M-Atrial Natriuretic Peptide

A Novel Atrial Natriuretic Peptide Based Therapeutic in Experimental Angiotensin II Mediated Acute Hypertension

Paul M. McKie, Alessandro Cataliotti, Guido Boerrigter, Horng H. Chen, S. Jeson Sangaralingham, Fernando L. Martin, Tomoko Ichiki, John C. Burnett, Jr

Abstract—M-atrial natriuretic peptide (ANP; M-ANP) is a novel next generation 40 amino acid peptide based on ANP, which is highly resistant to enzymatic degradation and has greater and more sustained beneficial actions compared with ANP. The current study was designed to advance our understanding of the therapeutic potential of M-ANP in a canine model of acute angiotensin II–induced hypertension with elevated cardiac filling pressures and aldosterone activation. We compare M-ANP with vehicle and equimolar human B-type natriuretic peptide, which possesses the most potent in vivo actions of the native natriuretic peptides. M-ANP significantly lowered mean arterial pressure and systemic vascular resistance. Importantly, despite a reduction in blood pressure, renal function was enhanced with significant increases in renal blood flow, glomerular filtration rate, diuresis, and natriuresis after M-ANP infusion. Although angiotensin II induced an acute increase in pulmonary capillary wedge pressure, M-ANP significantly lowered pulmonary capillary wedge pressure, pulmonary artery pressure, and right atrial pressure. Further, M-ANP significantly suppressed angiotensin II–induced activation of aldosterone. These cardiovascular and renal enhancing actions of M-ANP were accompanied by significant increases in plasma and urinary cGMP, the second messenger molecule of the natriuretic peptide system. When compared with human B-type natriuretic peptide, M-ANP had comparable cardiovascular actions but resulted in a greater natriuretic effect. These results suggest that M-ANP, which is more potent than ANP in normal canines, has potent blood pressure lowering and renal enhancing properties and may, therefore, serve as an ANP based therapeutic for acute hypertension. (Hypertension. 2010;56:1152-1159.) ● Online Data Supplement

Key Words: hypertension ■ atrial natriuretic factor ■ natriuretic peptides ■ angiotensin ■ aldosterone ■ kidney

Continuing research has established the importance of the cardiac hormone atrial natriuretic peptide (ANP) in the regulation of blood pressure. This role in blood pressure regulation reflects the pluripotent cardiorenal actions of ANP, which functions via the guanylyl cyclase (GC) A receptor and the second messenger cGMP. The actions of ANP include natriuresis, vasodilatation, suppression of aldosterone, inhibition of myocardial hypertrophy, and suppression of cardiac fibrosis.1,2 Importantly, Newton-Cheh and coworkers3,4 have reported recently that genetic variants of the ANP gene, which result in increased circulating ANP, are associated with lower blood pressure and reduced risk of hypertension. The clinical phenotype of human genetic variants of the ANP gene, as well as mouse models of ANP gene disruption5 or overexpression6 together with the cardiorenal enhancing actions of ANP lay the foundation for ANP based therapeutics for cardiovascular diseases, such as hypertension.

We recently reported the design of a novel ANP based therapeutic peptide, M-ANP, which is a 40 amino acid (AA) peptide consisting of the 28 AAs of native ANP with a 12-AA C-terminus extension.7 M-ANP possesses markedly greater and more sustained mean arterial pressure (MAP) lowering, diuretic and natriuretic, glomerular filtration rate (GFR) enhancing, renin-angiotensin (Ang) aldosterone system suppressing, and cGMP activating properties compared with native ANP, making it highly attractive as a cardiovascular therapeutic agent. Dickey et al8 have shown that M-ANP, compared with ANP, is highly resistant to degradation by neprilysin, while retaining GC-A–activating properties.7,9–11

To date, the actions of intravenous M-ANP in a model of cardiovascular disease, such as acute hypertension, remain undefined. Importantly, a recent report from the Studying the Treatment of Acute Hypertension12 registry reported a high incidence of acute kidney injury associated with severe acute hypertension, which was associated with greater risk for heart failure (HF), cardiac arrest, and death. These and other studies underscore the need for acute hypertension therapeutic agents which possess renal enhancing in addition to blood pressure lowering properties.

The current study was designed to advance our understanding of the therapeutic potential of M-ANP in a model of acute hypertension. We hypothesized that M-ANP would have potent MAP lowering, cardiac unloading, renal enhancing, and aldosterone suppressing properties in acute hypertension. We further hypothesized that M-ANP, with its extended C-terminus, would have equivalent biological actions in vivo.

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1152
Table 1. Cardiovascular and Renal Hemodynamics: M-ANP Compared With Vehicle

<table>
<thead>
<tr>
<th>Variable</th>
<th>Peptide</th>
<th>Baseline</th>
<th>Ang II</th>
<th>M-ANP or Vehicle Infusion</th>
<th>After M-ANP or Vehicle Infusion</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>30 min</td>
<td>60 min</td>
</tr>
<tr>
<td>CO, L/min</td>
<td>Vehicle</td>
<td>3.5±0.2</td>
<td>2.7±0.2*</td>
<td>2.8±0.3*</td>
<td>2.8±0.2*</td>
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<tr>
<td></td>
<td>M-ANP</td>
<td>3.1±0.2</td>
<td>2.6±0.2</td>
<td>2.9±0.1</td>
<td>2.5±0.1*</td>
</tr>
<tr>
<td>SVR, mm Hg·L⁻¹·min⁻¹</td>
<td>Vehicle</td>
<td>38.1±2.5</td>
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<td>56.0±3.7*</td>
<td>56.1±4.0*</td>
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<td>M-ANP</td>
<td>43.3±2.6</td>
<td>57.4±2.8*</td>
<td>46.2±2.0†</td>
<td>51.6±1.9*</td>
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<tr>
<td>PAP, mm Hg‡</td>
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<td>13.6±0.5</td>
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<td>12.0±0.8‡</td>
<td>11.4±0.8-special</td>
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<td>RAP, mm Hg‡</td>
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<td>1.3±0.5</td>
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<td>M-ANP</td>
<td>1.5±0.3</td>
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<td>-0.4±0.3†</td>
<td>-0.8±0.4†</td>
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<tr>
<td>PVR, mm Hg·L⁻¹·min⁻¹</td>
<td>Vehicle</td>
<td>2.4±0.2</td>
<td>2.8±0.3</td>
<td>2.8±0.2</td>
<td>2.9±0.2*</td>
</tr>
<tr>
<td></td>
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<td>2.9±0.2</td>
<td>2.9±0.3</td>
<td>3.3±2.0</td>
<td>3.7±0.2†</td>
</tr>
<tr>
<td>RVR, mm Hg·L⁻¹·min⁻¹‡</td>
<td>Vehicle</td>
<td>729±51</td>
<td>995±118*</td>
<td>936±117*</td>
<td>880±95*†</td>
</tr>
<tr>
<td></td>
<td>M-ANP</td>
<td>716±61</td>
<td>955±69*</td>
<td>642±147†</td>
<td>729±154†</td>
</tr>
<tr>
<td>Heart rate, bpm</td>
<td>Vehicle</td>
<td>100±5</td>
<td>115±4*</td>
<td>124±4*</td>
<td>129±5*‡</td>
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<tr>
<td></td>
<td>M-ANP</td>
<td>93±4</td>
<td>112±8*</td>
<td>130±10†</td>
<td>132±7†</td>
</tr>
</tbody>
</table>

CO indicates cardiac output; SVR, systemic vascular resistance; PAP, pulmonary artery pressure; RAP, right atrial pressure; RVR, renal vascular resistance; PVR, pulmonary vascular resistance.

*P<0.05 vs BL, 1-way ANOVA with Bonferroni multiple-comparison test.
†P<0.05 vs ANG II, 1-way ANOVA with Bonferroni multiple-comparison test.
‡P<0.05 for main group effect of M-ANP vs vehicle, 2-way ANOVA.

Methods

M-Atrial and BNP

M-ANP and human BNP were synthesized by Phoenix Laboratories. Structure was confirmed by mass spectrometry, and high-performance liquid chromatography analysis confirmed purity to be >95%.

Study Protocol

We investigated the effects of M-ANP, vehicle (0.9% normal saline), and human BNP (n=6 for each group) in a canine model of Ang II–induced acute hypertension. Studies were performed in accordance with the Animal Welfare Act and with approval of the Mayo Clinic Institutional Animal Care and Use Committee.

Experimental procedures have been extensively described previously.13 and are available in the online Data Supplement (please see http://hyper.ahajournals.org). After completion of the procedural setup, a baseline (BL) clearance was performed. All clearances lasted 30 minutes and consisted of arterial blood sampling, hemodynamic measurements, and urine collection over 30 minutes. Immediately after the BL clearance, the saline infusion was replaced by continuous Ang II infusion (20 pmol·kg⁻¹·min⁻¹; 1 mL/min; Phoenix Pharmaceuticals), which was continued throughout the experimental protocol. After a 15-minute lead-in of the Ang II infusion, a 30-minute Ang II clearance was performed. After the Ang II infusion lead-in period (15 minutes) and Ang II clearance (30 minutes), M-ANP (30 pmol/kg per min), vehicle (0.9% normal saline), or BNP (30 pmol/kg per min) was infused at a rate of 1 mL/min. M-ANP, vehicle, or BNP was infused for a total of 45 minutes, which included a 15-minute lead-in period followed by a 30-minute clearance. M-ANP, vehicle, or BNP infusion was then discontinued, and four 30-minute clearances were performed that were 0 to 30, 30 to 60, 61 to 90, and 91 to 120 minutes after M-ANP, vehicle, or BNP infusion.

Neurohormonal and Electrolyte Analysis

Plasma and urine ANP were measured by radioimmunoassay (Phoenix Pharmaceuticals), as described previously.12 The cross-reactivity for M-ANP with the above assay is 100%. Plasma/urine cGMP15 (PerkinElmer), renin16 (Diasorin), Ang II17 (Phoenix Pharmaceuticals), and aldosterone18 (Siemens Healthcare Diagnostics) were determined by radioimmunoassay, as described previously. Inulin concentrations were measured using the anthrone method, as described previously.19 Electrolytes including lithium were measured by flame photometry (IL943, Instrumentation Laboratory). GFR was measured by inulin clearance. Proximal fractional reabsorption of sodium and distal fractional reabsorption of sodium were calculated using the lithium clearance technique, as described previously.20 Net renal generation of cGMP and plasma filtered cGMP were calculated as described previously.21

Statistical Analysis

Descriptive statistics are reported as mean±SE. Comparisons within a group were made by 1-way ANOVA for repeated measures followed by the Bonferroni multiple comparison posttest analysis when the global test was significant. Two-way ANOVA was used to compare the main group effects of M-ANP, vehicle, and BNP, followed by Bonferroni posttests. GraphPad Prism 5 (GraphPad Software) was used for the above calculations. Statistical significance was accepted as P<0.05.

Results

Systemic Hemodynamics

All animals received a continuous infusion of Ang II resulting in a significant increase in systemic vascular resistance (SVR; Table 1) and MAP (Figure 1A) compared with BL measurements. There was a significant increase in pulmonary capil-
rate significantly increased in response to Ang II in both pretreatment as Ang II did not affect these parameters. Heart pressure and right atrial pressure were independent of Ang II vehicle infusion. The actions of M-ANP on pulmonary artery significantly decreased after M-ANP versus no change with tion, both pulmonary artery pressure and right atrial pressure significant change in pulmonary vascular resistance. In addi-

There was a significant and sustained decrease in cardiac filling pressures as there was no significant change in PCWP after M-ANP infusion (Figure 1B) consistent with the increase in urinary sodium and water excretion, both proximal fractional reabsorption of sodium and distal fractional reabsorption of sodium significantly decreased from 88.9±1.1% and 99.5±0.1%, respectively, at BL to 58.9±4.5% and 72.1±2.3%, respectively, with M-ANP infusion. Both proximal fractional reabsorption of sodium and distal fractional reabsorption of sodium returned to BL 60 minutes after M-ANP infusion. There was no significant change in either proximal fractional reabsorption of sodium or distal fractional reabsorption of sodium during or after vehicle infusion (data not shown).

Neurohumoral Analysis
Neurohumoral parameters are shown in Table 2 and Figure 3. Plasma Ang II and aldosterone levels were similarly increased and renin levels similarly suppressed in the M-ANP and vehicle groups in response to Ang II infusion. Plasma and urinary ANP and cGMP were not significantly changed by Ang II infusion. M-ANP infusion resulted in significant increases in plasma and urinary ANP immunoreactivity. Consistent with increased plasma and urinary ANP immunoreactivity levels, both plasma cGMP (Figure 3A) and urinary cGMP excretion (Figure 3B) were markedly increased after M-ANP infusion. Both renal generation and plasma filtration of cGMP were significantly increased (Table 2). The significant increase in plasma and urinary cGMP after M-ANP infusion persisted for 60 minutes after infusion. There was no increase in plasma or urinary cGMP with vehicle infusion. Despite the continuous infusion of Ang II and significant increase in plasma Ang II levels, aldosterone levels were significantly suppressed after M-ANP infusion (Figure 3C).

M-ANP and BNP Comparison
Equimolar (30 pmol/kg per minute) human recombinant BNP (nesiritide) infusion was compared with M-ANP. Important hemodynamic, renal, and neurohumoral data are shown in Table 3. M-ANP infusion resulted in a significantly greater natriuretic effect when compared with human BNP. Other cardiorenal parameters, including MAP, PCWP, RBF, GFR, SVR, and aldosterone were not significantly different after M-ANP and BNP infusion.
Importantly, M-ANP has more sustained and greater cardio-renal actions,\textsuperscript{7} together with greater resistance to enzymatic degradation,\textsuperscript{8} compared with native ANP. In the current study of Ang II–induced hypertension with elevated cardiac filling pressures M-ANP lowered MAP. Despite a reduction in

**Table 2. Neurohumoral Data: M-ANP Compared With Vehicle**

<table>
<thead>
<tr>
<th>Variable</th>
<th>Peptide</th>
<th>Baseline</th>
<th>Ang II CL</th>
<th>M-ANP or Vehicle Infusion</th>
<th>After M-ANP or Vehicle Infusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ang II, pg/mL</td>
<td>Vehicle</td>
<td>4.0±0.8</td>
<td>70.6±8.2*</td>
<td>66.7±8.1*</td>
<td>80.5±13.3*</td>
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<td>M-ANP</td>
<td>7.1±2.0</td>
<td>65.0±12.5*</td>
<td>82.2±15.8*</td>
<td>88.6±15.6*</td>
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<tr>
<td>Plasma renin, ng/mL per h</td>
<td>Vehicle</td>
<td>1.8±0.5</td>
<td>0.7±0.2</td>
<td>0.6±0.1</td>
<td>0.3±0.1*</td>
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<tr>
<td></td>
<td>M-ANP</td>
<td>2.2±0.3</td>
<td>1.1±0.1</td>
<td>0.8±0.1</td>
<td>0.7±0.1</td>
</tr>
<tr>
<td>Plasma ANP, pg/mL$§$</td>
<td>Vehicle</td>
<td>53.7</td>
<td>66±5</td>
<td>69±10</td>
<td>83±10</td>
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<td>M-ANP</td>
<td>30.2</td>
<td>48±1</td>
<td>1735±185*</td>
<td>426±66*</td>
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<td>Urine ANP, pg/min$§$</td>
<td>Vehicle</td>
<td>19±2</td>
<td>24±4</td>
<td>26.4</td>
<td>31±6*</td>
</tr>
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<td></td>
<td>M-ANP</td>
<td>14.2</td>
<td>17±2</td>
<td>134±28*†</td>
<td>122±12*†</td>
</tr>
<tr>
<td>Renal cGMP generation, pmol/min$§$</td>
<td>Vehicle</td>
<td>661±120</td>
<td>805±215</td>
<td>781±132</td>
<td>779±78</td>
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<td>M-ANP</td>
<td>486±62</td>
<td>550±114</td>
<td>5827±1246*†</td>
<td>8303±1769*†</td>
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<tr>
<td>Filtered cGMP, pmol/min$§$</td>
<td>Vehicle</td>
<td>303±77</td>
<td>322±58</td>
<td>314±65</td>
<td>393±81</td>
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<td>M-ANP</td>
<td>365±53</td>
<td>461±86</td>
<td>3304±187*†</td>
<td>1995±330*†</td>
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</tbody>
</table>

Ang II CL indicates Ang II clearance.
*P<0.05 vs BL, 1-way ANOVA with Bonferroni multiple-comparison test.
†P<0.05 vs Ang II, 1-way ANOVA with Bonferroni multiple-comparison test.
‡P<0.05 for main group effect of M-ANP vs vehicle, 2-way ANOVA.
§Data show ANP immunoreactivity.

**Figure 2.** RBF (A), GFR (B), urinary water excretion (UV; C), and urinary sodium excretion (UNaV; D) at BL, during Ang II clearance (Ang II CL); during M-ANP or vehicle infusion, and 30, 60, 90, and 120 minutes after M-ANP or vehicle infusion. *P<0.05 vs BL, †P<0.05 vs Ang II, 1-way ANOVA with Bonferroni multiple-comparison test. GFR, UV, and UNaV (2-way ANOVA). ‡P<0.05 for M-ANP vs vehicle at a specific time point, 2-way ANOVA with Bonferroni posttests.
M-ANP significantly lowered PCWP, MAP, renal function was enhanced with significant increases in RBF, GFR, natriuresis, and diuresis after M-ANP infusion. Although Ang II induced an acute increase in PCWP secondary to systemic vasoconstriction, M-ANP significantly lowered PCWP, pulmonary artery pressure, and right atrial pressure, as well as SVR. Furthermore, M-ANP significantly suppressed Ang II–induced activation of aldosterone. When compared with recombinant human BNP, M-ANP had comparable cardiovascular actions to human BNP but resulted in a greater natriuretic effect. These results suggest that M-ANP, which is more potent than ANP in normal canines,7 has blood pressure lowering and renal enhancing properties and may, therefore, serve as an ANP based therapeutic for acute hypertension.

The natriuretic peptides, specifically ANP, are increasingly recognized to play a fundamental role in blood pressure regulation and cardio renal homeostasis.3–6,23 These studies support the development of an ANP based therapeutic for hypertension. Indeed, ANP (carperitide) is already approved as an intravenous agent for HF treatment in Japan and was shown to have beneficial renoprotective actions after bypass surgery, reduced infarct size, and HF after myocardial infarction, and prevented contrast-induced nephropathy.24–26

M-ANP is novel ANP based peptide with greater diuretic, natriuretic, GFR enhancing, aldosterone-suppressing, and MAP-lowering properties when compared with native ANP in normal canines.7 When we compare the actions of M-ANP in the current study to the actions of ANP in historical studies of Ang II–induced acute hypertension, there are important differences.27 Specifically, GFR was not enhanced by ANP, whereas there was a significant increase in GFR by M-ANP in the current study. ANP did not suppress aldosterone activation, whereas M-ANP significantly suppresses aldosterone in the current study. Furthermore, the natriuretic effect of M-ANP was ~100% greater in the current study than with native ANP in the previous study. Recent studies8 have shown that M-ANP, compared with ANP, is highly more resistant to degradation by neprilysin, and this may serve as the principal mechanism for its enhanced in vivo cardio renal actions compared with ANP, because there was no change in affinity for its GC-A receptor.7–9–11

The mechanism of the blood pressure lowering properties observed in the current study of M-ANP was multifactorial. First, there was a significant reduction in SVR, and it is known that M-ANP activates GC-A,8 which mediates cGMP related vasodilatation.1 A second mechanism was the significant natriuretic and diuretic effects of M-ANP. This is an important property of M-ANP, which goes beyond conventional vasodilators, which lack diuretic and natriuretic actions. Also, M-ANP suppressed Ang II–induced aldosterone production from the zona glomerulosa where there is an abundance of GC-A.28,29 Finally, it is possible that M-ANP directly inhibits the vasoconstrictive effects of Ang II on the blood vessel wall; however, this is speculative and would require additional studies to confirm.

It is notable that, although there was significant reduction in MAP, both RBF and GFR significantly increased after M-ANP. These renal hemodynamic actions most likely were related to significant increases in plasma and urinary cGMP and net renal cGMP generation with subsequent activation of GC-A, which is widely distributed in the kidney. The increases in GFR and RBF are also consistent with M-ANP/GC-A mediated cGMP generation8 and inhibition of Ang II related renal arterial and afferent arteriolar vasoconstric-
The renin-angiotensin-aldosterone system in hypertension is well established. Indeed, this model of Ang II mediated acute hypertension has been used extensively in the past and in key studies in the development of antihypertensive agents, such as β-blockers, calcium channel blockers, and native natriuretic peptides.27,40–43

The clinical relevance of the current study is underscored by the recent report from the Studying the Treatment of Acute Hypertension registry, which showed that acute and chronic kidney diseases are common comorbidities in subjects with acute hypertension and are associated with greater risk for HF, cardiac arrest, and death.12 These findings underscore the need for therapeutics aimed at preventing or mitigating the severity of kidney injury to reduce subsequent increased morbidity and mortality. As recently emphasized by de Bold,44 the central importance of ANP as a therapeutic agent is its diverse pharmacological properties. Moreover, ANP exerts beneficial cardiorenal effects without activating the renin-angiotensin-aldosterone system.27,40–43

The therapeutic blood pressure lowering and diuretic/natriuretic agent that concomitantly enhances renal function is a highly attractive characteristic when compared with other conventional diuretic agents and vasodilators, which tend to reduce renal perfusion and activate the renin-angiotensin-aldosterone system.27,30,31

In the current study, we compared M-ANP with recombinant human BNP testing the hypothesis that M-ANP would have comparable biological actions to BNP. Importantly, in normal human subjects, equimolar BNP is 2 to 3 times more potent than ANP, which may be secondary to the much greater metabolic clearance of ANP compared with BNP.35 Furthermore, in experimental HF, BNP has more potent cardiorenal enhancing actions compared with ANP at equimolar doses.13 It should be noted that, whereas human and canine ANP are identical, there is ≈20% AA heterogeneity between human and canine BNP.36,37 Extensive previous studies have demonstrated the ability of human BNP to have potent cardiorenal and humoral actions in the dog.38 Indeed, human BNP activates cGMP generation in isolated canine glomeruli.39 Although our results suggest comparable actions between M-ANP and human BNP, our conclusions need to be cautious because of differences in human and canine BNP.

We used a model of Ang II–induced acute hypertension in the current study to investigate the cardiovascular, neurohumoral, and renal effects of M-ANP and compare with human BNP. This model can be viewed as a limitation, but because it has been widely used in the literature, it allows for reproducibility and has clinical relevance as the role of the renin-angiotensin-aldosterone system in hypertension is well

### Table 3. M-ANP and BNP

<table>
<thead>
<tr>
<th>Variable</th>
<th>Peptide</th>
<th>Baseline</th>
<th>Ang II CL</th>
<th>M-ANP or BNP Infusion</th>
<th>After M-ANP or BNP Infusion</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>30 min</td>
<td>60 min</td>
</tr>
<tr>
<td>MAP, mm Hg</td>
<td>M-ANP</td>
<td>131 ± 6</td>
<td>148 ± 6*†</td>
<td>133 ± 6†</td>
<td>129 ± 6†</td>
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<tr>
<td></td>
<td>BNP</td>
<td>125 ± 4</td>
<td>150 ± 4*</td>
<td>130 ± 3†</td>
<td>136 ± 4†</td>
</tr>
<tr>
<td>PCWP, mm Hg</td>
<td>M-ANP</td>
<td>5.4 ± 0.8</td>
<td>7.4 ± 0.8*†</td>
<td>2.6 ± 0.5†</td>
<td>2.1 ± 0.5†</td>
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<tr>
<td></td>
<td>BNP</td>
<td>5.2 ± 0.6</td>
<td>7.1 ± 0.8*</td>
<td>2.4 ± 0.6*†</td>
<td>2.1 ± 0.5†</td>
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<tr>
<td>SVR, mm Hg · L⁻¹ · min⁻¹</td>
<td>M-ANP</td>
<td>43.3 ± 2.6</td>
<td>57.4 ± 2.8*</td>
<td>46.2 ± 2.0†</td>
<td>51.6 ± 1.9*</td>
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<td>BNP</td>
<td>38.9 ± 2.5</td>
<td>57.1 ± 4.6*</td>
<td>45.2 ± 3.7†</td>
<td>53.6 ± 4.3*</td>
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<tr>
<td>RBF, mL/min</td>
<td>M-ANP</td>
<td>194 ± 9</td>
<td>155 ± 6*</td>
<td>267 ± 14†</td>
<td>226 ± 12†</td>
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<tr>
<td></td>
<td>BNP</td>
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<td>163 ± 14*</td>
<td>250 ± 10†</td>
<td>219 ± 12†</td>
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<tr>
<td>GFR, mL/min</td>
<td>M-ANP</td>
<td>30 ± 4</td>
<td>32 ± 7</td>
<td>57 ± 4†</td>
<td>41 ± 6</td>
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<tr>
<td></td>
<td>BNP</td>
<td>32 ± 2</td>
<td>40 ± 3</td>
<td>47 ± 2*</td>
<td>41 ± 5</td>
</tr>
<tr>
<td>UNaV, µEq/min†</td>
<td>M-ANP</td>
<td>3 ± 1</td>
<td>34 ± 20</td>
<td>891 ± 62†‡‡‡</td>
<td>395 ± 106†‡</td>
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<td></td>
<td>BNP</td>
<td>6 ± 2</td>
<td>16 ± 6</td>
<td>679 ± 33†</td>
<td>288 ± 64†</td>
</tr>
<tr>
<td>Aldosterone, ng/dL</td>
<td>M-ANP</td>
<td>4.8 ± 1.7</td>
<td>21.0 ± 1.8*</td>
<td>9.9 ± 1.5†</td>
<td>10.1 ± 2.0†</td>
</tr>
<tr>
<td></td>
<td>BNP</td>
<td>3.2 ± 0.4</td>
<td>13.0 ± 2.7*</td>
<td>8.7 ± 2.0</td>
<td>12.7 ± 2.3*</td>
</tr>
<tr>
<td>Plasma cGMP, pmol/mL</td>
<td>M-ANP</td>
<td>12.1 ± 0.7</td>
<td>14.6 ± 0.5</td>
<td>58.6 ± 3.1†</td>
<td>48.0 ± 3.5†</td>
</tr>
<tr>
<td></td>
<td>BNP</td>
<td>10.0 ± 2.9</td>
<td>11.7 ± 9.2</td>
<td>56.7 ± 8.2†</td>
<td>43.5 ± 5.1†</td>
</tr>
</tbody>
</table>

UnNaV indicates urinary sodium excretion; Ang II CL, Ang II clearance.

*P < 0.05 vs BL, 1-way ANOVA with Bonferroni multiple-comparison test.
†P < 0.05 vs Ang II, 1-way ANOVA with Bonferroni multiple-comparison test.
‡P < 0.05 for main group effect of M-ANP vs BNP at a specific time point, 2-way ANOVA with Bonferroni posttests.
§P < 0.05 for M-ANP vs BNP at a specific time point, 2-way ANOVA with Bonferroni posttests.
pared with ANP. In the current study, we report that M-ANP lowers blood pressure in a model of acute Ang II–induced hypertension and reduces cardiac filling pressures. Importantly, the cardiovascular properties of this advanced ANP were also associated with an improvement in renal function with significant increases in RBF, GFR, natriuresis, and diuresis together with suppression of aldosterone activation. These characteristics make M-ANP an attractive candidate for the treatment of acute hypertension warranting further studies.

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Disclosures
The Mayo Foundation holds a patent for M-ANP.

References


35. Pidgeon GB, Richards AM, Nicholls MG, Espiner EA, Yandle TG, Frampton C. Differing metabolism and bioactivity of atrial and brain


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A NOVEL ATRIAL NATRIURETIC PEPTIDE BASED THERAPEUTIC IN EXPERIMENTAL ANGIOTENSIN II MEDIATED ACUTE HYPERTENSION

Paul M McKie MD, Alessandro Cataliotti MD PhD, Guido Boerrigter MD, Horng H Chen MD, S. Jeson Sangaralingham PhD, Fernando L Martin, MD, Tomoko Ichiki MD PhD, and John C Burnett Jr MD

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Online Methods Supplement
Studies were performed in normal male mongrel dogs (21–27 kg) on a fixed sodium diet (58 mEq/day, Hill's ID, Topeka, KS) with free access to drinking water. The night before experimentation, dogs were fasted and given 300 mg of lithium carbonate for assessment of renal tubular function. Dogs were anesthetized with pentobarbital sodium (15 mg/kg intravenous), intubated, and mechanically ventilated with supplemental oxygen (Harvard respirator, Amersham, MA) at 12 cycles/min. A balloon-tipped thermodilution catheter was advanced to the pulmonary artery via the external jugular vein for measurement of cardiac filling pressures [pulmonary capillary wedge pressure (PCWP), pulmonary artery pressure (PAP), and right atrial pressure (RAP)] and cardiac output (CO). The femoral artery was cannulated for MAP monitoring and blood sampling. The femoral vein was cannulated for inulin, ANG II, and M-ANP or normal saline or BNP infusion. The left kidney was exposed and the ureter was cannulated for urine sampling. An electromagnetic renal artery flow probe (Carolina Medical Electronics, East Bend, NC) was used to measure renal blood flow (RBF). Systemic vascular resistance (SVR), pulmonary vascular resistance (PVR), and renal vascular resistance (RVR) were calculated as (MAP-RAP)/CO, (PAP-PCWP)/CO, and (MAP-RAP)/RBF respectively. Supplemental non-hypotensive doses of pentobarbital were administered as needed. After completion of the above procedural set up a weight-adjusted inulin bolus was administered followed immediately by continuous inulin infusion to maintain plasma inulin levels between 40 and 60 mg/dL for determination of GFR. The experiment began after a 60 minute equilibration period.