Obesity Surgery, 16, 870-878

Is Ghrelin the Culprit for Weight Loss after Gastric Bypass Surgery? A Negative Answer

Marta E. Couce[2]; Daniel Cottam[3]; James Esplen[2]; Phillip Schauer[3]; Bartolome Burguera[1]

[1]Division of Endocrinology, [2]Division of Neuropathology, [3]Department of Surgery, University of Pittsburgh School of Medicine, Pittsburgh, PA, USA

Background: Ghrelin is a potent appetite stimulator, mainly synthesized in the stomach. Paradoxically, obese subjects have lower plasma ghrelin than lean subjects and increase their weight in spite of low ghrelin levels. The role of ghrelin in weight regulation after bariatric surgery is still controversial. The aim of this study was to evaluate whether rapid weight loss after laparoscopic Roux-en-Y gastric bypass surgery (LRYGBP), was associated with changes in plasma ghrelin levels. In addition, we determined the acute impact of LRYGBP on insulin resistance and adiponectin levels.

Methods: 49 morbidly obese subjects who underwent LRYGBP were studied. 19 subjects who underwent other laparoscopic gastrointestinal surgeries acted as the control group. Fasting plasma levels of ghrelin, insulin and adiponectin were determined preoperatively and 2 hours, 10 days and 6 months postoperatively.

Results: At 2 hours after LRYGBP, there was a significant reduction in ghrelin and adiponectin levels, which coincided with elevated plasma glucose and insulin levels. Interestingly, once glucose and insulin levels normalized at 6 months after surgery, ghrelin also normalized. Adiponectin reached pre-surgical levels at 10 days after LRYGBP and continued to significantly rise until 6 months postoperatively.

Conclusion: Weight loss after LRYGBP occurs in spite of the absence of significant changes in plasma ghrelin levels. Improvement of insulin resistance occurred within 10 days after surgery, and could be related to the normalization of adiponectin levels. This data questions the role of peripheral ghrelin as a cause of weight loss in obese humans after LRYGBP.

Key words: Ghrelin, obesity, gastric bypass surgery, insulin, adiponectin, appetite

Current address and reprint requests to : B. Burguera, MD, PhD, Servicio de Endocrinologia, Hospital Universitario Son Dureta, C/ Andrea Doria 55, Palma de Mallorca 07014, Spain.
E-mail: burguerab@telefonica.net

Introduction

Obesity is an epidemic, and in the U.S.A. 7 million individuals are classified as severely or morbidly obese.[1,2] Complications of obesity include dyslipidemia, hypertension, type 2 diabetes mellitus, cardiovascular disease, obstructive sleep apnea, certain cancers, and degenerative joint disease.[2-4] The NIH and international agencies have determined that bariatric surgery may be the only effective treatment for severely obese individuals.[5] Laparoscopic Roux-en-Y gastric bypass (LRYGBP) results in dramatic weight loss, with an average loss of 45 kg during the first 12 to 24 months.[6-8] This substantial weight loss attained with bariatric surgery has major impacts upon the co-morbidities associated with obesity.[6-10]

Ghrelin is the endogenous ligand for the GH secretagogue receptor[11] and also an important appetite-stimulating hormone.[12-14] Ghrelin is produced predominantly by the stomach, but also in the hypothalamus.[11,15-17] Ghrelin administration increases food intake in rats[13,14] and in humans,[18] and ghrelin levels are reduced in obese subjects.[19-21] Other important peripheral signals, such as insulin and adiponectin, have been implicated in the regulation of appetite and plasma ghrelin levels.[22,23] In obese humans, plasma adiponectin is low,[24-26] and low levels of adiponectin have been proposed to contribute to insulin resistance associated with obesity.[27] Hyperinsulinemia, which is usually associated with obesity, has been suggested to be responsible for the reduced ghrelin levels seen in obese individuals.[28,29] Although the mechanism(s) by which LRYGBP induces long-term weight loss are not well under-

stood, it has recently been hypothesized that reduction of circulating ghrelin may contribute to the success of weight loss after LRYGBP.[30] Whereas several studies have shown that weight loss achieved after gastric bypass surgery was associated with decreased ghrelin levels,[30-33] a significant number of studies have also described that circulating ghrelin remained unchanged[34,35] or increased after gastric bypass surgery.[34,36]

In an effort to contribute to the better understanding of the specific role that ghrelin may play in the weight loss after gastric bypass, we have evaluated in a prospective study, the acute impact of LRYGBP on ghrelin, insulin and adiponectin in morbidly obese subjects.

Materials and Methods

In Vivo Studies

This study was carried out on 49 morbidly obese subjects (30 females and 19 males) who underwent LRYGBP for the treatment of morbid obesity at the University of Pittsburgh Medical Center (UPMC). The details of this surgery have been previously described.[8] These patients had a mean age of 46.3 ± 2.2 years, mean weight of 142.2 ± 4.4 kg and mean BMI of 50.0 ± 5.3 kg/m^2. Twenty of these patients were on medical treatment for type 2 diabetes, 8 received oral therapy (metformin and sulfanylureas), and 12 were treated with oral therapy plus insulin. These patients were on a clear liquid diet 48 hours prior to surgery, and their diabetic medications were tapered or discontinued 24 hours prior to surgery. In addition, 19 subjects (9 females and 10 males), with a mean age of 56.6 ± 3.6 years, mean weight of 88.3 ± 7.1 kg and mean BMI of 29.8 ± 3.1 kg/m^2, who underwent other laparoscopic gastrointestinal surgeries (13 cholecystectomies and 6 inguinal hernias) at the UPMC by the same surgeons, acted as a control group. The exclusion criteria for the study included history of acute infection or injury, current or past history of chronic liver or kidney disease, history of congestive heart failure, current pregnancy, or substance abuse. Blood samples from both groups were collected 1 hour preoperatively, then 2 hours, 10 days, and 6 months postoperatively. Subjects were recruited from our Bariatric and Surgical Clinics at UPMC. All study procedures were in accordance with the Helsinki convention and approved by the University of Pittsburgh Institutional Review Committee. All blood samples were collected between 7 and 8 AM in the fasting state. Sera were centrifugally separated, aliquoted and stored at -70°C until being analyzed for levels of ghrelin, glucose, insulin, and adiponectin. Pre- and postoperative anthropometric measurements were obtained in all patients at different time points and are summarized in Table 1.

Hormonal Assay

Serum ghrelin was measured in duplicate, using a commercially available radioimmunoassay (RIA) for total ghrelin, according to the manufacturer's instructions (Phoenix Pharmaceuticals). Phoenix's human ghrelin RIA kit detects full-length, desoctanoyl human ghrelin, including Ser3-octanoyl and Ser-desoctanoyl ghrelin. Ghrelin intra- and inter-assay coefficients of variation (CoV) were: low=13.4, high=3.0 and low=8.7, high=7.2, respectively. Plasma adiponectin was measured using an RIA for human adiponectin (Linco Research) with intra- and inter-assay CoV low=6.1, high=5.7 and low=14.5, high=12, respectively. Insulin was measured with a human insulin-specific RIA kit (Linco Research) with intra- and inter-assay CoV low=4.3, med=2.4, high=3.8 and low=6.4, med=7.5, high=6.7, respectively. Plasma glucose was determined by the glucose oxidase method (Autoanalyzer, Beckman Coulter). Fasting plasma glucose and insulin levels were used to calculate the insulin resistance by the Homeostasis Model Assessment index (HOMA-IR). HOMA-IR was calculated as previously described[36] using the formula: fasting serum insulin (U/ml) X fasting serum glucose (mmol/liter)/22.5.

Statistical Analysis

Results are expressed as mean ± SEM. Data is compared using a two-tailed, paired *t*-test (pre- vs postoperative) and two-sample Student's *t*-test. Statistical analysis was performed using SigmaStat with significance set at $P<0.05$ and a power of >80%.

Table 1. Demographic characteristics and hormonal profile of surgery subjects

Preop vs 2 Hours	LRYGBP (n=49, 30 female)		Control (n=19, 9 female)	
	Preoperative	2 Hours	Preoperative	2 Hours
Age (yr)	46.3 ± 2.2	+	56.3 ± 3.6	+
Body Mass Index†	50.0 ± 5.3	+	29.8±3.1	+
Ghrelin (pg/mL)	932.4 ± 52.2	713.3 ± 40.4**	1245.1 ± 128.2	891.6 ± 93.7*
Adiponectin (µg/mL)	9.0 ± 1.1	8.0 ± 1.1**	9.0 ± 5.2	7.9 ± 1.2**
Insulin (µU/mL)	25.5 ± 3.2	35.2 ± 3.4*	13.5 ± 2.6	16.0 ± 3.2
Glucose (mg/dL)	101.9 ± 8.4	154.2 ± 16.0*	80.4 ± 10.5	120.4 ± 11.5**
Preop vs 10 Days	LRYGBP (n=18, 14 female)		Control (n=8, 5 female)	
	Preoperative	10 Days	Preoperative	10 Days
Age (yr)	45.6 ± 2.3	+	48.8 ± 6.1	+
Body Mass Index†	48.8 ± 6.1	47.4 ± 6.4	28.7±2.8	28.2±2.5
Ghrelin (pg/mL)	1015.0 ± 67.7	875.2 ± 45.5*	1067.0 ± 159.3	1037.0 ± 171.1
Adiponectin (µg/mL)	8.2 ± 1.1	7.6 ± 1.0	7.8 ± 1.5	8.0 ± 2.0
Insulin (µU/mL)	29.9 ± 4.4	19.0 ± 2.9**	16.0 ± 5.0	12.7 ± 7.0
Glucose (mg/dL)	107.8 ± 11.8	83.1 ± 4.4*	79.6 ± 12.9	83.9 ± 19.6
HOMA Index	7.9± 0.7	3.8*± 0.3	3.1± 0.4	2.6± 0.3
Preop vs 6 Months	LRYGBP (n=11, 8 female)		Control (n=10, 8 female)	
	Preoperative	6 Months	Preoperative	6 Months
Age (yr)	47.2 ± 3.3	+	47.5 ± 4.5	+
Body Mass Index†	54.3 ± 6.0	39.8 ± 1.5**	29.1±2.6	30.8±3.9
Ghrelin (pg/mL)	802.3 ± 79.6	622.7 ± 59.4	1214.5 ± 81.3	1051.8 ± 135.5
Adiponectin (µg/mL)	7.9 ± 1.8	11.7 ± 1.5**	5.9 ± 4.6	5.0 ± 4.2
Insulin (µU/mL)	23.7 ± 4.8	8.4 ± 0.82*	14.4 ± 8.4	13.0 ± 10.1
Glucose (mg/dL)	94.8 ± 10.3	86.2 ± 1.5*	82.0 ± 16.7	81.3 ± 17.4
HOMA Index	5.5± 0.7	1.8**± 0.4	2.9± 0.5	2.6± 0.4

Mean values (±SE) are shown.
+Age, 2 hour weight, and 2 hour BMI did not change from preoperative and are therefore not shown.
†Body Mass Index (BMI) is calculated as the weight of the patient in kilograms divided by the square of the patients' height in meters (kg/m^2).
*Denotes ($P<0.05$) student's *t*-test
**Denotes ($P<0.01$) student's *t*-test

Results

Plasma Ghrelin Levels after LRYGBP

Fasting plasma ghrelin levels were measured in patients having LRYGBP and in obese control patients undergoing other laparoscopic abdominal surgeries, preoperatively and at 2 hours, 10 days and 6 months postoperatively. The preoperative plasma ghrelin was lower in the LRYGBP group compared to the control group (Figure 1). These results would be expected, given the higher BMI in the LRYGBP group (Table 1). At 2 hours postoperatively, there was a statistically significant drop in ghrelin levels in both the LRYGBP ($P<0.001$) and control groups ($P<0.001$). At 10 days after surgery, plasma ghrelin levels continued to be reduced in the LRYGBP group, whereas ghrelin levels had normalized in the control group. At 6 months after LRYGBP, morbidly obese patients had plasma levels of ghrelin that were not significantly different compared to pre-

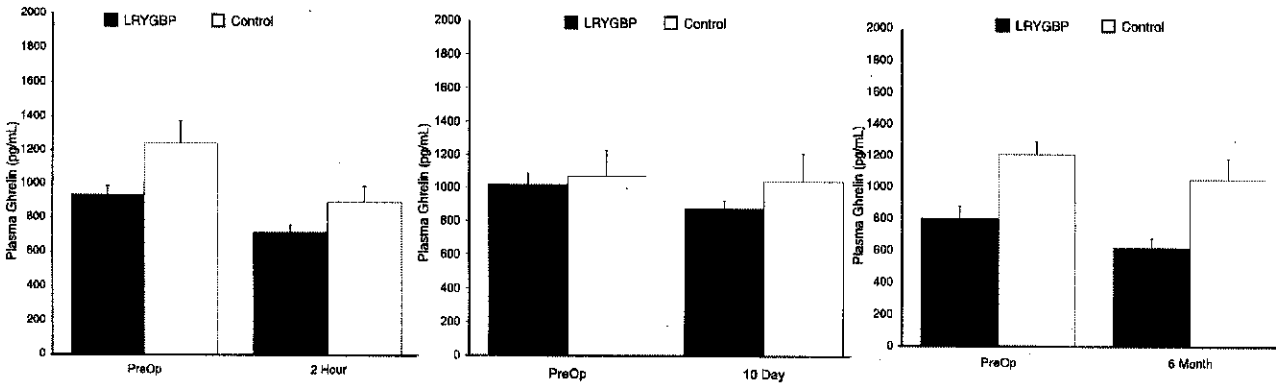

Figure 1. Radioimmunoassay Analysis of Plasma Ghrelin in LRYGBP and Control Subjects during a 6 months follow-up after surgery. Plasma ghrelin was measured in 48 LRYGBP and 19 control patients: preoperatively (baseline), and at 2 hours, 10 days, and 6 months after surgery. Data is expressed as mean ±SE. Paired student's *t*-tests were used to determine statistical significance in all LRYGBP comparisons and for 2 hour and 10 day comparisons in controls ($P<0.05^*$, $P<0.01^{**}$). Independent student's *t*-test was used for 6-month controls ($P<0.05^*$).

surgery values ($P<0.08$). This happened in spite of significant weight loss in these patients, in whom BMI had by decreased 20% (Figure 1).

Plasma Glucose and Insulin after LRYGBP

Morbidly obese patients showed a mean plasma glucose which was significantly higher compared to the control surgical group ($P<0.03$) (Figure 2A). There were 20 patients with type 2 diabetes who had their oral medications discontinued 24 hrs prior to surgery. The mean plasma glucose significantly increased in both the LRYGBP ($P<0.01$) and control groups ($P<0.001$) when measured 2 hours after surgery, but this is not unexpected as patients were given intravenous infusion of 5% dextrose and 1/2 normal saline during surgery. Ten days after surgery, the mean plasma glucose in the LRYGBP group had dropped to normal values similar to the control group. Mean plasma glucose in the LRYGBP group continued to be normal 6 months after surgery. Mean pre-surgical insulin levels were significantly higher in the LRYGBP group compared to the control group ($P<0.004$). Insulin levels increased in both groups of patients 2 hours after surgery ($P<0.03$) (Figure 2B); however, only the LRYGBP group showed a significant increase when compared to baseline insulin levels. Ten days after surgery, insulin had significantly dropped in the LRYGBP group (36.4%) ($P<0.001$), reaching normal fasting levels. This insulin reduction in the LRYGBP group was even more pronounced at 6 months after surgery (64.7%) ($P<0.01$), coinciding with a normalization of ghrelin levels at this point (622.7 ± 59.4 pg/mL).There was a significant improvement in insulin sensitivity as measured by HOMA index at 10 days and 6 months after surgery (Table 1).

Plasma Adiponectin Levels after LRYGBP

Fasting plasma adiponectin was similar in both surgical groups before surgery. Our data showed a significant drop in plasma adiponectin at 2 hours after surgery in both the LRYGBP ($P<0.004$) and control ($P<0.001$) groups (Figure 2C). Adiponectin levels were back to within normal range for both groups at 10 days. At 6 months, plasma adiponectin in LRYGBP was significantly higher than the preoperative level ($P<0.001$). Interestingly, we noticed that the changes in plasma adiponectin concentration after LRYGBP were a virtual mirror image of plasma fluctuations for insulin.

Discussion

Our studies provide the first time-course record of plasma ghrelin changes over a 6-month period after LRYGBP. In agreement with previous studies, we noticed that plasma ghrelin was significantly lower in morbidly obese compared to lean subjects.[19-21]

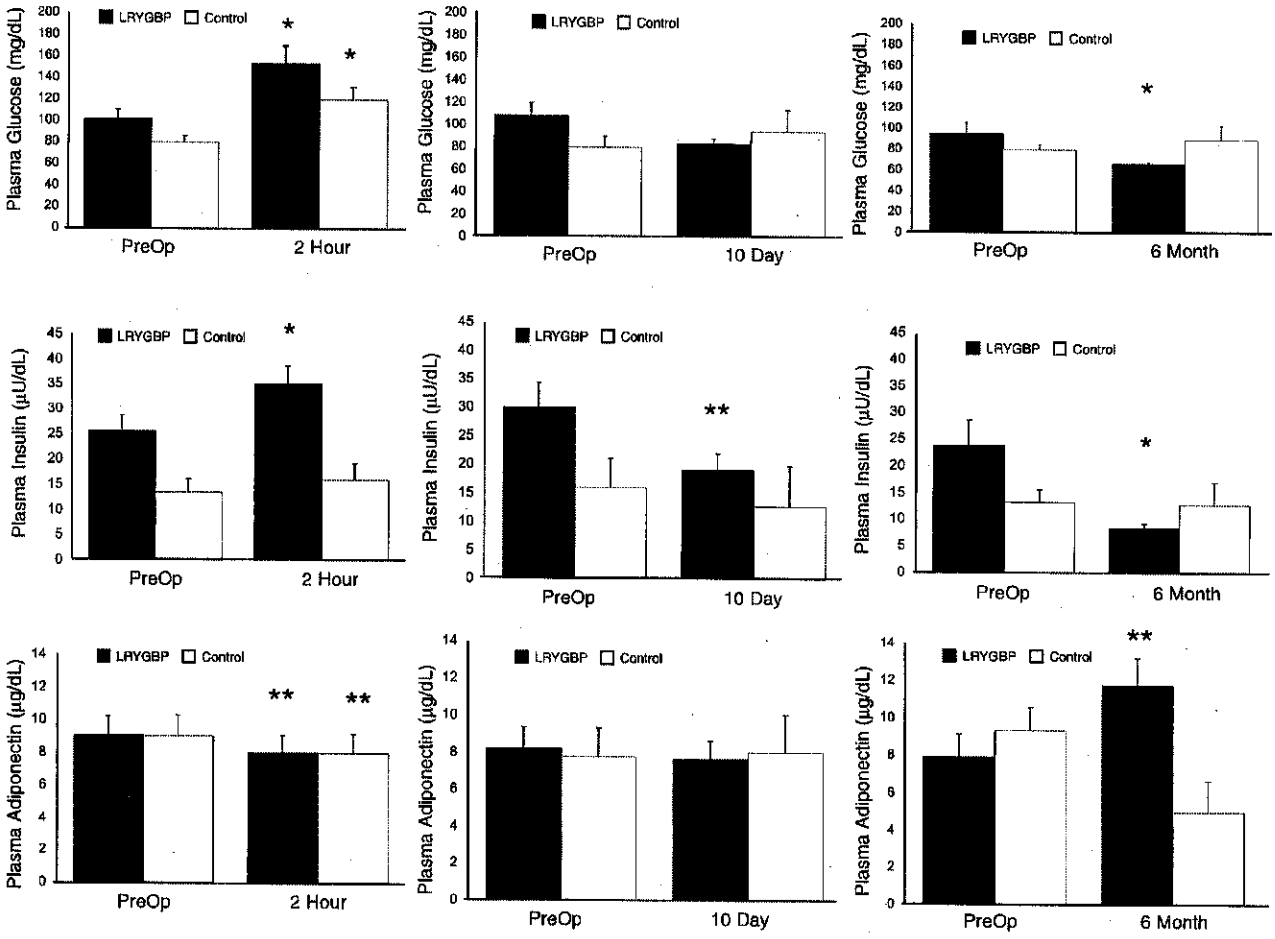

Figure 2. Radioimmunoassay Analysis of Plasma Glucose, Insulin, and Adiponectin in LRYGBP and Control Subjects during 6 months follow-up after surgery. Preoperative (baseline) plasma adiponectin (μg/mL) glucose (mg/dL), and insulin (μU/mL) from 49 LRYGBP and 17 control patients were compared to their respective levels at 2 hours, 10 days or 6 months after surgery. Data is expressed as mean ±SE. Paired student's *t*-test was used to determine statistical significance in all LRYGBP comparisons and for 2 hour and 10 day comparisons in controls ($P<0.05^*$, $P<0.01^{**}$). Independent student's *t*-test was used for 6 month contols ($P<0.05^*$).

Plasma ghrelin significantly fell 2 hours after LRYGBP. Interestingly, we saw a similar acute ghrelin drop in the control surgical group undergoing other gastrointestinal surgical operations. Plasma ghrelin continued to be low in the LRYGBP group 10 days after surgery, whereas they had almost returned to pre-surgical values by 6 months.

Ghrelin seems to be implicated, in both the regulation of appetite and body weight.[12,13] However, there is still significant controversy in the literature regarding the role that ghrelin might play in the regulation of appetite in patients who have undergone bariatric surgery (Table 2). Contrary to what would be expected, plasma levels of this orexigenic hormone are lower in obese individuals compared to lean subjects;[19-21] and circulating ghrelin increases under negative energy balance circumstances, such as low-calorie diets.[37] Cummings et al[30] showed that plasma ghrelin was suppressed 9-31 months after gastric bypass surgery. Based on this early data, it has been suggested that the weight-reducing efficacy of gastric bypass surgery could be partially explained by its effect reducing circulating ghrelin levels. Similarly, Tritos et al[31] showed that serum ghrelin was lower in obese subjects 18 months after gastric bypass surgery, compared to obese subjects

Table 2. Impact of the different types of bariatric operations on plasma ghrelin levels (GBP: Gastric bypass, BPD: Biliopancreatic diversion, GB: Gastric banding, LSG: Laparoscopic sleeve gastrectomy, VBG: Vertical banded gastropasty, NC: no change)

Type of surgery	Ghrelin levels	Short-term after surgery	Long-term after surgery	Type of study	Author
GBP	decreased		9-31 months	cross-sectional	Cummings[30]
GBP	decreased		18 months	cross-sectional	Tritos[31]
GBP	decreased		12 months	prospective	Geloneze[32]
GBP	decreased		6 months	prospective	Frühbeck[33]
GBP	decreased	6 weeks		prospective	Morinigo[38]
GBP	decreased	1 day		prospective	Lin[41]
GBP	NC		>12 months	cross-sectional	Korner[39]
GBP	NC		12 months	prospective	Stoeckli[40]
GBP	NC/increased		6 months	prospective	Faraj[34]
GBP	increased		6 months	prospective	Holdstock[35]
LSG	decreased	4 weeks	6 months	prospective	Langer[55]
BPD	decreased	5 days		prospective	Adami[58]
BPD	increased		12 moths	prospective	Adami[48]
BPD	increased		3-12 months	prospective	Garcia[49]
GB	decreased		9-15 months	cross-sectional	Leonetti[56]
GB	NC		6-12 months	prospective	Hanusch[44]
GB	NC		18-36 months	cross-sectional	Dixon[47]
GB	NC		6-14 months	prospective	Ram[57]
GB	increased		6 months	prospective	Schindler[45]
GB	increased		12-24 months	prospective	Nijhuis[46]
GB	increased		6 months	prospective	Frühbeck[33]
GB	increased		6-12 months	prospective	Stoeckli[40]
GB	increased		6 months	prospective	Langer[55]
VBG	increased		4 months	prospective	Foschi[54]

in response to an oral glucose tolerance test. In addition to these two cross-sectional studies, Geloneze et al[32] described in a prospective study, that the mean plasma ghrelin levels decreased significantly after surgery in obese non-diabetics and obese type 2 diabetic subjects, 12 months after gastric bypass surgery. Frühbeck et al[33] also showed that LRYGBP caused a significant reduction in ghrelin levels in morbidly obese patients 6 months after the surgery. The acute impact of gastric bypass surgery on plasma ghrelin levels was also evaluated by Morinigo et al,[38] who measured plasma ghrelin in eight morbidly obese patients before and 6 weeks after LRYGBP. These authors found that, despite a significant weight loss, fasting serum ghrelin was decreased in obese subjects 6 weeks after LRYGBP.

In contrast to these studies, Faraj et al[34] showed in a prospective study, that circulating ghrelin did not decrease 6 months after gastric bypass surgery. In fact, plasma ghrelin increased postoperatively in the subset of subjects undergoing active weight loss, whereas ghrelin remained unchanged in weight-stable subjects. Holdstock et al[35] also reported that serum ghrelin significantly increased (by +58%) 12 months after RYGBP. In agreement with these studies, Korner et al[39] showed that fasting plasma ghrelin was nearly identical between subjects who had undergone RYGBP and matched controls. Supporting these studies, Stoeckli et al[40] found that plasma ghrelin levels did not change during substantial weight loss 24 months after RYGBP.

The discrepancies among these different studies (Table 2) may be partially explained by analyzing three different issues. The first variable that needs to

be taken into consideration is the difference in follow-up time after the gastric bypass operation. It seems that short-term follow-up after LRYGBP is associated with acute reductions of plasma ghrelin as suggested by Morinigo et al,[38] Lin et al[41] and our studies (6 weeks, 1 day and 10 days after surgery, respectively). However, the long-term follow-up data collection after surgery (>3 months) varies among the different studies published (Table 2). Our studies showed that plasma ghrelin levels were not significantly different when measured 6 months after LRYGBP, in agreement with Faraj et al[34] and Holdstock et al[35].

Secondly, it has been shown that weight loss in obese individuals is associated with increasing plasma ghrelin levels. Cummings et al[30] showed that a diet-induced weight loss of 17% of initial body weight, was associated with a 24% increase in the area under the curve for the 24-hour ghrelin profile in obese subjects. However, 5% weight loss had no effect on plasma ghrelin levels in obese subjects that followed 3 weeks of an integrated weight loss program.[42] These findings could explain why we see low ghrelin levels 10 days after LRYGBP, when there is still little weight loss, compared to the higher ghrelin levels at 6 months after surgery when patients have lost between 10-30% of their original body weight. LRYGBP seems to be causing reduction of ghrelin levels; however, active weight loss "normalizes" plasma ghrelin.

Thirdly, the small variants within the gastric bypass surgical technique, such as the size of the gastric pouch, length of the Roux limb and additional vagotomy, may also be partially responsible for the differences in ghrelin levels documented in the published studies. Interestingly, truncal vagotomy is known to cause reduced food intake and weight loss in humans, and ghrelin administration did not stimulate food intake in patients with surgical procedures involving vagotomy.[43] Regardless of what is the explanation(s) responsible for these conflicting data, a clear evidence emerges from all these studies (Table 2): morbidly obese patients lose weight after undergoing gastric bypass regardless of their plasma ghrelin levels after surgery.

The notion that variations in ghrelin concentrations may have little impact on causing weight loss after gastric bypass is further supported by three different sets of studies: The first evidence comes from morbidly obese subjects who underwent laparoscopic adjustable gastric banding (LAGB). Several studies have shown that adjustable silicone gastric banding (ASGB) caused significant weight loss, in spite of causing an increase in plasma ghrelin concentrations after the operation.[33,44-6] It seems that changes in eating behavior, which promote reduction of food intake and not fasting ghrelin levels, determine weight loss achieved by adjustable gastric banding. Studies modifying the degree of band restriction around the stomach, showed that optimal LAGB restriction increased fasting and postprandial satiety. However, plasma ghrelin levels appeared unrelated to the LAGB satiety effect.[47]

A second set of evidence comes from studies using another bariatric surgical procedure, the Scopinaro biliopancreatic diversion (BPD). Adami et al[48] showed similar serum ghrelin concentrations before and 2 months following BPD, whereas plasma ghrelin increased significantly 1 year after BPD.[48,49] Lastly, if ghrelin were to play a significant role in the weight loss after bariatric surgery, it would be expected that the amount of weight lost after gastric bypass should correlate with plasma ghrelin levels. Interestingly, Christou et al[50] showed that the failure to lose weight after gastric bypass surgery did not correlate with pre- or post-prandial plasma ghrelin. They also reported that ghrelin levels did not correlate with satiety. Taken together, these different studies question the role of ghrelin as the main culprit for weight loss after RYGBP.

In addition to ghrelin, we also evaluated the impact of LRYGBP surgery on plasma insulin and adiponectin. Several lines of evidence suggest that insulin may regulate the production of ghrelin. Insulin concentrations are higher in obese individuals with insulin resistance, whereas ghrelin levels are low. Insulin administration has also been shown to decrease ghrelin concentrations in humans.[28,29] In addition, the changes in plasma insulin concentrations, before and after meals, are a virtual mirror image of fluctuations in ghrelin concentrations.[51]

Our studies show that there is a significant elevation of plasma insulin levels 2 hours after LRYGBP surgery, which is associated with low ghrelin and adiponectin levels. Interestingly, plasma insulin levels like ghrelin, normalized at 6 months after surgery. A similar situation occurred with adiponectin, which also normalized at 10 days after surgery and continued to significantly rise at 6 months after surgery.

Adiponectin therapy has been associated with improvement of insulin resistance[52,53] and reduction of appetite. To what degree this reduction in insulin after LRYGBP could be responsible for the maintenance of weight loss, rise of adiponectin and improvement of insulin sensitivity, is a matter of great interest which deserves further studies. Adami et al[48] showed a negative correlation between the postoperative changes of serum ghrelin concentration and insulin sensitivity 2 and 12 months after BPD. Based on our results, we hypothesize that insulin has a possible causal role in modulating ghrelin and adiponectin secretion after LRYGBP surgery. Further studies are necessary to investigate whether a negative energy balance or changes in other gastrointestinal hormones, such GLP or PYY, could also be involved in the normalization of plasma insulin soon after bariatric surgery and before a significant weight loss has been accomplished.

In summary, our study shows that weight loss after LRYGBP occurs in spite of the absence of significant changes in plasma ghrelin levels. This data seriously questions the potential role of ghrelin as responsible for the weight loss after bariatric surgery.

We thank the John F. & Nancy A. Emmerling Fund of The Pittsburgh Foundation and The University of Pittsburgh Obesity and Nutrition Research Center for their generous financial support.

References

1. Sugerman HJ. The epidemic of severe obesity: the value of surgical treatment. Mayo Clin Proc 2000; 75: 669-72.
2. Must A, Spadano J, Coakley EH et al. The disease burden associated with overweight and obesity. JAMA 1999; 282: 1523-9.
3. Jung RT. Obesity as a disease. Br Med Bull 1997; 53: 307-21.
4. Field AE, Coakley EH, Must A et al. Impact of overweight on the risk of developing common chronic diseases during a 10-year period. Arch Intern Med 2001; 161: 1581-6.
5. Gastrointestinal surgery for severe obesity: National Institutes of Health Consensus Development Conference Draft Statement. Obes Surg 1991; 1: 257-65.
6. Pories WJ, Swanson MS, MacDonald KG et al. Who would have thought it? An operation proves to be the most effective therapy for adult-onset diabetes mellitus. Ann Surg 1995; 222: 339-50; discussion 350-52.
7. Sjostrom CD, Lissner L, Wedel H et al. Reduction in incidence of diabetes, hypertension and lipid disturbances after intentional weight loss induced by bariatric surgery: the SOS Intervention Study. Obes Res 1999; 7: 477-84.
8. Schauer PR, Burguera B, Ikramuddin S et al. Effect of laparoscopic Roux-en-Y gastric bypass on type 2 diabetes mellitus. Ann Surg 2003; 238: 467-84.
9. Scopinaro N, Adami GF, Marinari GM et al. Biliopancreatic diversion. World J Surg 1998; 22: 936-46.
10. Brolin RE, Kenler HA, Gorman RC et al. The dilemma of outcome assessment after operations for morbid obesity. Surgery 1989; 105: 337-46.
11. Kojima M, Hosoda H, Date Y et al. Ghrelin is a growth-hormone-releasing acylated peptide from stomach. Nature 1999; 402: 656-60.
12. Wren AM, Small CJ, Ward HL et al. The novel hypothalamic peptide ghrelin stimulates food intake and growth hormone secretion. Endocrinology 2000; 141: 4325-8.
13. Tschop M, Smiley DL, Heiman ML. Ghrelin induces adiposity in rodents. Nature 2000; 407: 908-13.
14. Nakazato M, Murakami N, Date Y et al. A role for ghrelin in the central regulation of feeding. Nature 2001; 409: 194-8.
15. Date Y, Kojima M, Hosoda H et al. Ghrelin, a novel growth hormone-releasing acylated peptide, is synthesized in a distinct endocrine cell type in the gastrointestinal tracts of rats and humans. Endocrinology 2000; 141: 4255-61.
16. Ariyasu H, Takaya K, Tagami T et al. Stomach is a major source of circulating ghrelin, and feeding state determines plasma ghrelin-like immunoreactivity levels in humans. J Clin Endocrinol Metab 2001; 86: 4753-8.
17. Gnanapavan S, Kola B, Bustin SA et al. The tissue distribution of the mRNA of ghrelin and subtypes of its receptor, GHS-R, in humans. J Clin Endocrinol Metab 2002; 87: 2988-93.
18. Wren AM, Seal LJ, Cohen MA et al. Ghrelin enhances appetite and increases food intake in humans. J Clin Endocrinol Metab 2001; 86: 5992-8.
19. Shiiya T, Nakazato M, Mizuta M et al. Plasma ghrelin levels in lean and obese humans and the effect of glucose on ghrelin secretion. J Clin Endocrinol Metab 2002; 87: 240-4.
20. Ravussin E, Tschop M, Morales S et al. Plasma ghrelin concentration and energy balance: overfeeding and negative energy balance studies in twins. J Clin Endocrinol Metab 2001; 86: 4547-51.
21. Tschop M, Weyer C, Tataranni PA et al. Circulating ghrelin levels are decreased in human obesity. Diabetes 2001; 50: 707-9.
22. McCowen KC, Maykel JA, Bistrian BR et al. Circulating ghrelin concentrations are lowered by intravenous glucose or hyperinsulinemic euglycemic conditions in rodents. J Endocrinol 2002; 175: R7-11.
23. Scherer PE, Williams S, Fogliano M et al. A novel serum protein similar to C1q, produced exclusively in adipocytes. J Biol Chem 1995; 270: 26746-9.
24. Hu E, Liang P, Spiegelman BM. AdipoQ is a novel adipose-specific gene dysregulated in obesity. J Biol Chem 1996; 271: 10697-703.
25. Statnick MA, Beavers LS, Conner LJ et al. Decreased expression of apM1 in omental and subcutaneous adipose tissue of humans with type 2 diabetes. Int J Exp Diabetes Res 2000; 1: 81-8.
26. Weyer C, Funahashi T, Tanaka S et al. Hypoadipo-

nectinemia in obesity and type 2 diabetes: close association with insulin resistance and hyperinsulinemia. J Clin Endocrinol Metab 2001; 86: 1930-5.
27. Tschritter O, Fritsche A, Thamer C et al. Plasma adiponectin concentrations predict insulin sensitivity of both glucose and lipid metabolism. Diabetes 2003; 52: 239-43.
28. Mohlig M, Spranger J, Otto B et al. Euglycemic hyperinsulinemia, but not lipid infusion, decreases circulating ghrelin levels in humans. J Endocrinol Invest 2002; 25: RC36-8.
29. Saad MF, Bernaba B, Hwu CM et al. Insulin regulates plasma ghrelin concentration. J Clin Endocrinol Metab 2002; 87: 3997-4000.
30. Cummings DE, Weigle DS, Frayo RS et al. Plasma ghrelin levels after diet-induced weight loss or gastric bypass surgery. N Engl J Med 2002; 346:1623-30.
31. Tritos NA, Mun E, Bertkau A et al. Serum ghrelin levels in response to glucose load in obese subjects post-gastric bypass surgery. Obes Res 2003; 11: 919-24.
32. Geloneze B, Tambascia MA, Pilla VF et al. Ghrelin: a gut-brain hormone: effect of gastric bypass surgery. Obes Surg 2003; 13: 17-22.
33. Fruhbeck G, Rotellar F, Hernandez-Lizoain JL et al. Fasting plasma ghrelin concentrations 6 months after gastric bypass are not determined by weight loss or changes in insulinemia. Obes Surg 2004; 14: 1208-15.
34. Faraj M, Havel PJ, Phelis S et al. Plasma acylation-stimulating protein, adiponectin, leptin, and ghrelin before and after weight loss induced by gastric bypass surgery in morbidly obese subjects. J Clin Endocrinol Metab 2003; 88: 1594-602.
35. Holdstock C, Engstrom BE, Ohrvall M et al. Ghrelin and adipose tissue regulatory peptides: effect of gastric bypass surgery in obese humans. J Clin Endocrinol Metab 2003; 88: 3177-83.
36. Matthews DR, Hosker JP, Rudenski AS et al. Homeostasis model assessment: insulin resistance and [beta]-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia 1985; 28: 412-9.
37. Weigle DS, Cummings DE, Newby PD et al. Roles of leptin and ghrelin in the loss of body weight caused by a low fat, high carbohydrate diet. J Clin Endocrinol Metab 2003; 88: 1577-86.
38. Morinigo R, Casamitjana R, Moize V et al. Short-term effects of gastric bypass surgery on circulating ghrelin levels. Obes Res 2004; 12: 1108-16.
39. Korner J, Bessler M, Cirilo LJ et al. Effects of Roux-en-Y gastric bypass surgery on fasting and postprandial concentrations of plasma ghrelin, peptide YY, and insulin. J Clin Endocrinol Metab 2005; 90: 359-65.
40. Stoeckli R, Chanda R, Langer I et al. Changes of body weight and plasma ghrelin levels after gastric banding and gastric bypass. Obes Res 2004; 12: 346-50.
41. Lin E, Gletsu N, Fugate K et al. The effects of gastric surgery on systemic ghrelin levels in the morbidly obese. Arch Surg 2004; 139: 780-4.
42. Morpurgo PS, Resnik M, Agosti F et al. Ghrelin secretion in severely obese subjects before and after a 3-week integrated body mass reduction program. J Endocrinol Invest 2003; 26: 723-7.
43 le Roux CW, Neary NM, Halsey TJ et al. Ghrelin does not stimulate food intake in patients with surgical procedures involving vagotomy. J Clin Endocrinol Metab. 2005, 90: 4521-4.
44. Hanusch-Enserer U, Cauza E, Brabant G et al. Plasma ghrelin in obesity before and after weight loss after laparoscopical adjustable gastric banding. J Clin Endocrinol Metab 2004; 89: 3352-8.
45. Schindler K, Prager G, Ballaban T et al. Impact of laparoscopic adjustable gastric banding on plasma ghrelin, eating behaviour and body weight. Eur J Clin Invest 2004; 34: 549-54.
46. Nijhuis J, van Dielen FM, Buurman WA et al. Ghrelin, leptin and insulin levels after restrictive surgery: a 2-year follow-up study. Obes Surg 2004 14: 783-7.
47. Dixon AF, Dixon JB, O'Brien PE. Laparoscopic adjustable gastric banding induces prolonged satiety: a randomized blind crossover study. J Clin Endocrinol Metab. 2005; 90: 813-9.
48. Adami GF, Cordera R, Andraghetti G et al. Changes in serum ghrelin concentration following biliopancreatic diversion for obesity. Obes Res 2004; 12: 684-7.
49. Garcia-Unzueta MT, Fernandez-Santiago R, Dominguez-Diez A et al. Fasting plasma ghrelin levels increase progressively after biliopancreatic diversion: one-year follow-up. Obes Surg 2005; 15: 187-90.
50. Christou NV, Look D, McLean AP. Pre- and post-prandial plasma ghrelin levels do not correlate with satiety or failure to achieve a successful outcome after Roux-en-Y gastric bypass. Obes Surg 2005; 15: 1017-23.
51. Cummings DE, Purnell JQ, Frayo RS et al. A preprandial rise in plasma ghrelin levels suggests a role in meal initiation in humans. Diabetes 2001; 50: 1714-9.
52. Berg AH, Combs TP, Du X et al. The adipocyte-secreted protein Acrp30 enhances hepatic insulin action. Nat Med 2001; 7: 947-53.
53. Havel PJ. Update on adipocyte hormones: regulation of energy balance and carbohydrate/lipid metabolism. Diabetes 2004; 53: 143-51.
54. Foschi D, Corsi F, Rizzi A et al. Vertical banded gastroplasty modifies plasma ghrelin secretion in obese patients. Obes Surg 2005; 15: 1129-32.
55. Langer FB, Reza Hoda MA, Bohdjalian A et al. Sleeve gastrectomy and gastric banding: effects on plasma ghrelin levels. Obes Surg 2005; 15: 1024-9.
56. Leonetti F, Silecchia G, Iacobellis G et al. Different plasma ghrelin levels after laparoscopic gastric bypass and adjustable gastric banding in morbid obese subjects. J Clin Endocrinol Metab 2003; 88: 4227-31.
57. Ram E, Vishne T, Diker D et al. Impact of gastric banding on plasma ghrelin, growth hormone, cortisol, DHEA and DHEA-S levels. Obes Surg 2005; 15: 1118-23.
58. Adami GF, Cordera R, Marinari G et al. Plasma ghrelin concentration in the short-term following biliopancreatic diversion. Obes Surg 2003; 13: 889-92.

(Received February 2, 2006; accepted April 29, 2006)