Fourth ventricular administration of ghrelin induces relaxation of the proximal stomach in the rat

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Kobashi M, Yanagihara M, Fujita M, Mitoh Y, Matsuo R. Fourth ventricular administration of ghrelin induces relaxation of the proximal stomach in the rat. Am J Physiol Regul Integr Comp Physiol 296: R217–R223, 2009. First published November 26, 2008; doi:10.1152/ajpregu.00878.2007.—The effects of fourth ventricular administration of ghrelin on motility of the proximal stomach were examined in anesthetized rats. Intragastric pressure (IGP) was measured using a balloon situated in the proximal part of the stomach. Administration of ghrelin into the fourth ventricle induced relaxation of the proximal stomach in a dose-dependent manner. Significant reduction of IGP was observed at doses of 3, 10, or 30 pmol. The administration of ghrelin (10 or 30 pmol) with growth hormone secretagogue receptor (GHS-R) antagonist ([D-Lys3]GHRP-6; 1 nmol) into the fourth ventricle did not induce a significant change in IGP. The sole administration of [D-Lys3]GHRP-6 also did not induce a significant change in IGP. Bilateral sectioning of the vagi at the cervical level abolished the relaxation induced by the administration of ghrelin (10 or 30 pmol) into the fourth ventricle, suggesting that relaxation induced by ghrelin is mediated by vagal preganglionic neurons. Microinjections of ghrelin (200 fmol) into the caudal part of the dorsal vagal complex (DVC) induced obvious relaxation of the proximal stomach. Similar injections into the intermediate part of the DVC did not induce significant change. Dose-response analyses revealed that the microinjection of 2 fmol of ghrelin into the caudal DVC significantly reduced IGP. These results revealed that ghrelin induced relaxation in the proximal stomach via GHS-R situated in the caudal DVC.

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fifth ventricular administration of ghrelin (10 or 30 pmol) with growth hormone secretagogue receptor (GHS-R) antagonist ([D-Lys3] GHRP-6; 1 nmol) into the fourth ventricle did not induce a significant change in IGP. The sole administration of [D-Lys3] GHRP-6 also did not induce a significant change in IGP. Bilateral sectioning of the vagi at the cervical level abolished the relaxation induced by the administration of ghrelin (10 or 30 pmol) into the fourth ventricle, suggesting that relaxation induced by ghrelin is mediated by vagal preganglionic neurons. Microinjections of ghrelin (200 fmol) into the caudal part of the dorsal vagal complex (DVC) induced obvious relaxation of the proximal stomach. Similar injections into the intermediate part of the DVC did not induce significant change. Dose-response analyses revealed that the microinjection of 2 fmol of ghrelin into the caudal DVC significantly reduced IGP. These results revealed that ghrelin induced relaxation in the proximal stomach via GHS-R situated in the caudal DVC.

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on proximal stomach motility were investigated. Furthermore, the effect of the local administration of ghrelin was also investigated to demonstrate that the caudal DVC is responsible for relaxation of the proximal stomach induced by ghrelin.

**MATERIALS AND METHODS**

**Animals, preparations, and recording of intragastric pressure.**
Animal care was in accordance with the guidelines of the Physiological Society of Japan. The experimental protocols were approved by Okayama University Animal Use Committee.

Male Sprague-Dawley rats (280–320 g) were used. Each animal was anesthetized with an intraperitoneal injection of urethane-chloralose (urethane, 0.8 g/kg; chloralose, 65 mg/kg body wt). Subsequent anesthesia was administered through Silastic tubing (OD: 1.0 mm; ID: 0.5 mm) inserted into the right jugular vein. Each animal had a tracheal cannula made from polyethylene tubing (OD: 2.07 mm). After an abdominal incision was made, gastric contents were gently squeezed out of the tip of the fundus, and the stomach was washed out twice using Ringer solution. A balloon, created from thin latex rubber and plastic tubing (OD: 1.7 mm) and another tube (OD: 2.0 mm) to drain gastric juices were introduced into the stomach (15–18). When recording proximal stomach motility, a balloon was introduced into the proximal stomach from the tip of the fundus. The balloon was secured by purse sutures around the tubing. It was confirmed that the inflated balloon was correctly settled in the proximal part of the stomach. When recording distal stomach motility, a balloon was introduced into the distal stomach from the greater curvature just distal to the limiting ridge to drain gastric juices and was ligated to the gastric wall around the tubing. It was confirmed that the inflated balloon was correctly settled in the proximal part of the stomach. When recording distal stomach motility, a balloon was introduced into the distal stomach from the greater curvature just proximal to the limiting ridge toward the antrum. The balloon was secured by purse sutures around the gastric wall using 4–0 silk thread. The plastic tubing protruding through the abdomen was ligated to the gastric wall around the tubing. Another tube was introduced into the stomach from the greater curvature just distal to the limiting ridge to drain gastric juices and was ligated to the gastric wall around the tubing. It was confirmed that the inflated balloon was correctly settled in the proximal part of the stomach.

After closing the abdominal incision, each animal was mounted on a stereotaxic apparatus. The neck muscles were removed and the ligaments between the occipital bone and atlas were carefully removed. A small hole was made through the dura mater to administer drugs using a Hamilton syringe into the fourth ventricle, as previously described (15, 18). When the drugs were injected into the DVC using a glass micropipette, the occipital bone and dura mater were removed to expose the surface of the brain stem. Animals were placed in the prone position during the experiments. Body temperature was maintained at 36°C using a heating pad placed under the body (ATB-1100; Nihon Kohden, Tokyo, Japan).

The intragastric balloon was inflated with 0.2–0.5 ml water, maintaining initial intragastric pressure (IGP) of 0.5–0.7 kPa. A similar volume was used in our previous studies to measure gastric relaxation of the proximal stomach (15–18). After gastric balloon inflation, animals were left for at least 60 min until IGP stabilized. The distal end of this tubing was connected to a strain-gauge pressure meter (NEC-Sanei, 6M82; Tokyo, Japan) to measure IGP. Gastric response data were stored on a personal computer using the Power Lab system (ADInstruments, Colorado Springs, CO) for later analyses. At the end of each experiment, the abdomen was reopened, and the position of the balloon was confirmed again. The inflated balloon in each animal was always positioned in the correct part of the stomach.

**Fourth ventricular administration of drugs.**
Dose-dependent relaxation of the proximal stomach and enhanced contractile activity of the distal stomach were studied with fourth ventricular administration of ghrelin. All drugs were dissolved in Ringer solution. Because we confirmed that similar administration of Ringer solution (3 μl) did not induce any response of proximal and distal stomach motility in our previous study (15), we did not test the effect of Ringer solution on motility in the present study. Four different ghrelin doses [1, 3, 10, and 30 pmol (3 μl)] for proximal stomach motility and four different ghrelin doses [3, 10, 30, and 100 pmol (3 μl)] for distal stomach motility were used. Each rat received at least three different doses of ghrelin. The gastric response was observed for at least 1 h after the administration of each drug. Drug doses were always administered in ascending order. The previous study showed that intracerebroventricular injection of ghrelin at doses of 10 pmol and above significantly increased food intake in rats (25). Intracerebroventricular injection of 0.1 μg (30 pmol) ghrelin stimulated antral motility in conscious rats (8). Thus, the doses used in the present study are appropriate to investigate the phenomena associated with food intake, such as gastric motility.

To determine whether the ghrelin response is caused via growth hormone secretagogue receptor (GHS-R), the effects of [D-Lys3] GHRP-6, and its doses used in the present study are appropriate to investigate the phenomena associated with food intake, such as gastric motility.

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**Cervical vagotomy.**
To determine the descending pathway, a vagotomy was performed in five rats. Both vagi were carefully separated from the left and right carotid arteries. After observing the relaxation induced by the administration of ghrelin (10 pmol, 3 μl), cervical vagotomy was performed using seven animals. Additionally, simultaneous administration of a higher dose of ghrelin (30 pmol, 3 μl) and [D-Lys3] GHRP-6 (1 nmol, 3 μl) was performed using five animals. Further, it was confirmed using six rats that the administration of [D-Lys3] GHRP-6 alone had no effect on proximal stomach motility. A dose of 1 nmol [D-Lys3] GHRP-6 was sufficient to block the effect of the central injection of ghrelin on motor activity of the antrum, according to the previous study (8).

Ghrelin was purchased from the Peptide Institute (Osaka, Japan). [D-Lys3] GHRP-6 was purchased from Sigma-Aldrich (St. Louis, MO).

**Local administration of ghrelin.**
Commercially prepared glass micropipettes (10 μm in tip diameter; World Precision Instruments, Sarasota, FL) connected to a 50-μl Hamilton syringe were installed in a microinjector (XF-320J; Nihon Kohden, Japan) to inject ghrelin into the left DVC. Each glass pipette was first filled with fluid paraffin. The test solution was sucked into the tip of the pipette, which penetrated the caudal DVC or intermediate DVC at the left medulla using A stereotaxic apparatus. The coordinates were 0.3 mm posterior to the obex, 0.25 mm lateral to the midline, and 0.9 mm ventral from...
the surface of the brain stem for caudal DVC, and 0.5 mm anterior to the obex, 0.5 mm lateral to the midline, and 0.4 mm ventral from the surface of the brain stem for intermediate DVC. These locations corresponded with the boundary between the NST and the DMV, according to the atlas (27, 28). Just after penetration, ghrelin (200 fmol, 60 nl) was injected for 30 s. Each 60-nl volume formed a sphere of about 500 μm in diameter. The injection, therefore, filled the entire DMV and the adjacent NST and hypoglossal nucleus. To examine the effects of ghrelin, each animal received injections into either the caudal (6 rats) or intermediate (6 rats) part of the DVC. The stereotaxic coordinates, where the glass pipette penetrated, were identical to where we injected drugs in the previous study (18). According to this previous study, dye injections into these areas stained the ventral part of the NST, DMV, and dorsal part of the hypoglossal nucleus. We also confirmed that injections of vehicle solution (60 nl of Ringer solution) into the caudal or intermediate part of the DVC induced no significant change in intragastric pressure; therefore, in the present study, we did not attempt vehicle injection into the DVC. In addition, the dose response function of ghrelin was examined using 20 animals. One of 3 doses of ghrelin (0.2 fmol, 2.0 fmol, or 20 fmol) was administered into the caudal DVC in each animal. Each injected volume was 60 nl, the same as the other injections.

In the present study, the part of the DVC located caudal to the obex was defined as the “caudal DVC,” and the portion of the DVC located between the obex and anterior tip of the AP was defined as the “intermediate DVC,” according to a previous study (Fig. 4 B). According to this previous study, dye injections into these areas stained the ventral part of the NST, DMV, and dorsal part of the hypoglossal nucleus. We also confirmed that injections of vehicle solution (60 nl of Ringer solution) into the caudal or intermediate part of the DVC induced no significant change in intragastric pressure; therefore, in the present study, we did not attempt vehicle injection into the DVC. In addition, the dose response function of ghrelin was examined using 20 animals. One of 3 doses of ghrelin (0.2 fmol, 2.0 fmol, or 20 fmol) was administered into the caudal DVC in each animal. Each injected volume was 60 nl, the same as the other injections.

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Data analyses. To analyze relaxation of the proximal stomach, the minimum value (kPa) was measured once every minute from 10 min before drug administration to 90 min after drug administration using chart software (AD Instruments). To analyze the contractile response of the distal stomach, the area under the contraction wave (kPa × min) above minimum IGP in a 1-min period was calculated every minute from 10 min before drug administration to 90 min after drug administration using chart software (AD Instruments). The area for the 1-min period was used for the index of motility (motility index, MI). This method is suitable for minimizing the inclusion of the area formed by the slow change of the baseline. This analysis method was similar to that used in our previous study (9, 15, 18).

All numerical values are expressed as the means ± SE. Most statistical analyses were made between prestimulus values and poststimulus values using the paired t-test (P < 0.05 for significance). For statistical evaluation of the effects of fourth ventricular administration of drugs on proximal and distal stomach motility, the paired t-test (P < 0.05 for significance) was used. Differences between the mean of the minimum values of the IGP (or the area for 1-min period) just before administration of the solutions and those 21–22 min after the administration were examined, since mean IGP of proximal stomach motility showed the minimum value 21–22 min after the administration of 30 pmol ghrelin. Dose-related effect of ghrelin was evaluated by repeated-measures ANOVA (P < 0.05 for significance). To evaluate the effects of microinjection of drugs on proximal stomach motility, the paired t-test (P < 0.05 for significance) was used. Differences were examined between the mean of the minimum values just before solution injection and that 9–10 min after injection, since mean IGP of proximal stomach motility showed a minimum value at 9–10 min after the injection of 200 fmol ghrelin. Student’s t-test was used to examine the difference between postinjection values of the microinjection into the intermediate DVC and that into the caudal DVC, as shown in Fig. 4B, b.

RESULTS

Ghrelin induced relaxation of the proximal stomach and stimulated contractility of the distal stomach. The effects of fourth ventricular administration of ghrelin on motility of the proximal and distal stomach were examined. Administration of ghrelin induced relaxation of the proximal stomach in a dose-dependent manner (Fig. 1A, a). Low-dose (1 pmol) administration of ghrelin did not induce a significant decrease in IGP (t = 0.17, NS, n = 5); however, the administration of 3 pmol (t = 2.86, P < 0.05, n = 7), 10 pmol (t = 12.58, P < 0.05, n =
induce a significant change (Fig. 1, 3). ANOVA [F(3,12) = 4.14, P < 0.05, n = 5] for five rats which received four doses of ghrelin. Distal stomach contractility was conversely stimulated in response to ghrelin administration (Fig. 1B, a). A significant increase in the MI was observed after the administration of 100 pmol ghrelin (t = 2.47, P < 0.05, n = 8); however, the administration of 3 pmol (t = 0.65, NS, n = 7), 10 pmol (t = 0.81, NS, n = 8), and 30 pmol (t = 1.16, NS, n = 0.28) ghrelin did not induce a significant change (Fig. 1B, b). Dose-related effect of ghrelin was statistically proven using repeated-measures ANOVA [F(3,18) = 3.73, P < 0.05, n = 7] for seven rats that received four doses of ghrelin.

Ghrelin induced relaxation of the proximal stomach via GHS-R. To determine whether relaxation of the proximal stomach was caused by GHS-R, the effect of GHS-R antagonist ([D-Lys3] GHRP-6) on ghrelin-induced relaxation was examined.

Before testing the antagonist, the effect of repeated administrations of ghrelin was examined. More than 90 min after the first administration of ghrelin (10 pmol) into the fourth ventricle, ghrelin (10 pmol) was administered a second time into each animal. First, the administration of ghrelin induced a significant reduction in IGP (0.092 ± 0.016 kPa, t = 5.76, P < 0.05, n = 6). Second, the administration of ghrelin also induced a significant reduction in IGP (0.097 ± 0.026 kPa, t = 3.78, P < 0.05, n = 6). Thus, repeated administration of 10 pmol ghrelin caused a reproducible result.

Fourth ventricular administration of [D-Lys3] GHRP-6 (1 nmol) did not affect the IGP. Mean IGP before the administration of [D-Lys3] GHRP-6 (0.677 ± 0.018 kPa) was not significantly different (t = 2.39, NS, n = 6) from that after administration (0.653 ± 0.020 kPa); however, the ghrelin response was abolished by administration of [D-Lys3] GHRP-6. Each animal first received fourth ventricular administration of ghrelin (10 pmol), which caused a significant decrease in IGP (Fig. 2, A and B) (t = 7.85, P < 0.05, n = 7). More than 90 min after the first administration, simultaneous administration of ghrelin (10 pmol) and [D-Lys3] GHRP-6 (1 nmol) into the fourth ventricle was performed in each animal. The administration of ghrelin with [D-Lys3] GHRP-6 did not induce a significant change in IGP (Fig. 2, A and B) (t = 0.01, NS, n = 7). Furthermore, using the other five animals, simultaneous administration of a higher dose of ghrelin (30 pmol) and [D-Lys3] GHRP-6 (1 nmol) into the fourth ventricle was performed, which did not induce a significant change in IGP (Fig. 2B) (t = 1.20, NS, n = 5).

Contribution of vagal efferent neurons. To examine the contribution of parasympathetic preganglionic neurons, the effect of bilateral cervical vagotomy on relaxation induced by the administration of ghrelin (10 pmol) was examined. Vagotomy abolished the relaxation induced by the administration of ghrelin into the fourth ventricle (Fig. 3). The significant decrease in IGP observed before sectioning of the vagi (t = 11.07, P < 0.05, n = 5) was not observed after sectioning (t = 1.50, NS, n = 5) (Fig. 3). Furthermore, the effect of a higher dose of ghrelin (30 pmol, 3 μl) was examined in six additional animals, which preliminarily underwent bilateral cervical vagotomy.
IGP before the administration of 20 fmol ghrelin (0.67 ± 0.041 kPa) was significantly different (t = 5.92, P < 0.05, n = 6) from that after administration (0.62 ± 0.035 kPa). Mean IGP before the administration of 2.0 fmol ghrelin (0.66 ± 0.05 kPa) was significantly different (t = 3.81, P < 0.05, n = 6) from that after administration (0.63 ± 0.05 kPa). Mean IGP before the administration of 0.2 fmol ghrelin (0.66 ± 0.032 kPa) was not significantly different (t = 0.24, NS, n = 7) from that after administration (0.66 ± 0.028 kPa). The dose-response relationship of the mean change in IGP to four different doses (0.2, 2.0, 20, and 200 fmol) of ghrelin injected into the caudal DVC is shown in Fig. 5.

Fig. 3. Effect of bilateral cervical vagotomy on relaxation of the proximal stomach induced by ghrelin. Open circles denote mean IGP before vagotomy. Solid circles denote mean IGP after vagotomy. *Significant difference compared with the IGP before administration of ghrelin in the prevagotomized animal. Reduction in IGP induced by ghrelin administration (10 pmol) was not observed after vagotomy.

Fig. 4. Effect of microinjection of ghrelin into two different sites of the dorsal vagal complex (DVC) on IGP of the proximal stomach. A: a: typical response of the proximal stomach induced by microinjection of ghrelin into the intermediate DVC. A: b: mean IGP preinjection and postinjection of ghrelin. B: a: typical response of the proximal stomach induced by microinjection of ghrelin into the caudal DVC. B: b: mean IGP preinjection and postinjection of ghrelin. *Significant difference compared with the IGP before administration of ghrelin. †Significant difference compared with mean IGP after ghrelin injection into the intermediate DVC shown in A, b. C: schematic representation of injection sites. Two different sites are plotted on the horizontal plane of the medulla. DMV, dorsal motor nucleus of the vagus.

Fig. 5. Dose-response relationship of mean change in intragastric pressure to four different doses (0.2, 2.0, 20, 200 fmol) of ghrelin injected into the caudal DVC. *Significant differences between the IGP before ghrelin injection and that after ghrelin injection. Number of measurements at each dose is shown by each circle.
DISCUSSION

The present study demonstrates that ghrelin induced relaxation of the proximal stomach via GHS-R situated in the caudal DVC. Relaxation induced by ghrelin was achieved by vagal preganglionic neurons.

Relaxation of the proximal stomach induced by fourth ventricular administration of 10 pmol or 30 pmol ghrelin was reversed by fourth ventricular administration of 1 nmol [D-Lys³] GHRP-6 in the present study. Similarly, antral motility induced by the intravenous administration of 0.3 nmol and 3 nmol ghrelin was reversed by the intravenous administration of 100 nmol [D-Lys³] GHRP-6 in the previous study using arousal rats (8). A 30-fold dose of the receptor antagonist is required to disable the ghrelin effects. This can be explained by the lower affinity of [D-Lys³] GHRP-6 for GHS-R than ghrelin (11, 34).

The main purpose of the present study was to identify the control mechanism of reservoir functions; therefore, most experiments were devoted to proximal stomach motility. Previous studies have clarified that the intravenous or intracerebroventricular administration of ghrelin enhances distal stomach motility (8, 23). We have also indicated that a similar response of the distal stomach was observed in our experimental condition. Although systemic administration of ghrelin accelerates gastric emptying (6, 14), the simultaneous occurrence of enhanced distal stomach motility and proximal stomach relaxation must be critical to the functional outcome of accelerated gastric emptying. Because actual transit of food into the duodenum is necessary for relaxation of the pyloric sphincter, which coordinates with antral contraction during the emptying period (40–60 min after feeding) (13), temporal and precise analyses are required to know the effect of ghrelin on the actual transit of food into the duodenum.

Systemic administration of ghrelin stimulates distal stomach motility (8, 23) and gastric emptying (6, 14). Peripheral and central actions of ghrelin on distal stomach motility are almost the same. A few studies have investigated the effect of systemic administration of ghrelin on proximal stomach motility. An electrophysiological study using deprived mice revealed that the peripheral administration of ghrelin reduced afferent signals from gastric tension receptors (26). Signaling of the presence and amount of food in the stomach is reduced; that is, desensitization of mechanosensitive afferents reduces satiation and allows the stomach to receive a large amount of food. This neural response seems likely to cooperate with the effects of central ghrelin on the proximal stomach observed in the present study. In contrast, bath application of ghrelin agonists reduced electrical field stimulation-induced relaxation in fundic strips (6). Intravenous administration of ghrelin increased proximal stomach tone in deprived humans (32). Thus, the peripheral action of ghrelin does not necessarily correspond to the central action as to the reservoir functions. The species of subjects and/or feeding conditions might induce different results. More investigations are needed to resolve this problem.

The present study showed that microinjection of 200 fmol ghrelin (60 nl) into the caudal DVC induced marked relaxation of the proximal stomach. Although we could not determine whether ghrelin acted on just NST or DMV neurons, the caudal DVC is the most probable area inducing relaxation of the proximal stomach. Microinjections of a lower dose of ghrelin into the caudal DVC revealed that 2 fmol (60 nl of 33 nM solution) but not 0.2 fmol (60 nl of 3.3 nM solution) ghrelin induced a significant decrease in IGP of the proximal stomach. For electrophysiological analysis of hypothalamic neurons, 15 nM solution of ghrelin were used in the previous study (4). Thus, the effective concentration of proximal stomach relaxation was not so different from the previous study.

It is widely assumed that peptides and proteins cannot pass through the blood-brain barrier. A recent study using mice showed the existence of a saturable transporter in the blood-to-brain direction, which recognizes human ghrelin but not mouse ghrelin (1). Thus, the transportability of peripheral ghrelin into the CNS might depend on the structure of ghrelin. The AP, which is located just above the NTS, has a loose blood-brain barrier and senses substances in the blood. It is, therefore, possible that ghrelin affects AP neurons; however, immunohistochemical study revealed that GHS-R was not observed in AP neurons (22). Taken together, it is still debatable whether circulating ghrelin directly acts on neurons in the medulla to induce gastric motor response. The possibility that peripheral ghrelin affected caudal DVC neurons cannot be rejected.

Perspective and Significance

The present study showed for the first time that central ghrelin induced gastric relaxation of the proximal stomach. The result strongly suggests that ghrelin facilitates accommodation by the stomach of a meal without increasing IGP. Our previous studies also demonstrated that central orexin-A and NPY induced proximal stomach relaxation (15, 18). It is, therefore, hypothesized that anticipatory gastric relaxation to accommodate food might be a common characteristic of orexigenic neuropeptides. This shows new features of the role of orexigenic neuropeptides in autonomic regulation associated with food intake. Fasting is followed by feeding; therefore, gastric relaxation achieved by the actions of orexigenic neuropeptides, which are released by fasting, to enhance reservoir function of the stomach, is rational. Since impaired accommodation of the proximal stomach during and after ingestion of food may be accompanied by increased IGP and intense activation of a gastric mechanoreceptor, thus inducing pathological early satiation (31), orexigenic neuropeptides might be quite important to enable normal healthy satiation.

GRANT

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REFERENCES

5. Date Y, Kojima M, Hosoda H, Sawaguchi A, Mondal MS, Suganuma T, Matsukura S, Kangawa K, Nakazato M. Ghrelin, a novel growth hormone-releasing acylated peptide, is synthesized in a distinct endocrine


