Effect of cerebroventricular perfusion of bombesin on gastrointestinal myoelectric activity

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Hashmonai, M., and J. H. Szurszewski. Effect of cerebroventricular perfusion of bombesin (BBS) on gastrointestinal myoelectric activity. Am. J. Physiol. 274 (Gastrointest. Liver Physiol. 37): G677–G686, 1998.—The effect of intracerebroventricular (ICV) perfusion of bombesin (BBS) on the interdigestive migrating myoelectric complex (MMC) activity was examined in conscious dogs with electrodes implanted on the stomach and small intestine. Cannulas and a catheter were chronically positioned in the lateral and fourth cerebral ventricles, respectively. ICV perfusion of BBS, which failed to increase plasma BBS levels, replaced phase I activity in the stomach and duodenum by intense irregular spike activity and decreased the occurrence rate of MMCs, whereas intravenous infusion of BBS evoked phase II-like activity, mainly in the jejunum and ileum, and suppressed phase III activity. These data suggest that the effect of ICV administration of BBS was mediated by direct activation of central brain structures. During ICV perfusion of BBS, cycling in plasma levels of motilin persisted even when phase III activity was absent and plasma levels of epinephrine rose significantly. Epinephrine infusion, however, did not affect myoelectric gastrointestinal activity except for prolonging phase II. Thus it is unlikely that the central action of BBS is exerted by motilin or epinephrine.

epinephrine; intravenous infusion of bombesin

MATERIALS AND METHODS

Fourteen healthy female dogs (9.5–15.0 kg) were used in this study. The results described were obtained from four of these dogs. Premature occlusion of the ICV drainage system prevented completion of a full set of experiments in the other 10 dogs. These animals and the partial results of the experiments performed on them are therefore excluded from this report. The Mayo Institutional Animal Care and Use Committee approved the use of the animals and the experimental procedures.

Surgical procedure. All operations were performed under general anesthesia induced with Brevital (12.5 mg/kg iv). An endotracheal tube was inserted for a free airway, and anesthesia was maintained by assisted respiration with halothane (Halo-Carbon Laboratory) in oxygen supplied by a mechanical respirator. A prophylactic dose of 600,000 U of Flo-cillin (Bristol Laboratory, Syracuse, NY) was given at the beginning of each operation.

In the first operation, through a midline laparotomy, nine Ag-AgCl electrodes were sutured to the stomach and small intestine. Two electrodes were sutured to the distal antrum: one 3 cm and the other 1 cm orad to the pylorus. Two electrodes were sutured to the duodenum: one between the upper and middle thirds and the other between the middle and bottom thirds. The other five electrodes were sutured to the remaining small bowel at intervals of one-sixth the distance between the ligament of Treitz and the ileocecal junction. A stainless steel cannula containing the socket connector was positioned in the right anterior abdominal wall and anchored in place by 2-0 stainless sutures.

One week later, by a stereotaxic technique previously described (18), cannulas were inserted into the lateral ventricles of the brain and a polyethylene (PE-100) catheter with a Silastic tube was placed into the fourth ventricle to be used for drainage during perfusion experiments.

Experimental procedures. Experiments were carried out 1 wk after the dogs recovered from the second operation. None of the dogs showed any sign of discomfort or neurological damage. Dogs received one daily meal, at a regular hour, which consisted of 822 g of Alpo Beef Chunks (Allen Products, Allentown, PA) and received water ad libitum.

Recordings of myoelectric activity were made at the same time of the day on nonconsecutive days. The dogs were fasted for 20 h before each recording session. Recordings were made on an eight-channel rectilinear penwriting recorder (Gould 2800S, Gould, Cleveland, OH) using preamplifiers with low- and high-frequency filters set at 0.05 and 10 Hz, respectively. The recordings also were digitalized and stored on VHS.
3M T120 professional videocassettes using a pulse code modulator (PCM-8, Medical Science, Greenvale, NY). During recording sessions, the animals rested quietly in a supporting canvas sling.

Perfusion of the cerebral ventricular system was done by inserting needles into the lateral ventricles through the cannulas, which were placed during the second operation. One of these needles was used to monitor the ventricular pressure by a Tektronix 5113 Oscilloscope (Tektronix, Beaverton, OR), with a Gould pressure transducer. The other was used for perfusion of the ventricular system, using a Harvard pump, model 975 (Harvard Apparatus, South Natick, MA). Perfusion was at a constant rate of 0.15 ml/min, and the outflow was simultaneously collected from the fourth ventricle, thereby preventing increases in pressure in the ventricular system and escape of the cerebrospinal fluid (CSF) from the fourth ventricle into the subarachnoid space (18). Artificial CSF with or without BBS was used to perfuse the cerebral ventricular system. Artificial CSF was prepared daily by mixing three solutions: 7.46 g NaCl, 0.19 g KCl, 0.14 g CaCl₂, 0.19 g MgCl₂, and 182.11 ml distilled water (solution A); 1.7 g NaHCO₃ and 50 ml distilled water (solution B); and 1.7 g Na₂HPO₄ and 50 ml distilled water (solution C). The mixing ratio was 20:5:5, to which 70 ml of distilled water and 1 mg of bovine serum albumin (Sigma Chemical, St. Louis, MO) were added. It was filtered through an Acrodisc, 25-mm filter assembly (Gelman Sciences, Ann Arbor, MI). IV infusions were made through a 19-gauge butterfly needle positioned into one of the superficial veins in the limbs, with the same pump as for the cerebral ventricular perfusions. Perfusion was initiated after at least two complete MMCS were observed and while phase I activity was present in the stomach.

Four sets of experiments were conducted. Each experiment was performed on three dogs and repeated in each dog at least three times. The first set (group A) consisted of ICV perfusions of BBS (n = 10). BBS (mol wt 1,618.8, Peninsula Laboratories, Belmont, CA) was dissolved in CSF and administered for 2 h at a rate of 1.2 pmol·kg⁻¹·min⁻¹, followed by a further 1-h perfusion of CSF alone. BBS was administered only to the perfusion CSF to prevent any possible cumulative effect. The second set (group B), which served as a control study, consisted of ICV perfusions of CSF of 3-h duration (n = 9). The third set (group C) consisted of IV infusions of BBS (n = 9). BBS was dissolved in normal saline and infused for 2 h at a rate of 1.2 pmol·kg⁻¹·min⁻¹, followed by a further 1-h infusion of normal saline only. The last set (group D) consisted of IV infusion of epinephrine (n = 9). Epinephrine (Abbott Laboratories, Chicago, IL) was dissolved in normal saline with 1 mg/ml ascorbic acid (Baxter Healthcare, Deerfield, IL). Epinephrine was administered at a rate of 0.05 µg·kg⁻¹·min⁻¹ in a volume of 0.15 ml/min and infused for 2 h, followed by a 1-h infusion of normal saline only.

Blood samples and CSF samples from the fourth ventricle were collected before the initiation and at the end of each perfusion-infusion to determine BBS concentrations in the CSF and peripheral circulation, respectively. Plasma levels of motilin also were examined every 20 min. Blood samples to measure free catecholamine (i.e., norepinephrine, epinephrine, and dopamine) plasma levels were collected at 0, 10, 20, 40, 60, 90, 120, 150, and 180 min of each perfusion-infusion. Blood was sampled through a 19-gauge butterfly needle positioned in a superficial vein in one of the limbs. To determine the concentrations of BBS and motilin, 2 ml of blood were collected in sterile Vacutainer tubes (Becton Dickinson, Rutherford, NJ), containing 0.04 ml of liquid 7.5% EDTA (K3) solution (3 mg). The tubes were immediately placed on crushed ice. At the end of each experiment, the blood samples were spun for 30 min in a refrigerated centrifuge (4°C) at 2,500 rpm and the supernatants were transferred to other empty tubes and stored at −20°C. The radioimmunoassay procedures used to determine the concentration of BBS and motilin were similar to the technique described previously (13). The BBS antibody used in this study was rabbit N388. The sensitivity of the assay was 16–1,000 pg/ml with intra-assay and interassay variations of 7% and 12%, respectively. The radioimmunoassay procedures used in this study to determine the concentration of motilin were similar to the technique described previously (6). The antibody used was guinea pig 71 (gift of Dr. J. C. Brown). The sensitivity of the assay was 50 pg/ml with intra-assay and interassay variations of 11.4% and 8%, respectively. When the concentrations of free catecholamines were determined, 10 ml of blood were collected into sterile Venlet blood collection tubes (Terumo Medical, Elkton, MD) containing 143 IU sodium heparin. The tubes were placed on crushed ice, and the blood plasma was immediately spun for 30 min in a refrigerated centrifuge (4°C) at 2,500 rpm. Five milliliters of the supernatant plasma were transferred to other tubes containing 40 µl of sodium metabisulfite 5% solution and stored at −20°C. The catecholamine assay procedure used in this study was described previously (5).

The general condition of the experimental animals during the experiments was monitored by recording heart rate and temperature every 20 min. Observations were also made on their general behavior.

Electrical activity was analyzed by identifying MMCS at each recording electrode. The activity was divided into three phases as previously described (29). In the study described herein, an MMC was considered to be present at each recording site when phases I, II, and III were present in sequence. When phase III activity lasted >3 min, and when all three phases migrated past at least two consecutive electrode sites. The duration of each cycle was the time measured between the beginning of two consecutive phase I activities of complete MMCS at the same electrode site.

Data are expressed as means ± SD. The appropriate means between control and experimental data were compared using Student's t-test for paired data. Values of P ≤ 0.01 were considered significant.

RESULTS

In general, the data obtained in the various stages of the study were consistent between each trial in the same dog and between the trials of each dog in the same set of experiments. In the two exceptional instances in which inconsistent data were obtained within one set of experiments, the group was subdivided to examine each type of results separately.

Effects of ICV perfusion of BBS on MMCS. During a total of 2,306 min of recording time before ICV perfusions with BBS (10 experiments, 3 dogs), 24 MMCS were recorded (an average of 1 MMC/96 min). All MMCS originated in the stomach and propagated to the ileum. In 10 of 10 experiments (1,200 min total recording time), ICV perfusion with BBS disrupted the normal pattern of fasting myoelectric activity. The time lag between onset of ICV perfusion to change in MMC activity was 20.9 ± 7.4 min as measured in the stomach. Typically, phase I activity in the stomach and duodenum was replaced by a period of intense irregular
spike activity, more intense than that recorded during regular phase II activity but less intense and irregular than that recorded during normal, regular phase III activity. In the jejunum and ileum, clusters of intense spike activity were separated by short periods of phase I-like activity (Fig. 1). These results represent an average occurrence rate of 0 MMC/1,200 min in the stomach, 1 MMC/300 min in the duodenum, and 1 MMC/200 min in the jejunum. In 4 of 10 experiments, no phase III activity was observed at any electrode site during the ICV perfusion of BBS. In 4 of 10 experiments, phase III activity originated in the duodenum during ICV perfusion of BBS, and in 2 of 10 experiments, phase III activity originated in the jejunum. All phase III activity periods that originated in the small intestine migrated to the ileum. After ICV perfusion of BBS was stopped, the duration of intensive irregular spike activity in the stomach continued for 79.3 ± 72.6 min. MMC activity resumed in the stomach in 3 of 10 experiments 218.9 ± 61.0 min after BBS perfusion stopped. Phase III activity occurred in the duodenum in five experiments and in the jejunum in one experiment, 68.4 ± 44.9 min and 104.6 min, respectively, after ICV perfusion of BBS stopped. In the remaining experiment, no phase III activity was observed at any electrode site. ICV perfusion of BBS significantly elevated the CSF concentration of BBL immunoreactivity. However, there was no significant change in the level of BBL immunoreactivity in the peripheral circulation during ICV perfusion of BBS (Table 1).

Table 1. CSF and plasma levels of BBS

<table>
<thead>
<tr>
<th></th>
<th>ICV Perfusion of BBS (n = 10)</th>
<th>IV Infusion of BBS (n = 9)</th>
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<tr>
<td></td>
<td>CSF</td>
<td>Plasma</td>
</tr>
<tr>
<td>Before</td>
<td>57.7 ± 71.5</td>
<td>53.0 ± 52.2</td>
</tr>
<tr>
<td>After</td>
<td>1,133.2 ± 938.2</td>
<td>53.6 ± 53.4</td>
</tr>
<tr>
<td>P</td>
<td>&lt;0.005</td>
<td>&gt;0.3</td>
</tr>
</tbody>
</table>

Values are means ± SD in pg/ml; n = no. of experiments. BBS, bombesin; CSF, cerebrospinal fluid; ICV, intracerebroventricular; IV, intravenous.
ICV perfusion of CSF alone had no effect on the MMC cycling pattern in six of nine experiments (Fig. 2). In the remaining three experiments, the duration of phase II activity at all electrode sites was prolonged during the second MMC. Normal MMC activity was resumed immediately after the end of perfusion. All MMCs that occurred during ICV perfusion of CSF originated in the stomach and propagated to the ileum. During a total of 2,197 min of recording time of myoelectric activity before CSF perfusion (9 experiments, 3 dogs), 20 MMCs were recorded (an average of 1 MMC/109.8 min). During 1,620 min of CSF perfusion time, 12 MMCs were recorded (an average of 1 MMC/135.0 min).

The mean ± SD maximum ICV pressure recorded before and during ICV perfusion of BBS was 7.3 ± 3.5 and 5.4 ± 3.5 cmH\textsubscript{2}O, respectively. There was no statistically significant (P > 0.04) difference. ICV pressures measured before and during perfusion of CSF alone were 4.2 ± 2.3 and 3.7 ± 3.1 cmH\textsubscript{2}O, respectively (mean ± SD, P > 0.3).

Effect of IV infusion of BBS on MMCs. IV infusion of BBS disrupted the normal pattern of myoelectric activity (Fig. 3). During a total recording time of 2,346 min before IV infusion of BBS (9 experiments, 3 dogs), 19 MMC cycles were recorded (an average of 1 MMC/123.5 min). During 1,320 min of IV infusion time of BBS, only two periods of phase III activity occurred in the stomach, both of which propagated to the ileum (an average of 1 MMC/540 min). In seven of nine experiments, no phase III activity occurred during BBS infusion. Instead, only phase II-like spike activity was recorded in the jejunum and ileum, whereas only scant spike activity was observed in the stomach and duodenum. After IV infusion of BBS was stopped, the first MMCs occurred in the stomach in seven of nine experiments. In the remaining two experiments, MMC activity first occurred in the jejunum. IV infusion of BBS significantly elevated the circulating level of BBL immunoreactivity (Table 1).

Effect of ICV perfusion of BBS on motilin. The maximum and minimum plasma motilin concentrations during ICV perfusion of BBS are given in Table 2. The results are given separately for two subgroups: subgroup A1 included 6 of 10 experiments in which phase III activity occurred in the duodenum or jejunum and subgroup A2 included 4 of 10 experiments in which no phase III activity occurred at any recording site. In subgroup A1, the maximum and minimum plasma motilin concentrations during ICV perfusion of
BBS were not significantly \( P > 0.2 \) different from respective values observed during the control period. The corresponding values in subgroup A2 were significantly \( P = 0.01 \) higher during ICV perfusion of BBS than during the control period. An example of the effect of ICV perfusion of BBS on plasma motilin levels is illustrated in Fig. 4.

During ICV perfusion of CSF alone, the maximum plasma motilin concentration when phase III activity of an MMC occurred in the stomach was 106.0 ± 30.2 pg/ml. The motilin level decreased to 63.4 ± 22.6 pg/ml when phase III activity of the same MMC reached the distal ileum (mean ± SD, \( n = 9 \)). This difference was significant \( P < 0.001 \). The maximum and minimum levels of plasma motilin before ICV perfusion of CSF alone were 94.0 ± 23.9 and 55.2 ± 18.7 pg/ml, respectively (mean ± SD, \( n = 9 \), \( P < 0.0001 \)). These values

### Table 2. Plasma levels of motilin during ICV perfusion of BBS

<table>
<thead>
<tr>
<th>Subgroup</th>
<th>Maximum (pg/ml)</th>
<th>Minimum (pg/ml)</th>
<th>Maximum (pg/ml)</th>
<th>Minimum (pg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1</td>
<td>104.0 ± 34.0</td>
<td>64.8 ± 22.5</td>
<td>108.7 ± 33.5</td>
<td>72.7 ± 27.5</td>
</tr>
<tr>
<td>P</td>
<td>&lt;0.003</td>
<td>&lt;0.006</td>
<td>&lt;0.006</td>
<td>&lt;0.0005</td>
</tr>
<tr>
<td>A2</td>
<td>96.3 ± 21.7</td>
<td>48.8 ± 13.7</td>
<td>112.5 ± 26.5</td>
<td>79.8 ± 26.3</td>
</tr>
<tr>
<td>P</td>
<td>&lt;0.001</td>
<td>&lt;0.0005</td>
<td>&lt;0.0005</td>
<td>&lt;0.0005</td>
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</tbody>
</table>

Values are means ± SD in pg/ml. BBL, bombesin-like peptide. Subgroup A1, 6 of 10 experiments in which no phase III activity occurred at any recording site during ICV perfusion of BBS; Subgroup A2, 4 of 10 experiments in which phase III activity occurred during ICV perfusion of BBS in duodenum or jejunum but not in stomach.
Effects of BBS administration on endogenous catecholamines. The plasma concentration of epinephrine, norepinephrine, and dopamine before BBS administration and 60 min after onset of BBS administration are given in Table 3. Plasma levels of epinephrine, norepinephrine, and dopamine increased during IV perfusion of BBS and of CSF and during IV infusion of BBS. However, only the increase of epinephrine during ICV perfusion of BBS was statistically significant (P < 0.01). Nevertheless, it is possible that with a larger number of experiments and animals, statistical significance could be achieved also for the rise in norepinephrine and dopamine levels.

Effects of epinephrine infusion on MMCs, motilin, and catecholamines. During 2,161 min of total recording time of control myoelectric activity (9 experiments, 3 dogs), 19 MMCs were recorded before IV infusion of epinephrine (1 MMC/113.7 min). All originated in the stomach. IV infusion of epinephrine evoked a longer period of phase III-like activity in all recording sites and delayed induction of phase III activity in any recording site (Fig. 7). During 1,080 min of total recording time during IV infusion of epinephrine (9 experiments, 3 dogs), 4 MMCs were recorded (1 MMC/270 min) and all originated in the stomach. After IV infusion of epinephrine, the first MMC was recorded in the stomach in six of nine experiments, in the duodenum in two of nine experiments, and in the jejunum in one experiment. In all instances in which phase III activity of the first MMC did not originate in the stomach, the second did so.

Plasma concentrations of endogenous motilin were measured during recordings of nine MMCs which preceded IV infusion of epinephrine. There was periodic fluctuation in plasma motilin concentration. Peak levels of motilin occurred when phase III activity originated in the stomach. Plasma motilin concentration decreased during caudal propagation of MMCs. The same pattern of fluctuation of plasma motilin concentration was observed during IV infusion with epinephrine. During the control period before IV infusion of epinephrine, plasma motilin concentration was 98.9 ± 9.9 pg/ml (mean ± SD, n = 9) when phase III activity of an MMC occurred in the stomach and it decreased to 57.4 ± 14.9 pg/ml (mean ± SD, n = 9) when phase III activity of the same MMC reached the distal ileum. This difference was significant (P < 0.001). During the control period before IV infusion of epinephrine, equivalent values were 122.8 ± 45.0 and 65.2 ± 23.4 pg/ml, respectively (mean ± SD, n = 9, P < 0.001). These values were not statistically (P > 0.05) different from the respective values observed during IV infusion of BBS. An example of the effect of IV infusion of BBS on plasma motilin levels is illustrated in Fig. 6.

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**Fig. 4.** Effect of ICV perfusion of BBS on plasma motilin concentration in fasted dog. Occurrence of MMC activity is shown in schematic form. Overlapping line represents endogenous plasma motilin concentration. Cycling of motilin persisted despite suppression of phase III activity.

**Fig. 5.** Effect of ICV perfusion of only CSF on plasma motilin concentration in fasted dog. Occurrence of MMC activity is shown in schematic form. Overlapping line represents endogenous plasma motilin concentration. Peak levels were measured when phase III activity was present in stomach and duodenum.
In these five of nine experiments, the maximum plasma motilin level before IV infusion of epinephrine was 100.8 ± 7.5 pg/ml. The difference was statistically (P = 0.01) significant. An example of the effect of IV infusion of epinephrine on plasma motilin levels is illustrated in Fig. 8.

The plasma concentration of epinephrine during IV infusion of epinephrine increased from 59.2 ± 105.8 to 142.5 ± 173.0 pg/ml after 60 min of perfusion (mean ± SD, n = 9, P < 0.001). The equivalent values of norepinephrine during IV infusion of epinephrine were 169.5 ± 114.3 and 210.5 ± 138.8 pg/ml, respectively (mean ± SD, n = 9, P > 0.05). The equivalent values of dopamine were 12.0 ± 13.6 and 25.9 ± 15.7 pg/ml, respectively (mean ± SD, n = 9, P < 0.01).

Other observed effects of BBS administration. ICV perfusion of BBS increased pulse rate from 82.0 ± 11.3 beats per minute (bpm) at onset of perfusion to 108.0 ± 22.1 bpm after 60 min of perfusion (mean ± SD, n = 10, P < 0.001). ICV perfusion of CSF and IV infusion of BBS had no effect on pulse rate. IV infusion of epinephrine also increased the pulse rate from 85.8 ± 11.0 to 105.8 ± 14.0 bpm (mean ± SD, n = 9, P < 0.01). Core body temperature during ICV perfusion of BBS (38.3 ± 0.4°C), ICV perfusion of CSF (38.6 ± 0.6°C), IV infusion of BBS (38.3 ± 0.32°C), and IV infusion of epinephrine (38.5 ± 0.4°C) was not significantly different (P > 0.01) from control core body temperature (38.3 ± 0.2°C).

During each ICV perfusion of BBS, all dogs became restless, apprehensive, and agitated. This change in behavior appeared within 10 min of onset of ICV perfusion. No change in behavior was observed during ICV perfusion of CSF, IV infusion of BBS, or IV infusion of epinephrine.

**DISCUSSION**

ICV perfusion of BBS disrupted the fasting pattern of myoelectric activity of the stomach and small intestine and replaced MMCs with a pattern of irregular intense spike activity. This effect was particularly evident in the stomach, in which phase III activity of an MMC failed to appear during any of the experiments in which BBS was administered by ICV perfusion. ICV perfusion of BBS did not prevent the initiation of MMCs in the duodenum or jejunum and their aborad propagation to the distal ileum in 6 of 10 ICV perfusions.

It has been shown in previous studies that ICV BBS may appear in the peripheral circulation by leakage or via a peptide transport system (1, 22), thus raising the possibility that the effect seen in the present study during ICV perfusion of BBS may have been due to a peripheral site of action. However, in another study in which the same animal model was used as the one in the present study, test substances perfused from one lateral to fourth cerebral ventricles did not leak into the peripheral circulation (18). In the present study, there was no evidence that BBS had passed into the peripheral circulation because the plasma BBS concentrations were not affected during ICV perfusions of BBS. Additional supporting evidence that the effects of ICV administration of BBS were mediated by activation of central brain structures was the finding that IV administration of BBS evoked effects that were quite different from those observed during ICV perfusion of BBS. Also, the effect of ICV perfusion of BBS was not due to a change in intraventricular pressure, which may affect gastrointestinal activity (11), because no significant change in ICV pressures occurred during our experiments. Thus the disruption of the fasting pattern observed during ICV perfusions of BBS was due to a central, not peripheral, effect.

In previously reported studies, ICV administration of BBS was found to increase gastric intraluminal pres-

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**Table 3. Plasma levels of catecholamines**

<table>
<thead>
<tr>
<th></th>
<th>ICV Perfusion of BBS (n = 10)</th>
<th>ICV Perfusion of CSF (n = 9)</th>
<th>IV Infusion of BBS (n = 9)</th>
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<tbody>
<tr>
<td></td>
<td>At onset</td>
<td>After 60 min</td>
<td>At onset</td>
</tr>
<tr>
<td>Epi</td>
<td>44.3 ± 70.4</td>
<td>&lt;0.01</td>
<td>33.7 ± 19.5</td>
</tr>
<tr>
<td>NE</td>
<td>162.6 ± 94.5</td>
<td>&gt;0.03</td>
<td>181.6 ± 130.4</td>
</tr>
<tr>
<td>DA</td>
<td>15.1 ± 32.4</td>
<td>&gt;0.02</td>
<td>53.9 ± 122.5</td>
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| Values are means ± SD in pg/ml; n = no. of experiments. Epi, epinephrine; NE, norepinephrine; DA, dopamine.
sure (28), increase contraction in the duodenum and jejunum (9, 27), delay gastric emptying (25, 26), and delay small bowel transit (25–27). The antitransit effect of centrally administered BBS was explained as the result of an increase in frequency of nonpropulsive contractions or a disruption of the normal coordinated propulsive motor activity in the duodenum (27). The increase in intense and irregular spike activity of the stomach and of the small bowel observed in the present study during ICV perfusion of BBS may explain the increase in gastric luminal pressure, and the absence of gastric phase III activity and the decreased occurrence of MMCs in the small bowel may explain the delay in gastric emptying and small bowel transit.

Previous studies (25, 26) have shown that the effect of central administration of BBS on gastrointestinal motility requires intact vagal pathways. It also has been demonstrated that vagotomy primarily affects emptying of the stomach (31) and abolishes the gastric MMC (12). In the present study ICV perfusion of BBS affected the myoelectric activity of the stomach to a greater degree than that of the small intestine. It is therefore conceivable that ICV perfusion of BBS directly activates central brain structures, which then transmit signals to the gastrointestinal tract via the vagus nerves. This hypothesis is supported by the concentration of BBS receptors found in hypothalamic structures (23). The immediate vicinity of most of these structures to the ventricular system of the brain makes

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**Fig. 7.** Effect of IV infusion of epinephrine on migrating myoelectric activity in fasted dog. The 3 panels are consecutive recordings at 4 electrode sites (S, D, J, and I are described in legend for Fig. 1). Phase II was prolonged. Periods of gastric arrhythmia were recorded during phase I activity. Arrows indicate initiation (•) and cessation (•) of infusion.

**Fig. 8.** Effect of IV infusion of epinephrine on plasma motilin concentration in fasted dog. Occurrence of MMC activity is shown in schematic form. Overlapping line represents endogenous plasma motilin concentration. Peak levels of motilin occurred when phase III activity originated in stomach.
it likely that ICV perfused BBS reaches these receptors. The location of the vagal dorsal motor nuclei in the floor of the fourth ventricle also potentially exposes these structures to the action of BBS perfused ICV. Thus it is possible that BBS may affect MMC cycling through either the vagal or the splanchnic pathways. The results of previous studies (25, 26), which showed that subdiaphragmatic vagotomy abolishes the motor effect of ICV administered BBS, point to the vagal system as the probable pathway of the central action of BBS. To establish whether BBS acts directly on the vagal nuclei or via hypothalamic structures, further studies of direct injections of BBS into these neural centers are required.

Motilin is considered to be important in initiating phase III activity in the stomach and duodenum (14, 30). Cycling changes in motilin occur during fasting, with peak levels of motilin coinciding with phase III activity of a MMC in the stomach (14, 16). In the present study, in the experiments in which no MMC was initiated in the stomach, peak plasma motilin concentrations during ICV perfusion of BBS were significantly higher than the peak levels preceding perfusion. Although plasma motilin levels fluctuated in the absence of a MMC, the minimum levels were also significantly higher during perfusion in comparison to pre-ICV perfusion minimum levels and remained so until phase III activity of an MMC occurred. In the experiments in which phase III activity of an MMC originated in the duodenum or the jejunum during ICV perfusion of BBS, plasma motilin concentrations decreased during aboral migration of the MMC along the small bowel to concentrations similar to the minimum plasma motilin concentration observed before ICV perfusion of BBS. However, the decrease in plasma motilin concentration was not accompanied by decreased spike activity (phase I activity) in the stomach. This suggests that the increased level of motilin during ICV perfusion of BBS may have played only a partial role in the mechanism by which ICV perfusion of BBS disrupted the fasting pattern of myoelectric activity of the stomach. Activation of an inhibitory pathway during ICV perfusion of BBS, which prevents initiation of MMCs despite a high plasma motilin concentration, may be one possible mechanism by which centrally administered BBS disrupts gastric MMCs. If such an inhibitory mechanism was activated by BBS, it appears to have had a greater effect on the stomach than on the small intestine.

Previous studies have shown that centrally administered BBS affects the sympathetic system (2, 3, 19) and may activate the adrenal medulla and sympathetic nerves within the splanchnic region (20). Thus it is conceivable that changes in the peripheral circulating levels of epinephrine, norepinephrine, and dopamine could have mediated the effect of ICV perfusion of BBS. The significant rise in dopamine and in norepinephrine in the peripheral circulation during ICV perfusion of BBS limits the possibility that these substances mediated the effect of ICV perfusion of BBS on the fasting pattern in the stomach. Furthermore, the effect of dopamine on gastrointestinal myoelectric activity (17) is inverse to the changes observed in our experiments during central administration of BBS. During ICV perfusion of BBS, there was a significant increase in the circulating levels of epinephrine. The increase in epinephrine during ICV perfusion of BBS cannot be attributed to stress caused by the mechanical action of perfusion because there was no significant increase in the plasma levels of epinephrine during ICV perfusion of artificial CSF alone. However, during ICV perfusion of BBS, but not during ICV perfusion of CSF alone, the dogs were apprehensive and agitated. Thus the increased level of epinephrine could be related to stress induced by central administration of BBS. To examine the possibility that the changes in myoelectric activity observed during ICV perfusion of BBS were mediated by the increase in the level of epinephrine, experiments were performed in which epinephrine was infused intravenously to mimic the rise in epinephrine produced by ICV perfusions of BBS. The resulting change in myoelectric activity was a decrease in the occurrence rate of MMCs, a prolongation of phase II activity, and an aboral site of initiation of the first MMC when infusion of epinephrine was stopped. These alterations in myoelectric activity of the stomach and small intestine, although not identical, have some common aspects with those induced by ICV perfusion of BBS. It is therefore possible that epinephrine may play a role in mediating the central effect of BBS. Another common aspect of ICV perfusion of BBS and IV infusion of epinephrine, which was observed when phase III activity of an MMC was delayed by the administration of either substance, was the higher level of plasma motilin concentration compared with the maximum level observed during the control period. However, IV infusion of epinephrine did not produce cycling changes in plasma motilin levels in the absence of phase III activity. Thus it appears that central administration of BBS and peripheral infusion of epinephrine affect cycling of plasma motilin by different pathways.

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