/min compared with a vehicle control value of 12.4 ± 0.5 ml/min (p < 0.001) and 2.0 ± 0.1% compared with a vehicle control value of 1.1 ± 0.1% (p < 0.005), respectively. Urinary sodium excretion rose at 5 hours, 0.27 ± 0.02 mEq/min compared with a vehicle control value of 0.14 ± 0.01 mEq/min (p < 0.001), and remained elevated at 11 hours, 0.19 ± 0.02 mEq/min compared with 0.16 ± 0.02 mEq/min (p < 0.01). Renal plasma flow increased during fenoldopam at 5 hours, 505 ± 47 ml/min compared with a vehicle control value of 397 ± 25 ml/min (p < 0.01), and was sustained for 24 hours, 523 ± 31 ml/min compared with 432 ± 31 ml/min (p < 0.05). The distal sodium load increased and the percentage of distal sodium reabsorption decreased during fenoldopam. Glomerular filtration rate, blood pressure, heart rate, plasma aldosterone concentration, plasma renin activity, and fractional excretion of potassium were unchanged. Selective DA₁ receptor activation produced sustained 5-hour diuresis and 11-hour natriuresis without kaliuresis or a systemic hemodynamic effect. Calculation of the distal sodium load and the percentage of distal sodium reabsorption indicated both proximal and distal tubular DA₁ receptor activation. These findings support DA₁ receptor stimulation as a novel pharmacological approach to disorders of sodium and water retention. (Hypertension 11 [Suppl I]: I-69–I-74, 1988)

KEY WORDS • dopamine receptors • natriuresis • diuresis • fenoldopam mesylate

Subjects and Methods

Study Design

The protocol was approved by the Human Investigation Committee of the University of Virginia School of Medicine, and written informed consent was obtained from the study subjects. Ten normotensive men (ages 24–33 yrs) were studied in a single-blind, vehicle-controlled protocol. From Study Day 1 to 14, subjects ate an isocaloric-constant diet containing 150 mEq sodium and 60 mEq potassium daily. As outpatients on Days 1 to 5 and 8 to 12, blood pressure and heart rate were measured daily after subjects had been supine for 10 minutes and upright for 2 minutes; weight and temperature also were recorded.

The subjects were admitted to the Clinical Research Center on Study Day 5 and were supine until 1500 on Day 6. At 0600 on Day 6, an oral water load (tap water, 20 ml/kg) was ingested; urine samples then were collected every 30 minutes for the remainder of the study. After each urine collection, a volume of water equal to the volume of urine passed during the previous
30 minutes was ingested until 1500. In addition, at 0600 a priming dose of sterile inulin, 50 mg/kg (American Hospital Supply, McGraw Park, IL, USA) and p-aminohippurate (PAH), 8 mg/kg (Merck & Co., West Point, PA, USA) in 5% dextrose and water (D5W), was administered by intravenous bolus injection. Then a maintenance infusion of inulin and PAH, calculated on the basis of predicted glomerular filtration rate (GFR) and RPF, was initiated and continued at 0.3 ml/min until 1500. From 0830, four control urine measurements were obtained at 30-minute intervals; control blood samples were obtained at the midpoint of each urine collection. Urine specimens were analyzed for osmolality, sodium, potassium, inulin, and PAH. Plasma was analyzed for inulin, PAH, and osmolality, while serum sodium and potassium values were determined. Immediately after urine collection at 100, subjects were randomly assigned to receive vehicle at 20 ml/hr (vehicle control) or fenoldopam in D5W at 0.05 μg/kg/min for 24 hours. Urine for sodium and potassium was subsequently collected at 2-hour intervals until 2300 and then for the ensuing 7 hours until 0600 on Day 7. Blood pressure and heart rate were recorded every 15 minutes from 0600 to 1500, and then at 1-hour intervals until 2300 by a Dinamap automatic blood pressure and heart rate recorder (Model 845XT, Critikon, Inc., Tampa, FL, USA). Plasma aldosterone concentration (PAC) and plasma renin activity (PRA) were collected at 0945 and 1445.

Day 7 was identical to Day 6 except that at 1000 the infusion of D5W or fenoldopam was stopped. Inulin and PAH infusion, water loading, and blood and urine sampling were terminated at 1300. The PAC and PRA were sampled at 0815 and 1245.

The subjects were followed as outpatients until readmission on Day 12. The protocol was identical to that of Days 6 and 7, except that D5W or fenoldopam was infused depending on the randomization. No untoward effects occurred during the administration of fenoldopam.

**Analytical Methods**

Serum and urine sodium and potassium concentrations were measured by a Nova Biomedical analyzer (Model 1, Waltham, MA, USA). Plasma and urine osmolality were measured by a diagnostic osmometer (Model 302, Advanced Instruments, Needham Heights, MA, USA). Urine and plasma inulin concentrations were measured by the method of Heyrovsky. Urine and plasma PAH concentrations were determined by the method of Brun. Distal sodium exchange was calculated by the formula $C_{\text{in}} + C_{\text{out}}$, while the percentage of distal sodium reabsorption was determined by the equation $\left(\frac{C_{\text{in}} - C_{\text{out}}}{C_{\text{in}}}\right) \times 100$. Plasma renin activity was measured by the radioimmunoassay method of Sealey and Laragh. Plasma aldosterone concentration was determined by the method of Buhler et al.

**Statistical Analysis**

Paired t statistics were calculated to compare the differences in the sum of the values during fenoldopam and vehicle infusion for the control period (0830–1000 or −1.5–0 hours), experimental time period I (EI; 1000–1500 or 0–5 hours), experimental time period II (EII; 0830–1000 or 22.5 hours), and the postcontrol period (1000–1300 or 24–27 hours). One-way analysis of variance was determined to identify changeover time during fenoldopam and vehicle infusion. Data are reported as means ± SEM; p values below 0.05 are considered significant.

**Results**

Urine flow rate (Table 1, Figure 1) was unchanged during the control period and increased during EI to 16.9 ± 0.9 ml/min with fenoldopam infusion as compared to 12.4 ± 0.5 ml/min ($p < 0.001$) with vehicle infusion. By the beginning of EII, urine flow rate returned to the control level. By analysis of variance of

**Figure 1.** The response of urine flow rate (upper panel), renal plasma flow (middle panel), and urinary sodium excretion (lower panel) to vehicle (open circles) or fenoldopam infusion (solid circles).
urine flow rate, during fenoldopam infusion, the EI value was greater than control, EII, and postcontrol values (F = 9.28, p < 0.001), while during vehicle infusion no change occurred.

RPF (see Table 1, Figure 1) was unchanged in the control period and increased during fenoldopam infusion in EI as compared to the vehicle control (p < 0.01). The increase in RPF was sustained for 24 hours during fenoldopam infusion compared to the vehicle control value (p < 0.05) during EII. During postcontrol, RPF returned to the control level 90 minutes after cessation of fenoldopam. Analysis of variance of RPF demonstrated no change during either fenoldopam or vehicle infusion. The GFR (see Table 1) was unchanged during fenoldopam infusion as compared to the vehicle control (p < 0.001) with fenoldopam infusion compared to the vehicle control (p < 0.001) with fenoldopam, while during vehicle control no change occurred.

TABLE 1. Combined Results by Time Period

<table>
<thead>
<tr>
<th>Measurement</th>
<th>Control</th>
<th>EI</th>
<th>EII</th>
<th>Postcontrol</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urine flow</td>
<td>V</td>
<td>F</td>
<td>V</td>
<td>F</td>
</tr>
<tr>
<td>rate (ml/min)</td>
<td>12.7</td>
<td>±0.6</td>
<td>12.4</td>
<td>±0.5</td>
</tr>
<tr>
<td>RPF (ml/min)</td>
<td>421.8</td>
<td>±27.9</td>
<td>397.0</td>
<td>±25.3</td>
</tr>
<tr>
<td>GFR (ml/min)</td>
<td>102.9</td>
<td>±4.9</td>
<td>97.3</td>
<td>±4.5</td>
</tr>
<tr>
<td>FF</td>
<td>25.4</td>
<td>±1.9</td>
<td>25.3</td>
<td>±1.5</td>
</tr>
<tr>
<td>U_{Na}V (mEq/min)</td>
<td>0.12</td>
<td>±0.01</td>
<td>0.14</td>
<td>±0.01</td>
</tr>
<tr>
<td>FE_{Na} (%)</td>
<td>0.9</td>
<td>±0.1</td>
<td>1.1</td>
<td>±0.1</td>
</tr>
<tr>
<td>FE_{K} (%)</td>
<td>11.5</td>
<td>±1.1</td>
<td>10.9</td>
<td>±0.9</td>
</tr>
<tr>
<td>C_{osm} (mOsm/L)</td>
<td>3.0</td>
<td>±0.1</td>
<td>3.3</td>
<td>±0.1</td>
</tr>
<tr>
<td>C_{HCO3} (mOsm/L)</td>
<td>9.8</td>
<td>±0.6</td>
<td>10.6</td>
<td>±0.5</td>
</tr>
<tr>
<td>PAC (ng/dl)</td>
<td>8.0</td>
<td>±1.3</td>
<td>7.4</td>
<td>±1.3</td>
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<tr>
<td>PRA (pg/ml/hr)</td>
<td>0.8</td>
<td>±0.1</td>
<td>1.0</td>
<td>±0.2</td>
</tr>
<tr>
<td>Plasma osmolality (mOsm/kg)</td>
<td>281.0</td>
<td>±2.0</td>
<td>281.0</td>
<td>±2.0</td>
</tr>
<tr>
<td>Urine osmolality (mOsm/kg)</td>
<td>67.0</td>
<td>±4.0</td>
<td>67.0</td>
<td>±3.0</td>
</tr>
<tr>
<td>SBP (mm Hg)</td>
<td>115.0</td>
<td>±2.0</td>
<td>117.0</td>
<td>±3.0</td>
</tr>
<tr>
<td>DBP (mm Hg)</td>
<td>72.0</td>
<td>±2.0</td>
<td>72.0</td>
<td>±2.0</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>51.0</td>
<td>±1.0</td>
<td>52.0</td>
<td>±2.0</td>
</tr>
<tr>
<td>Distal Na load (mEq/min)</td>
<td>10.6</td>
<td>±1.0</td>
<td>11.6</td>
<td>±2.0</td>
</tr>
<tr>
<td>Distal Na re-absorption (%)</td>
<td>92.0</td>
<td>±0.7</td>
<td>91.0</td>
<td>±0.7</td>
</tr>
<tr>
<td>Distal Na⁺-K⁺ exchange (%)</td>
<td>26.5</td>
<td>±1.8</td>
<td>24.7</td>
<td>±2.3</td>
</tr>
<tr>
<td>Serum Na (mEq/L)</td>
<td>137.0</td>
<td>±1.0</td>
<td>137.0</td>
<td>±1.0</td>
</tr>
<tr>
<td>Serum K (mEq/L)</td>
<td>3.9</td>
<td>±0.1</td>
<td>4.0</td>
<td>±0.1</td>
</tr>
</tbody>
</table>

Values are mean ± SEM. EI = experimental time period 1; EII = experimental time period 2; V = vehicle infusion; F = fenoldopam infusion; RPF = renal plasma flow; GFR = glomerular filtration rate; FF = filtration fraction; U_{Na}V = urinary sodium excretion; FE_{Na} = fractional excretion of sodium; FE_{K} = fractional excretion of potassium; C_{osm} = osmolar clearance; C_{HCO3} = free water clearance; PAC = plasma aldosterone concentration; SBP = systolic blood pressure; DBP = diastolic blood pressure.

* p < 0.001, †p < 0.01, ‡p < 0.05, §p < 0.005, compared with vehicle infusion.
Fractional Excretion of Sodium

Distal Sodium Load

Percent Distal Sodium Reabsorption

Urinary Sodium Excretion

Figure 2. The response of fractional excretion of sodium (upper panel), distal sodium load (upper middle panel), and percentage of distal sodium reabsorption (lower middle panel) to vehicle (open circles) or fenoldopam infusion (solid circles). The lower panel shows urinary sodium excretion during specific experimental time periods.

Urinary sodium excretion ($U_{Na}V$; see Table 1, Figure 1) and fractional excretion of sodium ($FE_{Na}$; see Table 1; Figure 2) were stable during the control period. During fenoldopam infusion, $U_{Na}V$ rose in parallel with RPF in E1 ($p < 0.001$); however, in contrast to RPF, $U_{Na}V$ decreased to the control value in EII. By analysis of variance of $U_{Na}V$ excretion during fenoldopam infusion, the EI value was greater than control, EII, and postcontrol values ($F = 77.78, p < 0.0001$); during vehicle infusion, EI and postcontrol values were greater than control and EII values ($F = 3.29, p < 0.05$). The $U_{Na}V$ also was calculated for time periods when subjects were not water-loaded, 5 to 11 hours and 11 to 18 hours (see Figure 2). The $U_{Na}V$ excretion during fenoldopam infusion was elevated from 5 to 11 hours ($p < 0.01$) but was unchanged from 11 to 18 hours. The changes in $FE_{Na}$ were identical to those in $U_{Na}V$ and increased in EI in response to fenoldopam compared to the vehicle control value ($p < 0.005$) and returned to the control value during EII. By analysis of variance of $FE_{Na}$, during fenoldopam infusion the EI value was greater than control, EII, and postcontrol values ($F = 17.36, p < 0.001$), while with vehicle infusion no change occurred. Urine was collected during 21.5 hours of the 24 hours to determine cumulative sodium excretion; during fenoldopam infusion, sodium excretion was $192.4 \pm 13.7$ mEq compared to excretion during vehicle infusion of $138.2 \pm 9.5$ mEq ($p < 0.001$), for a net negative sodium balance of 54 mEq. The fractional excretion of potassium was unchanged throughout the protocol.

Distal sodium load (see Table 1, Figure 2) was constant during the control period and increased in EI during fenoldopam infusion as compared to the vehicle control value ($p < 0.005$), and returned to the control level for EII and the postcontrol period. The percentage of distal sodium reabsorption (see Table 1, Figure 2), after being stable in the control period, decreased during fenoldopam infusion in EI as compared to the vehicle value of $90.0 \pm 0.7\%$ ($p < 0.001$) and returned to the control level in EII. Analysis of variance demonstrated that neither distal sodium load nor percentage of distal sodium reabsorption changed during vehicle infusion; however, during fenoldopam infusion, distal sodium load increased, with EI greater than control, EII, and postcontrol values ($F = 5.55, p < 0.005$) and the percentage of distal sodium reabsorption decreased during EI compared with control, EII, and postcontrol values ($F = 10.32, p < 0.001$).

Systolic and diastolic blood pressures, heart rate, PAC, PRA, and plasma and urine osmolality (see Table 1) were unchanged during the protocol.

Discussion

Continuous DA$_A$, receptor stimulation for 24 hours produces sustained 5-hour diuresis and 11-hour natriuresis without concomitant kaliuresis or systemic hemodynamic effect. Moreover, the dissociation of RPF from urine flow rate and sodium excretion at 24 hours is of physiological interest.

Peripheral dopamine receptors may be classified as either DA$_A$, or DA$_B$. Activation of DA$_A$ receptors dilates renal, mesenteric, coronary, and cerebral vessels. In the kidney, DA$_A$ receptors are localized to the vasculature and proximal tubules. A physiological role for dopamine in sodium homeostasis has been postulated, since free dopamine is generated in the kidney. Moreover, urinary dopamine levels increase in response to a dietary change from low to high sodium. The importance of DA$_A$ receptors is demonstrated in dogs by blockade of dopamine-induced increases in RPF and sodium excretion by the selective DA$_A$ antagonist SCH 23390.
The selective DA	extsubscript{1} receptor agonist fenoldopam mesylate possesses peripheral and central nervous system dopaminergic activity and is four times more potent than dopamine as a renal vasodilator in anesthetized dogs.\textsuperscript{3} Fenoldopam does not cross the blood-brain barrier and has minimal \(\alpha\text{-}1\) or \(\beta\)-adrenergic receptor activity.\textsuperscript{13, 16} The selectivity of fenoldopam for DA	extsubscript{1} receptors has been demonstrated in experimental animals by reversal of fenoldopam-induced renal vasodilation and natriuresis by SCH 23390.\textsuperscript{3}

Bello-Ruess et al.\textsuperscript{17} and Pelayo et al.\textsuperscript{18} demonstrated a direct renal tubular effect for dopamine during in vitro studies at the straight portion of the proximal tubule in the rabbit and at a location beyond the proximal tubule in the rat. In water-loaded human subjects, calculation of distal sodium load approximates sodium that escapes reabsorption in the proximal tubule, while the percentage of distal sodium reabsorption approximates the fraction of sodium reabsorbed at the distal tubule.\textsuperscript{19, 20} The increase in distal sodium load supports activation of DA receptors in the proximal tubule, while the decrease in the percentage of distal sodium reabsorption supports distal tubular DA stimulation. Natriuresis occurs only after the capacity of distal segments to reabsorb an increased distal sodium load is exceeded. Thus, activation of DA receptors at both proximal and distal sites enhances the natriuretic effect by increasing distal sodium load and impairing distal reabsorption. Natriuresis generally is accompanied by kaliuresis, which is derived from increased sodium-potassium exchange in the distal tubule. In the present study, distal sodium-potassium exchange decreased in response to DA	extsubscript{1} receptor stimulation in EI and was unchanged in EI. The absence of kaliuresis demonstrates the importance of activation of distal tubular DA receptors in preventing sodium-potassium exchange. Therefore, the changes in distal sodium load and percentage of distal sodium reabsorption in the present study strongly suggest, but do not prove, activation of DA receptors at both proximal and distal tubular sites.

Renal hemodynamics and tubular sodium transport are the principal determinants of urinary sodium excretion. The rise in RPF, through changes in hydrostatic and oncotic pressures in the peritubular capillaries, could explain the initial 5-hour natriuresis; however, other mechanisms also may be active.\textsuperscript{21} Redistribution of intrarenal blood flow occurs during dopamine infusion and may contribute to increases in sodium excretion and urine flow rate.\textsuperscript{22} Changes in GFR also are associated with parallel alterations in sodium excretion, yet in our study GFR was unchanged. Aldosterone is the principal mediator of tubular sodium transport but was unchanged during the protocol. Thus, in addition to the rise in RPF, redistribution of intrarenal blood flow and stimulation of renal tubular DA	extsubscript{1} receptors may have contributed to the increase in sodium excretion.

In the present study, at 24 hours, sodium excretion and urine flow rate were dissociated from RPF. Several mechanisms may alter renal sodium excretion independent of RPF. Profound decreases in serum sodium of approximately 20 mEq/L during in vitro studies are associated with increases in sodium reabsorption, yet in our study the serum level was unchanged.\textsuperscript{23} A fall in blood pressure, by changes in renal interstitial and hydrostatic pressures, causes an increase in sodium reabsorption.\textsuperscript{24} During the protocol, systolic and diastolic blood pressures did not decrease. Total-body sodium depletion, through activation of the renin-angiotensin-aldosterone system, increases sodium reabsorption;\textsuperscript{25} however, PRA and PAC were not altered during the protocol. Studies with renal vasodilators have elucidated mechanisms for dissociation of RPF and sodium excretion. Infusion of acetylcholine increases RPF and sodium excretion. However, Hartupee et al.\textsuperscript{26} demonstrated in the decapsulated kidney that an increase in renal interstitial pressure, but not RPF, is essential for natriuresis. The gastrointestinal hormone secretin is a unique renal vasodilator, since RPF increases without natriuresis. Studies comparing renal responses to secretin and bradykinin have demonstrated the importance of increased papillary blood flow in natriuresis, since the bradykinin-induced rise in papillary blood flow was absent during secretin infusion.\textsuperscript{27} Finally, DA	extsubscript{1} receptors are located in both the renal vasculature and proximal tubules; tachyphylaxis for tubular, but not vascular, receptors could explain the dissociation. In summary, dissociation of RPF and sodium excretion at 24 hours may occur from a decrease in renal interstitial pressure, a fall in papillary blood flow, or a change in sensitivity of the tubular DA	extsubscript{1} receptors. However, the abrupt fall in sodium excretion after fenoldopam infusion was stopped indicates that RPF continues to contribute to sodium loss at 24 hours.

In conclusion, continuous DA	extsubscript{1} receptor stimulation produces sustained natriuresis and diuresis. The response is not associated with either systemic hemodynamic effects or kaliuresis. Moreover, the dissociation of RPF from sodium excretion supports the concept of stimulation of proximal and distal tubular DA receptors in dopamine-induced natriuresis. Finally, the findings support DA	extsubscript{1} receptor stimulation as a novel pharmacological approach to disorders of sodium and water balance.

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References
Diuresis and natriuresis during continuous dopamine-1 receptor stimulation.
J M Hughes, N V Ragsdale, R A Felder, R L Chevalier, B King and R M Carey

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