Diabetologia (2009) 52:873–881
DOI 10.1007/s00125-009-1288-9

ARTICLE

Circadian rhythms of GIP and GLP1 in glucose-tolerant and in type 2 diabetic patients after biliopancreatic diversion

G. Mingrone · G. Nolfe · G. Castagneto Gissey · A. Iaconelli · L. Leccesi · C. Guidone · G. Nanni · J. J. Holst

Received: 27 October 2008 / Accepted: 14 January 2009 / Published online: 20 February 2009
© Springer-Verlag 2009

Abstract
Aims/hypothesis We tested the hypothesis that the reversibility of insulin resistance and diabetes observed after biliopancreatic diversion (BPD) is related to changes in circadian rhythms of gastrointestinal hormones.
Methods Ten morbidly obese participants, five with normal glucose tolerance (NGT) and five with type 2 diabetes, were studied before and within 2 weeks after BPD. Within-day variations in glucose-dependent insulinotropic polypeptide (GIP) and glucagon-like peptide 1 (GLP1) levels were assessed using a single cosinor model. Insulin sensitivity was assessed by euglycaemic–hyperinsulinaemic clamp.
Results Basal GLP1 relative amplitude (amplitude/mesor×100) was 25.82–4.06% in NGT; it increased to 41.38–4.32% after BPD but was unchanged in diabetic patients. GLP1 and GIP mesor were shifted in time after surgery in diabetic patients but not in NGT participants. After BPD, the GLP1 AUC significantly increased from 775±94 to 846±161 pmol l^{-1} min in NGT, whereas GIP AUC decreased significantly from 1,373±565 to 513±186 pmol l^{-1} min in diabetic patients. Two-way ANOVA showed a strong influence of BPD on both GIP ($p=0.010$) and GLP1 AUCs ($p=0.033$), which was potentiated by the presence of diabetes, particularly for GIP (BPD×diabetes, $p=0.003$). Insulin sensitivity was markedly improved ($p<0.01$) in NGT (from 9.14±3.63 to 36.04±8.55 μmol [kg fat-free mass]$^{-1}$ min^{-1}) and diabetic patients (from 9.49±3.56 to 38.57±4.62 μmol [kg fat-free mass]$^{-1}$ min^{-1}).
Conclusions/interpretation An incretin circadian rhythm was shown for the first time in morbid obesity. The effect of BPD on the 24 h pattern of incretin differed between NGT and diabetic patients. GLP1 secretion impairment was reversed in NGT and could not be overcome by surgery in diabetes. On the other hand, GIP secretion was blunted after the operation only in diabetic patients, suggesting a role in insulin resistance and diabetes.

Keywords Bariatric surgery · Circadian rhythm · GIP · GLP1 · Morbid obesity

Abbreviations

BPD	Biliopancreatic diversion
FFM	Fat-free mass
GIP	Glucose-dependent insulinotropic polypeptide
GLP1	Glucagon-like peptide 1
NGT	Normal glucose tolerance
RYGB	Roux-en-Y gastric bypass

G. Mingrone (✉) · A. Iaconelli · L. Leccesi · C. Guidone
Department of Internal Medicine, Università Cattolica S. Cuore,
Largo A. Gemelli, 8,
00168 Rome, Italy
e-mail: gmingrone@rm.unicatt.it

G. Nolfe
CNR Institute of Cybernetics 'E. Caianiello',
Pozzuoli, Italy

G. Castagneto Gissey
Department of Economics, University of Kent,
Canterbury, UK

G. Nanni
Department of Surgery, Università Cattolica S. Cuore,
Rome, Italy

J. J. Holst
Department of Biomedical Sciences, Panum Institute,
University of Copenhagen,
Copenhagen, Denmark

Introduction

It has been clearly demonstrated that in diabetes the glucagon-like peptide 1 (GLP1) response is blunted [1] and that the response of pancreatic beta cells to glucose-dependent insulinotropic polypeptide (GIP) is severely impaired [2]. These and other observations have strengthened the hypothesis, advanced by Nauck et al. [3], that the beta cell incompetence observed in diabetes is, at least partially, due to an impaired effect of incretin.

The prevalence of obesity has recently reached epidemic proportions [4] in both Western and developing countries, with morbid obesity accounting for about 9% [5] of the whole obese population. On the other hand, obesity and type 2 diabetes are so strictly associated that the term 'diabesity' has been coined [6]. Because of such interdependence, it is difficult to discriminate between the separate roles of obesity and glucose tolerance in the effect of incretins on beta cell function. Recently, however, some light has been shed on this topic, showing that obesity and glucose tolerance each attenuate the effect of incretin and the GLP1 response independently of one another [7].

Lifestyle intervention programmes and pharmacotherapy fail to maintain long-term weight loss, particularly in severe obesity [8], whereas bariatric surgery is effective and associated with a significant reduction in mortality [9]. Relatively few data are available concerning the effect of bariatric surgery on incretin secretion [10–15] and, at least to our knowledge, no data exist regarding the response of incretin to multiple meals.

We have previously demonstrated that insulin resistance and type 2 diabetes are reversible after malabsorptive bariatric surgery [16–18]. However, we are far from understanding the mechanisms involved. In a previous study of ours [11], the time course of GIP and GLP1 after an OGTT showed a reduction in the GIP AUC and an increase in the GLP1 AUC early after the bariatric operation in obese diabetic people. In the present study, however, the fluctuations of incretins were studied over 24 h in near-physiological conditions.

To check the hypothesis that the reversibility of insulin resistance and diabetes observed after biliopancreatic diversion (BPD) is related to changes in the circadian rhythm of gastrointestinal hormones, the 24 h profiles of GLP1 and GIP were studied in ten morbidly obese participants, five with normal glucose tolerance (NGT) and five with type 2 diabetes, before and within 2 weeks after BPD.

Methods

Study protocol Ten morbidly obese women undergoing BPD were studied. Five were aged 44.0±8.6 years and had NGT as assessed by OGTT, and five were aged 42.6± 5.7 years (age difference not significant) and had type 2 diabetes mellitus with an onset 2–5 years before the study began. The glycosylated haemoglobin value was 7.2–9.0%. All the diabetic patients were being treated with oral hypoglycaemic agents. Immediately after BPD, medical therapy for diabetes was stopped.

All participants underwent the metabolic study at baseline and within 2 weeks after the bariatric surgery, spending 24 h (starting at 08:00 hours) on the metabolic ward. During this period, four meals were administered. The total daily energy intake of 125.6 kJ/kg fat-free mass (FFM) was distributed as follows: 13.4% was taken at breakfast (09:00 hours), 36% at 12:00–13:00 hours, 16.4% as an afternoon snack (16:00–16:30 hours) and 34.2% at dinner (19:00–20:00 hours). The average diet composition was 16.9% of energy as protein, 34.6% fat and 48.5% carbohydrates.

Blood samples were drawn from a central venous catheter each hour for the measurement of glucose, insulin, GIP and GLP1 concentrations.

The study protocol was approved by the Institutional Ethics Committee of the Catholic University of Rome. The nature and purpose of the study were carefully explained to all participants before they provided their written consent to participate.

BPD and body composition The BPD operation was as described previously [11]. Body weight was measured to the nearest 0.1 kg with a beam scale and height to the nearest 0.5 cm using a stadiometer (Holatin, Crosswell, UK). Total body water was measured by the labelled water dilution method and FFM and fat mass were calculated as described previously [11].

Euglycaemic–hyperinsulinaemic clamp Peripheral insulin sensitivity was evaluated by the euglycaemic–hyperinsulinaemic clamp technique [19] at baseline and within 2 weeks after surgery. Small boluses of insulin were administered subcutaneously to diabetic patients to achieve euglycaemia. Glucose disposal (M value) was calculated from the exogenous glucose infusion rate during the last 40 min of the 2 h clamp after correction for changes in glucose concentration in a total distribution volume of 250 ml/kg. Whole-body glucose disposal was normalised per kg FFM (M/kg_{FFM}).

Analytical methods Blood samples were drawn into EDTA-evacuated tubes. The plasma was immediately separated by centrifugation at 4°C and stored at −80°C until assay. The samples were not thawed until hormone assays were performed.

Plasma glucose was measured by the glucose oxidase method (Beckman, Fullerton, CA, USA).

Plasma insulin was assayed by microparticle enzyme immunoassay (Abbott, Pasadena, CA, USA) with a sensitivity of 6 pmol/l and an intra-assay CV of 6.6%.

GIP and GLP1 concentrations in plasma were measured after extraction of plasma with 70% ethanol (vol./vol., final concentration) according to techniques developed at the Panum Institute, University of Copenhagen [20, 21].

Rhythm analysis To compare intra-day time series of insulin, GIP and GLP1 obtained in the basal condition and after bariatric surgery, the following time averages were estimated:

$$\frac{1}{T}\int_0^T y(t)dt \equiv \langle y(t)\rangle \qquad \text{mean level}$$

$$\frac{1}{T}\int_0^T y^2(t)dt \equiv \langle y^2(t)\rangle \qquad \text{mean square (or intensity)}$$

where T=24 h. The mean intensity is proportional to the mean power of the time series.

Single cosinor models of the form:

$$z(t) = M + A\cos\left(\frac{2\pi t}{t} + \varphi\right) + e(t)$$

were used to model the variation in measured hormonal levels as a function of time t (hours). $z(t)$ and $e(t)$ are the measured concentration and error between the cosinor model and the measurement, respectively. The variable M denotes the mesor (value about which the variation occurs), A the amplitude (distance from mesor to peak) and φ (radians) the acrophase (the time of occurrence of the peak equals $\varphi T/2\pi$). T is the period of 24 h.

For a fixed value of T and known values of t, simple rearrangement of the model using trigonometric identities gives a linear model in the coefficients, M, γ and β,

$$z(t) = M + \gamma\cos\left(\frac{2\pi t}{T}\right) + \beta\sin\left(\frac{2\pi t}{T}\right) + e(t)$$

which can be fitted using conventional least-squares methods. Individual single cosinor models were fitted using least squares. The idea that the data are better explained by the null hypothesis (H0) of a constant value (mesor) than (H1) a sine was tested using a likelihood ratio (F) test: reject H0 for large values of GIP and GLP1. An F ratio $F_{2,69}$ (0.95) of 3.13 was considered significant. A group cosinor model was computed by averaging the coefficients from the individual fits.

Statistical analysis The participants were divided into two groups on the basis of the presence of normal glucose tolerance or diabetes.

Before statistical analysis, normal distribution and homogeneity of the variances were evaluated using Levene's test. Since the hormone data exhibited moderate right skewness, their square roots were taken to normalise the data set.

To determine how the hormonal change was affected by two factors (BPD and diabetes), two-way ANOVA was carried out; the sample size power was computed using α=0.05.

Regression analysis and ANOVA for multiple dependent variables by the two factor variables (BPD and diabetes) were obtained by using the general linear model multivariate procedure. Using this general linear model procedure, we tested null hypotheses about the effects of BPD and diabetes factor variables on the means of various groupings of a joint distribution of dependent variables, namely insulin sensitivity (M value), GIP AUC and GLP1 AUC. The interactions between factors as well as the effects of individual factors were also investigated.

Data are mean±SD unless otherwise specified. Statistical significance was assumed at $p<0.05$.

Statistical analyses were performed by using the statistical software package SPSS version 10.0 (SPSS, Chicago, IL, USA).

Results

As shown in Table 1, body composition did not change significantly in either NGT or diabetic participants after surgery.

Insulin sensitivity was markedly improved after BPD ($p<0.0001$), both in NGT participants (from 9.14±3.63 to 36.04±8.55 μmol kg_{FFM}^{-1} min^{-1}) and in diabetic patients (from 9.49±3.56 to 38.57±4.62 μmol kg_{FFM}^{-1} min^{-1}). No significant differences from before to after BPD were

Table 1 Patient characteristics

Characteristic	NGT		Type 2 diabetes	
	Before BPD	After BPD	Before BPD	After BPD
BMI (kg/m^2)	45.4±6.8	42.8±6.0	44.6±7.9	43.0±7.1
FFM (kg)	76.6±15.6	74.1±15.3	81.0±18.3	78.5±18.1
Fat mass (kg)	58.6±12.3	53.6±7.9	59.2±13.0	52.9±9.5

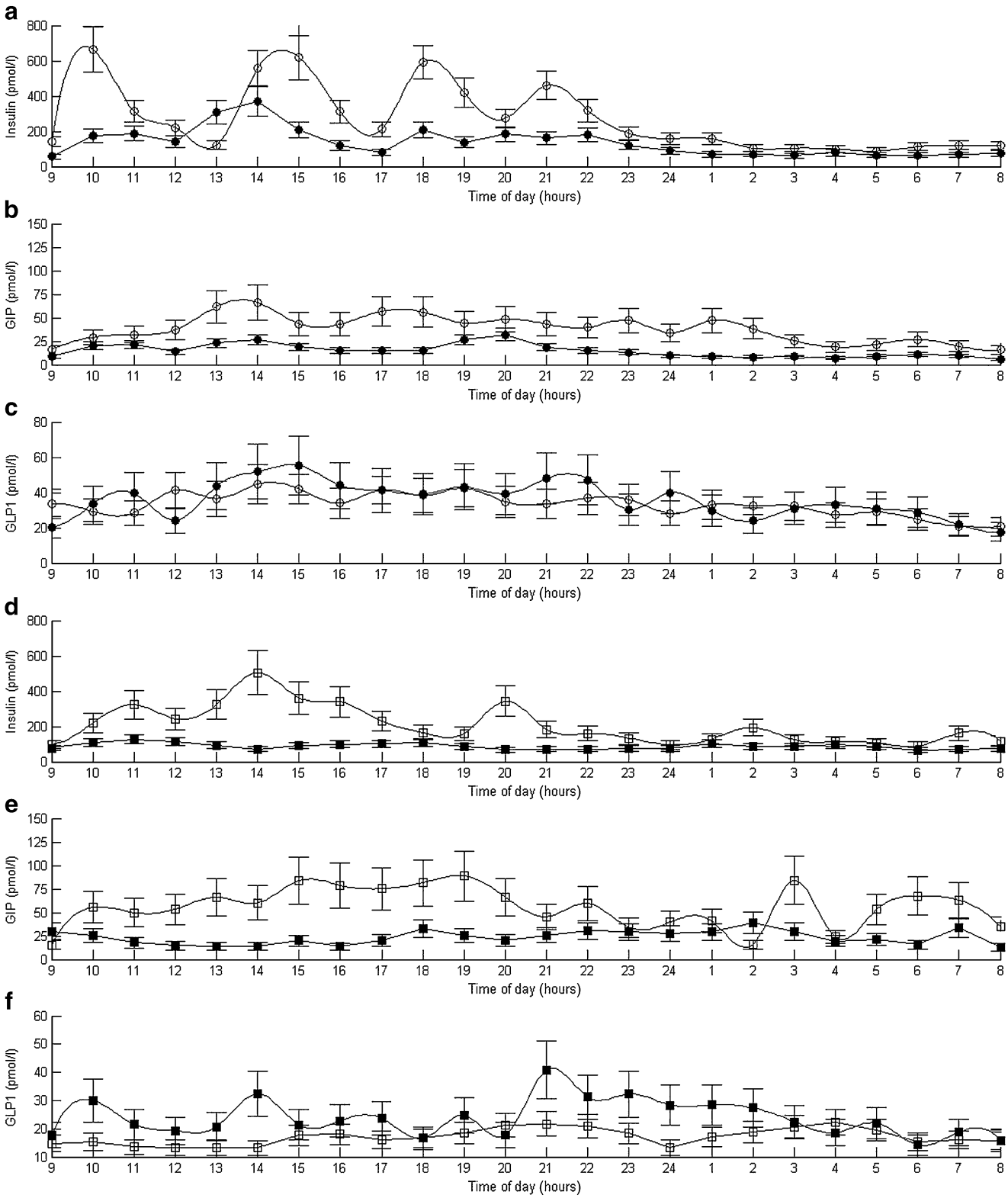

Fig. 1 Time courses of plasma insulin (**a**, **d**), GIP (**b**, **e**) and GLP1 (**c**, **f**) in NGT (circles) and type 2 diabetic (squares) participants before (white symbols) and after (black symbols) BPD. Values are means±SEM. Values on the *y*-axes represent time of day in the 24 h clock. The times of breakfast, lunch, snack and dinner were 09:00–09:30, 12:30–13:00, 16:30–17:00 and 19:30–20:00 hours, respectively. Sleeping time was between 22:30 and 08:00 hours

Table 2 Cosinor variables and AUC of incretins for NGT and type 2 diabetic participants

Variable	NGT				Type 2 diabetes			
	GLP1		GIP		GLP1		GIP	
	Before BPD	After BPD	Before BPD	After BPD	Before BPD	After BPD	Before BPD	After BPD
Acrophase[a]	17:56±1.08	17:14±1.31	18:30±1.54	16:48±0.19	07:22±3.51	11:14±2.53	13:56±2.42	11:22±3.07
Mesor	33.41±3.83	35.54±6.42	36.77±15.57	14.85±2.38	17.59±1.91	23.14±5.34	58.64±23.05	22.41±7.80
Amplitude	8.09±0.92	14.36±2.35	20.14±6.29	8.14±1.48	4.01±0.80	9.26±3.21	21.48±7.40	7.07±4.42
AUC (pmol l^{-1} min)	775±94	834±161	670±215	351±60	407±47	539±133	1,373±565	513±186

[a] Time of day in 24h clock ± time (h)

observed in plasma insulin levels during the clamp in each group (from 470.4±37.6 to 438.0±17.49 pmol/l in NGT participants and from 454.8±27.30 to 441.6±13.8 pmol/l in diabetic participants).

Figure 1 shows the 24 h concentration averaged profile of each hormone (mean±SEM) for diabetic and NGT participants. Visual inspection of the GIP and GLP1 time courses strongly supports the existence of a circadian rhythm. Based on this observation, we tested the hormone data for a periodic signal using single cosinor analysis, assuming a 24 h period.

Table 2 summarises the values of the cosinor variables for GLP1 and GIP. The mean±SEM percentage relative amplitude (amplitude/mesor×100) of GLP1 was 25.82± 4.06% in NGT participants before BPD and increased, but not significantly, to 41.38±4.32% after BPD. The relative amplitude of GLP1 did not change significantly in diabetic participants. In contrast, the relative amplitude of GIP was reduced more in type 2 diabetic patients (from 41.38±4.32 to 25.82±4.06%) than in NGT participants (63.00±16.27 to 51.24±8.26%).

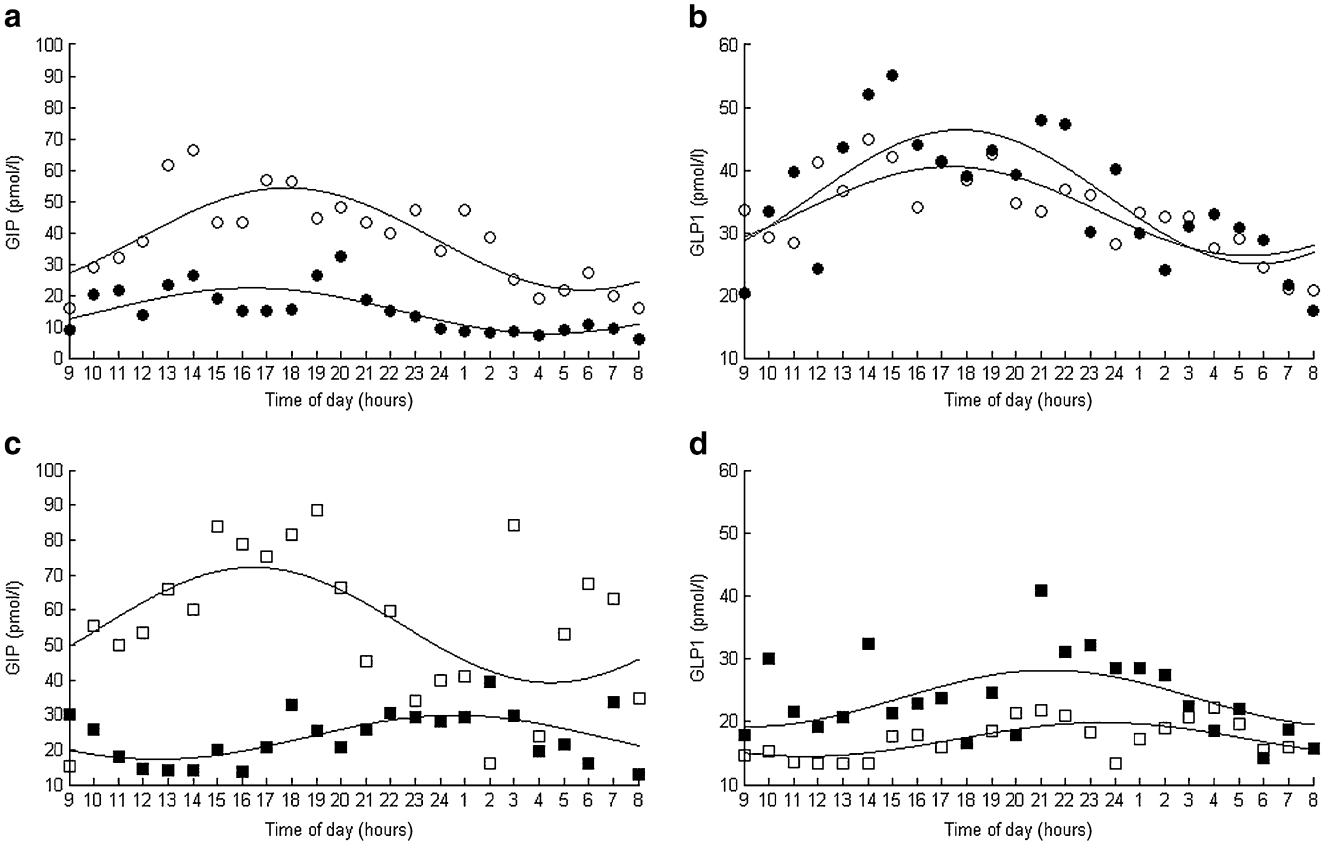

Fig. 2 Estimated cosinor (solid line) before (white symbols) and after (black symbols) BPD in NGT (**a**, **b**) and type 2 diabetic participants (**c**, **d**). Values on the *y*-axes represent time of day in the 24 h clock. The symbols are as for Fig. 1

Table 3 Results of the two-way ANOVA test for relevant measures

Factor	*F* value	*p* value	Observed power
GIP AUC (pmol l^{-1} min)			
BPD	8.636	0.010[a]	0.788[a]
Diabetes	5.064	0.043[a]	0.543[a]
BPD×diabetes	12.520	0.003[a]	0.913[a]
GLP1 AUC (pmol l^{-1} min)			
BPD	5.463	0.033[a]	0.697[a]
Diabetes	3.734	0.071	
BPD×diabetes	7.889	0.013[a]	0.823[a]
Insulin AUC (pmol l^{-1} min)			
BPD	10.070	0.006[a]	0.846[a]
Diabetes	0.383	0.545	
BPD×diabetes	0.101	0.755	
GLP1 acrophase (time of day)			
BPD	4.846	0.043[a]	0.543[a]
Diabetes	0.380	0.546	
BPD×diabetes	5.926	0.027[a]	0.628[a]
GLP1 mesor			
BPD	0.252	0.623	
Diabetes	21.832	0.000[a]	0.992[a]
BPD×diabetes	0.994	0.334	
Insulin sensitivity (*M*) (μmol kg_{FFM}^{-1} min^{-1})			
BPD	154.672	0.000[a]	1.000[a]
Diabetes	1.189	0.292	
BPD×diabetes	0.055	0.817	

Power analysis was computed using α=0.05

[a] Significant *p* values

As predicted by the model, GLP1 levels remained above the mesor between 16:48 and 19:04 hours in NGT participants before BPD, remaining almost similar after BPD; similar data were observed for GIP levels. Contrarily, in diabetic participants GLP1 levels remained above the mesor between 04:21 and 11:33 hours before BPD, and subsequently they shifted between 09:01 and 14:17 hours. In the same participants, GIP levels were above the mesor from 11:14 to 16:38 hours before BPD and from 08:15 to 14:29 hours after BPD.

Therefore, malabsorptive bariatric surgery had a strong influence on the rhythmicity of both GLP1 and GIP in type 2 diabetic participants, as shown in Fig. 2. After surgery the amplitude of the periodical components of the GIP time series was lower, suggesting a reduction in fluctuation around the mean level. The opposite was true for GLP1.

As shown in Table 3, which summarises the results of the two-way ANOVA, the BPD operation significantly influenced the reduction in the GIP AUC, although the combined action of BPD and diabetes had a larger effect on this change. The effect of BPD on the increase in the GLP1 AUC was significant, contrary to the effect of the factor diabetes alone; both factors, i.e. the combined effect of BPD and diabetes was stronger than that of BPD or diabetes individually. In contrast, the only factor affecting the insulin AUC value (from 6,524±1,175 to 3,112±987.9 pmol l^{-1} min in NGT participants and from 4,804±799 to 2,004±584 pmol l^{-1} min in diabetic patients) was represented by BPD.

The overall *F* value for the multivariate analysis model, whose variables are reported in Tables 4 and 5, was 2.27 (p=0.002). Using η^2 as the measure of effect size, the interaction between BPD and diabetes accounted for 46% of the total variability of the dependent variables (insulin sensitivity, GIP AUC and GLP1 AUC).

Discussion

The principal findings of this study are that: (1) GIP and GLP1 showed a circadian rhythm in both glucose-tolerant and diabetic, morbidly obese participants; (2) the GLP1 AUC significantly increased after BPD in NGT but not in diabetic participants; (3) the GIP AUC significantly diminished in diabetic patients, with a large combined effect of BPD and diabetes; (4) GLP1 levels remained above the mesor earlier during the day in diabetic patients than in NGT participants; (5) the circadian rhythm of incretin was shifted in time after BPD only in diabetic participants; (6) the interaction between BPD and diabetes accounted for 46% of the total variability of insulin sensitivity, GIP AUC and GLP1 AUC; (7) all the above changes happened independently of any significant modification of body weight, since the participants were studied a very short time after the operation.

Ultradian fluctuations in GLP1 secretion have been detected in young healthy men [22], suggesting the

Table 4 Summary of multivariate analyses

Effect	Pillai's trace value	*F* value	*p* value	Partial η^2
Intercept	0.99	595.40	0.000	0.99
BPD	0.91	79.05	0.000	0.91
Diabetes	0.26	1.69	0.216	0.27
BPD×diabetes	0.46	3.93	0.032	0.46

Design: intercept+BPD+DIAB +BPD × DIAB

Table 5 Multivariate analysis for between-participant effects

Source	Dependent variable	Type III sum of squares	Mean square	*F* value	*p* value	Partial η^2
Corrected model	*M* value	47.93[a]	15.98	51.93	0.000	0.907
	GLP1 AUC	12.92[b]	4.30	0.11	0.954	0.020
	GIP AUC	1,425.56[c]	475.19	8.43	0.001	0.612
Intercept	*M* value	407.4	407.40	1,324.08	0.000	0.988
	GLP1 AUC	11,883.68	11,883.68	297.36	0.000	0.949
	GIP AUC	11,719.91	11,719.91	207.81	0.000	0.929
BPD	*M* value	47.59	47.59	154.67	0.000	0.906
	GLP1 AUC	12.66	12.66	0.32	0.581	0.019
	GIP AUC	487.03	487.03	8.64	0.010	0.351
Diabetes	*M* value	0.37	0.37	1.19	0.292	0.069
	GLP1 AUC	0.02	0.02	0.00	0.983	0.000
	GIP AUC	251.76	251.76	4.46	0.051	0.218
BPD × diabetes	*M* value	0.02	0.02	0.06	0.817	0.003
	GLP1 AUC	0.19	0.19	0.00	0.946	0.000
	GIP AUC	706.07	706.07	12.52	0.003	0.439
Error	*M* value	4.92	0.31			
	GLP1 AUC	639.43	39.96			
	GIP AUC	902.35	56.40			
Total	*M* value	465.72				
	GLP1 AUC	12,776.92				
	GIP AUC	14,529.76				
Corrected total	*M* value	52.85				
	GLP1 AUC	652.35				
	GIP AUC	2,327.91				

[a] $R^2=0.907$ (adjusted $R^2=0.889$); [b] $R^2=0.020$ (adjusted $R^2=-0.164$); [c] $R^2=0.612$ (adjusted $R^2=0.540$)

possibility that these fluctuations might regulate insulin oscillations in secretion rate. In the present study we have demonstrated for the first time the presence of incretin circadian rhythms also in morbidly obese participants with either normal glucose tolerance or type 2 diabetes. After the operation, GLP1 secretion, which was basally higher in NGT participants than in diabetic patients, increased significantly only in the NGT group. In contrast, GIP secretion was significantly reduced after BPD in patients with type 2 diabetes but not in NGT participants. The BPD operation also changed the circadian rhythms of incretins by shifting them in time to earlier during the day.

In our series, 46% of the variation in insulin sensitivity, GIP and GLP1 was explained by the interaction between the BPD operation and the presence of diabetes.

Insulin-resistant diabetes, either in experimental animal models or in humans [23, 24], is associated with high circulating GIP levels secondary to an exaggerated K cell secretory response to nutrients. The findings of Flatt and colleagues [23, 25, 26] in rodents provide an interesting parallelism with our results in humans. Genetic knockout of the GIP receptor and blockade of GIP action were both associated with a large improvement in insulin resistance and diabetes [25, 26].

BPD bypasses a large area of the small intestine, including the duodenum, jejunum and the major part of the ileum, and allows nutrients to enter directly the last part of the ileum. Although a diverse anatomical modification of the small intestine takes place in the Roux-en-Y gastric bypass (RYGB), in which the stomach is anastomosed with the jejunum a few centimetres after Treitz's ligament, the effects of BPD and RYGB in terms of GIP suppression in diabetic patients seem to be comparable. After gastric bypass, Rubino et al. [10] found that GIP decreased to normal levels in diabetic patients, whereas it increased slightly but not significantly in obese non-diabetic patients. This similarity might depend on the distribution of GIP-secreting K cells, which are present in both the duodenum and the jejunum [27].

The role of GIP in inducing glucose intolerance and insulin resistance is supported by the evidence [28] that GIP receptor antagonism by (Pro(3))GIP[mPEG], a mPEGylated

antagonist of gastric inhibitory polypeptide, improves glucose tolerance and insulin secretory responses in both dietary and genetic diabesity.

In contrast, the effect of RYGB on GLP1 is quite different from that of BPD. After RYGB, an increase in the GLP1 level after OGTT or a meal was observed in people with NGT [14] and in diabetic patients [12, 13], and was associated with increased circulating levels of insulin.

While the diverse behaviours of the two types of bariatric surgery in terms of incretin secretion might be attributable to the bypass of different parts of the small intestine, the different effect of the same BPD operation in NGT and in type 2 diabetic participants requires further investigation.

Recently, an 'anti-incretin' theory has been postulated by Rubino [29], i.e. the existence of a counter-regulatory mechanism with opposite actions to those of incretins. According to this idea, this intestinal factor, which sustains GIP secretion, might be suppressed after BPD as a consequence of nutrient diversion.

In conclusion, the effect of BPD on the 24 h pattern of incretins differed markedly between NGT participants and patients with type 2 diabetes. Impairment of GLP1 secretion was reversed in NGT participants but could not be overcome by BPD in patients with diabetes. Contrarily, GIP secretion was blunted after the operation only in diabetic patients, suggesting a role of this incretin in insulin resistance and diabetes.

Duality of interest The authors declare that there is no duality of interest associated with this manuscript.

References

1. Vilsbøll T, Holst JJ (2004) Incretins, insulin secretion and type 2 diabetes mellitus. Diabetologia 47:357–366
2. Nauck M, Heimesaat MM, Orskov C, Holst JJ, Ebert R, Creutzfeldt W (1993) 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
3. Nauck M, Stöckmann F, Ebert R, Creutzfeldt W (1986) Reduced incretin effect in type 2 (non-insulin-dependent) diabetes. Diabetologia 29:46–52
4. Zimmet P, Alberti KG, Shaw J (2001) Global and societal implications of the diabetes epidemic. Nature 414:782–787
5. Leibson CL, Williamson DF, Melton LJ 3rd et al (2001) Temporal trends in BMI among adults with diabetes. Diabetes Care 24:1584–1589
6. Golay A, Ybarra J (2005) Link between obesity and type 2 diabetes. Best Pract Res Clin Endocrinol Metab 19:649–663
7. Muscelli E, Mari A, Casolaro A et al (2008) Separate impact of obesity and glucose tolerance on the incretin effect in normal subjects and type 2 diabetic patients. Diabetes 57:1340–1348
8. Anderson JW, Grant L, Gotthelf L, Stifler LT (2006) Weight loss and long-term follow-up of severely obese individuals treated with an intense behavioral program. Int J Obes 31:488–493
9. MacDonald KG, Long SD, Swanson MS et al (1997) The gastric bypass operation reduces the progression and mortality of NIDDM. J Gastrointest Surg 1:213–220
10. Rubino F, Gagner M, Gentileschi P et al (2004) 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
11. Guidone C, Manco M, Valera-Mora E et al (2006) Mechanisms of recovery from type 2 diabetes after malabsorptive bariatric surgery. Diabetes 55:2025–2031
12. Laferrère B, Teixeira J, McGinty J et al (2008) 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
13. Laferrère B, Heshka S, Wang K et al (2007) 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
14. Rodieux F, Giusti V, D'Alessio DA, Suter M, Tappy L (2008) Effects of gastric bypass and gastric banding on glucose kinetics and gut hormone release. Obesity 16:298–305
15. Pories WJ, Albrecht RJ (2001) Etiology of type II diabetes mellitus: role of the foregut. World J Surg 25:527–531
16. Mingrone G, DeGaetano A, Greco AV et al (1997) Reversibility of insulin resistance in obese diabetic patients: role of plasma lipids. Diabetologia 40:599–605
17. Camastra S, Manco M, Mari A et al (2007) Beta-cell function in severely obese type 2 diabetic patients: long-term effects of bariatric surgery. Diabetes Care 30:1002–1004
18. Mari A, Manco M, Guidone C et al (2006) Restoration of normal glucose tolerance in severely obese patients after bilio-pancreatic diversion: role of insulin sensitivity and beta cell function. Diabetologia 49:2136–2143
19. DeFronzo RA, Tobin JD, Andres R (1979) Glucose clamp technique: a method for quantifying insulin secretion and resistance. Am J Physiol 237:E214–E223
20. Krarup T, Madsbad S, Moody AJ et al (1983) Diminished gastric inhibitory polypeptide (GIP) response to a meal in newly diagnosed type I (insulin dependent) diabetics. J Clin Endocrinol Metab 56:1306–1312
21. Ørskov C, Rabenhøj L, Kofod H, Wettergren A, Holst JJ (1994) Production and secretion of amidated and glycine-extended glucagon-like peptide-1 (GLP-1) in man. Diabetes 43:535–539
22. Simon C, Brandenberger G (2002) Ultradian oscillations of insulin secretion in humans. Diabetes 51:S258–S261
23. Flatt PR, Bailey CJ, Kwasowski P, Page T, Marks V (1984) Plasma immunoreactive gastric inhibitory polypeptide in obese hyperglycaemic (ob/ob) mice. J Endocrinol 101:249–256
24. Vilsbøll T, Krarup T, Sonne J et al (2003) Incretin secretion in relation to meal size and body weight in healthy subjects and people with type 1 and type 2 diabetes mellitus. J Clin Endocrinol Metab 88:2706–2713
25. McClean PL, Irwin N, Cassidy RS, Holst JJ, Gault VA, Flatt PR (2007) GIP receptor antagonism reverses obesity, insulin resistance, and associated metabolic disturbances induced in mice by prolonged consumption of high-fat diet. Am J Physiol Endocrinol Metab 293:E1746–E1755
26. Gault VA, Irwin N, Green BD et al (2005) Chemical ablation of gastric inhibitory polypeptide receptor action by daily (Pro3)GIP administration improves glucose tolerance and ameliorates insulin

resistance and abnormalities of islet structure in obesity-related diabetes. Diabetes 54:2436–2446
27. Damholt AB, Kofod H, Buchan AM (1999) Immunocytochemical evidence for a paracrine interaction between GIP and GLP-1-producing cells in canine small intestine. Cell Tissue Res 298:287–293
28. McClean PL, Irwin N, Hunter K, Gault VA, Flatt PR (2008) (Pro (3))GIP[mPEG]: novel, long-acting, mPEGylated antagonist of gastric inhibitory polypeptide for obesity-diabetes (diabesity) therapy. Br J Pharmacol 155:690–701
29. Rubino F (2008) Is type 2 diabetes an operable intestinal disease? A provocative yet reasonable hypothesis. Diabetes Care 31:S290–S296