

# High-energy breakfast with low-energy dinner decreases overall daily hyperglycaemia in type 2 diabetic patients: a randomised clinical trial

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## Abstract

**Aims/hypothesis** High-energy breakfast and reduced-energy dinner (Bdiet) significantly reduces postprandial glycaemia in obese non-diabetic individuals. Our objective was to test whether this meal schedule reduces postprandial hyperglycaemia (PPHG) in patients with type 2 diabetes by enhancing incretin and insulin levels when compared with high-energy dinner and reduced-energy breakfast (Ddiet).

**Methods** In a randomised, open label, crossover design performed in a clinic setting, 18 individuals (aged 30–70 years with BMI 22–35 kg/m<sup>2</sup>) with type 2 diabetes (<10 years duration) treated with metformin and/or diet were given either Bdiet or Ddiet for 7 days. Participants were randomised by a person not involved in the study using a coin flip. Postprandial levels of plasma glucose, insulin, C-peptide and intact and total glucagon-like peptide-1 (iGLP-1 and tGLP-1) were assessed. The Bdiet included 2,946 kJ breakfast, 2,523 kJ lunch and 858 kJ dinner. The Ddiet comprised 858 kJ breakfast, 2,523 kJ lunch and 2,946 kJ dinner.

**Results** Twenty-two individuals were randomised and 18 analysed. The AUC for glucose (AUC<sub>glucose</sub>) throughout the day was 20% lower, whereas AUC<sub>insulin</sub>, AUC<sub>C-peptide</sub> and

AUC<sub>tGLP-1</sub> were 20% higher for the Bdiet than the Ddiet. Glucose AUC<sub>0–180min</sub> and its peak were both lower by 24%, whereas insulin AUC<sub>0–180min</sub> was 11% higher after the Bdiet than the Ddiet. This was accompanied by 30% higher tGLP-1 and 16% higher iGLP-1 levels. Despite the diets being isoenergetic, lunch resulted in lower glucose (by 21–25%) and higher insulin (by 23%) with the Bdiet vs Ddiet.

**Conclusions/interpretation** High energy intake at breakfast is associated with significant reduction in overall PPHG in diabetic patients over the entire day. This dietary adjustment may have a therapeutic advantage for the achievement of optimal metabolic control and may have the potential for being preventive for cardiovascular and other complications of type 2 diabetes.

**Trial registration** ClinicalTrials.gov NCT01977833

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**Keywords** Breakfast · Clock · Diabetes · Dinner · GLP-1 · Insulin · Timing

## Abbreviations

Bdiet	High-energy breakfast and reduced-energy dinner
Ddiet	High-energy dinner and reduced-energy breakfast
iGLP-1	Intact glucagon-like peptide-1
PPHG	Postprandial hyperglycaemia
tGLP-1	Total glucagon-like peptide-1

## Introduction

Postprandial hyperglycaemia (PPHG) contributes 31–70% to HbA<sub>1c</sub> values [1] and is strongly associated with increased cardiovascular risk in type 2 diabetes [2, 3]. Therefore, postprandial glycaemia is an important treatment target in type 2

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diabetes. Post-meal glycaemia displays a clear circadian pattern with a more prolonged and higher response to an identical meal in the evening vs the morning [4–8]. This is achieved in part by the circadian secretion and activity of enzymes and hormones involved in the regulation of postprandial glycaemia [9–13]. Indeed, the circadian clock controls glucagon-like peptide 1 (GLP-1) secretion in intestinal L cells [9], insulin secretion in beta cells [14, 15], hepatic insulin extraction [7] and insulin-dependent glucose transporter GLUT 4 expression in skeletal muscle [10, 13].

While the master clock is entrained to the light–dark cycles [16], meal timing and feeding schedules exert strong entraining effects on peripheral oscillators maintaining the internal synchrony of most metabolic processes [17, 18]. Studies in animals and humans have shown that altered meal timing, such as skipping breakfast or high energy intake at dinner, are associated with disrupted clock gene expression, increased lipogenesis, higher HbA<sub>1c</sub> and poor glycaemic control [4, 19–22]. In contrast, high-energy breakfast with no dinner or time-restricted feeding reversed the impaired clock gene expression, resulting in decreased plasma glucose and triacylglycerol levels and reduced body weight in animal models of obesity and type 2 diabetes [12, 22, 23]. The different effects of meals during the day may therefore be modulated by different energy loads to achieve optimal glucose homeostasis. However, the effect of different meal timing schedules on overall incretin, insulin and glycaemic excursion during the entire day has not been previously explored in individuals with type 2 diabetes. In this study, we assessed the differential influence of two isoenergetic diets with different meal timing schedules on PPHG over the entire day and the corresponding changes in insulin and incretin hormones in patients with type 2 diabetes. The two dietary schedules were as follows: one with high-energy breakfast and lower-energy dinner (Bdiet) and the other with high-energy dinner and lower-energy breakfast (Ddiet).

## Methods

**Participants** The study population initially included 22 individuals who had type 2 diabetes for less than 10 years and an HbA<sub>1c</sub> level of 7–9% (53–75 mmol/mol) on recruitment (Table 1). Individuals aged 30–70 years with BMI 22–35 kg/m<sup>2</sup> were included. None of the participants had impaired thyroid, renal or liver function, pulmonary disease, psychiatric, immunological or neoplastic diseases or severe diabetic complications, such as cardiovascular disease, cerebrovascular disease, proliferative diabetic retinopathy or gastroparesis and none had undergone bariatric surgery. All participants were insulin-naïve and patients taking oral hypoglycaemic agents other than metformin were excluded. Those on GLP-1 analogues or anorectic drugs or those on steroid treatment were not allowed to participate. The

**Table 1** Clinical and anthropometric characteristics

Characteristic	
No. of patients	18
Sex (no. male/no. female)	8/10
Age (years)	57.8±4.7
Weight (kg)	76.8±13.5
BMI (kg/m <sup>2</sup> )	28.1±2.9
Waist circumference (cm)	94±5.8
Systolic BP (mmHg)	130.7±7.2
Diastolic BP (mmHg)	80.6±7.46
HbA <sub>1c</sub> (%)	7.6±0.4
HbA <sub>1c</sub> (mmol/mol)	59±2
Time since diagnosis of diabetes (years)	9.3±5
Fasting blood glucose (mmol/l)	7.2±0.5
Fasting plasma insulin (pmol/l)	49.3±9

Data are shown as *n* or as mean ± SE

Helsinki Committee of the Wolfson Medical Center in Holon, Israel approved the study. All the participants gave their informed consent. The study was registered at ClinicalTrials.gov (NCT01977833).

**Study design** This was a randomised, open-label, crossover-within-subject clinical trial. Participants were randomised by a person not involved in the study using a coin flip. We performed two separate testing days each over the course of 14 h. Test meals were provided in the clinic as breakfast, lunch and dinner with energy and composition according to the assigned diet schedule (Table 2). The participants consumed their meals within 15 min, with breakfast at 08:00 hours, lunch at 13:00 hours and dinner at 19:00 hours. The diet during both testing days provided a total daily energy intake of 6,276±105 kJ (23% fat, 46% carbohydrates, 31% protein) with identical macronutrient content and composition, but different meal timing distribution. The two meal timing schedules were either breakfast diet (Bdiet) or dinner diet (Ddiet). The Bdiet consisted of a large breakfast (2,946 kJ; 22% fat, 47% carbohydrates, 31% protein), a medium-sized lunch (2,523 kJ; 23% fat, 50% carbohydrates, 27% protein) and a small dinner (858 kJ; 30% fat, 27% carbohydrates, 43% protein). This was reversed in the Ddiet (858 kJ breakfast, 2,523 kJ lunch and 2,946 kJ dinner). Participants followed the Bdiet and Ddiet meal plans for 6 days at home prior to the intervention day to prevent the pre-study meal pattern from influencing the results. Participants were also asked to avoid alcohol and excessive physical activity 6 days before each test day. Participants ingested their last oral therapy 24 h before the test day. Following a washout period of 2 weeks, the same procedure was repeated on the second test day, with each participant now crossed over to the opposite diet schedule.

**Table 2** Diet composition on the meal test days

Meal type	Energy content (kJ)	Composition			
		Fat (g)	Carbohydrate (g)	Protein (g)	Fibre (g)
<b>Large meal</b>					
Whole-wheat bread: two slices	569	2.4	25.8	5.4	3.9
Caffè latte with non-fat milk: one tall	418	0.0	15.0	10.0	0.0
Tuna, light in water: 115 g	502	1.0	1.0	26.0	0.4
Scrambled egg: one egg	159	1.5	0.5	5.5	0.0
Olive oil: two teaspoons	335	9.0	0.0	0.0	0.0
Yogurt and cereal: one container	460	2.0	20.0	5.0	3.0
Granola bar, dark chocolate: one bar	502	2.0	24.0	5.0	4.0
Total	2,946	17.9	86.3	56.9	11.3
Percentage of energy		22	47	31	-
<b>Lunch</b>					
Apple: one apple	339	0.5	21.0	0.3	3.7
Roasted chicken breast: 115 g	782	4.0	0.0	35.0	0.0
Diet drink: 236 ml	0	0.0	0.0	0.0	0.0
Salad, tossed greens: one salad	42	0.0	2.0	0.0	0.5
Vegetable soup: one cup	418	2.0	20.0	4.0	4.0
Olive oil: two teaspoons	335	9.0	0.0	0.0	0.0
Baked potato: one potato	607	0.2	33.6	3.1	2.3
Total	2,523	15.7	76.6	42.4	10.5
Percentage of energy		23	50	27	-
<b>Small meal</b>					
Caffè Americano: one large	63	0.0	3.0	1.0	0.0
Salad, fresh tomato and mozzarella: one order	469	7.0	6.0	6.2	1.0
Salad, tossed greens: one salad	42	0.0	2.0	0.0	0.5
Turkey breast: three slices	251	0.0	1.0	14.0	-
Salad, mixed greens: one cup	33	0.0	2.0	1.0	1.0
Total	858	7.0	14.0	22.2	2.5
Percentage of energy		30	27	43	-

**Meal challenges** On the day of the meal challenge, each participant reported to the laboratory at 07:00 hours after an overnight fast. Participants were asked not to take glucose-lowering medication during the test days and were instructed to keep dietary logs. A dietitian reviewed these logs on the day of the meal tests and only those who had 80% adherence to the diet participated in the meal tests. Anthropometric data were collected in the morning and each group consumed their assigned meal plan: breakfast at 08:00 hours, lunch at 13:00 hours and dinner at 19:00 hours. At 07:30 hours, a catheter was placed in the antecubital vein of the non-dominant arm and remained in place until 22:00 hours. Venous blood samples were collected just before breakfast ( $t=0$  min) and at 15, 30, 60, 90, 120, 150 and 180 min after eating commenced. Blood sampling was repeated at the same time points after lunch and dinner. The primary outcome was to assess the overall postprandial GLP-1 levels in both meal

schedules. The secondary outcome was to assess the overall postprandial glycaemia. Other outcomes were to assess the overall postprandial response of plasma insulin and C-peptide in both meal schedules.

**Biochemical and hormonal blood analyses** Plasma glucose was immediately analysed on an Olympus AU 2700 analyser (Beckman Coulter, Brea, CA, USA). Serum and plasma EDTA tubes for insulin and C-peptide were left on ice to rest for approximately 30 min. Blood samples for determining iGLP-1 and tGLP-1 were collected into chilled tubes containing EDTA, aprotinin and diprotin A (0.1 mmol/l). Samples were centrifuged immediately at 2,000g at 4°C for 10 min and stored at -80°C. Insulin and C-peptide were determined by electrochemiluminescence using a Cobas 601 Roche Diagnostic analyser (Madison, WI, USA) according to the manufacturer's instructions.

Plasma tGLP-1 and iGLP-1 were quantified using ELISA (Millipore, Billerica, MA, USA).

**Sample size and power analysis** Power analysis revealed that a sample size of 18 participants (18 in each treatment group; Bdiet and Ddiet) is required to provide 80% power to detect 5% difference between groups in overall postprandial plasma AUC for iGLP-1, insulin and glucose assessed after three meals in each diet intervention. To allow discontinuation during the course of the study, 22 participants were recruited.

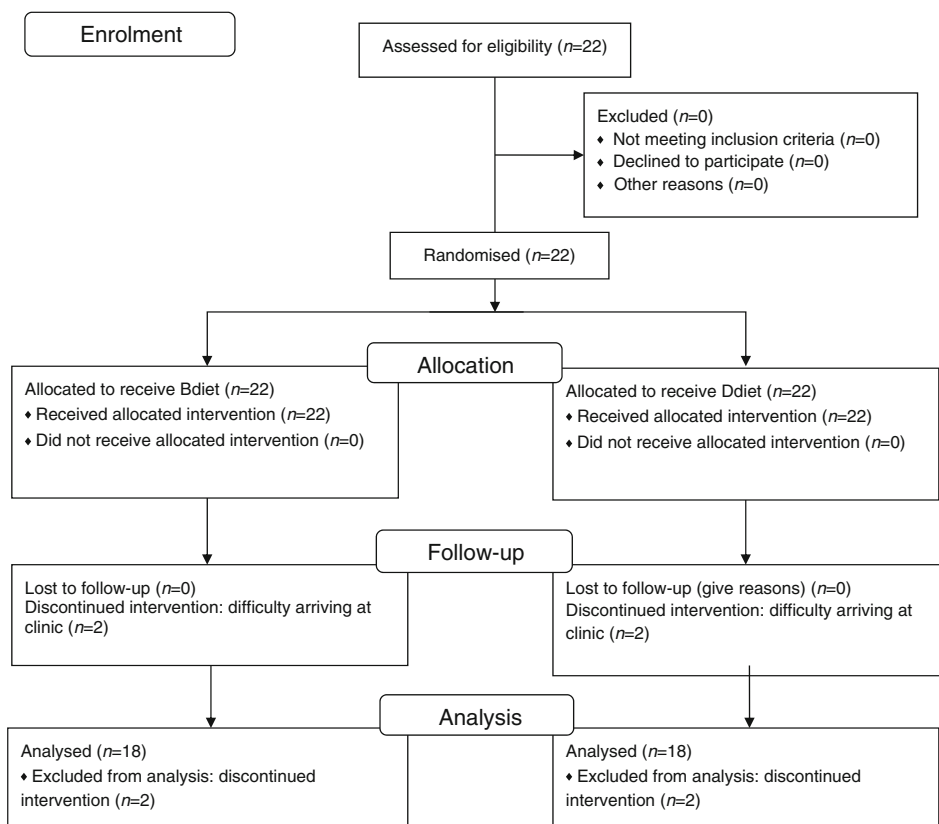
**Statistical analyses** The results are expressed as mean±SEM. For time series, a two-way ANOVA (time×diet) was performed and a least-significant difference *t* test post hoc analysis was used for comparison between the diets at each time point. Means comparisons were made using the CI of the estimators. AUCs were calculated using the trapezoidal rule. A Student's *t* test for paired data was used for comparing the AUC at different time intervals. In addition, a multivariate ANOVA for repeated measurements was performed to assess between- and within-subject effects for diet and time. A *p* value ≤0.05 was considered statistically significant. Statistical analysis was performed using SPSS (version 18) software ([www14.software.ibm.com/download/data/web/en\\_US/trialprograms/W110742E06714B29.html](http://www14.software.ibm.com/download/data/web/en_US/trialprograms/W110742E06714B29.html)).

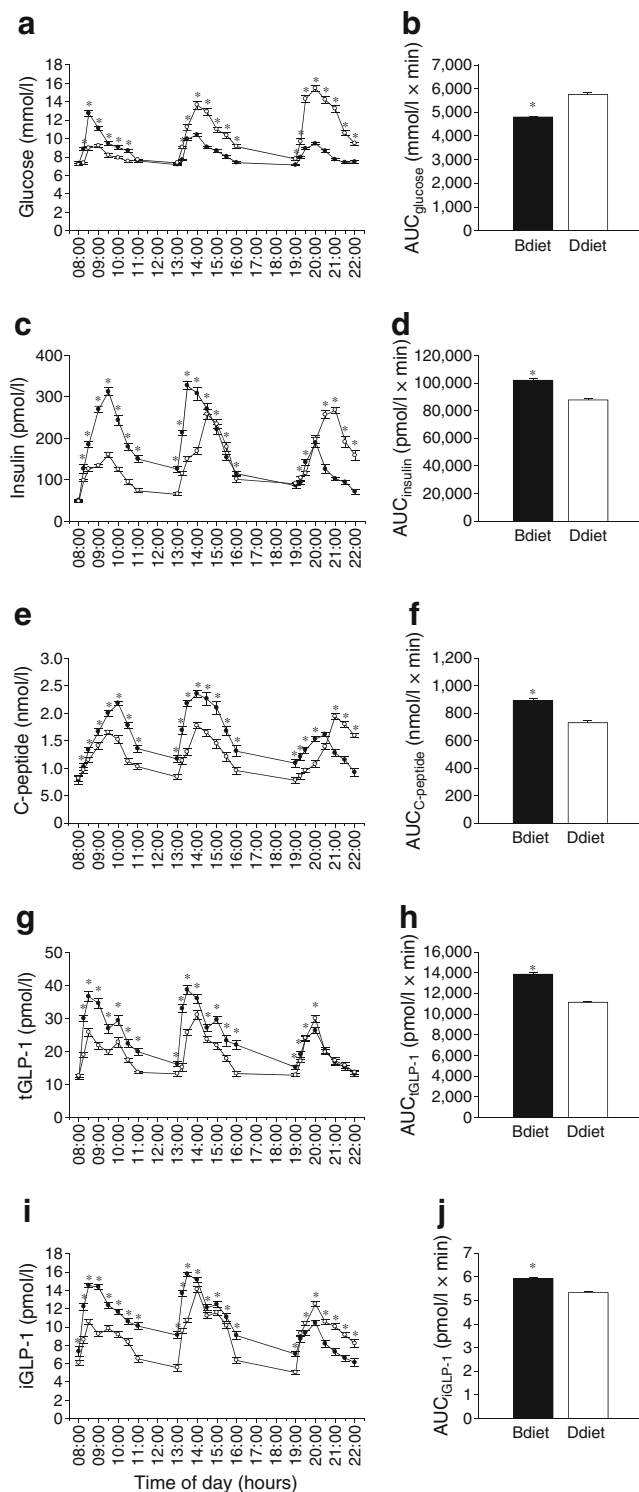
## Results

**Participants** Twenty-two individuals from the Wolfson Diabetes Unit outpatient clinic were enrolled into the study (Fig. 1). The study period from recruitment and including follow-up was December 2013 to April 2014. After completing the first all-meals testing day, four participants dropped out (two from the Bdiet group and two from the Ddiet group), because of difficulties getting to the clinic. Eighteen individuals (eight men, ten women) completed the study. These patients had an average age of 57.8±4.7 years, controlled type 2 diabetes of 9.3±5.0 years duration, HbA<sub>1c</sub> values of 7.6±0.4% (59±2 mmol/mol) and BMI of 28.1±2.9 kg/m<sup>2</sup> (Table 1). Eight patients were treated with diet alone, whereas ten were treated with diet and metformin. Five patients had a history of hypertension and were treated with thiazides, angiotensin-converting enzyme inhibitors and/or calcium antagonists.

**Plasma glucose and hormonal profiles of Bdiet vs Ddiet group** Fasting plasma glucose, insulin, C-peptide, tGLP-1 and iGLP-1 did not differ significantly between the two test days or the groups (Fig. 2). Integrated AUC<sub>glucose</sub> after breakfast, lunch and dinner was 20% lower in the Bdiet group than in the Ddiet (*p*<0.001, *t* test) group (Fig. 2). Integrated

**Fig. 1** Flow diagram of recruitment





**Fig. 2** All-day graphs for glucose, insulin, C-peptide, tGLP-1 and iGLP-1. White circles, Ddiet group; black circles, Bdiet group. \* $p < 0.05$

$AUC_{\text{insulin}}$ ,  $AUC_{\text{C-peptide}}$  and  $AUC_{\text{tGLP-1}}$  after breakfast, lunch and dinner were higher by 20% during the Bdiet than during the Ddiet ( $p < 0.001$ ,  $t$  test). Integrated  $AUC_{\text{iGLP-1}}$  after breakfast, lunch and dinner was higher by 10% during the Bdiet than during the Ddiet ( $p < 0.001$ ,  $t$  test).

**Plasma glucose and hormonal profiles after high-energy breakfast vs high-energy dinner** After high-energy breakfast, glucose  $AUC_{0-180\text{min}}$  and its peak were reduced by 24% compared with the glucose response after high-energy dinner ( $p < 0.001$ ,  $t$  test) (Table 3, Fig. 2). During the early interval,  $AUC_{0-30\text{min}}$ , plasma glucose was lower by 10% after high-energy breakfast vs high-energy dinner ( $p < 0.006$ ,  $t$  test). After reaching their peak, plasma glucose levels decreased rapidly after the high-energy breakfast whereas after the high-energy dinner, plasma glucose persisted and remained significantly higher.

The insulin response to the meal challenge was more rapid and higher after high-energy breakfast compared with high-energy dinner (Table 3, Fig. 2). Insulin  $AUC_{0-180\text{min}}$  was 11% higher after high-energy breakfast than after high-energy dinner ( $p < 0.001$ ,  $t$  test) (Table 3). During the first 30 min, plasma insulin levels increased more rapidly after high-energy breakfast than after high-energy dinner ( $p < 0.008$ ,  $t$  test). During the late interval,  $AUC_{60-180\text{min}}$ , insulin secretion did not differ ( $p < 0.11$ ) between the meals. After high-energy breakfast, peak insulin secretion occurred at 60 min, 12% higher ( $p < 0.002$ ,  $t$  test) than after high-energy dinner. After high-energy dinner, peak insulin secretion occurred at 120 min. After meal ingestion, patterns of C-peptide levels mirrored those of insulin with a more rapid and higher increase after high-energy breakfast. High-energy breakfast resulted in a higher C-peptide early response and  $AUC_{0-180\text{min}}$ , by 17% ( $p < 0.001$ ,  $t$  test) compared with high-energy dinner (Table 3).

The  $AUC_{0-180\text{min}}$  for tGLP-1 was 29% higher after high-energy breakfast than after high-energy dinner ( $p < 0.001$ ,  $t$  test) (Table 3, Fig. 2). Plasma tGLP-1 increased rapidly after high-energy breakfast and peaked at 30 min. In contrast, after high-energy dinner, the increase in plasma tGLP-1 was more gradual and peaked at 60 min. Compared with high-energy dinner, the high-energy breakfast resulted in 21% higher tGLP-1 levels ( $p < 0.001$ , post hoc  $t$  test). The high-energy breakfast elicited 30–35% higher early and late interval for tGLP-1 ( $p < 0.001$ ,  $t$  test). Similarly, iGLP-1 plasma levels peaked at 30 min after high-energy breakfast compared with a peak at 60 min after high-energy dinner. The peak level of iGLP-1 was higher by 16% after high-energy breakfast than after high-energy dinner ( $p < 0.001$ , post hoc  $t$  test). The  $AUC_{0-180\text{min}}$ , as well as the early and late intervals for tGLP-1 and iGLP-1, were 14–35% higher after high-energy breakfast than after high-energy dinner ( $p < 0.001$ ,  $t$  test).

**Plasma glucose and hormonal profiles after Bdiet lunch vs Ddiet lunch** Glucose  $AUC_{0-180\text{min}}$  and its peak were lower by 21–25% after Bdiet lunch vs Ddiet lunch ( $p < 0.001$ ,  $t$  test) (Table 3, Fig. 2). The glucose early and late interval was lower by 13% and 24%, respectively, after Bdiet lunch than after Ddiet lunch ( $p < 0.006$ ,  $t$  test). Insulin  $AUC_{0-180\text{min}}$  was higher by 23% after Bdiet lunch than after Ddiet lunch. In addition,

**Table 3** AUC at different time intervals of breakfast vs dinner and HE lunch of Bdiet vs Ddiet

AUC	Bdiet HE breakfast	Ddiet HE dinner	<i>p</i> value	% change	Bdiet lunch	Ddiet lunch	<i>p</i> value	% change
Glucose (mmol/l×min)								
0–30 min	283±16	311±27	<0.006	–10	243±6	275±16	<0.001	–13
60–180 min	1,094±56	1,513±125	<0.001	–27	1,038±55	1,368±124	<0.001	–24
0–180 min	1,735±76	2,270±167	<0.001	–24	1,586±9	2,015±67	<0.001	–21
Insulin (pmol/l×min)								
0–30 min	3,653±757	3,104±326	<0.008	19	6,587±590	3,327±382	<0.001	49
60–180 min	28,308±2,952	26,780±3,702	0.111	8	25,683±3,841	24,155±3,118	0.140	8
0–180 min	38,753±3,695	34,572±4,257	<0.001	11	41,781±4,424	32,260±3,792	<0.001	23
C-peptide (nmol/l×min)								
0–30 min	31±7	26±5	0.042	17	50±7	33±5	<0.001	35
60–180 min	224±19	194±15	<0.001	16	236±36	170±25	<0.001	28
0–180 min	300±29	250±20	<0.001	17	354±43	249±23	<0.001	30
tGLP-1 (pmol/l×min)								
0–30 min	817±104	533±51	<0.001	35	906.1±89	519.7±115	<0.001	43
60–180 min	3,178±388	2,238±241	<0.001	30	3,271±453	2,566±352	<0.001	22
0–180 min	5,063±489	3,581±256	<0.001	29	5,298±523	3,938±490	<0.001	26
iGLP-1 (pmol/l×min)								
0–30 min	347±36	255±27	<0.001	27	391.3±26	265.5±22	<0.001	32
60–180 min	1,403±124	1,202±133	<0.001	14	1,431±161	1,294±104	<0.002	11
0–180 min	2,183±154	1,799±161	<0.001	18	2,286±64	1,931±113	<0.001	16

the early prandial insulin AUC<sub>0–30min</sub> was higher by almost 50% after Bdiet lunch vs Ddiet lunch ( $p<0.001$ ,  $t$  test). Insulin reached peak levels 30 min after the Bdiet lunch and 90 min after the Ddiet lunch, with the Bdiet lunch producing higher peak levels ( $p<0.001$ ,  $t$  test) (Table 3, Fig. 2). Throughout the 180 min of analysis, C-peptide levels were 30% higher after the Bdiet lunch than after the Ddiet lunch ( $p<0.001$ ,  $t$  test) and tGLP-1 and iGLP-1 levels were 16–26% higher after the Bdiet lunch than after the Ddiet lunch ( $p<0.001$ ,  $t$  test). In addition, tGLP-1 and iGLP-1 peaked 30 min earlier after Bdiet lunch compared with Ddiet lunch.

## Discussion

In this study, we demonstrate in individuals with type 2 diabetes that a meal schedule of high-energy breakfast and low-energy dinner (Bdiet) leads to overall increased GLP-1 and insulin levels and to reduced hyperglycaemia throughout the day compared with a reverse meal schedule (Ddiet). Although both diets had an identical day-long energy composition, the difference in timing led to a significant reduction in overall glucose excursions in the Bdiet compared with the Ddiet. In parallel, the integrated AUCs for insulin, C-peptide, iGLP-1 and tGLP-1 secretion were significantly enhanced along the entire day in the Bdiet compared with the Ddiet schedule. These observations suggest that a change in meal timing

influences the overall daily rhythm of postprandial insulin, incretin and glycaemic excursions. Our findings showing a significantly reduced overall glycaemia with the Bdiet schedule are consistent with studies in animal models. Impaired peripheral clock gene expression as a result of breakfast skipping or reduced food intake in the first meal of the day along with high-energy dinner, has been associated with increased lipogenesis and higher overall daily glucose excursions, despite no differences in daily total or fat-derived energy [12, 22]. Alternatively, a specific change in the meal timing (i.e. increasing breakfast and reducing dinner energy content) may also restore clock gene expression in obese and diabetic animals, resulting in reduced plasma glucose and triacylglycerol levels and body weight [12, 22]. Recently, it was reported that high-energy breakfast with reduced dinner improved insulin sensitivity and decreased body weight, glucose excursions and HbA<sub>1c</sub> among obese and diabetic patients [24–27]. The omission of breakfast was also associated with increased risk of type 2 diabetes, poor glycaemic control, higher HbA<sub>1c</sub>, increased lipogenesis, visceral adiposity and high blood pressure and increased cardiovascular risk despite the same daily energy intake in individuals with type 2 diabetes [20, 21]. Although this study investigated the acute effect of meals, long-term interventions should determine whether adhering to this diet would successfully lower HbA<sub>1c</sub> levels. This issue was addressed in our previous study, in which eating a large breakfast each day for 3 months led to a 5% reduction in

HbA<sub>1c</sub> levels [27]. Thus, meal timing schedule may be a crucial factor in the improvement of glucose balance and prevention of complications in type 2 diabetes and lends further support to the role of the circadian system in metabolic regulation.

The postprandial glucose response after the high-energy breakfast was significantly lower than after the low-energy breakfast, and decreased rapidly after the peak at 30 min. In contrast, the plasma glucose after high-energy dinner peaked higher than after the low-energy dinner, and remained elevated until the end of the postprandial response. Decline in carbohydrate tolerance toward the evening, with more prolonged and higher postprandial glycaemic response to identical meals in the evening than in the morning has been shown in healthy individuals and those with type 2 diabetes [4–8]. Notably, the 2 h post-meal glycaemia was significantly lower after the high-energy breakfast than after the high-energy dinner, and was below the upper limit (10 mmol/l) of 2 h post-meal glycaemia recommended in the recent ADA guidelines for the prevention of cardiovascular events in patients with type 2 diabetes. In parallel, significantly higher insulin and C-peptide responses were observed after the high-energy breakfast than after the high-energy dinner. This greater and more rapid early prandial insulin secretion after the high-energy breakfast has further importance since in type 2 diabetes the deficiency of early prandial insulin is the major contributor to postprandial hyperglycaemia and cardiovascular risk [2, 3]. The mechanism of better glucose tolerance after high-energy breakfast than after an identical dinner may be in part the result of clock regulation that triggers higher beta cell responsiveness in the morning, lower hepatic insulin extraction after breakfast than after dinner [7] and the increase in insulin-mediated muscle glucose uptake in the morning [7, 10]. Thus, the assignment of major energy load at breakfast, when beta cell responsiveness and insulin-mediated muscle glucose uptake are at optimal levels, seems an adequate strategy to decrease PPHG in patients with type 2 diabetes. The contribution to our results made by different clearance of insulin cannot be excluded, although ratios of insulin to C-peptide (a rough estimate of hepatic extraction) did not differ between the diets.

The early prandial increase in GLP-1 during the first 30 min after high-energy breakfast coincided with the early prandial insulin response. These results are supported by previous findings in which AUC<sub>30</sub><sub>iGLP-1</sub> correlated significantly with AUC<sub>30</sub><sub>insulin</sub> [6]. This suggests that the more rapid and higher early insulin response after the high-energy breakfast may be a result of a more rapid early prandial GLP-1 level, which mediates the potentiation of beta cell function. Recently, the circadian rhythm of GLP-1 secretory responses was shown in animals, with increased release after the first meal at the beginning of the normal feeding period and this profile was correlated with insulin secretion [9]. Moreover,

when this first meal was omitted, both GLP-1 and insulin rhythms were completely inverted along with disruption of clock gene expression. Collectively, these results indicate that the peripheral clock in intestinal L cells, which drives the circadian expression of GLP-1, can be modulated by different meal timing.

Glucose excursions were reduced after lunch preceded by high-energy breakfast (Bdiet) compared with after an identical lunch preceded by low-energy breakfast (Ddiet). This reduced glucose response was associated with rapid and significantly enhanced insulin, tGLP-1 and iGLP-1 responses. This second-meal phenomenon suggests that by changing meal timing schedule, it is possible to enhance the incretin and insulinotropic effects, achieving significant reduction in PPHG through the day in patients with type 2 diabetes. Several theories have been suggested to explain the second-meal phenomenon in type 2 diabetes, including differences in the amount and the pattern of insulin secretion, augmentation of beta cell function and increased incretin response [26, 28]. The high GLP-1 response to lunch after high-energy breakfast suggests that incretin hormones may play an important role in the second-meal phenomenon. Indeed, this phenomenon was absent after a repeated intravenous glucose tolerance test [29], but not after a meal tolerance test [26, 30, 31].

One limitation of our study is that the macronutrient composition varied according to the time of day. Thus, some of the results may also be due to the timing of macronutrient ingestion. Another limitation is that a group of healthy individuals was not included. Although we show the effect of two different meal timing schedules on the overall hyperglycaemia and on the potentiation of insulin and incretin secretion throughout the day and the effect of second-meal phenomenon in diabetic individuals, we cannot determine whether these effects also occur in healthy people. In addition, insulin sensitivity, suppression of endogenous glucose production and gastric emptying were not examined.

In conclusion, the results of this study support a role for diurnal regulation in glycaemic control in patients with type 2 diabetes. We demonstrated that a larger percentage of daily energy consumed at breakfast is associated with significant reduction in overall PPHG in patients with type 2 diabetes. The findings also suggest the possibility of applying the second-meal phenomenon as a way of reducing postprandial glucose excursion, especially at lunch. This, along with the benefits of the Bdiet schedule in reducing overall hyperglycaemia, may have a therapeutic advantage for the achievement of optimal metabolic control and may prevent cardiovascular and other complications of type 2 diabetes.

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**Contribution statement** JW contributed to the conception and design of the study, acquired and interpreted data and drafted the article. OF and DJ contributed to the conception and design of the study, acquired, analysed and interpreted data and drafted and revised the article. BA researched data, contributed to the interpretation of the data and drafted and revised the article. HRR, YB and ZL contributed to the conception and design of the study, acquired and interpreted data, organised the randomisation and drafted the article. All listed authors approved the final version of the manuscript. DJ is the guarantor of this work.

## References

- Monnier L, Lapinski H, Colette C (2003) Contributions of fasting and postprandial plasma glucose increments to the overall diurnal hyperglycemia of type 2 diabetic patients: variations with increasing levels of HbA(1c). *Diabetes Care* 26:881–885
- Ceriello A, Colagiuri S, Gerich J, Tuomilehto J, Guideline Development G (2008) Guideline for management of postmeal glucose. Nutrition, metabolism, and cardiovascular diseases. *Nutr Metab Cardiovasc Dis* 18:S17–S33
- Szuskiewicz-Garcia MM, Davidson JA (2014) Cardiovascular disease in diabetes mellitus: risk factors and medical therapy. *Endocrinol Metab Clin N Am* 43:25–40
- Morgan LM, Shi JW, Hampton SM, Frost G (2012) Effect of meal timing and glycaemic index on glucose control and insulin secretion in healthy volunteers. *Br J Nutr* 108:1286–1291
- van Cauter E, Shapiro ET, Tillil H, Polonsky KS (1992) Circadian modulation of glucose and insulin responses to meals: relationship to cortisol rhythm. *Am J Physiol* 262:E467–E475
- Lindgren O, Mari A, Deacon CF et al (2009) Differential islet and incretin hormone responses in morning versus afternoon after standardized meal in healthy men. *J Clin Endocrinol Metab* 94:2887–2892
- Saad A, Dalla Man C, Nandy DK et al (2012) Diurnal pattern to insulin secretion and insulin action in healthy individuals. *Diabetes* 61:2691–2700
- Gibbs M, Harrington D, Starkey S, Williams P, Hampton S (2014) Diurnal postprandial responses to low and high glycaemic index mixed meals. *Clin Nutr* 33:889–894
- Gil-Lozano M, Mingomataj EL, Wu WK, Ridout SA, Brubaker PL (2014) Circadian secretion of the intestinal hormone GLP-1 by the rodent L cell. *Diabetes* 63:3674–3685
- Dyar KA, Ciciliot S, Wright LE et al (2014) Muscle insulin sensitivity and glucose metabolism are controlled by the intrinsic muscle clock. *Mol Metab* 3:29–41
- Froy O (2010) Metabolism and circadian rhythms—implications for obesity. *Endocr Rev* 31:1–24
- Wu T, Sun L, ZhuGe F et al (2011) Differential roles of breakfast and supper in rats of a daily three-meal schedule upon circadian regulation and physiology. *Chronobiol Int* 28:890–903
- Prasai MJ, Mughal RS, Wheatcroft SB, Kearney MT, Grant PJ, Scott EM (2013) Diurnal variation in vascular and metabolic function in diet-induced obesity: divergence of insulin resistance and loss of clock rhythm. *Diabetes* 62:1981–1989
- Yoshino J, Imai S (2010) A clock ticks in pancreatic beta cells. *Cell Metab* 12:107–108
- Sadacca LA, Lamia KA, deLemos AS, Blum B, Weitz CJ (2011) An intrinsic circadian clock of the pancreas is required for normal insulin release and glucose homeostasis in mice. *Diabetologia* 54:120–124
- Reppert SM, Weaver DR (2002) Coordination of circadian timing in mammals. *Nature* 418:935–941
- Damiola F, Le Minh N, Preitner N, Kornmann B, Fleury-Olela F, Schibler U (2000) Restricted feeding uncouples circadian oscillators in peripheral tissues from the central pacemaker in the suprachiasmatic nucleus. *Genes Dev* 14:2950–2961
- Hara R, Wan K, Wakamatsu H et al (2001) Restricted feeding entrains liver clock without participation of the suprachiasmatic nucleus. *Genes Cells* 6:269–278
- Loboda A, Kraft WK, Fine B et al (2009) Diurnal variation of the human adipose transcriptome and the link to metabolic disease. *BMC Med Genomics* 2:7
- Reutrakul S, Hood MM, Crowley SJ, Morgan MK, Teodori M, Knutson KL (2014) The relationship between breakfast skipping, chronotype, and glycemic control in type 2 diabetes. *Chronobiol Int* 31:64–71
- Mekary RA, Giovannucci E, Willett WC, van Dam RM, Hu FB (2012) Eating patterns and type 2 diabetes risk in men: breakfast omission, eating frequency, and snacking. *Am J Clin Nutr* 95:1182–1189
- Fuse Y, Hirao A, Kuroda H, Otsuka M, Tahara Y, Shibata S (2012) Differential roles of breakfast only (one meal per day) and a bigger breakfast with a small dinner (two meals per day) in mice fed a high-fat diet with regard to induced obesity and lipid metabolism. *J Circadian Rhythms* 10:4
- Sherman H, Genzer Y, Cohen R, Chapnik N, Madar Z, Froy O (2012) Timed high-fat diet resets circadian metabolism and prevents obesity. *FASEB J* 26:3493–3502
- Jakubowicz D, Froy O, Wainstein J, Boaz M (2012) Meal timing and composition influence ghrelin levels, appetite scores and weight loss maintenance in overweight and obese adults. *Steroids* 77:323–331
- Jakubowicz D, Barnea M, Wainstein J, Froy O (2013) High caloric intake at breakfast vs. dinner differentially influences weight loss of overweight and obese women. *Obesity* 21:2504–2512
- Jakubowicz D, Froy O, Ahrén B et al (2014) Incretin, insulinotropic and glucose-lowering effects of whey protein pre-load in type 2 diabetes: a randomised clinical trial. *Diabetologia* 57:1807–1811
- Rabinovitz HR, Boaz M, Ganz T et al (2014) Big breakfast rich in protein and fat improves glycemic control in type 2 diabetics. *Obesity* 22:E46–E54
- Lee SH, Tura A, Mari A et al (2011) Potentiation of the early-phase insulin response by a prior meal contributes to the second-meal phenomenon in type 2 diabetes. *Am J Physiol Endocrinol Metab* 301:E984–E990
- Ravanam A, Jeffery J, Nehlawi M, Abiraira C (1991) Improvement of glucose-primed intravenous glucose tolerance and correction of acute insulin decrement by glipizide in type II diabetes. *Metab Clin Exp* 40:1173–1177
- Jovanovic A, Gerrard J, Taylor R (2009) The second-meal phenomenon in type 2 diabetes. *Diabetes Care* 32:1199–1201
- Bonuccelli S, Muscelli E, Gastaldelli A et al (2009) Improved tolerance to sequential glucose loading (Staub-Traugott effect): size and mechanisms. *Am J Physiol Endocrinol Metab* 297:E532–E537