Effects of Brain Natriuretic Peptide and C-Type Natriuretic Peptide Infusion on Urine Flow and Jejunal Absorption in Anesthetized Dogs

Hironobu Morita, Masanobu Hagiike, Takao Horiba, Keisuke Miyake, Hideo Ohyama, Hideo Yamanouchi, Hiroshi Hosomi, Kenji Kangawa,* Naoto Minamino,** and Hisayuki Matsuo**

Department of Physiology, Kagawa Medical School, Kagawa, 761-07 Japan
*Department of Biochemistry, Miyazaki Medical College, Miyazaki, 886-16 Japan
**National Cardiovascular Center Research Institute, Suita, 565 Japan

Summary  Effects of brain natriuretic peptide (BNP) or C-type natriuretic peptide (CNP) on urinary excretion and jejunal absorption of fluid and electrolytes were examined in anesthetized dogs. Intravenous infusion of BNP increased urinary fluid and electrolyte excretion and decreased jejunal fluid and electrolyte absorption. CNP had a similar effect on jejunal absorption as BNP. However, CNP had no significant effect on renal fluid or electrolyte excretion. These results indicate that: 1) BNP is a powerful natriuretic peptide comparable to ANP and; 2) CNP may also contribute to the regulation of body fluid homeostasis by way of inhibiting net jejunal fluid and electrolyte absorption.

Key words : BNP, CNP, jejunal NaCl absorption.

In 1988 Sudoh et al. [1] found a novel peptide in a porcine brain. They designated the peptide “brain natriuretic peptide (BNP).” This peptide has 26 amino acid residues and is very similar in structure and pharmacological effect of atrial natriuretic peptide (ANP). More recently, Sudoh et al. [2] found a new peptide of 22 amino acid residues, whose sequences have a remarkable similarity to ANP and BNP. This peptide was designated “C-type natriuretic peptide (CNP).” With the identification of CNP, the natriuretic peptide family is shown to comprise three different peptides. However, the physiological role or pharmacological effect of BNP and CNP are not well known. Therefore, the purpose of this study was to examine effects of the intravenous infusion of BNP or CNP on urinary excretion and intestinal absorption of electrolytes.

All experiments were conducted in 16 mongrel adult dogs weighing 7–10 kg.

Received on July 5, 1991; Accepted on January 10, 1992
The dogs were fed on commercial dog food at 30 g/kg per day (Oriental Yeast Co. Ltd., type DS, Na 0.47 g/100 g) and water ad libitum. Dogs were deprived of food for 24 h prior to the experiments. Water remained available throughout the food deprivation period. The dogs were anesthetized with pentobarbital sodium (30 mg/kg, i.v.). Arterial and venous catheters were inserted into the abdominal aorta and inferior vena cava for pressure measurement and administration of BNP or CNP. Through the central laparotomy, the jejunal loop, which was 30 cm long and started from 10 cm distal to the ligament of Treitz, was made. The loop was washed with warm saline and intubated in both ends of the loop. The ureters were cannulated bilaterally. The intestine was returned to the abdominal cavity. The tubes and catheters were exteriorized, and the incision was closed.

To examine the jejunal electrolyte absorption, the test solution (30 ml, 37°C) was injected into the jejunal loop from the proximal tube and allowed to remain there for 15 min, then collected from the distal tube. The test solution had the following composition (mEq/l): Na, 130; K, 4; Ca, 3; Cl, 109; CH₃COO, 28; glucose, 50 g/l; and pH, 4.0–6.5. Na, K, and Cl concentrations of the injected and collected fluid were measured by flame photometer and Cl counter (Hitachi, No. 750, Tokyo). The volume of the collected fluid was also measured. Fluid and electrolyte net absorption were calculated as the difference between the absolute values of the injected solution and the absolute values of the collected solution. Positive numbers represented net absorption and negative numbers expressed net secretion.

A 30–60 min equilibration period was observed before the experiment. The intravenous infusion of physiological saline (0.02 ml/(kg·min)) was initiated 10 min before pouring of the test solution into the jejunal loop and lasted for 25 min. The test solution was allowed to remain in the jejunal loop for 15 min. During that 15 min period, urine was collected by gravity drainage from the previously inserted ureter catheters. After a 30 min equilibration period, synthetic porcine BNP [1] (n = 8) or CNP [2] (n = 8) was infused intravenously at a rate of 97 pmol/(kg·min). BNP and CNP were dissolved into the physiological saline, and infused volume was 0.02 ml/(kg·min). Ten minutes after the start of infusion, urinary excretion and jejunal absorption was examined for 15 min. Arterial pressure was measured by connecting the previously implanted catheter to a Statham P23ID transducer.

All values presented here are reported as mean ± SE. For statistical analysis, paired t-test was used. A p < 0.05 was taken as the criterion for the significant difference.

Mean arterial pressure tended to decrease during the infusion of BNP (from 135 ± 10 to 121 ± 8 mmHg) or CNP (from 135 ± 10 to 130 ± 11 mmHg), but the changes were not statistically significant (Table 1). Figure 1 shows effects of BNP or CNP on urinary fluid and electrolyte excretion during a 15 min clearance period. BNP significantly increased urine volume, urinary Na, Cl, and K excretion, whereas CNP had no significant effect on urinary fluid and electrolyte excretion.
Table 1. Effects of BNP and CNP on mean arterial pressure and heart rate.

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>At the end of infusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean arterial pressure (mmHg)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>BNP (n = 8)</td>
<td>135 ± 10</td>
<td>121 ± 8</td>
</tr>
<tr>
<td>CNP (n = 8)</td>
<td>135 ± 10</td>
<td>130 ± 11</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>BNP (n = 8)</td>
<td>182 ± 15</td>
<td>182 ± 14</td>
</tr>
<tr>
<td>CNP (n = 8)</td>
<td>171 ± 12</td>
<td>177 ± 13</td>
</tr>
</tbody>
</table>

![Graphs showing effects of BNP and CNP](image)

Fig. 1. Effects of BNP and CNP on urine volume, urinary Na, Cl, and K excretion (n = 8). Urine was collected for 15 min. Data are shown by mean ± SE. *p < 0.05, significantly different from control.

Figure 2 shows the effects of BNP or CNP on jejunal fluid and electrolyte absorption. The jejunal effects of BNP and CNP were very similar. Both BNP and CNP significantly depressed jejunal net fluid, Na, and Cl absorption.

The amino acid sequences of BNP and CNP are very similar to those of ANP, suggesting that those peptides have similar pharmacological effects to ANP, i.e., natriuretic effect. In fact, BNP has potent natriuretic effect comparable to ANP.
Fig. 2. Effects of BNP and CNP on jejunal net fluid, Na, Cl, and K absorption (n=8). Data are shown mean±SE. *p<0.05, significantly different from control.

[1]. However, diuretic and natriuretic activities of CNP are very weak. In the present study, the same amount of CNP as BNP (97 pmol/(kg·min)) did not elicit diuresis or natriuresis. Concerning this, it is interesting to note that BNP increases cyclic GMP in the kidney epithelial cell line [3], but CNP fails to increase cyclic GMP in rat renal glomeruli [4]. These results suggest that CNP may not have renal effect in regulating body fluid homeostasis.

Recently, Matsushita et al. [5] demonstrated that ANP had an important role in regulating body fluid homeostasis by controlling not only urinary excretion but also intestinal absorption, i.e., ANP depressed jejunal Na, Cl, and fluid absorption. In fact, the intestine is the important organ for controlling body fluid homeostasis. A decrease in arterial pressure and/or left atrial pressure elicits an increase in intestinal absorption of electrolytes and fluid, then restores pressures [6, 7]. These findings indicate that the small intestine is involved in the feedback control of extracellular fluid volume. In the present study, CNP did not increase urinary excretion but decreased intestinal NaCl absorption, and BNP increased urinary NaCl excretion and decreased intestinal NaCl absorption. Although BNP and
CNP were originally identified in the porcine brain, the presence of BNP-like immunoreactivity has been demonstrated in plasma [8] and small intestine [9], and the presence of CNP-like immunoreactivity has been demonstrated in the lower part of gastrointestinal tract [10]. These data suggest the possibility that BNP and CNP act for controlling body fluid homeostasis as neuromodulator or humoral factor.

In conclusion, BNP exerts pharmacological renal and intestinal effects similar to ANP. Although CNP has no or little renal effect, CNP exerts intestinal effect similar to ANP and BNP. Thus, CNP may also participate in regulating homeostatic balance of body fluid by selectively controlling the uptake from the intestine.

Dr. Morita was supported by a Grant-in-Aid for Encouragement of Young Scientists of Japan (No. 03770043). This study was supported in part by Research Grant for Cardiovascular Diseases 3A-1 from the Ministry of Health and Welfare, Japan.

REFERENCES