2018

Sustained elevation of postprandial GLP-1 after bariatric surgery

BOSTON UNIVERSITY

SCHOOL OF MEDICINE

Thesis

SUSTAINED ELEVATION OF POSTPRANDIAL GLP-1 AFTER BARIATRIC SURGERY

by

JUSTIN R. PUCKETT

B.S., Michigan State University, 2013

Submitted in partial fulfillment of the

requirements for the degree of

Master of Science

2018

Approved by

First Reader _____

Donald Hess, M.D.
Assistant Professor of Surgery

Second Reader _____

John Weinstein, Ph.D.
Assistant Professor of Medicine

ACKNOWLEDGMENTS

I would like to acknowledge my advisors, family, and friends for their help in my completion of this thesis. I appreciate the support you all have given me.

SUSTAINED ELEVATION OF POSTPRANDIAL GLP-1 AFTER BARIATRIC SURGERY

JUSTIN R. PUCKETT

ABSTRACT

The incidence of obesity is on the rise globally and is associated with many comorbidities, especially type 2 diabetes mellitus (T2DM). Bariatric surgery is the most effective intervention for weight loss and reducing obesity-associated morbidity. The most common bariatric surgeries are roux-en-Y gastric bypass (RYGB) and sleeve gastrectomy (SG). RYGB and SG are equally efficacious at long-term reduction of weight in obese individuals and amelioration of T2DM. Interestingly, the improvement of glucose regulation is noted before weight loss is observed. The most likely mechanism underlying glucose homeostasis after bariatric surgery is hormonal changes in the intestine. Enteroendrocrine changes favorable of an anti-diabetic profile are noted after only a few days of receiving either RYGB or SG surgery. Most consistently, elevated postprandial GLP-1, a potent regulator of appetite and glucose control, is observed in post-bariatric surgery patients. However, data is limited regarding post-prandial GLP-1 levels beyond two years after surgery. This study will address the gap in literature by assessing postprandial elevations of GLP-1 following RYGB or SG for up to five years. We will recruit obese type-2 diabetics from an outpatient bariatric surgery clinic at Boston Medical Center

scheduled to receive RYGB or SG and periodically assess postprandial GLP-1 levels to determine if they remain elevated after 5 years. Additionally, we will provide evidence if there is a correlation among changes in postprandial GLP-1, weight loss, and hemoglobin A1c at five years. Our proposed study will help direct researchers to develop safer and more efficacious interventions for obesity and T2DM.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF ABBREVIATIONS

CCK- Cholecystokinin

GIP- Gastric inhibitory peptide

GLP-1-Glucagon-like peptide-1

PYY-Peptide YY3

RYGB- Roux-en-Y gastric bypass

SG- Sleeve gastrectomy

T2DM- Type-2 diabetes mellitus

INTRODUCTION

Background

With type 2 diabetes mellitus (T2DM) on the rise, a search for more successful interventions is warranted. First-line interventions for T2DM, such as diet and exercise, are not always efficacious in those with severe obesity.[1] Improvement in diet and exercise only result in a loss of 10% of one's initial weight, which nearly all individuals regain after 5 years.[1] Bariatric surgery is usually performed on obese individuals who are refractory to more conservative weight-loss interventions. The most common bariatric surgeries are Roux-en-Y gastric bypass (RYGB) and sleeve gastrectomy (SG).[2] RYGB and SG have been shown to result in profound weight loss and improvement in chronic co-morbidities, especially T2DM.[2,3] Up to 30% of patients who receive bariatric surgery have T2DM and usually discontinue their anti-diabetic medications post-operatively.[4,5] Up to 80% of those with T2DM who receive RYGB or SG experience complete remission.[2,5–7] Interestingly, improvement in glycemic control is noted after bariatric surgery, independent of weight loss.[8] The rapid and marked improvement in glucose regulation after surgery has raised questions as to the underlying mechanism.

Statement of the Problem

Existing literature has identified changes in gut hormones following bariatric surgery as a possible explanation for the improvement of glucose regulation but research regarding long-term follow-up is limited. Specifically, postprandial GLP-1, an enteroendocrine hormone believed to be responsible for weight-loss-independent improvement of glucose homeostasis, has not been evaluated beyond two years after surgery. Further evidence is needed regarding the sustained elevations of postprandial GLP-1 levels following either RYGB or SG.

Hypothesis

We hypothesize elevations of postprandial GLP-1 observed following bariatric surgery will be sustained over five years.

Objectives and specific aims

Additional studies are needed to better understand the role of hormonal changes in the gut following bariatric surgery and their effect on glucose regulation. This information is critical in creating novel interventions for both obesity and T2DM. The objective of this study is to evaluate the role changes in gut hormones following bariatric surgery have on the amelioration of type 2 diabetes mellitus. Specific aims of this study are as follows:

1.) Provide a literature review regarding obesity, diabetes, and changes in gut hormones following bariatric surgery.
2.) Provide further evidence RYGB and SG are effective at inducing weight loss and amelioration of T2DM for up to 5 years.
3.) Provide further evidence that GLP-1 remains elevated up to five years following RYGB or SG.
4.) Determine if there is any correlation between changes of postprandial GLP-1 from baseline compared to changes in weight or A1c from baseline.

REVIEW OF THE LITERATURE

Overview

Obesity

Obesity (BMI >30 kg/m^2) is on the rise worldwide, especially in the United States. Over 30% of those older than 18 years old in the United States are obese and it's estimated this number may reach 50% by 2030.[9] Additionally, around 20% of adolescents are obese in the United States.[10] In 2009 the CDC estimated healthcare costs of obesity were around $147 billion. Healthcare costs of obesity are comprised of direct, such as preventive, diagnostic, and treatment services, and indirect costs, associated with morbidity and mortality. With costs of

healthcare and incidence of obesity increasing, the United States must find a way to better address these issues.

One potential cause for the increased rates of obesity is an adoption of a Western lifestyle, a decrease in physical activity and increase in consumption of carbohydrate-dense foods.[10] Obesity develops when individuals remain in a state of positive calorie balance, where consumption of calories exceeds expenditure.[10] Genetics are also responsible for the development of obesity.[11] Studies searching for specific genes associated with obesity have found genes associated with brain signaling, suggesting obesity develops from a disruption in neurocognition rather than adiposity disease.[11] Hormonal changes may also attribute to development of obesity.[12] Many metabolic disturbances, such as impaired leptin and insulin signaling, are observed in those who are obese.[10] Impairment in these hormones results in dysfunction of appetite control and leads to significant obesity-associated metabolic disease.[10] Aside from rare obesity-causing diseases, such as leptin deficiency, there is little evidence regarding the mechanism underlying development of hazardous obesity (BMI>40).[10]

Interventions to treat obesity

First-line interventions, such as improving diet or increasing physical activity, can offer significant weight loss of about 10% initial weight to obese individuals, which is almost always transient.[10] Pharmacologic interventions, such

as absorption inhibitors or appetite suppressants, are minimally efficacious and are commonly accompanied with side effects such as fatigue, depression, nausea, abdominal pain, and insomnia.[10] Absorption inhibitors work by reducing the amount of fat our body can absorb from food by decreasing the amount of lipase our pancreas secretes. Researchers developing novel pharmacologic interventions have not yet found a safe way to overcome the body's robust response to a caloric deficit.[10] Many who lose weight struggle with strong drives to eat, resulting in weight regain to their original weight.[10] The underlying mechanisms of the strong desire to eat arise from the physiology of the gut.[10]

Energy homeostasis

The liver, adipose tissue, and gastrointestinal tract produce many short and long acting hormones which inform the body of its energy status.[10] Energy status may be thought of as a positive or negative caloric balance. Multiple hormones (Table 1) act throughout the body, influencing food intake and energy expenditure. Normally, consumption of food will generate signals to inhibit feeding and promote digestion or energy utilization.[10] However, those with a BMI >35 have impairments in these hormonal signals resulting in increased appetite and decreased energy utilization.[10] Additionally, obese individuals have a dysfunctional entero-insular axis, or impairment in communication between the intestines and pancreas, increasing their risk of developing insulin resistance.[10]

Table 1. Characteristics of hormones affecting glucose regulation

Hormone	Origin	Stimuli	Appetite Effect	Function
GLP-1	L-cells	Food ingestion	Decrease	-Insulin secretion -Delays gastric emptying -Increases beta-cell mass
PYY	L-cells	Food ingestion	Decrease	-Delays gastric emptying
CCK	I-cells	Fat and protein consumption	Decrease	-Gallbladder contraction -Delays gastric emptying -Pancreatic enzyme secretion
Ghrelin	Fundus and body of stomach	Fasting	Increase	-Increases gastric emptying
GIP	K-cells in GI mucosa	Nutrients in gut lumen	Decrease	-Promotes insulin and glucagon secretion

Endocrine signaling

GLP-1

GLP-1 is a hormone released from L-cells which are mostly found in the distal intestine.[13] The L-cells are activated by contact with nutrients passing by the lumen.[13] GLP-1 is categorized as an incretin, which helps pancreatic beta-cells secrete insulin in response to glucose.[14] Additional roles of GLP-1 include reducing appetite, delaying gastric emptying, and increasing the size of pancreatic beta-cells.[15] Obese individuals with T2DM have decreased postprandial GLP-1 levels compared to those with a normal BMI and without T2DM.[16–18] Toft-Nielsen et al. have demonstrated that continuous infusion of GLP-1 in T2DM individuals significantly improves glucose homeostasis, reduces appetite, does not affect blood pressure, and has no gastrointestinal side effects.[17]

PYY

Peptide YY (PYY) is also released from L-cells in the distal intestine following ingestion of food and is known to have appetite suppressing properties similar to GLP-1.[19] Unlike GLP-1, PYY is not known to have any effect on insulin secretion[19] but may have a role in restoration of impaired beta-cell function.[20] PYY levels have been shown to be decreased in obesity compared to normal BMI individuals.[20]

CCK

CCK is released from I cells which predominate the duodenum and jejunum following ingestion of food.[21] CCK is known to decrease appetite, stimulate contraction of the gallbladder, delay gastric emptying, and promote secretion of pancreatic enzymes.[21] Postprandial levels of CCK are decreased in those who are obese and diabetic compared to healthy controls.[19]

Ghrelin

Ghrelin is secreted from glands in the fundus and body of the stomach.[22] Its primary role in the body is to promote the desire to ingest food.[22] Ghrelin is elevated before consumption of a meal and is decreased after.[23] Obese individuals are known to have difficulty suppressing ghrelin levels following a meal compared to non-obese individuals.[24] After individuals lose a significant amount of weight from lifestyle changes, ghrelin has been shown to be increased both before and after meals.[24] The increase in ghrelin following weight loss suggests the body employs a compensatory mechanism to resist weight loss.

Glucagon

Glucagon is released from the alpha-cells of the pancreas and functions by raising blood concentrations of glucose.[25] Those with T2DM have inappropriate elevations in glucagon both before and after meals.[26] Elevated levels of glucagon in T2DM are considered inappropriate because the elevation occurs in the setting

of hyperglycemia.[26] Appropriate elevations of glucagon are typically seen when blood sugar is low.[26] Type 2 diabetics usually have a higher fasting baseline blood sugar, which may account for changes in the activation threshold of pancreatic alpha-cells.[26]

GIP

Gastric inhibitory polypeptide (GIP) is known to stimulate secretion of glucagon but only at basal glucose levels.[27] When blood glucose levels are elevated beyond basal levels, GIP has no effect on glucagon secretion and instead stimulates release of insulin.[27] Furthermore, GIP has been shown to have a reduction in incretin effect in those with T2DM in contrast to GLP-1, which has preserved functionality.[28]

Diabetes and Obesity

Those who are obese are at greater risk for acquiring chronic diseases, increasing the risk of mortality and morbidity.[29] Specifically, obesity increases the risk for the development of type 2 diabetes mellitus (T2DM), heart failure, liver disease, hypertension, hyperlipidemia, cancer, depression, obstructive sleep apnea, and dementia.[5] A rise in the prevalence of T2DM may be a result of an increasing prevalence of obesity.[30]

Development of T2DM in obese individuals is related to a metabolic dysfunction affecting insulin signaling and appetite control.[31] Adipocytes release proinflammatory cytokines and free fatty acids, which are damaging to the liver, pancreas, and intestines.[31] Compared to those with a normal BMI, obese individuals have a greater number of adipocytes, resulting in greater damage to the organs responsible for appetite and glucose regulation.[31]

Type 2 diabetes mellitus

The prevalence of diabetes in the United States ranges from 6-12% of adults with T2DM accounting for around 90% of these cases.[32] The diagnosis of T2DM is made when an individual has a glycosylated hemoglobin A1c of greater than 6.5%, a three month reflection of the body's ability to regulate glucose.[32] Additionally, a two hour oral glucose tolerance test greater than 200 mg/dL or two fasting blood sugars greater than 126 mg/dL will make the diagnosis.[32]

T2DM is commonly seen in obese adults, not genetically inherited, and characterized by an insulin insensitivity.[32] Insulin insensitivity occurs when the body's insulin receptors require more insulin stimulus for glucose to enter cells, compared to nondiabetic individuals.[32] Only about 16% of T2DM require insulin.[32]

T2DM Interventions

First-line interventions for T2DM are similar to interventions already discussed for obesity, such as diet and exercise modifications.[33] Weight loss provides significant physiologic improvement of glucose regulation in those with T2DM but is not shown to be effective in the long-term in preventing acquisition of T2DM.[30] For example, obese individuals may lose weight, improving their ability to regulate glucose, but still have significant ongoing adipocyte-induced organ damage, which eventually will lead to insulin insensitivity.

There are a wide variety of pharmacologic interventions for T2DM. Generally, individuals are first started on metformin, which acts to reduce hepatic gluconeogenesis.[32] Additionally, individuals may be started on medications which help the body secrete insulin, inhibit intestinal reabsorption of glucose, increase urinary glucose excretion, or improve sensitivity to insulin.[32] If these medication do not help the individual achieve a goal HgbA1c of less than 7.0%, then insulin therapy may be initiated.[32]

Bariatric surgery and obesity

Failure of first line therapies and limited pharmacologic interventions call for more efficacious treatments, such as surgery. The most effective treatment for both losing and maintaining healthy weight is bariatric surgery.[33] Generally, individuals who undergo bariatric surgery lose around 50-70% of excess body

weight.[2] However, around 20% of bariatric surgery patients fail to lose a significant amount of weight; the underlying reason remains unclear.[34] The mechanism for surgically-induced weight loss is primarily attributed to appetite and caloric intake reduction.[35] Despite patients' negative calorie balance following bariatric surgery, they report greater degrees of satiety.[15]

The most common bariatric surgeries are the roux-en-Y gastric bypass (RYGB), and sleeve gastrectomy (SG).[35] The RYGB incorporates both restrictive and malabsorptive components, while the while the SG is considered only restrictive.[30] The restrictive component, shared by both RYGB and SG, arises from resection of the stomach. The stomach resection results in less space for food and earlier sensation of satiety. RYGB provides an additional malabsorptive component by bypassing the duodenum where significant amounts of nutrient absorption takes place.

Indications/contraindications for bariatric surgery

Generally, bariatric surgery is indicated for those who have a BMI >40 kg/m2 or BMI >35 kg/m2 with at least one associated comorbidity, such as hypertension, hyperlipidemia, obstructive sleep apnea, T2DM, asthma, or gastroesophageal reflux disease.[36] Additionally, bariatric surgery is indicated for those with a BMI between 30-35 with uncontrolled T2DM or metabolic syndrome. Relative and absolute contraindications for bariatric surgery include

age of less than 18 or greater than 65, severe coagulopathy, severe cardiac disease, current drug or alcohol abuse, untreated major depression or psychosis, or uncontrolled eating disorders. Furthermore, individuals who are unable to adhere to a strict diet rich in vitamins and nutrients should not receive the surgery. Generally, individuals with a BMI <27 are not offered the surgery.

Preoperative predictive factors for T2DM remission

The ABCD score is a validated tool used to assess the likelihood of T2DM remission after bariatric surgery.[30] It is comprised of the patient's age, BMI, C-peptide level, and duration of disease.[30] Those who have been diabetic for shorter lengths of time are more likely to receive improved glucose regulation from the surgery.[40] Bariatric surgery for individuals with a BMI <27 provides minimal benefits and is contraindicated for this population, as previously mentioned.[30] C-peptide is a byproduct of insulin and can be measured to assess how much insulin a person is producing or give an estimate of pancreatic beta-cell mass.[30] Those with advanced age usually do not receive as much improvement in glucose regulation than those who are younger.[30]

Bariatric Surgery and T2DM

Bariatric surgery has also been shown to be the most effective treatment for T2DM in obese individuals with remission rates up to 80%.[5–7] The mechanisms for T2DM resolution in bariatric surgery patients are not clearly understood, as this effect is observed independent of weight loss.[37–39] There are two major hypotheses to explain this phenomenon: the hindgut hypothesis and the foregut hypothesis.[30] The hindgut hypothesis proposes the weight loss-independent improvement of glucose regulation comes from increased production of anti-diabetic hormones in the distal intestine.[30] After surgically bypassing the foregut, more undigested food reaches the hindgut stimulating enteroendocrine cells to produce anti-diabetic hormones, such as GLP-1.[30] The foregut hypothesis proposes less anti-incretin and appetite-stimulating hormones are produced in the proximal intestine when bypassed by bariatric surgery.[30]

Existing Research

RYGB vs SG efficacy in weight loss and remission of T2DM

Literature regarding the comparative efficacies of RYGB and SG is extensive and summarized in Table 2. Generally, RYGB and SG do not differ significantly in weight loss or remission of T2DM, suggesting the surgeries are equally efficacious.[35,41–48] One meta-analysis showed no difference in weight loss or remission of T2DM among RYGB compared to SG.[45] However, there are two studies which demonstrate differences in efficacy, suggesting potential superiority of RYGB.[49,50] Lee et al. conducted a 1-year randomized controlled trial involving 60 moderately obese (25 < BMI < 35) subjects with poorly controlled T2DM (HbA1c >7.5%) who were randomly assigned to RYGB (n=30) or SG (n=30). They demonstrated a significantly greater number achieving remission of T2DM 12 months after RYGB compared to SG and significantly more weight loss in the RYGB group.[50] Kashyae et al. showed a similar superiority of T2DM remission 4 weeks after RYGB (n=9) compared to SG (n=7) but without any significant difference in weight loss among the two surgical groups in a prospective cohort study.[49] The largest prospective study involving 558 subjects by Zhang et al followed obese T2DM subjects for 1 year after either RYGB (n=358) or SG (n=200), however, revealed no significant difference in weight loss or remission of T2DM.[47]

Table 2. A comparison of significant weight loss and amelioration of type 2 diabetes mellitus following roux-en-Y gastric bypass vs sleeve gastrectomy

Method	Author	Study	Follow-up	Subject #	Wt loss	T2DM
RYGB SG	Lee et al.	RCT	1 year	30 30	↓↓ ↓	↓↓ ↓
RYGB SG	Kashyap et al.	Prospective	4 weeks	9 7	↓ ↓	↓↓ ↓
RYGB SG	Abbatini et al.	Retrospective	3 years	16 20	↓ ↓	↓ ↓
RYGB SG	Vidal et al. (2007)	Prospective	4 months	50 35	↓ ↓	↓ ↓
RYGB SG	Cutolo et al.	Retrospective	2 years	16 15	↓ ↓	↓ ↓
RYGB SG	Yip et al.	Meta-analysis	Up to 3 years	998 179	↓ ↓	↓ ↓
RYGB SG	Leyba et al.	Prospective	1 year	75 42	↓ ↓	N/A
RYGB SG	Zhang et al.	Prospective	1 year	358 200	↓ ↓	↓ ↓
RYGB SG	Kokkinos et al.	Prospective	6 months	14 23	↓ ↓	N/A
RYGB SG	Nocca et al.	Prospective	1 year	35 33	↓ ↓	↓ ↓
RYGB SG	Vidal et al. (2008)	Prospective	1 year	52 39	↓ ↓	↓ ↓

One arrow is used to indicate a significant decrease while two arrows are used to indicate a significantly greater significant decrease. N/A= not assessed.

Elevated GLP-1 as a mechanism for improved glucose homeostasis

One potential mechanism for the significant improvement in glucose regulation is a hormonal change in the gut following bariatric surgery.[10,30,38] There

is a large amount of research demonstrating elevated postprandial levels of GLP-1 following either RYGB or SG[5,6,29,33,51–59] (Table 3). The largest study, by Jimenez et al., recruited 153 subjects with T2DM who were scheduled to undergo RYGB or SG.[52] Among study participants, 75% experienced remission of T2DM for up to one year, defined as hemoglobin A1c <6.5%, without a statistical significant difference between surgical groups.[52] In both surgical groups, postprandial GLP-1 was elevated compared to baseline after surgery while fasting levels remained unchanged.[52] Additionally, Jimenez et al. demonstrated an association between T2DM remission and elevated postprandial GLP-1 levels.[52] The evidence provided by the study is consistent with others and suggests GLP-1 plays a role in improvement of glucose regulation following bariatric surgery.

Two studies have demonstrated a significantly greater postprandial GLP-1 response following RYGB compared to SG[33,56]. One of the studies, a randomized prospective study by Peterli et al., assigned either RYGB (n=13) or SG (n=14) to patients.[33] Their study showed early improvement in glucose regulation at one week in both groups without a statistical significant difference among the two surgeries.[33] However, there was a greater postprandial GLP-1 response in the RYGB group.[33] Of note, this study is likely underpowered given their small sample size. Consistent with other studies, both groups did not show any statistical difference compared to controls in their fasting GLP-1 levels.[29,33,54,56] A present limitation of research regarding GLP-1 is a lack of long-term follow-up to

provide evidence that the elevations of postprandial GLP-1 are sustained beyond two years.

Table 3. Comparison of fasting and postprandial GLP-1 and PYY following roux-en-Y or sleeve gastrectomy

Method	Author	Study Methodology	Sample size	Fasting GLP-1	Fasting PYY	Postprandial GLP-1	Postprandial PYY
RYGB	Rodieux et al.	CS	8	NC	NC	↑	↑
RYGB	Le Roux et al.	Prosp	12	NC	NC	↑	↑
RYGB	Borg et al.	Prosp	6	NC	NC	↑	↑
RYGB SG	Peterli et al.	Prosp Rand	12 11	NC	NC	↑↑ ↑	↑ ↑
RYGB SG	Yousseif et al.	Prosp	10 8	NC	NC	↑↑ ↑	↑↑ ↑
RYGB SG	Ramon et al.	Prosp Rand	7 8	NC	NC	↑ ↑	↑ ↑
RYGB	Vidal et al.	CS	24	NC	N/A	↑	N/A
RYGB SG	Jimenez et al.	Prosp	98 55	NC	N/A	↑ ↑	N/A
RYGB SG	Nannipieri et al.	Prosp	23 12	NC	NC	↑ ↑	↑ ↑
RYGB	Bose et al.	Prosp	11	NC	N/A	↑	N/A
RYGB	Laferrere et al.	Prosp	8	NC	N/A	↑	N/A

RYGB SG	Karamanakos et al.	Prosp	16 16	N/A	NC	N/A	↑ ↑↑

CS=Cross-sectional; Prosp=prospective cohort; Rand=randomized controlled trial; NC=no change; N/A=not assessed; ↑=significant increase; ↑↑=significantly greater significant increase

PYY in weight reduction and glucose regulation

Peptide YY3 (PYY) is another gut hormone believed to be involved in the reduction of weight and possibly improvement in glucose regulation following bariatric surgery.[14] Following RYGB or SG, levels of PYY have been shown to be elevated after ingestion of a meal while fasting levels remain unchanged (Table 3).[29,54–60] One of the studies showed a greater postprandial PYY response following RYGB compared to SG,[56] while seven other studies showed no significant difference among the two surgical groups.[29,33,54] Yousseif et al. recruited 18 obese non-diabetic females in a prospective study undergoing RYGB (n=10) or SG (n=8), matched subject's adiposity among the two groups, and found a significantly greater postprandial PYY response in the RYGB group compared to SG.[56] Despite this one study showing superiority in RYGB, it likely was underpowered given the small sample size. The majority of studies show no difference in PYY following bariatric surgery.[29,54–56,58,60] It has also been shown those who fail to lose a significant amount of weight following RYGB have lower circulating levels of PYY.[57]

Ghrelin is significantly reduced following SG

Given the intimate relationship of obesity and T2DM, it is important to further examine the effects of ghrelin, before and after bariatric surgery. Studies regarding ghrelin levels following RYGB or SG are inconclusive but generally show decreased levels after SG but not after RYGB.[22,54,55,60] Karamanakos et al. performed a prospective double blind study which examined 6 month postoperative ghrelin levels following bariatric surgery in 32 patients.[54] Subjects were assigned to each surgical group (16 each) with a significant decrease in hunger stimulating ghrelin levels in those who underwent SG but no difference among those who received RYGB.[54] Conversely, Holdstock et al. recruited 66 subjects undergoing RYGB in a prospective study and revealed significantly increased ghrelin levels following surgery.[61] Similarly, Ramon et al. demonstrated decreased fasting and postprandial ghrelin levels in those undergoing SG but no difference in those who received RYGB.[29] Other studies have shown significantly decreased ghrelin levels following RYGB compared to controls.[22,58] In summary, levels of ghrelin are typically decreased following bariatric surgery although there have been some reported elevations after RYGB.

CCK levels after bariatric surgery are inconclusive

There are few studies evaluating the effect bariatric surgery has on CCK levels and the results remain inconclusive. Rubino et al. demonstrated no change

in circulating levels of CCK following RYGB at 6 weeks follow-up in 10 morbidly obese patients.[62] Similarly, another study by Kellum et al. recruited morbidly obese subjects assigned to either RYGB (n=9) or vertical banded gastroplasty (n=7) and found no difference in postprandial CCK levels at 6 months compared to controls or among surgical groups.[63] A more recent study by Jacobsen et al. recruited 8 non-diabetic obese subjects and found a significantly increased secretion of CCK after ingestion of a meal within two weeks of RYGB.[64] A 2013 cross-sectional study by Dirksen et al. of RYGB subjects revealed significantly increased postprandial CCK levels in 17 individuals who failed to lose significant weight (defined as <50% excess BMI) compared to 16 individuals who experienced significant weight loss (defined as >60% excess BMI).[65] In a study comparing postprandial CCK, obese subjects were randomly assigned to either RYGB (n=12) or SG (n=11) and demonstrated increased levels at 1 year follow-ups but to a greater degree in the SG group.[60]

GIP is increased or unchanged following bariatric surgery

Research regarding GIP levels after bariatric surgery is limited and inconclusive; studies have reported increases following surgery, whereas others have shown no change.[5,6,38,49,66,67] Romero et al. conducted a prospective study in 12 obese diabetic subjects who were receiving either RYGB (n=6) or SG (n=6) and revealed a significant increase in postprandial GIP levels in the SG group six

weeks after surgery but no change in the RYGB group.[38] Conversely, Laferrere et al. conducted two studies in subjects undergoing RYGB (N= 9 and 8) and showed increased levels of postprandial GIP one month following the surgery in both studies.[5,66] A prospective study by Kashyap et al. found no difference in postprandial GIP levels 1 month following either RYGB (n=9) or SG (n=7) in obese T2DM subjects.[49] Hansen et al. also found no difference in postprandial GIP levels in 9 diabetic obese subjects six weeks following RYGB.[67] Bose et al. conducted a prospective study with 11 obese subjects with T2DM, which showed elevated postprandial GIP levels one month following surgery with no change at one year follow-ups.[6] More research is needed to help determine the affect bariatric surgery has on GIP levels given that the current literature is limited and yields conflicting results.

Glucagon levels are unchanged after bariatric surgery

Shah et al. have shown an association between elevated levels of glucagon and a dysfunction in glucose homeostasis in T2DM.[26] Glucagon levels following bariatric surgery have not been extensively studied but generally are not changed following RYGB.[5,38,51,66,68,69] A study by Romero et al. compared RYGB (n=6) to SG (n=6) and found no significant difference in fasting or postprandial levels compared to controls.[38] Other studies which looked solely at RYGB subjects similarly revealed a failure to suppress glucagon levels following surgery.[5,51,66,69]

Conversely, Swabrick et al. demonstrated a decrease in glucagon levels following RYGB in 19 diabetic obese subjects.

Calorie restriction improves glucose regulation

Another mechanism proposed for improvement of glucose regulation following bariatric surgery is calorie restriction.[39] Isbell et al. conducted a study which recruited 18 obese subjects with T2DM and divided them in two groups, RYGB (n=9) and caloric restriction diet (n=9).[39] The RYGB group had the same caloric restriction as the diet control group and all subjects were matched for age, sex, weight, and diabetes duration.[39] They found improvement in insulin resistance among both groups without any significant difference between the groups.[39] Thus, the study suggests that early improvement in glucose regulation is not dependent on gut hormonal changes but, rather, the anti-diabetic effect is due to calorie restriction.

Although there are clear post-surgical associations with hormones such as GLP-1 and weight loss or improvement in glucose regulation, the underlying mechanism is likely multifactorial. Additionally, the current research is limited regarding long-term follow-up to assess the levels of the hormones discussed beyond two years. Our study aims to bridge the gap in literature by assessing the

most consistent change noted after bariatric surgery, elevations in postprandial GLP-1, and evaluating it for five years.

METHODS

Study Design

This will be a prospective cohort study assessing sustained elevations of postprandial GLP-1 levels following RYGB or SG in individuals with T2DM.

Study Population and Sampling

Subjects will be recruited over a period of 6 months from the outpatient bariatric surgery clinic at Boston Medical Center. Inclusion and exclusion criteria (Table 4) have been established to minimize modifying factors. The estimated sample size for repeated measures of ANOVA is 32 with an alpha <0.05, power of 80%, and effect size of 0.28 in postprandial GLP-1. This effect size was estimated from the results of a previous study conducted by Peterli et al. assessing difference in postsurgical postprandial GLP-1 levels and pre-bariatric surgical values[60] The sample size is based on 5 measures which are conservatively assumed to be non-correlated. Of note, correlation would increase the power of the study. Accounting for a loss to follow-up rate of about 20%, we will recruit 40 subjects in our study.

Table 4. Inclusion and exclusion criteria

Inclusion Criteria	Exclusion Criteria
• BMI of 35-45 • Scheduled for RYGB or SG • Age 25-65 • Using oral anti-diabetic medication • 10%> A1c >7.5%	• Use of insulin, GLP-1 agonist, or DPP4 inhibitors • Hx of severe psychiatric disease, uncontrolled HTN, cancer, CHF, cirrhosis, alcoholism, drug abuse, or severe pulmonary disease

Surgical Technique

RYGB

The laparoscopic RYGB will create a vertical gastric pouch of 20-30 ml with surgical staples in the cardia of the stomach.[60] The Roux limb will be approximately 100 cm and the biliopancreatic limb will be approximately 50 cm from the ligament of Treitz, as described in a previous study.[60]

SG

The laparoscopic SG will be done along a 36 French bougie from the angle of His to approximately 3-4 cm proximal to the pylorus, as described in a previous study.[60]

Recruitment

Individuals who meet inclusion and exclusion criteria will be recruited at an outpatient bariatric center affiliated with Boston Medical Center over a 6-month period. Type of surgery will be assigned per the surgeon's discretion and patient preference. We will aim to recruit an equal number of RYGB and SG participants. Subjects interested in participating will be briefed by one of the investigators to fully inform them of study details, including risks and benefits. Informed consent will be obtained during the visit with the investigator.

Data Collection

Individuals will be assessed within one month of surgery then postoperatively at three months, one year, two years, three years, and five years. Blood will be drawn at each of these encounters after a 12 hour fast to assess GLP-1 and HgbA1c. Individuals will be instructed to not take their anti-diabetic medications during the fast. Weight and height will be assessed at each visit to calculate BMI. A standard liquid meal of 350 kcals (46% carbs, 32% protein, and 12% lipids) will be given to the subjects. Blood will be drawn at 0, 30, 60, 90, and 120 minutes for assessment of GLP-1. GLP-1 will be measured with commercially-available kits, as previously described, to obtain a maximum plasma concentration (Pmax).[60] Blood for hemoglobin A1c will be drawn at each visit and sent to Quest Diagnostics for evaluation.

Data Analysis

Data analysis will be performed using SPSS 15.0. Values will be reported as means +/- standard error of the mean (SEM). Repeated measures analysis of variance (ANOVA) will be used to assess for significant differences in longitudinal levels of GLP-1 after surgery, overall and stratifying for type of surgery. If a significant difference is noted, then post hoc analysis using a pairwise comparison of all measures will be conducted, adjusting alpha using the Bonferroni correction. To verify there is a significant change of pre- and post-operative postprandial GLP-1, a paired t test will be performed at 1 year. At 5 years, Spearman correlations between the change in GLP-1 from baseline to the change in weight and A1c from baseline will be assessed as secondary outcomes.

Timeline

We aim to have our research proposal approved by the IRB in the Fall of 2018 so subjects can be recruited over a six-month period from January 2019 to June 2019 (Table 5). We will collect and analyze data over the subsequent five years from July 2019 to July 2024. Our manuscript will be submitted for peer review in the Fall of 2024. Resources will include investigators, statisticians, and surgeons.

Table 5. Timeline of research project

Fall 2018	Submission and Approval by IRB
January 2019- June 2019	Recruitment of subjects and surgery
July 2019- July 2024	Data collection and analysis
Fall 2024	Manuscript submitted for peer review

Institutional Review Board

This study will be submitted for full IRB review to the Boston University Medical Campus IRB under INSPIR II criteria and to the IRB of Boston Medical Center.

CONCLUSION

Discussion

With global incidence rates for both obesity and T2DM on the rise, the need to develop more efficacious interventions is warranted to reduce the associated morbidity.[10] First-line therapies are not always successful in providing sustained weight loss.[30] Bariatric surgery, however, is highly effective at inducing sustainable weight loss and amelioration of T2DM.[34]

Interestingly, the improvement in glucose homeostasis occurs before significant weight loss is observed.[8] The underlying mechanisms for the profound improvement of glucose regulation are not clearly understood but believed to be largely attributed to hormonal changes seen in the intestine after surgery.[4] Just days after surgery, hormonal changes favorable of an antidiabetic profile are observed.[4] The most consistent change noted after surgery are elevated postprandial GLP-1 levels.[52]

GLP-1 serves multiple functions in improved glucose homeostasis. GLP-1 acts as an incretin to restore functionality of pancreatic beta cells to appropriately release insulin when blood sugars are elevated.[13] Additionally, GLP-1 acts to delay gastric emptying resulting in increased satiety.[14] The elevation of postprandial GLP-1 observed after bariatric surgery is one of many mechanisms proposed for amelioration of T2DM and weight loss.

Bariatric surgery reorganizes the intestine in a way which alters the secretion of hormones in response to meals. One theory, the hindgut hypothesis, is more undigested food reaches the L-cells after surgery, resulting in proliferation of the cells and a more robust GLP-1 response.[30] The restrictive and malabsorptive nature of the surgeries are responsible for the increase in nutrient signaling occurring in the intestine by altering the structure.[10] The hindgut hypothesis supports the proposition that GLP-1 is responsible for the improvement in glucose regulation observed independent of weight loss.

Mechanisms underlying weight regain following bariatric surgery remain elusive but are believed to be attributed to lifestyle, psychiatric, hormonal, and surgical factors.[70] Individuals who are minimally physically active or do not adhere to a postsurgical diet are more likely to experience weight regain than those who exercise 30 minutes per day and consume a healthy diet rich in protein.[70] Psychological disorders, particularly eating disorders, have been associated with weight regain.[70] One study showed one half of those who experienced weight regain suffered from a binge-eating disorder.[71] Hormonal changes after surgery are also believed to contribute to weight regain.[70] One study demonstrated low levels of postprandial GLP-1 to be associated with weight regain.[72] Surgical factors such as pouch dilatation, increased stoma size, and gastro-gastric fistulas have been recognized as mechanisms for weight regain following RYGB.[70]

In our present study, we propose to address the gap in existing literature regarding elevated postprandial GLP-1 levels beyond 2 years after surgery. There has been a significant weight regain observed 2-5 years after surgery.[70] It is possible there is an underlying hormonal mechanism responsible for the regain of weight. Our study will measure postprandial GLP-1 levels up to 5 years after surgery and determine if there is any correlation with changes in weight or hgbA1c.

Further studies should assess the other hormones discussed in the literature review such as PYY, glucagon, GIP, ghrelin, and CCK. These hormones control glucose regulation and appetite control and will be helpful in understanding the mechanisms involved in weight regain and amelioration of T2DM.[14] Specifically, levels of these hormones should similarly be assessed over a period of 5-years to determine if there is a correlation between change in weight or hgbA1c. Further information quantifying the hormonal changes following bariatric surgery will provide insight as to why individuals regain weight and maintain remission of T2DM.

It is of interest for future studies to also include an analysis of adipokines not mentioned in this study, such as TNF-alpha, IL-6, and leptin. These adipokines are involved in obesity-related insulin impairment.[31] Although weight regain is relatively common in 20% of individuals, recurrence of T2DM is much less so.[34] Further research is needed to better understand why individuals may

regain weight but do not appear to have a concomitant impairment of glucose regulation.

Strengths of this present study include a sample size with greater than 80% power, consistency in surgical technique, and an ethnically diverse community. This study provides insight into the mechanisms underlying weight loss and glucose homeostasis over a longer period than most current studies. One limitation is not providing an analysis of additional hormones, which may be acting as modifying factors; however this also serves as a strength by reducing the likelihood of alpha errors.

Summary

The incidence of obesity and T2DM are on the rise and are usually refractory to first-line interventions. Bariatric surgery is highly efficacious at inducing sustained weight loss and improvement in blood sugar regulation. Enteroendocrine changes in postprandial GLP-1 levels following bariatric surgery have been associated with weight loss and amelioration of T2DM. Whether these changes are sustained beyond 2 years after surgery has not been adequately addressed. Our study will address this by assessing postprandial GLP-1 levels for up to 5 years following surgery. Additionally, we will assess the findings at 5 years to determine if there is an association with change in weight or A1c. Given a period of weight regain typically observed 2-5 years following surgery, we

suspect postprandial GLP-1 will be elevated during the first two years of weight loss and improvement in glucose regulation will then significantly decrease during a period of weight regain. However, if weight regain does not occur, then we suspect the elevations of postprandial GLP-1 will be sustained and associated with improved HgbA1c.

Clinical and/or public health significance

Understanding the mechanisms responsible for improvement in glucose regulation or weight loss helps researchers develop more efficacious interventions for obesity and T2DM, two significant public health issues. Additionally, assessing postsurgical enteroendocrine beyond 2 years specifically may unmask a potential mechanism for the period of weight regain typically observed 2-5 years following bariatric surgery.

LIST OF JOURNAL ABBREVIATIONS

Am. J. Physiol.-Endocrinol. Metab.	American Journal of Physiology- Endocrinology and Metabolism
Am. J. Physiol.-Regul. Integr. Comp. Physiol	American Journal of Physiology- Regulatory, Integrative, and Comparative Physiology
Am. J. Physiol.-Gastrointest. Liver Physiol	American Journal of Physiology- Gastrointestinal and Liver Physiology
Ann. Surg.	Annals of Surgery
Cell Rep.	Cell Reports
Endocr. Oncol. Metab.	Endocrine, Oncology, and Metabolism
Int. J. Obes.	International Journal of Obesity
Int. J. Obes. Relat. Metab. Disord.	International Journal of Obesity and Related Metabolic Disorders
J. Clin. Endocrinol. Metab.	Journal of Clinical Endocrinology and Metabolism
J. Clin. Invest.	Journal of Clinical Investigation
J. Diabetes	Journal of Diabetes
J. Gastrointest. Surg.	Journal of Gastrointestinal Surgery

JAMA	The Journal of the American Medical Association
N. Engl. J. Med.	New England Journal of Medicine
Physiol. Rev.	Physiological Reviews
Surg. Endosc	Surgical Endoscopy
World J. Gastroenterol	World Journal of Gastroenterology

REFERENCES

1. Umeda, L. M. *et al.* Early improvement in glycemic control after bariatric surgery and its relationships with insulin, GLP-1, and glucagon secretion in type 2 diabetic patients. *Obes. Surg.* **21,** 896–901 (2011).
2. Buchwald, H. *et al.* Bariatric Surgery: A Systematic Review and Meta-analysis. *JAMA* **292,** 1724–1737 (2004).
3. Thaler, J. P. & Cummings, D. E. Minireview: Hormonal and metabolic mechanisms of diabetes remission after gastrointestinal surgery. *Endocrinology* **150,** 2518–2525 (2009).
4. Roux, C. W. le *et al.* Gut Hormone Profiles Following Bariatric Surgery Favor an Anorectic State, Facilitate Weight Loss, and Improve Metabolic Parameters. *Ann. Surg.* **243,** 108–114 (2006).
5. Laferrère, B. *et al.* Incretin Levels and Effect Are Markedly Enhanced 1 Month After Roux-en-Y Gastric Bypass Surgery in Obese Patients With Type 2 Diabetes. *Diabetes Care* **30,** 1709–1716 (2007).
6. Bose, M. *et al.* Weight loss and incretin responsiveness improve glucose control independently after gastric bypass surgery. *J. Diabetes* **2,** 47–55 (2010).
7. Sjöström, L. *et al.* Lifestyle, Diabetes, and Cardiovascular Risk Factors 10 Years after Bariatric Surgery. *N. Engl. J. Med.* **351,** 2683–2693 (2004).
8. Jiménez, A. *et al.* GLP-1 and the Long-Term Outcome of Type 2 Diabetes Mellitus After Roux-en-Y Gastric Bypass Surgery in Morbidly Obese Subjects. *Ann. Surg.* **257,** 894 (2013).

9. Wang, Y., Beydoun, M. A., Liang, L., Caballero, B. & Kumanyika, S. K. Will all Americans become overweight or obese? estimating the progression and cost of the US obesity epidemic. *Obesity. Silver Spring Md* **16,** 2323–2330 (2008).
10. Scott, W. R. & Batterham, R. L. Roux-en-Y gastric bypass and laparoscopic sleeve gastrectomy: understanding weight loss and improvements in type 2 diabetes after bariatric surgery. *Am. J. Physiol.-Regul. Integr. Comp. Physiol.* **301,** R15–R27 (2011).
11. O'Rahilly, S. Human genetics illuminates the paths to metabolic disease. *Nature* **462,** 307 (2009).
12. Marti, A., Moreno-Aliaga, M. J., Hebebrand, J. & Martínez, J. A. Genes, lifestyles and obesity. *Int. J. Obes. Relat. Metab. Disord. J. Int. Assoc. Study Obes.* **28 Suppl 3,** S29-36 (2004).
13. Gautier, J.-F., Choukem, S.-P. & Girard, J. Physiology of incretins (GIP and GLP-1) and abnormalities in type 2 diabetes. *Diabetes & Metabolism* **34 Suppl 2,** S65-72 (2008).
14. Ilic, M., Pašić, F., Kirigin, L., Mirosević, G. & Bekavac-Beslin, M. Changes in Gut Hormones After Roux en Y Gastric bypass, Sleeve Gastrectomy, and Adjustable Gastric Banding. *Endocr. Oncol. Metab.* **1,** 25–36 (2016).
15. Finelli, C., Padula, M. C., Martelli, G. & Tarantino, G. Could the improvement of obesity-related co-morbidities depend on modified gut hormones secretion? *World J. Gastroenterol.* **20,** 16649–16664 (2014).

16. Nauck, M., Stöckmann, F., Ebert, R. & Creutzfeldt, W. Reduced incretin effect in type 2 (non-insulin-dependent) diabetes. *Diabetologia* **29,** 46–52 (1986).
17. Toft-Nielsen, M. B., Madsbad, S. & Holst, J. J. Continuous subcutaneous infusion of glucagon-like peptide 1 lowers plasma glucose and reduces appetite in type 2 diabetic patients. *Diabetes Care* **22,** 1137–1143 (1999).
18. Lund, A., Vilsbøll, T., Bagger, J. I., Holst, J. J. & Knop, F. K. The separate and combined impact of the intestinal hormones, GIP, GLP-1, and GLP-2, on glucagon secretion in type 2 diabetes. *Am. J. Physiol.-Endocrinol. Metab.* **300,** E1038–E1046 (2011).
19. Steinert, R. E. *et al.* Ghrelin, CCK, GLP-1, and PYY(3–36): Secretory Controls and Physiological Roles in Eating and Glycemia in Health, Obesity, and After RYGB. *Physiol. Rev.* **97,** 411–463 (2016).
20. Ramracheya, R. D. *et al.* PYY-Dependent Restoration of Impaired Insulin and Glucagon Secretion in Type 2 Diabetes following Roux-En-Y Gastric Bypass Surgery. *Cell Rep.* **15,** 944–950 (2016).
21. Ritter, R. C., Covasa, M. & Matson, C. A. Cholecystokinin: proofs and prospects for involvement in control of food intake and body weight. *Neuropeptides* **33,** 387–399 (1999).
22. Cummings, D. E., Foster-Schubert, K. E. & Overduin, J. Ghrelin and energy balance: focus on current controversies. *Current. Drug Targets* **6,** 153–169 (2005).

23. Vincent, R. P. & le Roux, C. W. Changes in gut hormones after bariatric surgery. *Clinical. Endocrinology. (Oxf.)* **69,** 173–179 (2008).
24. le Roux, C. W. *et al.* Postprandial plasma ghrelin is suppressed proportional to meal calorie content in normal-weight but not obese subjects. *J. Clin. Endocrinol. Metab.* **90,** 1068–1071 (2005).
25. Unger, R. H. & Cherrington, A. D. Glucagonocentric restructuring of diabetes: a pathophysiologic and therapeutic makeover. *J. Clin. Invest.* **122,** 4–12 (2012).
26. Shah, P. *et al.* Lack of Suppression of Glucagon Contributes to Postprandial Hyperglycemia in Subjects with Type 2 Diabetes Mellitus. *J. Clin. Endocrinol. Metab.* **85,** 4053–4059 (2000).
27. Meier, J. J. *et al.* Gastric inhibitory polypeptide (GIP) dose-dependently stimulates glucagon secretion in healthy human subjects at euglycaemia. *Diabetologia* **46,** 798–801 (2003).
28. Nauck, M. A. *et al.* Preserved incretin activity of glucagon-like peptide 1 [7-36 amide] but not of synthetic human gastric inhibitory polypeptide in patients with type-2 diabetes mellitus. *J. Clin. Invest.* **91,** 301–307 (1993).
29. Ramón, J. M. *et al.* Effect of Roux-en-Y Gastric Bypass vs Sleeve Gastrectomy on Glucose and Gut Hormones: a Prospective Randomised Trial. *J. Gastrointest. Surg.* **16,** 1116–1122 (2012).
30. Kassem, M. A. M. *et al.* The Impact of Bariatric Surgery on Type 2 Diabetes Mellitus and the Management of Hypoglycemic Events. *Frontiers in Endocrinology.* **8,** (2017).

31. Antuna-Puente, B., Feve, B., Fellahi, S. & Bastard, J.-P. Adipokines: the missing link between insulin resistance and obesity. *Diabetes Metabolism.* **34,** 2–11 (2008).
32. Kahn, S. E., Cooper, M. E. & Del Prato, S. PATHOPHYSIOLOGY AND TREATMENT OF TYPE 2 DIABETES: PERSPECTIVES ON THE PAST, PRESENT AND FUTURE. *Lancet* **383,** 1068–1083 (2014).
33. Peterli, R. *et al.* Improvement in Glucose Metabolism After Bariatric Surgery: Comparison of Laparoscopic Roux-en-Y Gastric Bypass and Laparoscopic Sleeve Gastrectomy: A Prospective Randomized Trial. *Ann. Surg.* **250,** 234–241 (2009).
34. Butner, K. L., Nickols-Richardson, S. M., Clark, S. F., Ramp, W. K. & Herbert, W. G. A review of weight loss following Roux-en-Y gastric bypass vs restrictive bariatric surgery: impact on adiponectin and insulin. *Obes. Surg.* **20,** 559–568 (2010).
35. Kokkinos, A. *et al.* Improvement in Cardiovascular Indices After Roux-en-Y Gastric Bypass or Sleeve Gastrectomy for Morbid Obesity. *Obes. Surg.* **23,** 31–38 (2013).
36. Bult, M. J. F., van Dalen, T. & Muller, A. F. Surgical treatment of obesity. *European. Journal of Endocrinology.* **158,** 135–145 (2008).
37. Rizzello, M. *et al.* Early Postoperative Insulin-Resistance Changes After Sleeve Gastrectomy. *Obes. Surg.* **20,** 50–55 (2010).

38. Romero, F. *et al.* Comparable early changes in gastrointestinal hormones after sleeve gastrectomy and Roux-En-Y gastric bypass surgery for morbidly obese type 2 diabetic subjects. *Surgical. Endoscopy.* **26,** 2231–2239 (2012).
39. Isbell, J. M. *et al.* The Importance of Caloric Restriction in the Early Improvements in Insulin Sensitivity After Roux-en-Y Gastric Bypass Surgery. *Diabetes Care* **33,** 1438–1442 (2010).
40. Dixon J. B., Zimmet P., Alberti K. G., Rubino F. & null null. Bariatric surgery: an IDF statement for obese Type 2 diabetes. *Diabetic. Medicine.* **28,** 628–642 (2011).
41. Abbatini, F. *et al.* Long-term effects of laparoscopic sleeve gastrectomy, gastric bypass, and adjustable gastric banding on type 2 diabetes. *Surg. Endosc.* **24,** 1005–1010 (2010).
42. Vidal, J. *et al.* Short-term Effects of Sleeve Gastrectomy on Type 2 Diabetes Mellitus in Severely Obese Subjects. *Obes. Surg.* **17,** 1069–1074 (2007).
43. Vidal, J. *et al.* Type 2 Diabetes Mellitus and the Metabolic Syndrome Following Sleeve Gastrectomy in Severely Obese Subjects. *Obes. Surg.* **18,** 1077 (2008).
44. Cutolo, P. P. *et al.* Clinical Efficacy of Laparoscopic Sleeve Gastrectomy vs Laparoscopic Gastric Bypass in Obese Type 2 Diabetic Patients: a Retrospective Comparison. *Obes. Surg.* **22,** 1535–1539 (2012).
45. Yip, S., Plank, L. D. & Murphy, R. Gastric Bypass and Sleeve Gastrectomy for Type 2 Diabetes: A Systematic Review and Meta-analysis of Outcomes. *Obes. Surg.* **23,** 1994–2003 (2013).

46. Leyba, J. L., Aulestia, S. N. & Llopis, S. N. Laparoscopic Roux-en-Y Gastric Bypass Versus Laparoscopic Sleeve Gastrectomy for the Treatment of Morbid Obesity. A Prospective Study of 117 Patients. *Obes. Surg.* **21,** 212–216 (2011).
47. Zhang, N. *et al.* Reduction in obesity-related comorbidities: is gastric bypass better than sleeve gastrectomy? *Surg. Endosc.* **27,** 1273–1280 (2013).
48. Nocca, D. *et al.* Impact of Laparoscopic Sleeve Gastrectomy and Laparoscopic Gastric Bypass on HbA1c Blood Level and Pharmacological Treatment of Type 2 Diabetes Mellitus in Severe or Morbidly Obese Patients. Results of a Multicenter Prospective Study at 1 Year. *Obes. Surg.* **21,** 738–743 (2011).
49. Kashyap, S. R. *et al.* Acute effects of gastric bypass versus gastric restrictive surgery on β-cell function and insulinotropic hormones in severely obese patients with type 2 diabetes. *Int. J. Obes.* **34,** 462 (2010).
50. Lee, W.-J. *et al.* Gastric Bypass vs Sleeve Gastrectomy for Type 2 Diabetes Mellitus: A Randomized Controlled Trial. *Archives of Surgery* **146,** 143–148 (2011).
51. Vidal, J. *et al.* Long-Term Effects of Roux-en-Y Gastric Bypass Surgery on Plasma Glucagon-Like Peptide-1 and Islet Function in Morbidly Obese Subjects. *J. Clin. Endocrinol. Metab.* **94,** 884–891 (2009).
52. Jiménez, A. *et al.* Long-Term Effects of Sleeve Gastrectomy and Roux-en-Y Gastric Bypass Surgery on Type 2 Diabetes Mellitus in Morbidly Obese Subjects: *Ann. Surg.* **256,** 1023–1029 (2012).

53. Nannipieri, M. *et al.* Roux-en-Y gastric bypass and sleeve gastrectomy: mechanisms of diabetes remission and role of gut hormones. *J. Clin. Endocrinol. Metab.* **98,** 4391–4399 (2013).
54. Karamanakos, S. N., Vagenas, K., Kalfarentzos, F. & Alexandrides, T. K. Weight Loss, Appetite Suppression, and Changes in Fasting and Postprandial Ghrelin and Peptide-YY Levels After Roux-en-Y Gastric Bypass and Sleeve Gastrectomy: A Prospective, Double Blind Study. *Ann. Surg.* **247,** 401–407 (2008).
55. Borg, C. M. *et al.* Progressive rise in gut hormone levels after Roux-en-Y gastric bypass suggests gut adaptation and explains altered satiety. *British Journal of Surgery.* **93,** 210–215 (2006).
56. Yousseif, A. *et al.* Differential Effects of Laparoscopic Sleeve Gastrectomy and Laparoscopic Gastric Bypass on Appetite, Circulating Acyl-ghrelin, Peptide YY3-36 and Active GLP-1 Levels in Non-diabetic Humans. *Obes. Surg.* **24,** 241–252 (2014).
57. le Roux, C. W. *et al.* Gut hormones as mediators of appetite and weight loss after Roux-en-Y gastric bypass. *Ann. Surg.* **246,** 780–785 (2007).
58. Korner, J. *et al.* Prospective study of gut hormone and metabolic changes after adjustable gastric banding and Roux-en-Y gastric bypass. *Int. J. Obes. 2005* **33,** 786–795 (2009).
59. Rodieux, F., Giusti, V., D'Alessio, D. A., Suter, M. & Tappy, L. Effects of gastric bypass and gastric banding on glucose kinetics and gut hormone release. *Obes. Silver Spring Md* **16,** 298–305 (2008).

60. Peterli, R. *et al.* Metabolic and Hormonal Changes After Laparoscopic Roux-en-Y Gastric Bypass and Sleeve Gastrectomy: a Randomized, Prospective Trial. *Obes. Surg.* **22,** 740–748 (2012).
61. Holdstock, C. *et al.* Ghrelin and adipose tissue regulatory peptides: effect of gastric bypass surgery in obese humans. *J. Clin. Endocrinol. Metab.* **88,** 3177–3183 (2003).
62. Rubino, F. *et al.* The early effect of the Roux-en-Y gastric bypass on hormones involved in body weight regulation and glucose metabolism. *Ann. Surg.* **240,** 236–242 (2004).
63. Kellum, J. M. *et al.* Gastrointestinal hormone responses to meals before and after gastric bypass and vertical banded gastroplasty. *Ann. Surg.* **211,** 763-770; discussion 770-771 (1990).
64. Jacobsen, S. H. *et al.* Changes in gastrointestinal hormone responses, insulin sensitivity, and beta-cell function within 2 weeks after gastric bypass in non-diabetic subjects. *Obes. Surg.* **22,** 1084–1096 (2012).
65. Dirksen, C. *et al.* Gut hormones, early dumping and resting energy expenditure in patients with good and poor weight loss response after Roux-en-Y gastric bypass. *Int. J. Obes. 2005* **37,** 1452–1459 (2013).
66. Laferrère, B. *et al.* Effect of Weight Loss by Gastric Bypass Surgery Versus Hypocaloric Diet on Glucose and Incretin Levels in Patients with Type 2 Diabetes. *J. Clin. Endocrinol. Metab.* **93,** 2479–2485 (2008).

67. Hansen, E. N. *et al.* Role of the foregut in the early improvement in glucose tolerance and insulin sensitivity following Roux-en-Y gastric bypass surgery. *Am. J. Physiol. Gastrointest. Liver Physiol.* **300,** G795-802 (2011).
68. Swarbrick, M. M. *et al.* Longitudinal changes in pancreatic and adipocyte hormones following Roux-en-Y gastric bypass surgery. *Diabetologia* **51,** 1901–1911 (2008).
69. Goldfine, A. B. *et al.* Patients with Neuroglycopenia after Gastric Bypass Surgery Have Exaggerated Incretin and Insulin Secretory Responses to a Mixed Meal. *J. Clin. Endocrinol. Metab.* **92,** 4678–4685 (2007).
70. Maleckas, A., Gudaitytė, R., Petereit, R., Venclauskas, L. & Veličkienė, D. Weight regain after gastric bypass: etiology and treatment options. *Gland Surgery.* **5,** 617–624 (2016).
71. Kofman, M. D., Lent, M. R. & Swencionis, C. Maladaptive eating patterns, quality of life, and weight outcomes following gastric bypass: results of an Internet survey. *Obes. Silver Spring Md* **18,** 1938–1943 (2010).
72. Santo, M. A. *et al.* Weight Regain After Gastric Bypass: Influence of Gut Hormones. *Obes. Surg.* **26,** 919–925 (2016).

CURRICULUM VITAE