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Plasma ghrelin and gastric pacing in morbidly obese patients

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Abstract

Gastric pacing is a new treatment of morbid obesity. Patients experience increased satiety, the ability to reduce food intake, and a resultant weight loss. We hypothesized that the appetite-stimulating hormone ghrelin is involved in the changed appetite and eating behavior resulting from gastric pacing. Eleven morbidly obese patients (mean body mass index of 46 kg/m^2) were treated with gastric pacing. The peripheral blood levels of ghrelin were studied 1 month before gastric pacer implantation, 1 month after implantation, and 6 months after activation of electrical stimulation. Blood samples were drawn 12 hours after fasting and in response to a hypoenergetic meal (1130 kJ [270 kcal]). Patients were followed monthly for vital signs and weight level. Gastric pacing resulted in a significant weight loss of a mean 10.4 kg or 4.4 body mass index units after 6 months of treatment. No negative side effects or complications were observed during the treatment. Ghrelin levels decreased significantly in response to food intake at all visits. After activation of the pacemaker, levels of ghrelin were significantly increased ($P < .01$) as compared with before activation. Weight loss correlated significantly with increased ghrelin levels ($R = 0.69$, $P < .05$). Gastric pacing is a promising therapy for morbid obesity. It is suggested that increased ghrelin after gastric pacing is an adaptation to negative energy balance without a causal role in weight loss or body weight maintenance.

1. Introduction

Obesity is an increasing worldwide health problem leading to increased morbidity and mortality, particularly with respect to cardiovascular disease and type 2 diabetes mellitus [1]. Diet and antiobesity medication have shown limited efficacy to achieve and sustain weight loss. Surgical procedures such as laparoscopic gastric banding and gastric bypass have shown to be more effective in obtaining weight loss [2]. However, these methods are burdened with complications and side effects [2].

An implantable gastric stimulator system providing myoelectrical stimulation of the stomach has been developed to treat morbid obesity [3-5]. Patients treated with gastric pacing experience increased satiety, the ability to reduce food intake, and a resultant weight loss [5,6]. Recent data in morbidly obese patients confirm that gastric pacing is an effective and safe method avoiding nutritional side effects that are associated with other bariatric operations [7,8]. However, the mechanisms behind changed eating behavior with gastric pacing are still under investigation. We have previously studied satiety signals and demonstrated decreased levels of cholecystokinin, somatostatin, glucagon-like peptide 1, and leptin after 6 months of gastric pacing in obese patients [9].

Ghrelin is a 28–amino acid peptide that was first described as a potent endogenous ligand of the growth hormone secretagogue receptor. The primary source of ghrelin is the stomach and the duodenum, but it is also produced in the pituitary, hypothalamus, liver, kidneys, and placenta [10-13]. Ghrelin is an orexigenic hormone leading to increased food intake both in animals and humans [14]. The mechanisms of action involve the vagus nerve and activation of neurons in the hypothalamus, as supported by animal studies [15,16]. Circulating ghrelin rises before a meal and falls after a meal, suggesting a possible role in the induction of a meal [17]. Furthermore, there is evidence that ghrelin plays a role in regulating long-term energy balance. Ghrelin levels are negatively correlated with body weight [18]. Obese individuals have decreased ghrelin levels [19], which normalize after diet-induced weight loss [20]. These findings suggest that increased ghrelin may serve as a signal to the brain to stimulate eating in conditions of negative energy balance.

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To further investigate possible mechanisms for changed appetite and reduced food intake by gastric pacing, we studied basal and meal-related plasma levels of ghrelin before and after gastric stimulation in heavily obese humans.

2. Materials and methods

2.1. Subjects

Eleven obese patients (9 women and 2 men) were recruited from the Health District of Venice at "Umberto 1st" Hospital in Venice and University of Verona, Italy. The weight inclusion criterion was body mass index (BMI; kg/m^2) of greater than 40. All patients had previously received instruction on diet and lifestyle modifications and were unsuccessful in achieving and maintaining weight loss. Detailed information about the subjects has previously been reported [9]. Briefly, they were all healthy besides obesity and had no history of previous gastrointestinal surgery. None of the patients were taking any medication or had been taking oral contraceptives at least 2 months preceding the investigation. Smokers were excluded. The research protocol was approved by the Ethics Committee of Venice, Italy, and informed consent was obtained from each patient.

2.2. Surgical procedure and gastric pacing

The electrical stimulator system (implantable gastric stimulator [IGS]) is composed of a bipolar electrocatheter (the gastric lead), tunnelized in the gastric wall, and a gastric pacemaker or gastric pacer (battery with a microcircuit) connected to the lead and located outside the abdomen. The IGS was implanted by laparoscopic or open surgery under general anesthesia. The lead was tunnelized intramuscularly at the lesser curve of the anterior gastric wall (upper third of antrum). Intraoperative fiber-optic flexible endogastroscopy was performed to ensure that the mucosa was undamaged during electrode implantation and no intracavity penetration had occurred. After placement of the lead, the gastric pacer was located in a subcutaneous pocket created in the anterior abdominal wall. Gastric pacers and leads were supplied by Transneuronix (Mt Arlington, NJ). The IGS was interrogated using transcutaneous radiofrequency telemetry, which linked the implanted device to a computerized programmer. Telemetric data included programmed electrical stimulation parameters, the impedance of the circuit, and residual battery charge.

Postoperatively, patients were not prescribed any specific diet but encouraged to eat 3 main meals per day or less and to avoid snacks between meals. Ingestion of high-energy beverages and alcohol was discouraged.

2.3. Experimental design

Weight and vital signs were measured before implant and monthly postimplant. Standardized meal tests were performed approximately 1 month before implantation (visit 1), 1 month after implantation just before the activation was started (visit 2), and after about 6 months of electrical stimulation (visit 3). The experiments started in the morning after 12 hours of overnight fasting. An indwelling catheter was inserted into a forearm vein to collect 5 blood samples (10 mL each) during the 5-hour course of the experiment. The first blood sample was drawn at 8:00 AM in a fasting state, then the patients were fed a standardized hypoenergetic meal consisting of 1130 kJ (270 kcal) composed of 12% proteins, 39% lipids, and 49% carbohydrates. Additional blood samples were drawn 30, 60, 120, and 300 minutes after starting to eat. Blood samples were collected in chilled tubes containing aprotinin (500 IU/mL) and EDTA (1 mg/mL). Samples were centrifuged for 10 minutes at 1500*g* and 0°C, and plasma was removed and stored at −80°C in 0.5-mL aliquots until analyzed.

2.4. Analytical methods

Plasma concentrations of ghrelin were measured with a commercial radioimmunoassay (RIA) using iodine 125–labeled bioactive human ghrelin and a rabbit polyclonal antibody raised against full-length octanoylated human ghrelin that recognizes the acylated and des-acyl forms of ghrelin (Phoenix Pharmaceuticals, Belmont, CA). There is no cross-reactivity with human secretin, vasoactive intestinal peptide, prolactin-releasing peptide 31, galanin, growth hormone–releasing hormone, or neuropeptide Y. The detection limit of the assay was 80 pg/mL, the intra-assay variation was 5.4%, and the interassay variation was 9.2%.

2.5. Calculations and statistics

Values are expressed as the mean ± SEM. Ghrelin levels were analyzed by a 2-way analysis of variance with repeated measures on 2 factors, visits (before implantation, after implantation, pace activation) and minutes after food intake (0, 30, 60, 120, and 300 minutes). The total area under the curve (AUC) for ghrelin was also determined. A specific hypothesis about the first 2 visits (before and after implantation) vs the last (after pace activation) is referred to as a priori contrast because the main objective was to study the effects of gastric stimulation on ghrelin levels. Student *t* test was used for planned comparisons between visits. The per-contrast error rate was set to $\alpha = .05$ (the family error rate) as the factor "visit" has only 3 levels. The Pearson coefficient of correlation was calculated to evaluate the association between variables. $P < .05$ was considered statistically significant.

3. Results

The patients' age was 39.4 ± 3.4 years. Gastric pacing resulted in a significant weight loss and drop in BMI 6 months after electrical stimulation ($P < .01$) (Table 1). No clinical complications occurred during the course of the study, and there were no failures of the gastric stimulator

Table 1
Weight and BMI of the patients at the different visits

	Visit 1 before implantation	Visit 2 after implantation	Visit 3 after pace activation
Weight (kg)	121.7 ± 5.1	117.6 ± 4.7	110.2 ± 5.4 *
BMI kg/m^2)	46.0 ± 2.5	44.5 ± 2.4	41.3 ± 1.8 *

Values are mean ± SEM.
* $P < .01$, significant differences between visit 3 (6 months of pace activation) and visit 1 (before implantation).

system or the lead. Surgically, there was 1 reoperation for bleeding and 1 to revise the generator pocket.

Ghrelin levels were significantly decreased in response to food intake at all visits ($P < .001$) (Fig. 1), and there was no difference in the meal-related response between visits because the interaction visit × minute was not statistically significant ($P = .50$). Furthermore, fasting levels of ghrelin were not significantly different between visits ($P = .14$). However, repeated-measure curves of ghrelin were significantly increased 6 months after activation of the pacemaker compared with before activation ($P < .01$), as well as compared with the combined responses at the first 2 visits ($P < .05$) (Fig. 1). The integrated ghrelin response (AUC) was also significantly increased after pace activation than before activation ($P < .01$) and compared with the combined responses at the first 2 visits ($P < .05$) (Fig. 2). Weight loss (visit 1 − visit 3) correlated significantly with increased ghrelin levels (total AUC for ghrelin at visit 1 − visit 3) ($R = 0.69$, $P < .05$).

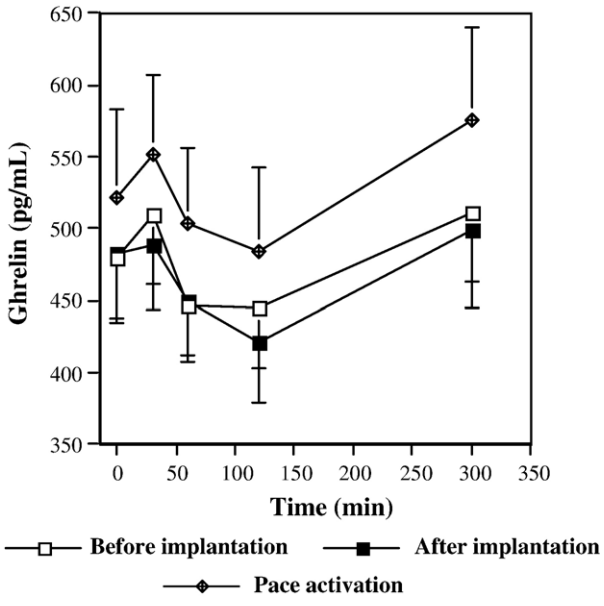

Fig. 1. Plasma ghrelin levels in response to a test meal in patients before implantation, after implantation, and after activation of the gastric pacemaker. The meal begins at 0 minute. Values are mean ± SEM. Repeated-measure curves were significantly increased after activation of the pacemaker than before activation ($P < .01$).

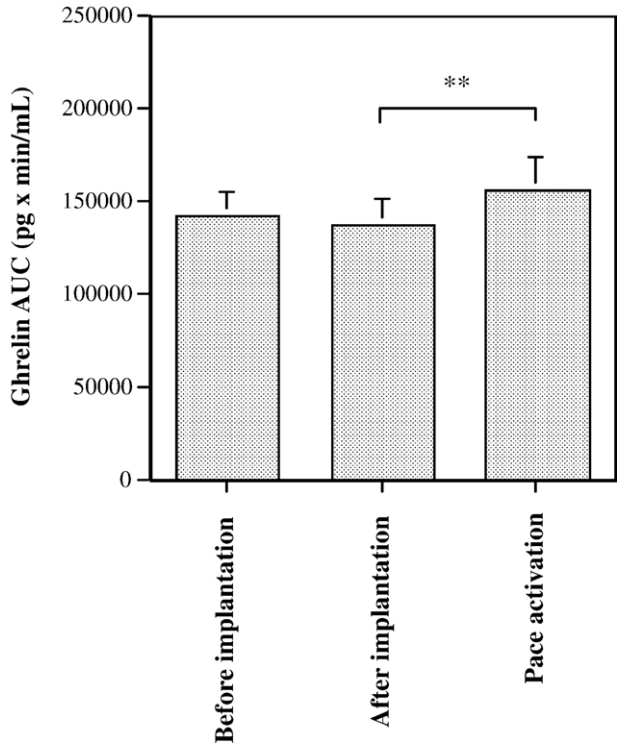

Fig. 2. Integrated response (AUC) of ghrelin in patients before implantation, after implantation, and after activation of the gastric pacemaker. Values are mean ± SEM. The integrated response was significantly increased after pace activation than before activation ($P < .01$).

4. Discussion

In this study, we demonstrate a significant general increase of plasma ghrelin levels in relation to weight loss after 6 months of gastric pacing in morbidly obese patients. It has previously been reported that the postprandial decrease in ghrelin is attenuated in obese humans and normalize after weight loss [21]. However, we found a significant decrease in the response of ghrelin at all visits.

Our findings of increased ghrelin levels are not in agreement with the recent report by De Luca et al [22] showing unchanged ghrelin levels after weight loss by gastric pacing. We have no clear explanation for the discrepant results, but it may be explained by the use of different analytical methods for ghrelin determination. We used a ghrelin RIA kit detecting total ghrelin, including acylated ghrelin and the nonacylated form, whereas in the article of De Luca et al [22], they refer to an RIA detecting only the acylated form of ghrelin. The acylated form of ghrelin is considered to exert the pituitary and pancreatic endocrine activities of human ghrelin. Recently, it was also shown that nonacylated ghrelin is metabolically active and is likely to counterbalance the influence of acylated ghrelin on

insulin secretion and glucose metabolism [23]. It remains to be determined to what extent the changes in ghrelin levels with weight loss represents changes in the acylated and/or the nonacylated form.

The present study cannot determine whether the increased levels of ghrelin are a result of gastric pacing per se or if they are secondary to weight loss. Both animal and human studies have demonstrated that ghrelin secretion is influenced by vagal nerve activity [15,24-26]. Although gastric pacing does not directly stimulate the vagus nerve, there are indications of changed parasympathetic activity by gastric pacing. Thus, treatment of patients has shown increased gastric mucous production and reduced blood pressure to a larger extent than expected from weight loss [7]. Furthermore, animal studies support an increased vagal activity by short-pulse gastric electrical stimulation [27]. It is therefore possible that gastric pacing might directly regulate the secretion of ghrelin.

Most weight reduction procedures are accompanied by an increase in ghrelin, such as after diet [20], gastric banding [28-30], and vertical banded gastroplasty [31], which has been suggested to represent a normal response of ghrelin to weight loss as an adaptation to negative energy balance. This could also be the mechanism by which gastric pacing results in increased ghrelin levels. In contrast, plasma ghrelin normally decreases after gastric bypass including both restrictive and malabsorptive functions [20,31-35]. Gastric bypass disconnects the ghrelin-producing cells in fundus from contact with enteral nutrients, which may explain why this treatment results in lower ghrelin levels.

Because ghrelin is suggested to play a role in long-term weight maintenance [19], it has been hypothesized that a decrease in ghrelin could contribute to maintaining weight loss after intervention. In this study, we followed the patients for 6 months, but the same patients were also included in a long-term follow-up. At 44 months of follow-up, the mean percentage of excess BMI (>25) loss was 25% [7]. Thus, increased ghrelin levels after 6 months of gastric pacing seem not to negatively influence the long-term efficacy of gastric pacing as treatment of morbid obesity.

The mechanisms leading to weight loss after gastric pacing remain under investigation. Enhanced neuroendocrine satiety mechanisms [36] or changed gastrointestinal motor activity have been suggested. We have previously investigated the involvement of certain satiety signals and found reduced plasma levels of cholecystokinin, somatostatin, glucagon-like peptide 1, and leptin levels associated with activation of the gastric pacemaker in heavily obese patients [9]. These responses were probably secondary to reduced food intake and weight loss and do not determine weight loss achieved by gastric pacing. In this study, we investigated if the appetite-stimulating hormone ghrelin could be of importance for weight loss by gastric pacing. An increase in ghrelin levels correlated with weight loss. However, increased ghrelin cannot explain increased satiety in patients treated with gastric pacing. Because it has been demonstrated that ghrelin stimulates gastric motility [37], it cannot be ruled out that such effect could be involved in the mechanism by which gastric pacing results in weight loss.

In conclusion, we found increased plasma levels of ghrelin by gastric pacing in morbidly obese patients after weight loss. This might be a normal response of ghrelin to body weight loss as an adaptation to negative energy balance to stimulate food intake. Because long-term follow-up of our patients has demonstrated persistent weight reduction, ghrelin seems not to have a causal role in weight loss or body weight maintenance by gastric pacing. Further studies are needed to elucidate the mechanisms by which gastric pacing induces increased satiety and weight loss in obese patients.

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