

Effect of weight loss by gastric bypass surgery versus hypocaloric diet on glucose and incretin levels in patients with type 2 diabetes

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Abstract

Context: Gastric bypass surgery (GBP) results in rapid weight loss, improvement of type 2 diabetes (T2DM) and increase in incretins levels. Diet-induced weight loss also improves T2DM and may increase incretin levels.

Objective: To determine whether the magnitude of the change of the incretin levels and effect is greater after GBP compared to a low caloric diet, after equivalent weight loss.

Design and Methods: Obese women with T2DM studied before and 1 month after GBP (n=9) or after a diet-induced equivalent weight loss (n=10). Patients from both groups were matched for age, body weight, BMI, diabetes duration and control, and amount of weight loss.

Setting: Outpatient GCRC.

Main Outcome Measures: Glucose, insulin, proinsulin, glucagon, GIP and GLP-1 levels were measured after 50 gr oral glucose. The incretin effect was measured as the difference in insulin levels in response to oral and to an isoglycemic iv glucose load.

Results: At baseline, none of the outcome variables (fasting and stimulated values) were different between the GBP and the diet group. Total GLP-1 levels after oral glucose markedly increased 6 times (peak: 17 ± 6 to 112 ± 54 pmol/L, $p < 0.001$) and the incretin effect increased 5 times (9.4 ± 27.5 % to 44.8 ± 12.7 %, $p < 0.001$) after GBP but not after diet. Post-prandial glucose levels ($p = 0.001$) decreased more after GBP.

Conclusions: These data suggest that early after GBP, the greater GLP-1 and GIP release and improvement of incretin effect are related not to weight loss but rather to the surgical procedure. This could be responsible for better diabetes outcome after GBP.

Introduction

Together with the epidemic of obesity (1), the number of weight loss surgeries has surged in the last decade (2). Roux-en-Y gastric bypass surgery (GBP) results in significant and prolonged weight loss with resolution of T2DM in 80% of cases (3). The mechanism by which T2DM improves rapidly after GBP, often prior to significant weight loss, has not yet been elucidated. The hormonal changes described after GBP suggest a possible endocrine effect of this surgery. We (4) and others (5-10) have shown that the meal- or glucose-stimulated incretin levels, which are blunted in T2DM, increase after GBP. In parallel with the increased levels of glucagon like peptide-1 (GLP-1) and gastric inhibitory peptide (GIP), the incretin effect on insulin secretion, impaired in patients with T2DM, markedly increased to levels similar to that of matched controls without T2DM, one month after GBP (4).

The two main incretins, GIP and GLP-1 (11), are secreted by the endocrine cells of the intestinal mucosa (12) in response to food and are responsible for 50-60% of insulin secretion after meals (12;13). The incretin effect is impaired in patients with T2DM (14). GLP-1 levels are blunted (15), but the effect of administered GLP-1 on insulin secretion persists (16). GIP levels are usually normal in patients with T2DM but the effect of administered GIP on insulin secretion is blunted (17), although can be restored under normal glycemic conditions (18). GIP and GLP-1 are rapidly degraded by the enzyme dipeptidyl peptidase-IV (DPP-IV) (19). GLP-1 and GIP analogues and DPP-IV inhibitors are in use or currently being developed as anti-diabetic agents (20).

The markedly increased incretin levels and effect observed after GBP could be one of the key mediators of the anti-diabetic effects of the surgery. However, weight loss occurs very rapidly after GBP and it is unclear whether the early changes in incretin levels and effect are a result of the surgery or could be attributed to weight loss *per se*. The blunted post-prandial response of GLP-1 observed in severely obese individuals (21) has been shown to improve after diet-induced weight loss (22). Other studies suggest that it is not the weight loss, but the surgical procedure that is responsible for the improvement of glucose tolerance. Ileal transposition, which increases GLP-1 levels, results in improved glucose control (23) independently of weight loss in rodent models (24) as well as in humans (25).

The goal of this study was to determine whether the magnitude of the change of the incretin levels and effect is greater after GBP compared to a low caloric diet, under conditions of short-term equivalent weight loss, in morbidly obese patients with T2DM. Specifically, we measured the changes in GLP-1 and GIP levels after oral glucose stimulation, and their incretin effect on insulin, in obese patients with T2DM before and one month after GBP, or after an equivalent diet-induced weight loss. A second goal was to determine whether an equivalent weight loss, achieved by GBP or by diet, would result in the same improvement of blood glucose levels in patients with T2DM.

Subjects and Methods

Subjects

Obese patients with BMI >35 kg/m², eligible candidates for GBP surgery, less than 60 years of age, of both genders and all ethnic groups, with T2DM diagnosed for

less than five years, not on insulin, thiazolidinedione, exenatide or DPP-IV inhibitors, with an HbA1C less than 8 %, were invited to participate in the study. All participants signed an informed consent, approved by our institution, prior to enrolling in the study. One group of patients was studied prior to and one month after GBP (surgical group). A second group of patients, fulfilling the same recruitment criteria, was studied before and after a 10kg diet-induced weight loss (diet group). In addition, patients in the diet group were matched for age, weight, BMI, T2DM duration and control (HbA1C) to patients from the surgical group.

Diet-induced weight loss and diabetes treatment

The diet consisted of a meal replacement plan of 1000 kcal/day. One week supply of meal replacement products (Robard Corporation, Mt. Laurel, NJ), including high-protein shakes, bars, fruit drinks and soups, was given to each patient during individual weekly visit at the GCRC. Fresh fruits and vegetables were allowed. Body weight was measured weekly and the diet adjusted when necessary. If no weight loss, or if weight gain occurred at two consecutive weekly visits, the patients were excluded from the study. Patients were kept on the 1000 kcal diet and in negative energy balance (active weight loss) while retested for incretin levels and effect after a 10kg weight loss.

Although there was no time limit, the expectation was that patients would lose 10 kg in 4 to 8 weeks. During the weight loss, patients were asked to monitor blood glucose levels by finger stick and keep logs. Diabetes medications were adjusted by a nurse educator or a diabetologist to avoid hypoglycemia and to fulfill the American Diabetes Association standard of treatment, based on fasting and post-prandial glucose levels. In most cases,

patients on sulfonylureas had their medication decreased or discontinued to avoid hypoglycemia.

Roux-en-Y Gastric Bypass (GBP) Protocol

All patients underwent a laparoscopic GBP. In brief, the jejunum was divided 30cm from the ligament of Treitz and anastomosed to a 30ml proximal gastric pouch. The jejunum was reanastomosed 150 cm distal to the gastrojejunostomy. All mesenteric defects were closed. The post-GBP diet recommendations included a daily intake of 600-800 kcal, 70 g of protein and 1.8 L of fluid. This was achieved, on an individual basis, with multiple small meals and snacks with various commercial protein supplements. The diet after GBP was monitored by food records but not directly supervised. The diet in the few days preceding the testing in surgical or diet patients before weight loss was not controlled for.

Incretin effect: insulin secretion after oral and isoglycemic iv glucose load (IsoG IVGT)

Subjects were studied for the oral glucose tolerance test (OGTT) and the isoglycemic iv glucose test (IsoG IVGT) in the morning after a 12-h overnight fast, on two different days, separated by less than 5 days.

3-h oral glucose tolerance test (OGTT)

All patients underwent first a 3-h OGTT with 50 g of glucose (non-carbonated, in a total volume of 200ml). After iv insertion, at 8:00 A.M., subjects received orally 50 g of glucose. Blood samples, collected on chilled EDTA tubes with added aprotinin (500 KIU

(kallikrein inhibitory units)/ml of blood) and dipeptidyl-peptidase IV (DPPIV) inhibitor (Linco St. Charles, MO) (10µl/ml of blood), were centrifuged at 4°C prior to storage at -70°C.

Isoglycemic iv glucose test (IsoG IVGT)

The goal of the IsoG IVGT was to expose the pancreas to blood glucose levels matched to the ones obtained during the OGTT in the same subject. Glucose (sterile 20% dextrose solution in water) was infused intravenously over 3 hours using a Gemini pump (Gemini, Inc.). A sample of blood was collected every 5 minutes, using a contra lateral antecubital iv catheter, then transferred in a microcentrifuge tube without any additive and centrifuged at bed side for immediate measure of glucose levels. The glucose infusion rate was adjusted in order to match the glucose concentrations obtained for the same patient during the OGTT at each time point for 3 hours. For insulin levels, blood samples were collected every 15 min for the first 90 min then every 30 min until 180'. During the OGTT and the IsoG IVGT, the arm used for blood sampling was kept warm with a heating pad.

Incretin Effect (INC)

The difference in β-cell responses (insulin total area under the curve or INS AUC (0-180') to the oral and the isoglycemic iv glucose stimuli represents the action of the incretin factor expressed as the percentage of the physiological response to oral glucose, which is taken as the denominator (100%) (26). The formula is:

$$INC = \frac{INS AUC_{oral} - INS AUC_{isoglycemic\ iv}}{INS AUC_{oral}} \times 100\%$$

Assays

Total GLP-1, an indicator of GLP secretion, was measured by radioimmunoassay (RIA) (Linco St.Charles, MO) after plasma ethanol extraction. The intra-assay and inter-assay CV were 3-6.5% and 4.7-8.8% respectively. This assay cross reacts 100% with GLP-1₇₋₃₆, GLP-1₉₋₃₆ and GLP-1₇₋₃₇ but does not cross react with glucagon (0.2%), GLP-2 (<0.01%) or exendin (<0.01%). Active GLP-1, an indicator of GLP potential action, was measured by ELISA (Linco St. Charles, MO). The intra-assay and inter-assay CV were 3-7% and 7-8% respectively. The assay cross reacts 100% with GLP-1₇₋₃₆ and GLP-1₇₋₃₇ but does not cross react with GLP-1₉₋₃₆, glucagon or GLP-2. Total GIP was measured by ELISA (Linco St. Charles, MO). The assay cross reacts 100% with GIP 1-42 and GIP 3-42 but does not cross react with GLP-1, GLP-2, oxyntomodulin or glucagon. The intra-assay and inter-assay CV were 3.0 -8.8% and 1.8-6.1% respectively. Plasma insulin, C-peptide, proinsulin and glucagon concentrations were measured by RIA (Linco St. Charles, MO) with intra-assay CV of 3-8%, and inter-assay CV of 5.5 % to 9%. The glucagon assay cross reacts 100% with glucagon but cross reacts less than 0.1% with oxyntomodulin. Glucose concentration was measured at the bedside by the glucose oxidase method (Beckman glucose analyzer, Fullerton, CA). All hormonal and metabolites assays were performed at the Hormone and Metabolite Core Laboratory of the New York Obesity Research Center.

Statistical analysis

Outcome variables were plasma glucose and plasma insulin, C-peptide, glucagon, pro-insulin, GLP-1 and GIP concentrations. Total areas under the curve 0-180' (AUC) for outcome variables were calculated using the trapezoidal method. ANOVA with repeated

measures was used to detect glucose and hormonal changes over time during the OGTT within each condition and for comparison before and after GBP and before and after diet, or between diet and surgical groups with T2DM. Paired t-tests were used to compare data between before and after GBP or diet. Data are expressed as the mean \pm SD except in the figures where SEM are used. Statistical significance was set at $p < 0.05$. Statistical analyses were performed with SPSS 14.0 (SPSS Inc., Chicago, IL).

Results

Subject characteristics

Subject characteristics are shown in Table 1. Obese women with T2DM and normal liver enzymes, thyroid function tests and blood pressure, were studied before and 1 month after GBP (n=9) and before and after an equivalent diet-induced weight loss (n=10). Of the 12 women recruited in the diet group 2 women did not complete the weight loss due to pregnancy or breast cancer and data from 10 diet completers are presented. Diabetes medications, sulfonylureas and/or metformin, were discontinued 3 days prior to being studied at baseline in all patients and were adjusted during the diet-induced weight loss to avoid hypoglycemia. Patients from the diet and surgical group were matched for age, body weight, BMI, diabetes duration and control (HbA1C) (Table 1). Prior to weight loss, neither fasting glucose ($p=0.874$), proinsulin ($p=0.797$), insulin ($p=0.629$), C-peptide ($p=0.589$), glucagon ($p=0.363$), GLP-1 ($p=0.832$) and GIP ($p=0.414$) and incretin effect ($p=0.245$) nor stimulated variables during the OGTT were significantly different between the diet and the GBP group.

Side effects

Although 50 g of glucose drink was used rather than 75g to minimize the risk of dumping syndrome after GBP, four patients experienced stomach cramping and discomfort, nausea, sweating, flushing and palpitations 5 to 20 minutes into the OGTT. No severe adverse effects were observed. There was no adverse effect from the diet.

Effect of weight loss

All patients in the surgical group discontinued their diabetes medications the day of the surgery. In the diet group, diabetes medications were either discontinued (n=2) or the dosage decreased, with patients taking only low doses of metformin (n= 8) at the completion of the weight loss. The duration of weight loss was shorter for the GBP group (32.3 ± 13.1 days) compared to the diet group (55.0 ± 9.9 days, $p=0.001$). Body weight, BMI, fasting glucose, insulin, C-peptide, proinsulin, proinsulin/insulin ratio and HOMA-IR decreased significantly and equally in the surgical and the diet group (Table 1). Fasting incretins did not change with either weight loss treatment.

Glucose AUC and glucose levels at 120' were significantly lower after GBP compared to diet ($p= 0.014$ and $p=0.001$ respectively) (Table 1). Although the changes of insulin with weight loss (fasting, AUC, peak response) were not different between GBP and diet, the pattern of secretion of insulin changed considerably after GBP ($p=0.001$), with recovery of the early phase with a peak at 30 min and a return to baseline after 180 min (Figure 1).

Stimulated levels of incretins increased significantly after surgery by a factor of 6 (peak GLP-1: 17 ± 6 to 112 ± 54 pmol/L, $p<0.001$), 2.1 (peak GLP-1 active, $p=0.038$), and

1.5 (peak GIP, $p=0.006$). (Table1, Figure 1). On the contrary, after diet, GLP-1 levels (total and active) tended to decrease ($p=NS$) and GIP did not change significantly (Table1). There was no change in GLP-1 or GIP levels during the IsoG IVGT (data not shown). A significant linear relationship was found between total GLP-1 release and insulin release during the OGTT, only in patients after GBP ($r=0.762$, $p=0.028$).

The glucose concentrations were well matched between the IsoG IVGT and the OGTT in the surgical and the diet group before and after the weight loss intervention (Table 2). The insulin response was not greater after oral than iv glucose before GBP or before diet, with a resulting blunted incretin effect (Table 2). The incretin effect increased significantly by a factor of 3.8 after GBP ($+35.4\pm 22.7\%$, $p=0.009$) but only minimally after diet ($+7.15\pm 18.13\%$, $p=0.244$).

Glucagon levels were mildly suppressed during the OGTT from 90 to 180 min ($p<0.001$) with no difference between the GBP and the diet group before intervention ($p=0.351$). After diet, glucagon levels remained similarly suppressed during the OGTT. However, after GBP, there was a paradoxical increase of glucagon levels during the OGTT from 15 to 120 min ($p=0.001$) (Figure 1).

Discussion

The magnitude of the effect of GBP on T2DM resolution has thus far baffled scientist and clinicians. Many studies (5;6;8;10;20;27-30), including ours (4), have shown an increase of incretin levels after GBP. It is unclear whether the caloric restriction and/or the rapid weight loss contribute to the change of the incretins levels, as diet-induced weight loss has also been associated with an increase, although of lesser magnitude, of

GLP-1 levels (22). In this study, we compared the effect of an equivalent weight loss by GBP or by diet, in two groups of matched morbidly obese patients with T2DM. The intensive diet intervention with meal replacements and weekly outpatient visits resulted in a weight loss equivalent to that lost by patients one month after the GBP. Although the well matched surgical and diet groups lost the same amount of weight, their changes in incretin levels were strikingly different. As shown previously (4), GLP-1 response to oral glucose markedly increased one month after GBP. GLP-1 levels tended to decrease after diet intervention, although this decrease was not significant. This is contrary to the results by Verdich et al (22) who showed an increase of GLP-1 levels, although of smaller magnitude, during a mixed meal after weight reduction in men. These differences between studies could be due to gender differences, the absence of T2DM, a greater weight loss (18.8 kg) and/or the use of a solid mixed meal as a stimulus in the study by Verdich (22). Our data on increase GIP levels after GBP are consistent with data after jejunio-ileal bypass (JIB) (31) but contrary to other study showing a decrease of GIP levels in patients without T2DM 6 months after GBP (30). The time of testing after surgery, which varied between studies, could be an important variable as we have shown that the increase of GIP levels is transient and does not persist 6 to 12 months after GBP (32). In parallel with the increase in incretin levels, the incretin effect markedly increased after GBP, but not after diet. These results need to be interpreted with caution as the sample size was small with large individual variation.

The data from this study suggest that the effect of GBP on the incretins is likely not weight loss related. However, the mechanism by which the incretins increase after GBP remains unclear. Whether it is the rapid and direct stimulation of the L cells of the

distal ileum, referred to as the hindgut mechanism, or the bypass of the duodenum (the foregut hypothesis) is still unclear. Elegant studies in rodents support the foregut hypothesis. In these studies in Goto-Kakizaki type 2 diabetic rats, the improvement of glucose tolerance after surgical exclusion of the duodenum, but not after gastro jejunostomy, is independent of calorie restriction or weight loss (33). The hindgut hypothesis is based on the results of experimental ileal transposition, a surgical procedure that improves diabetes in rodents (23) independently of weight loss (24). The foregut/hindgut hypothesis has not been tested in humans. Recent data demonstrate that ileal transposition with sleeve gastrectomy can improve diabetes, even after minimal weight loss, in patient with BMI less than 35 kg/m² (34). Our experiment was designed to address the effect of weight loss on incretins and did not allow us to separate out the effect of duodenal bypass versus rapid stimulation of the distal gut on incretin stimulation after GBP. Gastric emptying and intestinal transit time have been shown to increase after GBP (27;35), but not after diet (22). The release of GIP and GLP-1 is related to the rate of carbohydrates entry into the small intestine (36). Faster small intestinal glucose delivery increases plasma GIP and GLP-1 levels (37). The rapid delivery of nutrient after GBP could represent a mechanism by which incretins are markedly released after the surgery. We did not measure gastric emptying or intestinal transit time in our study. We therefore cannot exclude the possibility that, in the diet group, the glucose solution was absorbed entirely in the duodenum and did not reach the lower part of the ileum to exert its stimulating effect directly on the L cells to release GLP-1. However, recent data showed that the enteroendocrine K and L cells, which secrete GIP and GLP-1

respectively, are distributed all along the gut (38). It is therefore unlikely that the L cells would have had no contact with the glucose solution in the diet group.

We cannot exclude that gut adaptation played a role in the increased incretin response after GBP as hyperplasia of intestinal endocrine cells have been described three months after JIB (39). We have shown persistent increased GLP-1 levels one year after GBP (32) and others have shown increased incretin levels 20 years after JIB (31). Adaptative changes in gut motility have also been shown after a period of energy restriction and weight loss (40). We cannot exclude a role of calorie restriction per se, independently of weight loss, in the improvement of incretins after GBP. The daily calorie intake of patients after GBP was 600-800 kcals (data not shown) compared to 1000 kcal in the diet group. Although the calorie restriction was not matched between the 2 groups from day to day, the overall calorie deficit and weight loss were identical.

Diet-induced (41-46) or surgical weight loss (3;47) improve T2DM control. In this study, the effect of an equivalent weight loss on diabetes control was greater after GBP than after diet. Patients in the surgical group had a better clinical outcome and did not require diabetes medications after the weight loss. Additionally, post-prandial glucose levels were lower after GBP compared to diet. In the fed state, gastric emptying (GE) (36;37), glucose absorption (48) and the release of incretins (12) are key determinants of postprandial glucose levels. In patients with T2DM, many components of the gut physiology are impaired, such as gastric emptying (40;49) and incretin release and effect (50), resulting in postprandial hyperglycemia, a predictor of cardiovascular complications and mortality (51). Our data show that a diet-equivalent weight loss does not lower post-prandial glucose levels to the same extent as GBP in patients with T2DM. This may

indicate that the marked increase of incretins associated with GBP, and not with the diet, could be responsible for the better post-prandial glucose control. The administration of the synthetic exendin-4, a compound that binds to the GLP-1 receptor and exerts similar effects as the native GLP-1 (52), or of vildagliptin, an inhibitor of DPP-IV, the enzyme that rapidly inactivates the endogenous incretins (53;54), have been shown to reduce post-prandial glucose levels and improve diabetes control.

Patients with T2DM have typically hyper-glucagonemia (55), which contributes to the post-prandial hyperglycemia. It is puzzling to see that the decrease of post-prandial glucose levels after GBP is associated with a paradoxical increase of glucagon levels during the OGTT. The rise in glucagon is seen in spite of a marked increase in GLP-1, a gut hormone that inhibits glucagon release (56). This rise of glucagon levels was previously shown after GBP (2007) and ileal transposition in dogs (57). The source of this increase in glucagon is unclear. Although the commercial assay used in this study is specific for pancreatic glucagon, cross reactivity with enteroglucagon or oxyntomodulin cannot be entirely excluded. Our study group was small and limited to women. Future studies will need to address gender differences in incretin levels and effect after bariatric surgery.

In summary, our data suggest that it is the surgical procedure per se, rather than weight loss, which stimulates incretin release and effect after GBP. The rapid and marked increase GLP-1 levels after GBP plays an important role in insulin secretion and could be a key determinant in the decrease of postprandial glycemia and the resolution of T2DM after GBP.

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LEGENDS

Figure 1

Glucose, insulin, C-peptide, glucagon, total and active GLP-1 and GIP levels during the OGTT in patients before (diamond) and after (square) GBP and before (triangle) and after (circle) diet. Data are mean \pm SEM. * $p < 0.05$ between groups after weight loss by GBP or by diet.

Table 1- Subject characteristics before and after weight loss, by either diet or GBP^a

| | Pre-Diet | Post-Diet | Pre-GBP | Post-GBP | P |
|--|-------------|-------------|-------------|--------------|-------|
| Age (years) | 46.80±7.24 | | 44.56±9.65 | | |
| Weight (kg) | 110.9±10.2 | 101.1±9.5** | 113.2±15.5 | 103.2±16.6** | 0.816 |
| BMI (kg/m ²) | 43.3±3.6 | 39.6±3.5** | 43.3±6.2 | 39.5±6.5** | 0.939 |
| T2DM duration (months) | 22.2±17.7 | | 31.4±26.4 | | |
| HbA1C (%) | 6.68±0.88 | | 6.51±0.78 | | |
| Fasting glucose (mmol/L) | 7.84±1.09 | 6.34±1.00** | 7.95±1.74 | 6.42±0.80* | 0.957 |
| 120' glucose (mmol/L) | 10.17±2.40 | 9.62±2.34 | 11.12±1.64 | 7.22±1.62** | 0.001 |
| Peak glucose (mmol/L) | 12.83±1.36 | 11.45±3.04 | 14.24±3.23 | 12.02±1.38 | 0.514 |
| AUC glucose (mmol.L ⁻¹ . min ⁻¹) | 10.29±1.54 | 9.13±2.09* | 11.15±1.80 | 8.51±1.22** | 0.014 |
| Fasting insulin (pmol/L) | 192±108 | 110±50** | 172±69 | 127±51* | 0.195 |
| Peak insulin (pmol/L) | 545±357 | 575±522 | 492±288 | 769±335* | 0.286 |
| AUC insulin (pmol.L ⁻¹ . min ⁻¹) | 363±237 | 350±268** | 349±188 | 341±138 | 0.974 |
| Fasting C-peptide (pmol/L) | 4.09±2.27 | 3.47±2.35 | 4.04±1.71 | 3.71±1.01 | 0.656 |
| AUC C-peptide (pmol.L ⁻¹ . min ⁻¹) | 2.20±1.01 | 2.19±1.26 | 2.12±0.77 | 2.48±0.62 | 0.304 |
| Fasting glucagon (ng/L) | 71.6±10.6 | 58.5±16.0* | 65.9±15.0 | 67.0±21.8 | 0.002 |
| AUC glucagon (ng. L ⁻¹ .min ⁻¹) | 58.1±14.5 | 49.2±12.4* | 58.4±10.8 | 77.3±17.5* | 0.002 |
| Peak glucagon (ng/L) | 84.7±24.2 | 68.5±19.7* | 81.1±14.8 | 96.9±21.5* | 0.002 |
| Fasting Proinsulin (pmol/L) | 34.6±26.1 | 16.3±9.6* | 31.8±17.0 | 19.2±22.5** | 0.876 |
| Proinsulin/insulin | 0.18±0.11 | 0.16±0.11 | 0.19±0.07 | 0.16±0.19** | 0.082 |
| Fasting Total GLP-1 (pmol/L) | 6.18±2.87 | 6.34±4.36 | 6.52±4.06 | 6.69±3.28 | 0.998 |
| Peak Total GLP-1 (pmol/L) | 18.20±16.33 | 9.80±5.83 | 17.49±6.02 | 112.5±54.3** | 0.001 |
| AUC Total GLP-1 (pmol.L ⁻¹ .min ⁻¹) | 8.20±7.29 | 4.94±1.96 | 7.55±2.80 | 31.82±8.10** | 0.000 |
| Fasting Active GLP-1 (pmol/L) | 6.04±3.55 | 4.28±0.90 | 7.91±3.77 | 8.45±4.41 | 0.216 |
| Peak Active GLP-1 (pmol/L) | 10.85±9.73 | 5.27±1.74 | 11.21±3.94 | 24.13±19.31 | 0.038 |
| AUC Active GLP-1 (pmol.L ⁻¹ . min ⁻¹) | 6.43±3.69 | 4.25±0.96 | 7.38±2.98 | 10.88±4.94 | 0.029 |
| Fasting GIP (ng/L) | 34.18±11.41 | 33.84±33.11 | 39.27±15.05 | 40.54±29.87 | 0.901 |
| Peak GIP (ng/L) | 175±60 | 208±115 | 204±56 | 316±124** | 0.090 |
| AUC GIP (ng.L ⁻¹ .min ⁻¹) | 40.96±12.71 | 54.00±31.85 | 48.67±11.35 | 51.56±18.54 | 0.399 |
| HOMA-IR | 7.96±4.17 | 4.04±1.91** | 8.11±3.61 | 5.16±2.88* | 0.476 |

^aAUC= area under the curve. GLP-1T = GLP-1 total; GLP-1A= GLP-1 active.

T2DM=diabetes. HbA1C= hemoglobin A1C. Data are expressed as mean ± SD.

Fasting, peak, 120 min, AUC (total area under the curve, 180') values are obtained

during the OGTT. * $p < 0.05$, ** $p < 0.001$, effect of weight loss within each group (Diet, $n=10$ or GBP, $n=9$). The reported P value represents the difference between the changes occurring with either weight loss intervention.

Table 2- Mean Glucose, Insulin and C-peptide levels, Area Under the Curve for glucose and insulin during OGTT and Iso IVGT, before and after weight loss by either diet (n=10) or GBP (n=9)^a.

| | Before GBP | | After GBP | | Before Diet | | After Diet | |
|------------------------|------------|-----------|--------------|-----------|--------------|-----------|-------------|-----------|
| | Oral | IV | Oral | IV | Oral | IV | Oral | IV |
| GAUC mmol/L/min | 11.2±1.8 | 11.4±1.8 | 8.5±1.2 | 9.8±1.2* | 10.3±1.5 | 10.1±1.6 | 9.1±2.1 | 8.9±2.3 |
| Glucose mmol/L | 10.7±1.9 | 10.9±1.8 | 8.5±1.0 | 9.4±0.9 | 9.8±1.2 | 9.6±1.3 | 8.4±1.8 | 8.3±1.9 |
| IAUC pmol/L/min | 349±188 | 296±154 | 340±145 | 183±75* | 363±237 | 250±131 | 350±268 | 220±165 |
| Insulin pmol/L | 315±152 | 261±127 | 347±138 | 170±62* | 330±210 | 218±113 | 302±191 | 195±130 |
| C-Peptide pmol/L | 1.95±0.7 | 1.97±0.57 | 2.31±0.54 | 1.58±0.4* | 1.98±0.92 | 1.62±0.81 | 1.89±1.05 | 1.55±0.95 |
| Incretin effect (%) | 9.42±27.46 | | 44.84±12.73# | | 27.44±14.909 | | 34.59±15.15 | |

^a Data are expressed as mean ± SD. * p<0.05 between oral and IV glucose challenges within each group. The incretin effect was calculated by comparing the insulin response to oral and matched iv glucose load; #p=0.002 between before and after weight loss by either GBP or diet.

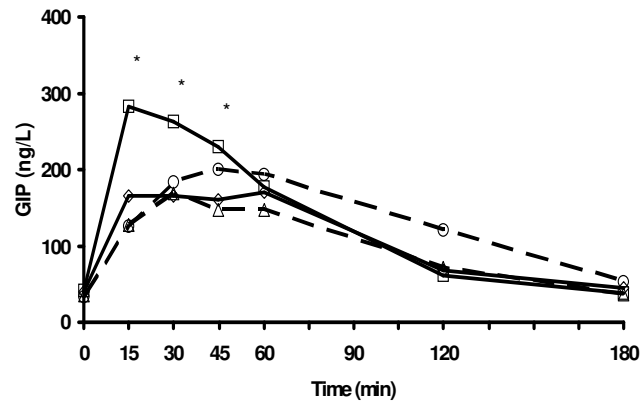
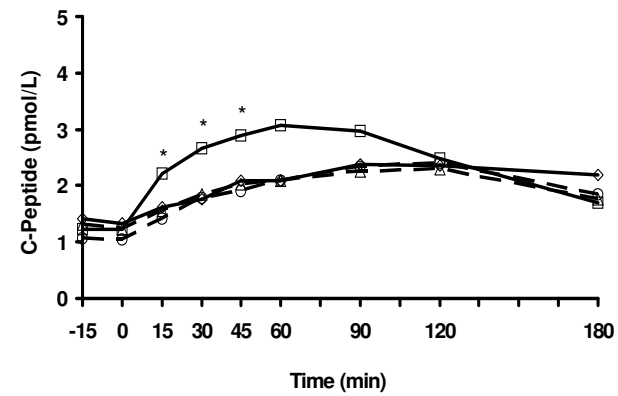
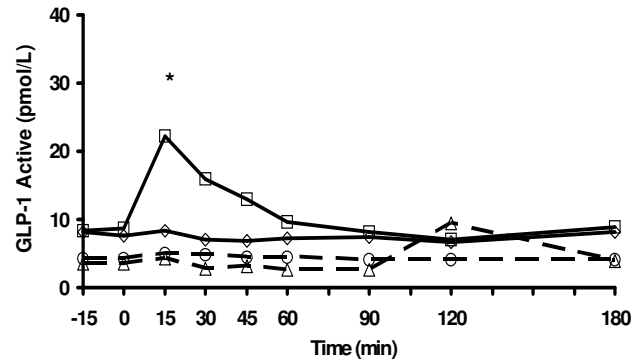
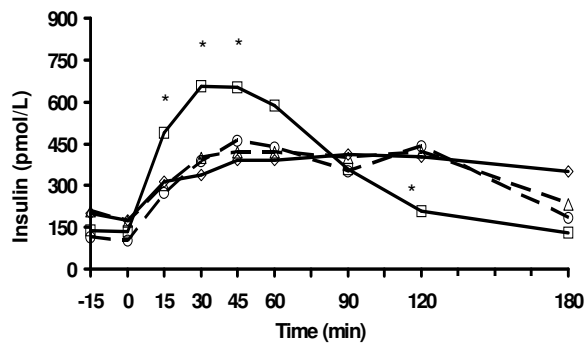
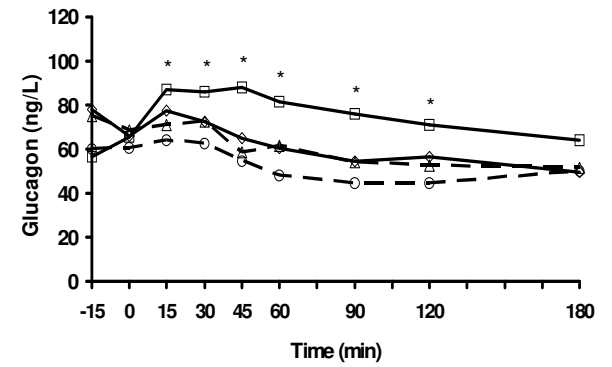
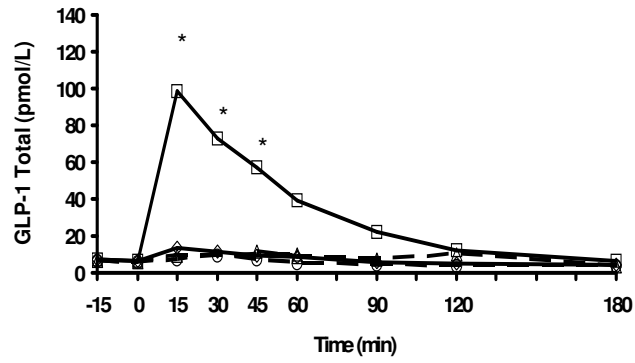
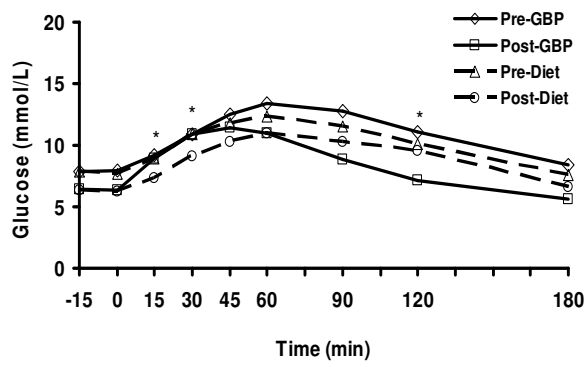


Figure 1