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Glycaemic, insulin and ghrelin responses to traditional South Asian flatbreads in diabetic and healthy subjects

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Abstract

In the South-East Asian subcontinent, flatbreads contribute the main portion of carbohydrate to a meal. There are no specific data on the effect of different flatbreads on satiety and recurrent hunger, as indicated by the duration of ghrelin suppression after a meal. The present study was designed to examine the glycaemic, insulin and ghrelin responses to traditional subcontinental breads in type 2 diabetic subjects and healthy volunteers. For this purpose, twelve normoglycaemic healthy volunteers and ten type 2 diabetic patients, in the fasting state, consumed one of five common flatbreads on consecutive days. Capillary blood glucose was examined in the fasting state and serially for 5 h after a meal. Serum insulin and ghrelin levels were determined at hourly intervals for 5 h after the consumption of bran and plain chapatti flatbreads. The incremental area under the curve (iAUC) was calculated for glycaemic and insulin responses, while the net AUC was used to assess the ghrelin response. The results showed that glycaemic and insulin iAUC were lowest for bran chapatti, and highest for plain chapatti. Furthermore, bran chapatti showed maximum ghrelin suppression in both normal and diabetic groups. In conclusion, the low-glycaemic index bran chapatti flatbread had a lower postprandial glycaemic excursion and insulin response, and a more prolonged suppression of ghrelin levels, compared with the plain chapatti flatbread, and in each case, the difference was greater for the diabetic subjects than for the normal subjects. The inclusion of these flatbreads in the diabetic/weight-reducing diet may help weight loss by promoting satiety and reducing hyperinsulinaemia.

Key words: Type 2 diabetes: Glycaemic index: Area under the curve: Insulin: Ghrelin: South Asian flatbreads

While the prevalence of type 2 diabetes has increased tremendously globally, a disproportionate rise has been observed in the South Asian subcontinent (Pakistan, India and Bangladesh), linked to an increasing trend towards a sedentary lifestyle and obesity[1,2]. Furthermore, it has been forecast that the number of diabetics in this region could double by 2025, with a much larger number of individuals with pre-diabetic states underlying the tip of the iceberg[3]. This is of particular concern due to the inadequate infrastructure to deal with a health crisis of this scale in developing economies of the region. National strategies focusing on diabetes prevention are clearly the answer, and as demonstrated by studies such as the Diabetes Prevention Programme, targeting the underlying cause through lifestyle change is the most successful intervention to achieve this[4].

Nutritional intervention in any community must take into account the prevalent dietary practices. In the South Asian subcontinent, meals traditionally contain some form of flatbread as a staple food. In most cases, this consists of unleavened bread known as 'roti' or 'chapatti', although leavened breads like 'naan' are also used. These breads supply the main source of carbohydrate in a traditional subcontinental meal, and this dietary pattern is generally maintained in South Asian immigrant populations in Europe and North America, in whom there is a markedly increased diabetes risk over the native population[5,6]. In the nutritional management of diabetes, the composition of these breads is often modified by the addition of bran or legume flour.

Type 2 diabetes is marked by the loss of early-phase insulin response that is reflected in postprandial hyperglycaemia[7].

Abbreviations: GI, glycaemic index; iAUC, incremental area under the curve.

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Postprandial hyperglycaemia is an independent cardiovascular risk factor, and is the main contributor to glycated Hb in the initial stages of diabetes[8,9]. Diets that decrease postprandial glucose rise can be expected to result in improvement in HbA1c, and also diminish the long-term cardiometabolic risk.

The glycaemic response to a meal depends primarily upon the quantity and quality of its carbohydrate content, and is quantified by indices such as the glycaemic index (GI) and load. The GI of food compares the glycaemic response to a test meal with that seen after ingestion of a standard meal containing a specified amount (usually 50 g) of carbohydrate, while the glycaemic load is a product of the absolute carbohydrate content and the GI[10]. In addition to the GI, the total load of carbohydrate consumed contributes to the glycaemic response of different foods, which is thus dependent on both the type and amount of carbohydrate contained. Dietary intervention for controlling postprandial hyperglycaemia may target either or both of these factors.

Whereas the quality of the carbohydrate content is modulated by altering the GI of a meal, the absolute amount consumed depends largely upon the satiety-promoting effect of a meal. Ghrelin, a peptide hormone released by the neuroendocrine P/D1cells in the mucosa of the gastric fundus, is a major orexigenic stimulus that acts on hypothalamic appetite pathways to promote hunger[11–13]. Low levels of ghrelin are seen after the ingestion of a meal containing carbohydrate, and a rise is observed after a variable interval depending upon the type of food[14]. The differential effect of carbohydrate meals with different glycaemic indices on ghrelin levels has not been ascertained. Food that keeps ghrelin levels suppressed for a longer period after a meal can be expected to promote satiety and prevent recurrent hunger after a meal. It is of interest to determine whether there is a difference between the effect of various sources of carbohydrate in the degree and duration of suppression of ghrelin levels after a meal, as it can be expected to correlate with an increase in postprandial fullness and interval before subsequent food intake.

Although GI data and the effect of using low-GI flour in bread-making have been studied for breads consumed in Western diets, the effect of altering the composition of bread consumed in the South Asian subcontinent on GI is not known. The satiety-promoting effect of different breads has also not been studied in subcontinental breads. To address these questions, we designed a series of experiments to study the glycaemic, insulin and ghrelin responses to traditional subcontinental breads in type 2 diabetic patients and healthy volunteers. We hypothesised that the use of low-GI flour and the addition of fibre to standard flour in flatbread-making would decrease the glycaemic response and increase satiety compared with an ordinary bread.

Research design and methods

Setting

The research proposal was registered with the Services Hospital research registry (unique identifier SHL07/2009). All clinical procedures were carried out at the Endocrinology Unit and Diabetes Management Centre, Services Hospital, Lahore. ELISA was done at the Pathology Department of the Services Institute of Medical Sciences, Lahore. The study was conducted in accordance with the ethical guidelines set out in the Declaration of Helsinki and all clinical procedures were approved by the Institutional Review Board/Ethics Committee, Services Hospital.

Subjects

Participants included twelve normoglycaemic healthy volunteers (eight males and four females; mean age 25·7 (SD 7·5) years and mean BMI 23·2 (SD 4·8) kg/m^2) and ten patients with diet-controlled type 2 diabetes (seven males and three females; mean age 47·9 (SD 7·7) years and mean BMI 27·4 (SD 10·5) kg/m^2). All volunteers gave written informed consent for participation. Exclusion criteria included pregnancy, any comorbid conditions or concomitant medication (including oral contraceptives), which could interfere with the results or be deleterious to the safety of the study participants. Baseline anthropometric measurements, physical examination and electrocardiogram were done before GI testing.

Methods

Meal selection and preparation. To determine the common breads consumed by the diabetic subjects, 100 patients attending the diabetic clinic were asked to fill in a questionnaire listing the three more common staples they consumed in their everyday diet. Based on the results of this survey, the common breads consumed were unleavened chapatti flatbread made from milled wheat flour (plain chapatti) and unleavened chapatti made in a 50:50 ratio with milled wheat flour and wheat bran (bran chapatti). Other common breads

Table 1. Amounts, macronutrient composition and energy content of the tested staples*

Meals	Serving size (g)	Energy content (kJ)	Carbohydrates (g)	Fats (g)	Proteins (g)	Dietary fibre (g)
WB	99	1112·9	50·0	3·3	7·5	2·4
CPF	142	1733·2	50·2	15·7	17·1	9·0
NAAN	91	1002·3	50·2	0·6	6·8	1·8
PLAIN C	115	988·9	50·4	1·3	9·6	8·6
BRAN C	165	826·9	50·4	0·6	9·4	19·3

WB, white bread (standard); CPF, chickpea flour chapatti; NAAN, naan flatbread; PLAIN C, plain chapatti; BRAN C, bran chapatti.
* Macronutrient composition calculated using ESHA Food Processor software version 10.2.3 (ESHA Research).

were chickpea flour chapatti and naan (leavened bread made from white flour). These four commonly consumed breads were selected for GI testing (details of the macronutrient content of all breads are shown in Table 1). The selected meals were prepared on-site in the metabolic kitchen and their weight was standardised to yield 50 g available carbohydrate. Plain white bread (Metro C&C) equivalent to 50 g available carbohydrate was used as the standard meal, to offset the concerns of the diabetic patients about the use of glucose as the standard meal. To improve palatability, all meals were served with 250 ml tea prepared with 30 ml milk (sweetened with aspartame, Canderel; Searle).

Glycaemic index testing. The participants were advised to include unrestricted carbohydrate in their daily diet, in the 3-d period immediately preceding the consumption of the test diet. This was done to ensure adequate carbohydrate intake in the days preceding the test, as a carbohydrate-restricted diet before the test can interfere with the glycaemic response to an oral challenge[15]. On each of the five consecutive days, the participants attended the clinic at 08.00 hours after an overnight (8 h) fast. Capillary blood glucose was tested in the fasting state using a glucometer (Accucheck Go, GmBH). The subjects were given the test meals in the following order: day 1, white bread (standard meal); day 2, chickpea flour chapatti; day 3, naan; day 4, plain chapatti; day 5, bran chapatti. The subjects were asked to consume the meal fully within 15 min. Capillary blood glucose was tested at 15, 30, 45, 60, 90, 120, 180, 240 and 300 min after the end of the meal. The subjects did not smoke and remained seated throughout the test. The postprandial glycaemic response was measured serially for 5 h after each test meal, to compensate for the delayed return to baseline glycaemia expected to be seen in the diabetic patients.

Insulin and ghrelin measurements. On days 4 and 5 (plain chapatti and bran chapatti) along with the glucose measurements, 3 ml venous blood were drawn into two serum separator vacutainers (BD) at 0, 60, 120, 180 and 300 min for the determination of insulin and ghrelin levels. Insulin and ghrelin levels were only tested on these 2 d (with the most commonly consumed flatbreads) and at hourly intervals rather than following the pattern of glucose estimation due to ethical considerations, because of the large cumulative volume of blood required.

Blood was allowed to clot at room temperature for 30 min, and then centrifuged (2500 rpm, 4°C, 15 min), to separate the serum. An inhibitor (phenylmethanesulfonyl fluoride (PMSF); Sigma) was added to the tubes for ghrelin estimation, to prevent the degradation of ghrelin. Serum was transferred to duplicate microcentrifuge tubes for insulin and ghrelin measurements, and then stored at −20°C to be tested later as a batch. Ghrelin was assayed by a sandwich ELISA method on a microtitre plate reader, using a commercial kit (Human Ghrelin (Total) ELISA kit; Millipore), which has a range of 100–5000 pg/ml, and an intra-assay and inter-assay variation of 1 and 2·6 %, respectively. Insulin was similarly assayed using an insulin ELISA kit (NovaTec, Immunodiagnostika, GmBH), with a lower sensitivity of 12 pmol/l, and an intra-assay and inter-assay variation of 2 and 6 %, respectively.

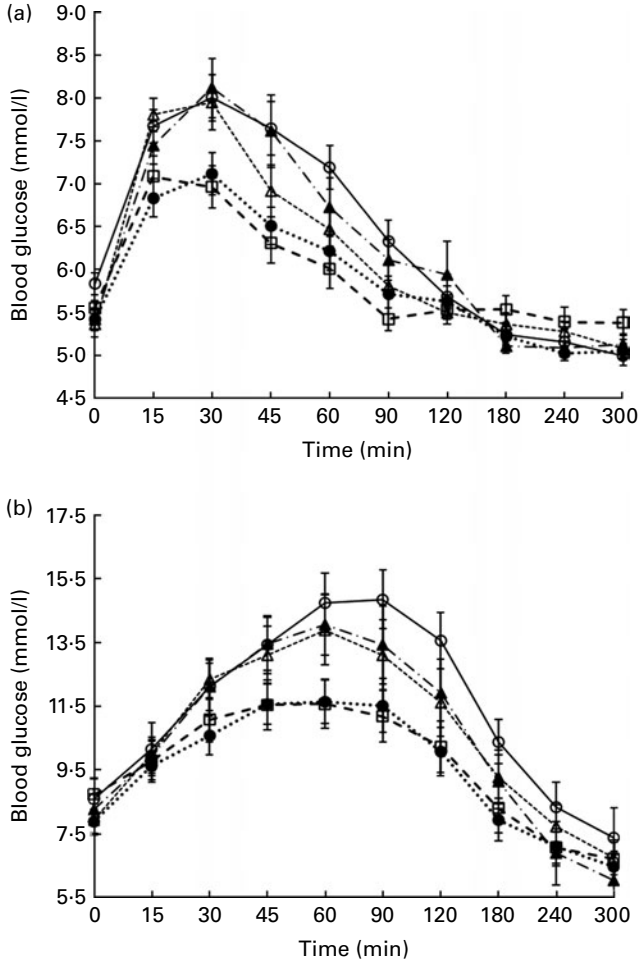

Fig. 1. Mean blood glucose concentrations in the normal and diabetic subjects in response to different flatbreads. (a) Mean blood glucose data from twelve normal subjects. The peak glycaemic level is reached at 30 min and values return to fasting levels at 120 min. (b) Mean blood glucose values from ten diabetic subjects. The peak glycaemic level is reached at 60–90 min and values return to fasting levels at 180–240 min. Values are means, with standard errors represented by vertical bars. —○—, White bread (standard); ··●··, chickpea flour chapatti; ··△··, naan flatbread; –▲–, plain chapatti; – ⊟ –, bran chapatti.

Area under the curve calculations and statistical analysis. Incremental areas under the curve for glycaemia and insulin (iAUC) were calculated using the trapezoidal rule, after subtracting the baseline value from all subsequent readings and ignoring values below the baseline. For AUC calculations involving ghrelin levels, the net AUC was calculated, in which a negative value was assigned to values below the baseline, and was thus suited for measurements that show a declining trend from the baseline[16]. Results are expressed as mean values with their standard errors for the normal or control subjects on each of the test days. Two-way repeated-measures ANOVA was used to determine any statistical difference among the AUC values within the type of meals and between the treatment groups, at the 5 % level of significance. A paired *t* test was applied to compare the results of day 4 (high-GI) and day 5 (low-GI) meals within the treatment groups.

Table 2. Incremental area under the glycaemic curve (iAUC) for the test breads
(Mean values with their standard errors and 95 % confidence intervals)

Groups	Meals	Mean* (mg × min/dl (mmol × min/l))	SE	95 % CI†
Normal	WB	2616·3 (145·3)	1727·3	−986·9, 6219·4
	CPF	2203·1 (122·4)	863·8	401·3, 4005·0
	NAAN	3084·1 (171·3)	1184·4	1436·4, 6377·8
	PLAIN C	3330·0 (185·0)	1114·0	1006·3, 5653·7
	BRAN C	1920·0 (106·7)	630·7	604·3, 3235·7
Diabetic	WB	15 627·8 (868·2)	1892·2	11 680·7, 19 574·8
	CPF	8079·8 (448·9)	946·2	6105·9, 10 053·6
	NAAN	12 916·2 (717·6)	1297·5	10 209·7, 15 622·7
	PLAIN C	12 261·8 (681·2)	1220·3	9716·3, 14 807·2
	BRAN C	5790·8 (321·7)	690·9	4349·5, 7232·0

WB, white bread (standard); CPF, chickpea flour chapatti; NAAN, naan flatbread; PLAIN C, plain chapatti; BRAN C, bran chapatti.

*Values are in mg × min/dl, with SI units in brackets.

† Statistical significance was tested using two-way repeated-measures ANOVA ($F_{cal} = 29{\cdot}85$; $P = 0{\cdot}000$). Mauchly's test of sphericity was found to be significant in this case, and thus violated the assumption ($P = 0{\cdot}038$) of sphericity. As the best choice for the analysis, the Greenhouse–Geisser correction had to be used to estimate epsilon[32]. The statistical difference between the pairs of meals was determined by the multiple comparison test (Bonferroni test). It was found that glycaemic iAUC for both CPF and BRAN C was statistically lower than the standard meal of white bread with P values of 0·002 and 0·000, respectively. Repeated-measures ANOVA showed that iAUC is also significantly different ($F_{cal} = 29{\cdot}85$; $P < 0{\cdot}01$) between the two groups (i.e. diabetic and normal).

Results

Glycaemic response

The glycaemic response was determined for a 5 h period after each test meal (Fig. 1). The mean fasting capillary glucose was 5·4 (SEM 0·1) mmol/l (976 (SEM 2·1) mg/l) and 8·3 (SEM 0·5) mmol/l (148·7 (SEM 9·8) mg/dl) in the healthy volunteers and diabetic patients, respectively. The glycaemic responses for the selected flatbreads in the normal and diabetic subjects are shown in Fig. 1(a) and (b), respectively.

To compare the glycaemic responses to different meals, the iAUC was calculated (Table 2). The mean iAUC values were then used to determine the GI over a 5 h test period for each of the test meals, compared with the reference meal of white bread. The results are shown in Fig. 2. The breads with the highest and lowest GI in the normal and diabetic subjects were plain chapatti/bran chapatti and naan/bran chapatti, respectively. The GI values in the diabetic and normal subjects were different for all the test meals, but approached statistical significance only in the case of plain chapatti ($P < 0{\cdot}05$). The assigned GI category of the different meals was as follows: normal subjects – high GI (≥70), white bread, naan and plain chapatti; medium GI (56–69), chickpea flour chapatti; low GI (≤55), bran chapatti; diabetic subjects – high GI, white bread, naan and plain chapatti; medium GI, none; low GI, chickpea flour chapatti and bran chapatti. Thus, the assigned GI category differed for one test meal, namely chickpea flour chapatti between the normal and diabetic subjects (Fig. 2).

The glycaemic responses to the highest-GI (plain chapatti) and lowest-GI (bran chapatti) flatbreads in the normal and diabetic subjects were compared. In both categories of subjects, the low-GI flatbread shifted the glycaemic response curve downwards, and the effect was greater in the diabetic subjects than in the normal subjects (Fig. 3).

Insulin response

Insulin levels were determined on day 4 (plain chapatti, the highest-GI bread) and day 5 (bran chapatti, the lowest-GI bread). The fasting insulin levels were higher in the diabetic subjects than in the normal subjects (mean 96·0 (SEM 17·4) *v.* 28·8 (SEM 8·4) pmol/l). The insulin response to low- and high-GI breads in both normal and diabetic subjects is shown in Fig. 4. Insulin levels returned to pre-meal levels 60–120 min earlier with the low-GI meal compared with the high-GI meal. The insulin response curve for the diabetic subjects showed prolonged hyperinsulinaemia in subjects consuming high-GI chapatti flatbread made from whole-wheat

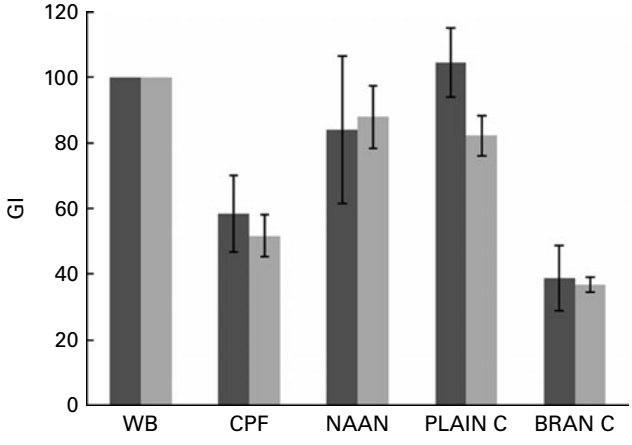

Fig. 2. Glycaemic index (GI) of four traditional breads compared with a standard meal. The 5 h GI ($GI_{wb(5\,h)}$) was calculated using white bread containing 50 g available carbohydrate as the reference meal, taken as GI 100, against which the GI of the other test breads was measured. Plain chapatti had the highest and bran chapatti had the least GI. Values are means, with standard errors represented by vertical bars. WB, white bread (standard); CPF, chickpea flour chapatti; NAAN, naan flatbread; PLAIN C, plain chapatti; BRAN C, bran chapatti. ■, Non-diabetic; ■, diabetic.

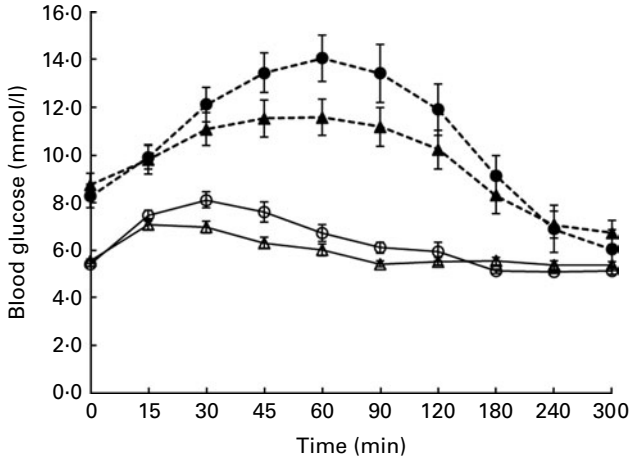

Fig. 3. Glycaemic response to the consumption of a low-glycaemic index (GI) bread compared with a high-GI bread. The mean glycaemic response of the normal (N; *n* 12) and diabetic (D; *n* 10) subjects to the highest- and lowest-GI flatbreads, both containing 50 g available carbohydrate. The glycaemic curve shifts downwards and to the left for the latter, and the shift effect is greater in the D subjects (··●·· (high GI), ··▲·· (low GI)) compared with the N subjects (—○— (high GI), —△— (low GI)). Values are means, with standard errors represented by vertical bars.

flour, and a marked reduction in this trend when the subjects consumed low-GI bran chapatti.

A paired *t* test was applied to see the difference in insulin iAUC in both normal and diabetic groups for high- and low-GI breads separately. The mean iAUC for insulin in response to the low-GI meal was significantly lower than the iAUC for the high-GI meal in the normal ($P=0{\cdot}002$) and diabetic groups ($P=0{\cdot}003$) (Table 3).

Ghrelin response

The mean fasting serum ghrelin concentrations were higher in the diabetic subjects than in the normal subjects, although the difference was of marginal significance statistically (1316 (SEM 128·4) *v.* 966 (SEM 133·9) pg/ml, $P=0{\cdot}07$). The ghrelin levels began to rise after 60 min with the high-GI bread (plain chapatti), but remained suppressed for up to 5 h with the low-GI bread (bran chapatti). The net ghrelin response to these two breads in the diabetic and normal subjects is shown in Fig. 5. Ghrelin net AUC was calculated in the normal and diabetic subjects, for both kinds of bread. Results from the paired *t* test indicate that ghrelin net AUC within the groups was significantly lower with the low-GI meal than with the high-GI meal, in both normal and diabetic groups, at the 5% level of significance (Table 4).

Table 3. 5 h Insulin incremental AUC (iAUC) for the low- and high-glycaemic index test meals in the normal and diabetic subjects
(Mean values with their standard errors and standard deviations)

Groups	Meals	Mean 5 h iAUC (pmol × h/l)	SD	SEM	*P*
Normal	PLAIN C	271·6	117·2	35·3	0·002
	BRAN C	35·4	110·7	33·4	
Diabetic	PLAIN C	398·3	215·9	71·9	0·003
	BRAN C	113·3	170·8	56·9	

PLAIN C, plain chapatti; BRAN C, bran chapatti.

Discussion

The burgeoning epidemic of obesity and type 2 diabetes in the South-East Asian subcontinent has been attributed to an increasingly sedentary lifestyle and changing dietary patterns, with increased intake of energy-dense food containing a large proportion of refined carbohydrates and saturated fats[17]. Although flatbreads form the principal source of carbohydrate in a traditional subcontinental meal, GI data for these are limited. Using calculated GI based on the GI of the ingredients is

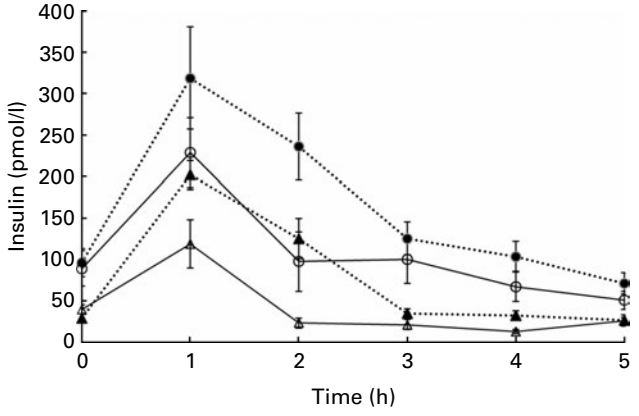

Fig. 4. Insulin response to high- and low-glycaemic index (GI) breads in the normal (N) and diabetic (D) subjects. In both N and D subjects, peak insulin levels were observed at 60 min in response to either low-GI (—△— (N), ··○·· (D); bran chapatti) or high-GI (—▲— (N), ··●·· (D); plain chapatti) breads; however, the insulin response extended to the 4th hour with the high-GI meal, compared with the low-GI meal, in which fasting levels were regained at 120 min. Values are means, with standard errors represented by vertical bars.

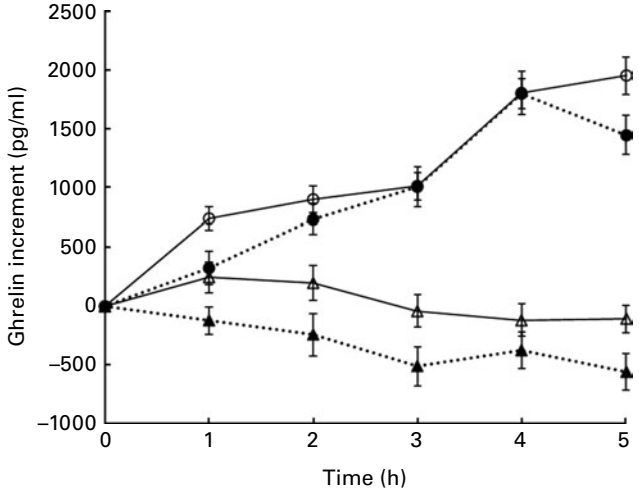

Fig. 5. Change in ghrelin levels from baseline after the high- and low-glycaemic index (GI) meals. Serum ghrelin levels were measured at hourly intervals. There is a steady rise in ghrelin levels after a high-GI (—○— (N), ··●·· (D)) meal in both categories of subjects, while prolonged suppression of ghrelin levels is seen in both normal (N) and diabetic (D) subjects after a low-GI (—△— (N), ··▲·· (D)) meal. Values are means, with standard errors represented by vertical bars.

Table 4. Comparison of ghrelin net area under the curve (AUC) after the high-glycaemic index (GI) and low-GI breads in the normal and diabetic subjects

(Mean values with their standard errors and standard deviations)

Groups	Meals*	Mean 5 h net AUC (pg × h/ml)	SD	SEM	*P*
Normal	High GI	5468·2	1539·1	444·3	0·000
	Low GI	143·9	2372·1	684·8	
Diabetic	High GI	4509·1	1403·2	467·7	0·000
	Low GI	−1546·6	2079·4	693·1	

* High GI, plain chapatti; low GI, bran chapatti.

one way of overcoming this deficiency[18], but this method has its limitations, as it does not take into account the effect of cooking method, and how the different ingredients act together to produce a cumulative glycaemic response[19]. This dearth of reliable data prompted us to test for the GI of four traditional flatbreads, of which two were traditionally advocated as 'diabetic' breads (chickpea flour chapatti and bran chapatti)[20], while the other two were the most commonly consumed household breads (plain whole-wheat chapatti and naan). To elucidate the effect of these breads on postprandial satiety, serial postprandial insulin and ghrelin levels after the consumption of one low-GI and one high-GI flatbread were compared in both healthy volunteers and diabetic patients.

We included a diabetic group among the study participants because we expected the glycaemic response to carbohydrates, and, by derivation, the GI, in the diabetic subjects to be different from that seen in the normal volunteers. The applicability of GI values obtained from the normal subjects to patients with diabetes has been debated: some authors[21] have been able to demonstrate significant correlation between the GI in normal and diabetic subjects, while others have argued against this due to the greater amplitude and duration of the glycaemic excursion in diabetics[22]. We were able to demonstrate that in the diabetic patients, the glycaemic response to all of the tested flatbreads was prolonged and of higher amplitude, compared with the normal volunteers (Fig. 1). On the other hand, in most cases, the assigned GI category for the flatbreads was the same in the normal and diabetic subjects, suggesting that GI data from the healthy volunteers may reasonably be extrapolated to the diabetic patients.

An objective of the present study was to determine whether consumption of a high-GI flatbread had an effect on appetite, as this could be a mechanism by which a high-GI meal could contribute to weight gain and obesity. In this context, we measured insulin and ghrelin responses to low- and high-GI breads. The rationale for measuring insulin levels was that the greater amplitude and duration of insulin response with high-GI foods can result in postprandial hypoglycaemia, which in turn leads to increased appetite and increased energy intake at the next meal (glucostatic theory)[23]. High insulin levels can also promote weight gain due to the lipogenic effect of insulin. Serial measurements of ghrelin levels were done to assess the duration of postprandial ghrelin suppression, which may be considered as a marker of satiety.

We were able to demonstrate that postprandial insulin response was shorter and of lower amplitude with the low-GI flatbread compared with the high-GI flatbread (Fig. 4). The present results were in agreement with the findings reported by Galgani *et al.*[24] and Krog-Mikkelsen *et al.*[25] who noted a similar difference in the amplitude and duration of insulin response after low-GI mixed meals.

The significantly lower ghrelin response to the low-GI bread compared with the high-GI bread (Fig. 5) was an important finding, as a significant relationship between postprandial ghrelin levels and the GI of the meal has not been demonstrated previously. Taking into consideration the role of ghrelin in the regulation of appetite, this prompts the question whether low-GI flatbreads promote satiety, and can thus have a role in a weight-reducing diet.

The effect of the GI of a meal on appetite and satiety is unsettled[26]. Ludwig[27] reviewed studies which had examined the effect of the GI of a meal on subjective sensations of satiety and hunger after its consumption: a majority of these reported that a high-GI meal was inversely related to postprandial satiety scores, subsequent desire for food was experienced earlier and the amount of food consumed *ad libitum* in the following meal was more after the consumption of high-GI meals compared with low-GI meals. A higher insulin response to high-GI meals was the proposed explanation for this effect on postprandial hunger. Other researchers, however, were unable to corroborate this effect of high-GI foods on satiety. Flint *et al.*[28] studied the effect of meals with varying GI and macronutrient composition on appetite, but were unable to show a correlation between the GI and subsequent appetite sensations, although they too found that a low-GI meal resulted in a lower energy intake at the next meal.

More recent studies have tried to document the effect of the GI on hunger pathways by looking at serial measurements of ghrelin levels after a meal. Krog-Mikkelsen *et al.*[25] studied the effect of two standardised isoenergetic, macronutrient-matched diets, which differed only in the GI, on several metabolic parameters including ghrelin; they found that although the low-GI meal induced greater postprandial fullness compared with the matched high-GI meal, the postprandial ghrelin response did not differ significantly between the groups. The present findings, however, were in contrast to this, as we showed a significant difference in the ghrelin response between the low-GI and high-GI meals. It is possible that this difference between the present findings and their results was due to the difference in the composition of the breads investigated: the test breads used in the present study were based on traditional recipes, and although equilibrated to 50 g carbohydrate, differed in energy and macronutrient content, unlike the standardised meals used by this group.

An unexpected finding regarding the ghrelin trends was that an initial fall below the baseline in mean ghrelin levels was not seen after the consumption of the high-GI bread (although it was observed in a few individual subjects; Fig. 5). This was surprising as ghrelin levels are expected to fall after carbohydrate-based meals[14]. It has also been reported that the duration of postprandial ghrelin suppression is

proportional to the energy content of the meal[29]. We therefore considered the possibility that this difference in postprandial ghrelin nadir after the high- and low-GI flatbreads may be due to the difference in their energy content. However, we discounted this explanation, because if this was the case, the high-GI bread, which had higher energy content, would be expected to suppress ghrelin for a longer duration than the low-GI bread, while the converse was true in the present results. We finally surmised that this finding only reflected individual variation in time to ghrelin nadir and its return to baseline, as it has previously been noted that the time to recovery of ghrelin levels after the initial postprandial fall tends to be variable in different subjects[29]. It is possible that if the first postprandial sample (which was obtained 75 min after the start of the meal) had been taken earlier, an initial dip in ghrelin level could have been seen more universally. We recommend more frequent sampling in the first hour when looking at postprandial ghrelin trends, to avoid missing these fluctuations.

The present study requires some methodological explanation. Traditionally, the GI of a meal is calculated considering the glycaemic response over 2 h in normal subjects, and over a 3 h period in diabetics[30]. However, according to Wolever[16], there is no 'right' method for testing the glycaemic response, and researchers have opted to study the GI over different time spans when needed[31]. The conventional 3 h test duration in the diabetic patients was originally 'chosen as a compromise between what is ideal to allow the blood glucose response to return to baseline (5 h) and what is practical for subjects testing many foods'[30]. We selected a longer test duration, as we wanted to study the prolonged metabolic response to the test breads, to see the effect on serial postprandial insulin and ghrelin levels, and to compare these with the glycaemic response over a similar period. Another difference in our method of determining the GI from the usual practice was the choice of white bread as the standard, while most studies have used 50 g glucose as the reference meal. This was due to the reservation most diabetic patients have about ingesting a large glucose load. Consequently, as all our readings are based on white bread, the GI values are different from values reported in nutritional databases, where available.

Conclusions

Bran and chickpea flour chapatti traditionally advocated as part of the diabetic diet have a lower GI compared with everyday flatbreads such as naan and plain flour chapatti. Low-GI bran chapatti flatbread has a lower postprandial glycaemic excursion and insulin response, and a more prolonged suppression of ghrelin levels, compared with plain chapatti flatbread, and in each case, the difference is greater for the diabetic subjects than for the normal subjects.

Recommendations

The substitution of low-GI flatbreads such as chickpea flour chapatti and bran chapatti for the more commonly consumed high-GI household breads may be advocated as part of the diabetic or weight-reducing diet, and can be expected to promote weight loss by promoting satiety and reducing insulin levels.

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