The effect of low glycaemic index diet on reducing daylong glycaemia in healthy young adults: a randomized crossover trial

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Abstract

Aims: We compared the effect of a low glycaemic index (LGI) diet on reducing daylong glycaemia with a macronutrient-match high glycaemic index (HGI) diet, using customized meal delivery to ensure compliance.
Materials and Methods: We conducted a single-blinded randomized crossover trial in 14 healthy adults (57% female) with a mean ± SD age of 21.6 ± 1.7 years. A flash glucose monitoring sensor was installed on the subjects in day 1 to capture interstitial glucose level every 15 minutes for 14 days. Subjects were randomized to receive an LGI (dietary GI = 40) or HGI (dietary GI = 60) diet (three meals and two snacks) from day 2 for 5 consecutive days, followed by a 2-day washout, and switched to the alternative diet for another 5 days. Paired t -test was used to test the differences in incremental area-under-curve (iAUC) of glucose, postprandial glucose concentration (PPG) and maximum postprandial glucose rise (MPGR) between the LGI and HGI periods.
Results: Subjects had lower iAUC for average daylong glycaemia during the LGI intervention period compared with the HGI period (mean ± SD, 865 ± 297 *vs.* 1024 ± 267 mmol * min/L; $p = 0.047$). PPG for breakfast and snack 2; and MPGR for breakfast, snack 2 and dinner were lower in the LGI period.
Conclusions: In young healthy adults, following an LGI diet resulted in lower average daylong glycaemia compared with a macronutrient matched HGI diet. Our results support the use of LGI diets to reduce the risk of developing glucose intolerance.
Keywords: glycaemic index; flash glucose monitoring; daylong glycaemia; postprandial glycaemia
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INTRODUCTION

Glycaemic Index (GI) is the term introduced by Jenkins *et al*[1] to indicate how much the available carbohydrates in foods raised blood glucose on a gram for gram basis. Foods with a higher GI are digested and absorbed in a quicker manner, causing a higher postprandial blood glucose excursion in a shorter duration [1].

The shift from traditional diets which are low GI (LGI) to industrialized diets, which are fiber-depleted, energy-dense, and nutrient poor[2], coincides with a drastic increase in the prevalence of obesity, insulin resistance and cardiovascular diseases (CVD)[3]. Accumulating evidence from acute and long-term studies have confirmed the health benefits of LGI diets, including but not limited to increased satiety[4], and improvements in postprandial glycaemia[5], insulin sensitivity[6], weight control[4,7] and inflammatory biomarkers[8].

Studies have shown that 70% of the total daytime hyperglycaemia is postprandial hyperglycaemia[9]. Non-diabetic individuals are able to response to postprandial hyperglycaemia and excursion by producing more insulin. When excessive glucose excursion occurs regularly in the long term, i.e. high glycaemic variability, the pancreas may become exhausted because of the need to secrete high levels of insulin frequently, eventually resulting in β cell dysfunction[10,11]. High glycaemic variability has also been shown to induce oxidative stress[12,13] and inflammation[14], and cause glycation of proteins and enzymes responsible for the insulin signaling pathways[15]. Dietary strategies to limit postprandial glycaemia and glycaemic excursions may therefore be beneficial to health. As carbohydrates are the main determinant of postprandial glycaemia, such strategies commonly work on the principles of reducing carbohydrate quantity and improving carbohydrate quality (i.e. GI)[16]. Consuming an LGI meal has also been shown to improve glycaemic control in the subsequent meal (i.e. the second meal effect)[17,18]. Therefore, following a low GI diet should theoretically improve day-long glycaemic control. A meta-analysis concluded that an LGI diet improves HbA_{1c} levels in patients with type 2 diabetes mellitus (T2DM), which suggest improvement in day-long glycaemia[19].

Despite the above, data on day-long glycaemic profile are rarely reported. Furthermore, in previous studies subjects were commonly only given education and instructions on how to follow an LGI diet, potentially leading to low compliance, which has been the major criticisms from opponents of the GI concept[20]. The aim of this study is therefore to examine the effect of an LGI diet on reducing day-long glycaemia for 5 days under controlled condition where all foods are provided, using a flash-glucose monitoring system (FGMS). We hypothesized that an LGI diet, compared with a macronutrient matched HGI diet, would results in lower day-long glycaemia.

MATERIAL AND METHODS

Study design

The study is a crossover and randomized controlled trial conducted between Oct 2018 – Mar 2019 at the University of Hong Kong (HKU). The study was approved by the Institutional Review Board of the HKU/Hospital Authority Hong Kong West Cluster (reference no. UW18-313), and prospectively registered at anzctr.org.au (registration number: ACTRN12618001576213). All subjects provided written informed consent before commencing the study.

Subject recruitment

Healthy adults were recruited through poster advertisements in HKU and mass email. The inclusion criteria of the participants include aged 18-40 years, with a body mass index (BMI) between 18.5-23 kg/m^2, non-smoker, non-regular alcohol drinker, generally healthy with no chronic diseases (e.g. non-diabetic), no skin sensitivity issues, able to wear the subcutaneous FGMS sensor for 14 days, able to receive, store and reheat the study meals according to the instructions, no special dietary requirement (e.g. non-vegetarian or vegan), no vigorous exercise during the study, and ownership of a smartphone with camera function. The exclusion criteria include doctor's diagnosis of any form of impaired glucose tolerance or any chronic diseases, individuals with impaired immunity, regularly taking any form of medication and women who are pregnant or planning to be pregnant during the study. Screening session was done to assess the eligibility, to clearly explain the study, the obligations and the right of being the participant after the enrolment.

Study diets

Both study diets were designed to be matched for macronutrient and fiber content for each meal – around half of the energy intake was contributed by carbohydrates, while fat and protein each contributed a quarter. Both diets were formulated to provide 15 g dietary fiber per 1000 kcal. The only difference between the two

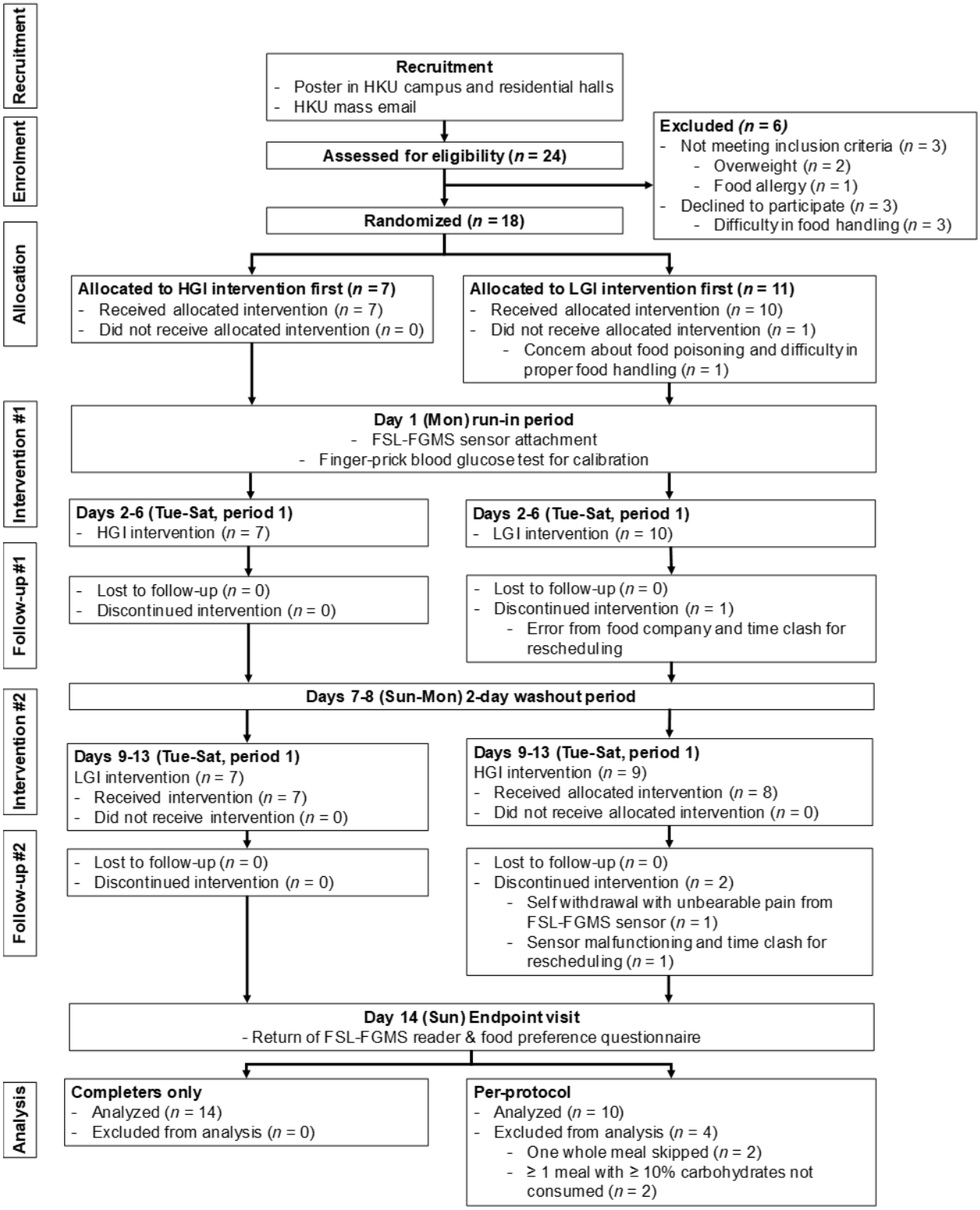

Figure 1 - The flow of the study, adapted from CONSORT.

diets was the GI – the HGI diet had 20-unit difference in GI when compared with the LGI diet. A sample menu was available in the **Online Supplementary Appendix 1**. Breakfast, lunch, dinner and 2 snacks were provided to participants during the intervention periods (2 × 5 days). The foods were purchased from a meal-delivery service (EatologyAsia, Hong Kong SAR), designed by the dietitian of the company according to the specifications of this study. The United States Department of Agriculture Food Composition Databases[21] and the nutritional label from the food packaging were referred to for the nutritional composition of foods in the study diets. The international tables of GI from Atkinson *et al*[22] were used to assign the GI values when designing the study diets. GI of a meal was calculated by the following formula: GL of a meal * 100 / total available carbohydrates of the meal in grams. Dietary GI values of the test diets were calculated by the following formula: sum of GL of all test meals in a day *100 / total available carbohydrates in a day in grams. The amount of food provided to each participant varied to meet the individual estimated energy requirements, the calculation of which was described in the subsequent section. However, the ratio of macronutrients in the diet was the same for all participants. Drinks were not provided but a drink list for both LGI and HGI diet marked by the dates of the intervention period was given (**Online Supplementary Appendix 2**).

Study protocol

The flow of study was shown in **Figure 1**. At enrolment, the weight and height of the subjects were measured with an electronic scale and the age and sex were recorded for the calculation of the basal metabolic rate, using Liu's equation[23] which is Chinese-, age-, weight- and height-specific. The estimated energy requirement was then calculated by multiplying

Table 1 – characteristics of study participants

Variables	CO (n = 14)	PP (n = 10)	P†
Females, *n* (%)	8 (57.1)	6 (60.0)	1.000
Age (y)	21.6 (1.7)	21.2 (1.5)	0.596
Weight (kg)	55 (4.9)	56 (5.5)	0.797
Height (m)	1.64 (0.06)	1.65 (0.06)	0.670
Body mass index (kg/m^2)	20.3 (0.9)	20.3 (1.05)	0.864
EER (kcal)	2044.4 (185.8)	2059.9 (199.8)	0.848
Preference on LGI‡	58.7 (16.9)	60.8 (16.7)	0.778
Preference on HGI‡	71.7 (14.7)	73.5 (16.4)	0.791
Fasting glucose concentration (mmol/L)	4.2 (0.4)	4.3 (0.5)	0.934
Mean time of awake (min)	959 (49)	968 (45)	0.686
Time of meal consumption (min from awake)			
Breakfast	10 (3)	11 (3)	0.429
Snack 1	138 (68)	159 (62)	0.448
Lunch	272 (52)	271 (53)	0.964
Snack 2	456 (86)	474 (74)	0.598
Dinner	632 (63)	646 (48)	0.561

Values are presented as mean (SD) for continuous variables or *n* (%) for categorical variables. CO, completers only; EER, estimated energy requirement (calories); HGI, high glycaemic index; LGI, low glycaemic index; PP, Per-protocol.
†P-values of continuous variables were obtained via t-tests, while those of categorical variables were obtained via chi-square tests.
‡Preference was assessed using a 5-point Likert scale and was analyzed in the orthodox way, such that a value of 71.1 represents that the subject had drawn a vertical line at 71.1% of the total length of the scale, counting from the left.

the basal metabolic rate by physical activity level and was used as the daily energy intake. Subjects were then randomized into two groups regarding the order of the treatment (i.e. LGI diet first or HGI diet first), using computer-generated random numbers before the intervention. The recruiter and participants were blinded to the assignment.

The study was conducted over fifteen days during which ten 24-hour periods of glucose measurements were captured by an FGMS device (FreeStyle Libre, Abbott Hong Kong; FSL-FGMS). This device was validated for its ability to capture postprandial changes in blood glucose concentrations in healthy adults[24]. A run-in was conducted on the first day, right before the intervention, in order to insert the FSL-FGMS sensor into the upper arm of the subject with a one-time blood glucose test by finger-prick for correction of the data if necessary. Each participant was educated on how to scan the sensor and were instructed to scan the sensor around 4 times a day, with the first scan conducted once the participant woke up, indicating the wake up time, the last scan conducted right before the participant sleeps, indicating the sleep time, and 2-3 times or every 6-7 hours during the day. Participants were instructed to have no medium to intense physical activity during the intervention in order to avoid the interference on blood glucose level[25].

The dietary intervention involved participants consuming an LGI or HGI diet for 5 consecutive 24-hour periods, followed by the alternative intervention for another 5 consecutive 24-hour periods, with a two-day wash-out period between interventions (**Figure 1**). Subjects were instructed to eat only the food provided and drink according to the drinks list during the intervention period. They were allowed to resume their normal diet during the run-in and wash-out period.

In order to gauge the participants' preference towards the two diets, they were asked the following question at the end of each intervention: "How much do you like your food in these five days?". They were asked to provide their answers in a five-point Likert scale, with 1 meaning "not at all" and 5 meaning "very much" (the Likert scale used in the study is shown in the **Online Supplementary Appendix 3**). The sensor was removed by the subjects after receiving both interventions (i.e. day 14, Sunday) following the instructions from the researchers for convenience. The FSL-FGMS reader and the food-preference questionnaire were collected on the endpoint visit. Subjects who decided to withdraw from the study did the endpoint visit on the day of withdrawal. The withdrawal reason was recorded, and the sensor was removed by the observer. The intervention was then discontinued.

Study outcomes

The blood glucose data were automatically sampled by the FSL-FGMS sensor every 15 minutes, and up to 8-hours' worth of data can be stored. Every time the subjects scanned the sensor using the hand-held reader, the data stored in the sensor were automatically transferred to the reader and the sensor could continue storing new data. Data recorded by the FSL-FGMS sensor were downloaded as Excel spreadsheets. Only the data during the awake time indicated by the first and the last scan of the day were included to reflect the day-long glycaemia. Incremental area under curve (iAUC) of each day were calculated by the trapezoidal rule using Microsoft Excel, and the average daily iAUC during intervention was the primary outcome of this study. The glucose level measured immediately after waking up was regarded as the baseline level for that day. The secondary outcomes include the average maximum peak glucose rise (MPGR) following each meal (breakfast, snack 1, lunch, snack 2 and dinner), reflected by the highest blood glucose level above the daily baseline after each meal; and the average peak postprandial glucose concentration (PPG) following each meal, reflected by the absolute value of the highest blood glucose level after each meal. PPG and MPGR of each meal (breakfast, snack 1, lunch, snack 2 & dinner) were identified from the raw FSL-FGMS data by the first author. Results of the Likert scale were analyzed in the orthodox way, for example a value of 71.1 represents that the subject had drawn a vertical line at 71.1% of the total length of the scale, counting from the left.

The mealtime was reported by the participants for at least one day in each 5-day period. The reported mealtime was used as the counter check of spotting the peak of the blood glucose level in each meal and the awake time (e.g. the first data, indicating the wakeup time, was used as the baseline value). The mealtime and wakeup time were further confirmed with the participant when anomaly was suspected (e.g. the last scan, indicating the sleep time, was immediately after the reported dinner time).

Details on how missing data were handled, and adjustments made to the FSL-FGMS values prior to PPG analysis were given in **Online Supplementary Appendices 4 and 5**.

Determination of compliance

Photo record was captured by the participants when they cannot finish the meal or when extra food was accidentally eaten. The total food intake was then re-calculated with food weight estimation by the first author. It is regarded as non-compliant when the food left contains more than 10% of the total carbohydrates in that meal or when the extra food eaten contains more than 10 g of carbohydrates. The American Diabetes Association considers 6-10 g of carbohydrates as ½ serving in its guideline[26], suggesting that this amount would likely impact on the blood glucose level. Given that the mean amount of carbohydrate in each meal is 52 ± 12 g, the cut-off was set at 10%.

Sample size calculation

Our study was designed *a priori* to utilize a sample size of 14 in a cross-over design to achieve 80% power to detect an effect size of 0.82 at $p < 0.05$, based on the data from the study by Camps *et al* which suggested the effect size to be 0.86[27]. Since this is a study comparing the difference in daylong glycaemia between two dietary interventions in healthy adults, effect sizes smaller than 0.8 is unlikely to be considered as clinically relevant. We recruited 18 subjects to allow for a 30% dropout.

Table 2 – Actual dietary intakes of completers (n = 14, 8 females)

Parameters	LGI	HGI
Energy (kcal/d)	F: 1946 (118); M: 2205 (139)	
Carbohydrates (%kcal/d)	49.8 (0.4)	50.0 (0.2)
Protein (%kcal/d)	24.9 (0.4)	24.8 (0.4)
Total fats (%kcal/d)	25.1 (0.7)	24.9 (0.3)
Fibre (g/1000kcal)	20.9 (1.4)	18.2 (1.1)
Average meal GI†		
Breakfast	44 (5)	61 (11)
Snack 1	39 (7)	63 (13)
Lunch	38 (6)	57 (9)
Snack 2	32 (8)	57 (4)
Dinner	45 (6)	59 (6)
Average dietary GI†	40 (4)	60 (3)

Values were mean (SD). Carbohydrates were available carbohydrates.
† Both meal and dietary GI presented were average values of those of the five test days. GI of a meal or a day was calculated by the following formula: sum of meal or daily GL * 100 / sum of available carbohydrates in a meal or day.

Statistical analysis

All statistical analyses were conducted by both completers only (CO) and per-protocol (PP) approaches using SPSS (version 25, IBM Corp Ltd., New York USA). The CO analysis included all subjects who completed the study. The PP analysis excluded participants who were non-adherent to the allocated dietary program. Intention-to-treat analysis was not performed in this study as the aim was to evaluate the effect of LGI *vs.* HGI diet under controlled conditions. Paired *t*-test was performed to test the differences in iAUC, PPG and MPGR between the LGI and HGI periods. A $p < 0.05$ was considered statistically significant.

RESULTS

Of 24 individuals assessed for eligibility, 18 subjects met all inclusion criteria. During the intervention, one participant did not receive the intervention because of concerns about food poisoning and difficulty in food handling right before the intervention, two participants decided to withdraw from the study during the first intervention period and one participant discontinued the intervention during the second intervention period, leaving the data from 14 participants to be included in the CO analysis. Four participants were excluded from the PP analysis because of non-compliance in consuming the provided foods. Subject characteristics in CO and PP analyses were shown **Table 1**. There were no significance differences observed between two groups in sex, age, anthropometric measurements, daily estimated energy requirements and the average preference on both diets. There was significant difference in preference between the LGI and HGI diets (mean ± SD preference: 58.7 ± 17.6 *vs.* 71.7 ± 15.2%, $p = 0.037$) in CO analysis but no significant differences in PP analysis (60.8 ± 17.6 *vs.* 73.5 ± 17.3%, $p = 0.102$). The overall actual mean dietary intakes achieved by the 14 participants are summarized in **Table 2**. The foods were assumed to be fully consumed by the participants unless reported.

A sample day-long glycaemic profile of a participant during a day each of LGI and HGI intervention periods is shown in **Online Supplementary Appendix 6.** Comparing the daylong glycaemic profiles of all subjects between the two intervention periods, CO analysis revealed that subjects had lower mean ± SD iAUC for daylong glycaemia during the LGI intervention period compared with the HGI period (865 ± 297 *vs.* 1024 ± 267 mmol * min/L; $p = 0.047$), but the statistical significance was lost in the PP analysis (859 ± 312 *vs.* 1036 ± 297 mmol * min/L; $p = 0.146$).

The adjusted PPG in both CO and PP analyses, with the use of pairwise deletion and listwise deletion methods in handling the missing data were shown in **Figure 2**. The LGI diet resulted in lower PPG in breakfast and snack 2 in both CO (7.51 ± 0.58 *vs.* 8.05 ± 0.50 mmol/L; $p = 0.005$ and 7.35 ± 0.33 *vs.* 7.62 ± 0.39 mmol/L; $p = 0.027$ respectively) and PP (7.55 ± 0.51 *vs.* 7.96 ± 0.57 mmol/L; $p = 0.014$ and 7.27 ± 0.27 *vs.* 7.69 ± 0.32 mmol/L; $p = 0.003$ respectively) analyses when pairwise deletion was used. When listwise deletion was used, the LGI diet resulted in lower PPG in breakfast (7.48 ± 0.71 *vs.* 7.97 ± 0.72 mmol/L; $p = 0.014$) in the CO analysis only.

Figure 3 illustrates the results of the MPGR. The LGI diet resulted in lower PPG in breakfast and snack 2 in both CO (1.90 ± 0.84 *vs.* 3.01 ± 0.76 mmol/L; $p < 0.001$ and 1.70 ± 0.62 *vs.* 2.31 ± 0.66 mmol/L; $p = 0.01$ respectively) and PP (1.91 ± 0.80 *vs.* 2.79

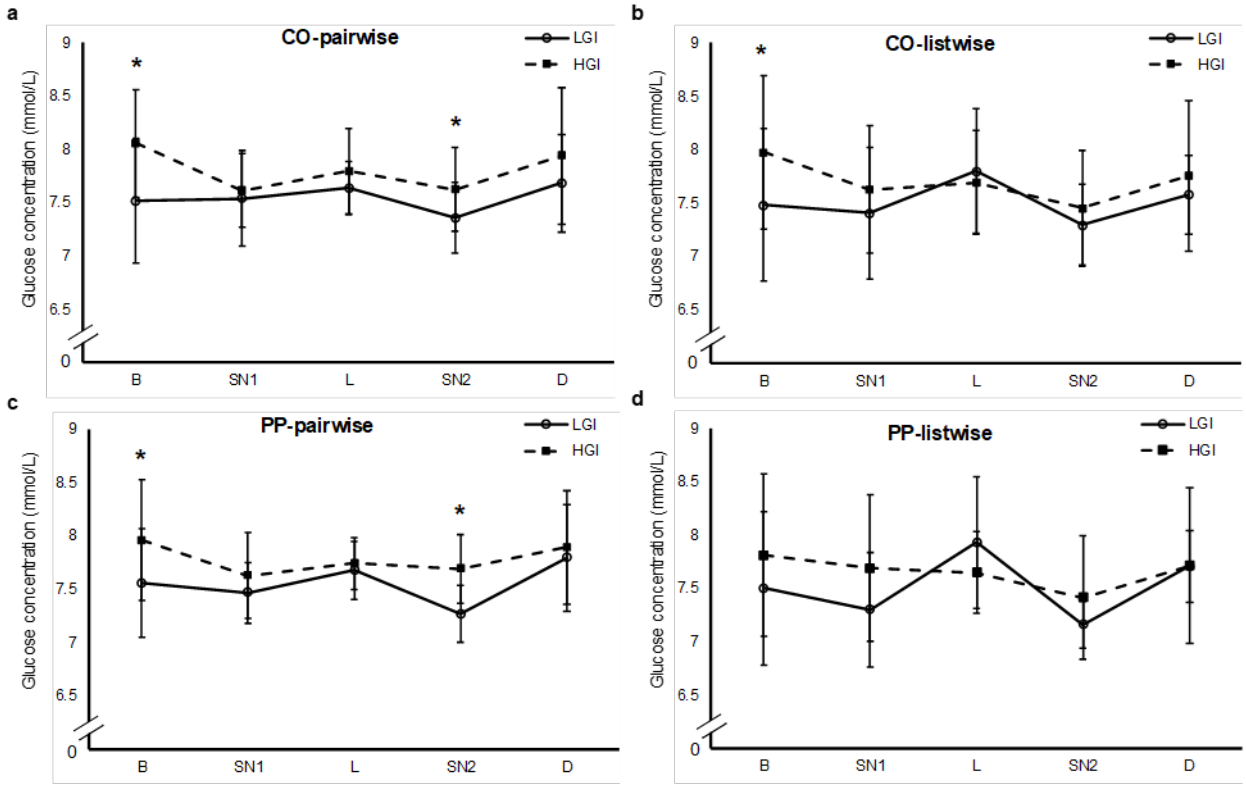

Figure 2 – Comparison of peak postprandial glucose concentrations of each meal between LGI and HGI diets, different analyses, and different methods of handling missing data. (a) Completers only, pairwise deletion (n = 14) (b) Completers only, listwise deletion (n = 14) (c) Per-protocol, pairwise deletion (n = 10) (d) Per-protocol, listwise deletion (n = 10). Values are mean and error bars are SD. Asterisks depict statistically significant difference between LGI and HGI diet in a single meal. The difference between pairwise and listwise deletion was explained in online supplementary appendix 4 and 5. CO, completers only. PP, per-protocol. LGI, low glycaemic index. HGI, high glycaemic index. B, breakfast. SN1, 1st snack. L, lunch. SN2, 2nd snack. D, dinner.

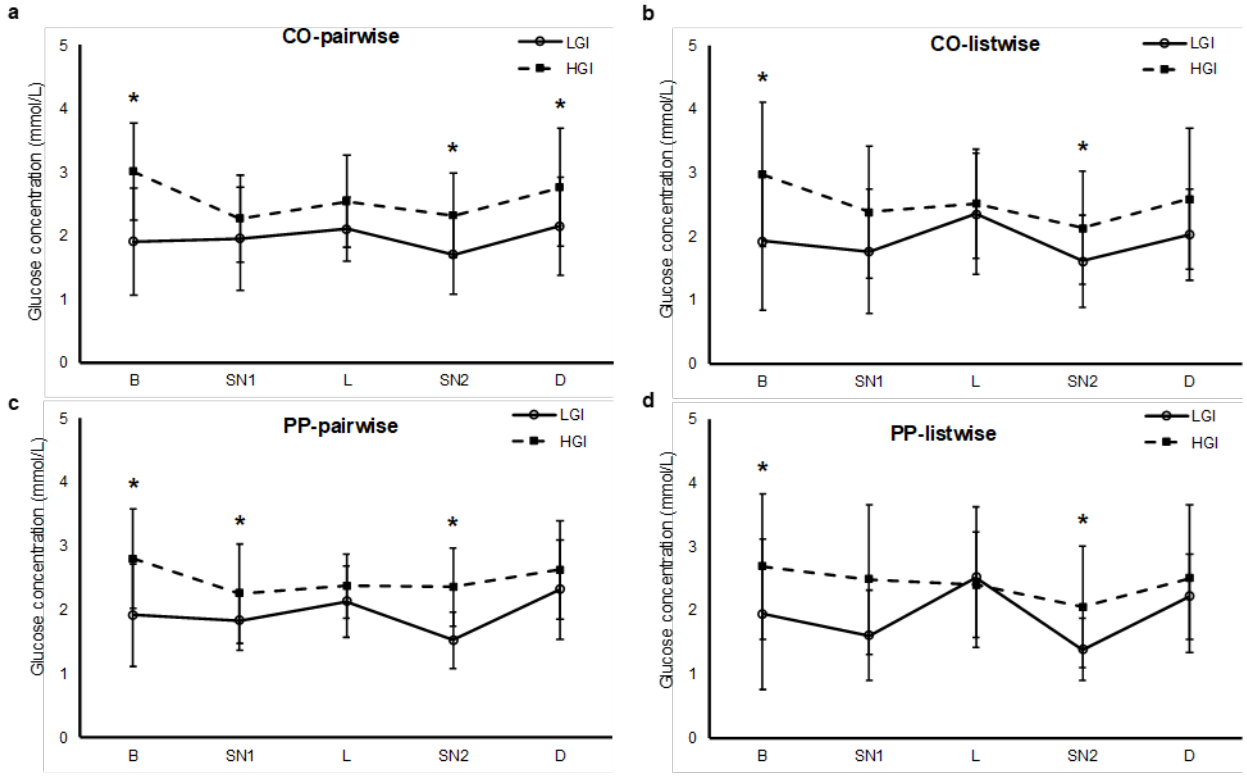

Figure 3 – Comparison of maximum postprandial glucose rise of each meal between LGI and HGI diets, different analyses, and different methods of handling missing data. (a) Completers only, pairwise deletion (n = 14) (b) Completers only, listwise deletion (n = 14) (c) Per-protocol, pairwise deletion (n = 10) (d) Per-protocol, listwise deletion (n = 10) Values are mean and error bars are SD. Asterisks depict statistically significant difference between LGI and HGI diet in a single meal. The difference between pairwise and listwise deletion was explained in online supplementary appendix 4 and 5. CO, completers only. PP, per-protocol. LGI, low glycaemic index. HGI, high glycaemic index. B, breakfast. SN1, 1st snack. L, lunch. SN2, 2nd snack. D, dinner.

± 0.78 mmol/L; p = 0.002 and 1.50 ± 0.44 *vs.* 2.35 ± 0.61 mmol/L; p = 0.004 respectively) analyses when pairwise deletion was used. The key differences between CO and PP analysis were that there were significant differences in dinner in CO analysis (2.15 ± 0.78 *vs.* 2.76 ± 0.93 mmol/L; p = 0.034) only and in snack 1 in PP analysis (1.82 ± 0.48 *vs.* 2.25 ± 0.78 mmol/L; p = 0.04) only. With the use of listwise deletion, only the differences between LGI and HGI diet in breakfast and snack 2 remained significant in both CO and PP analysis although the statistical significance and absolute difference were slightly attenuated.

DISCUSSION

In our trial with very good dietary compliance, a 5-day LGI dietary intervention resulted in a 17% lower daylong glycaemia compared with a macronutrient matched HGI dietary intervention. Specifically, lower PPG levels were observed after LGI breakfasts and snacks compared with their HGI counterparts, while no statistically significant differences between the two diets were observed for lunch and dinner.

While the statistical significance was lost in the PP analyses for some outcome variables, it is likely to be due to a reduction in sample size rather than a true absence in the treatment effect. This is supported by the similar iAUC values between CO and PP analyses, as well as the similar subject characteristics between those included in the two analyses. In fact, the subjects removed from the PP analyses still provided individual datapoints that were resulted from adhering to the experiment protocol. Removing these datapoints could lead to unnecessary exclusion of datapoints and biased conclusions[28]. Nonetheless, the results of the two analyses were presented to let the readers have a more informed conclusion.

Our findings are consistent with that of previous studies[27,29]. In a study of 12 healthy males of normal weight by Reynolds *et al*[29], an LGI diet (four meals in 10 hours) resulted in significantly lower glucose mean iAUC on daylong (10 h) glycaemia compared with the HGI diet (mean ± SE: LGI: 221 ± 36 *vs.* HGI: 356 ± 49 mmol * min/L; p = 0.027)[29]. Similar to our findings, the study by Camps *et al* [27] in 13 healthy, normal weight Asian men showed that consumption of LGI meals consisted of Singaporean Asian foods resulted in a 35% reduction (p = 0.014) in 24-hour blood glucose iAUC compared with macronutrient matched HGI meals. Our results expand on these by showing similar effects on average daylong glycaemia for 5 days, with more variation in the foods used to construct the LGI and HGI meals. This further strengthens the evidence base to support of the use of LGI diets as a lifestyle modification to hinder the development of glucose intolerance, as high glycaemic variability have been shown to increase oxidative stress and contribute to the etiology of glucose intolerance[30].

In order to control for the potential effects of other macronutrients on daylong glycaemia, the LGI and HGI meals in our study were macronutrient matched. Practically, an LGI diet is often associated with a higher fiber content[31], leading difficulties in lowering the fiber content to match with the HGI diet. The fiber contents in the 2000-kcal LGI and HGI diets of this study are 43.9 g/day and 36.4 g/day respectively, which are above the Institute of Medicine recommended value of 25 g per day[32]. However, it is unclear whether the differences in the absolute amount of the dietary fiber consumed by each participant, which is based on the total energy intake of their prescribed diets, may have an effect on PPG, as both GI and dietary fiber (especially viscous fiber) consumption can influence postprandial glycaemic excursion[26].

One of the possible reasons why significant differences were only shown in certain meals (most in breakfast and snack 2) but not all meals is the differences in the type and amount of carbohydrates. Although the macronutrients were well matched between LGI and HGI diets, macronutrient matching was not done between meals because of practical considerations regarding the dietary habits of most individuals (i.e. the portion is generally higher in lunch and dinner than snacks), which may have contributed to the difference in effect on glycaemia. Given that cooking method affects the GI[33-35], it is possible that the rice congee in the HGI diet which was fully cooked to give a refined texture when it was prepared, may have been over-cooked when it was reheated by the participants before consumption, thus increasing the GI value more than it is calculated. Pancakes and seeded bread in the LGI diet may be less affected because of the shorter heating period required. Another possible reason that LGI breakfasts result in significantly lower PPG compared with their HGI counterpart is that insulin sensitivity is generally higher in the morning[36], which may have enhanced the effect of a LGI meal.

It is interesting to note that the food preference score was lower when all completers were included, compared with those included in the PP analysis only. This suggests that those who were non-compliant did not enjoy the foods provided, and since they were instructed to only consume the foods provided but nothing else, they opted to either skip the meal or not finish all foods provided. Furthermore, the LGI diet received a lower preference score than the HGI diet. This could be due to the extensive inclusion of

legume dishes in the LGI menu which was not well-received among the participants. This implied that local food preference should be considered when designing LGI menus in the future to enhance compliance.

One of the strengths of this study is that all foods consumed by the subjects were provided, and all subjects had standardized diets, ensuring good compliance to the assigned dietary GI. In previous studies e.g.[37], the compliance to the assigned diet may be suboptimal when the participants were only given education and instructions on how to follow an LGI diet, as the foods eaten were not standardized, and foods that do not correspond to the assigned GI intervention may be inadvertently chosen. It has been shown that the effect of a LGI diet intervention was better when most of the foods were provided[38], compared with that when only dietary education was provided.

We caution the readers to several limitations of our study. First, although there was a large theoretical difference in the average dietary GI (20 units) between LGI and HGI diets compared with previous trials, the exact GI of the included foods may actually be lower than the value obtained from the international table[22]. This was because of the formation of resistant starch due to retrogradation when the foods were refrigerated, which lowers the GI (and hence GL) value[39]. Hence, the HGI diet may have become medium GI, reducing the difference in iAUC. Second, we were unable to ensure full compliance of the study participants and some of the test meals were not consumed. However, while this is a common limitation of dietary trials in humans[40], the interpretation of the outcomes did not change materially after excluding the non-compliant participants, despite the differences were no longer significant due to the reduction in sample size. Third, despite our attempt to recruit healthy adults between 18-40 years old, the mean age of the subjects was in the lower end of this range with a small SD, and all subjects were students. Hence, our findings may not be generalizable to other individuals. Moreover, we only examined blood glucose level but not other metabolic biomarkers, such as insulin level and fructosamine. Therefore, the mechanisms behind the physiological response to LGI and HGI diets could not be fully elucidated in the current study, although these have already been well documented in previous trials[41,42]. Fourth, drinks were not provided in this study due to logistic difficulties. However, participants were advised on the drinks they were allowed or forbidden to have during each intervention period. Fifth, mealtimes were not standardized in this study, which was done to increase compliance and create a condition similar to the real-life. Sixth, the readings of the FSL-FGMS meters were calibrated against the reading of hand-held glucometers by one-point calibrations, instead of multiple points, due to logistic limitations. Finally, the small number of participants involved in this study, despite achieving adequate statistical power, means that the results from this study could not be directly translated to an advice at a population level.

To conclude, in young healthy adults, following an LGI diet resulted in lower average daylong glycaemia compared with a macronutrient matched HGI diet, and the differences in postprandial glycaemia were more pronounced during breakfast and snack time. Our results further support the use of LGI diets for slowing the development of glucose intolerance.

Acknowledgements

N/A.

Conflicts of interest

No potential conflicts of interest were reported in this study. The meals used in this study were purchased at full retail price from the meal delivery service, who had no influence in the study design and writing of the manuscript.

Authors' contribution

JCYL designed and supervised the study. HWHH conducted the trial, collected, processed and analyzed the data, and drafted the original manuscript. IMYT assisted in trial management and data processing and collection. THTW performed additional analyses and revised the manuscript after peer-review. All authors were involved in the interpretation of data and subsequent edits of the manuscript and have read and approved the final manuscript. JCYL has primary responsibility of the content presented.

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Online Supplementary Appendix 1 – a sample menu for both LGI and HGI diet, as well as the nutritional composition, glycemic index, and glycemic load of the food.

		Energy (kcal)	CHO (g)	Protein (g)	Fat (g)	Dietary Fiber (g)	Meal GI* / Dietary GI †	Meal GL^ / Dietary GL#
LGI diet								
Breakfast	Buckwheat & Oatmeal Pancakes with Honeydew Melon	342.5	53.3	20.3	5.4	6.50	42.26*	20.08^
Snack 1	Sweet Potato, carrot and zucchini Noodles with Kale, Spinach and mushroom	369.9	35.6	13.9	19.1	7.84	34.90*	9.68^
Lunch	Buckwheat and broccoli Salad with Chicken Breast	512.2	59.0	35.5	14.9	7.85	42.93*	21.96^
Snack 2	Vanilla Panna Cotta with Strawberries	149.3	28.1	8.1	0.5	3.40	24.56*	6.06^
Dinner	Veggie Spaghetti with Tiger Prawns	427.7	49.5	34.4	10.2	12.26	40.43*	15.06^
Total		**1801.6**	**225.4**	**112.3**	**50.1**	**37.86**	**32.32†**	**72.84#**
HGI diet								
Breakfast	White Rice Porridge with Pork and edamame	355.3	47.7	26.2	6.0	3.04	73.19*	32.70^
Snack 1	Sweet Corn Soup with Chickpeas	343.3	40.0	10.3	15.7	4.49	57.07*	20.30^
Lunch	Swordfish and veggies with Couscous and White Rice	490.6	57.4	37.1	12.2	7.33	63.07*	31.57^

		Energy (kcal)	CHO (g)	Protein (g)	Fat (g)	Dietary Fiber (g)	Meal GI* / Dietary GI †	Meal GL^ / Dietary GL#
Snack 2	Tropical Fruit Salad	134.8	30.6	1.6	0.7	5.55	58.59*	14.69^
Dinner	Paleo Stuffed Peppers, mashed potato and Spinach Salad	490.9	50.4	36.7	15.8	12.00	51.15*	19.65^
Total		**1815.0**	**226.2**	**111.9**	**50.5**	**32.40**	**52.56†**	**118.91#**

LGI, low glycaemic index. HGI, high glycaemic index. CHO, available carbohydrate. GI, glycemic index. GL, glycemic load. Meal or dietary GI was calculated by the formula: meal or dietary GL / available CHO in meal or diet in grams * 100. Meal or dietary GL was calculated by summing the dietary GL values of each individual ingredient. GL of individual ingredient was calculated by the following formula: ingredient GI * available CHO in the ingredient in grams.

Online Supplementary Appendix 2 – The drink lists provided to the subjects

Drink list during low glycaemic index intervention

Drink list (Date:)

Thirsty? No worries! Here's what you can drink! :P
Forbidden fruits tastes so much sweeter…??? <- That's NOT the truth!!!
When you have so many options here, why you have to seek those are not on the list!?

	Beverages
YES! Enjoy~ ☺ 	Drinks below; **NO added sugar** - **Plain water [your best friend to quench your thirst!!!]** - Sparkling (mineral) Water - Tea (Black, White, Green, buckwheat, corn silk, etc.) - Black Coffee (Long black, Espresso, Americano) - Lemon water - Coca-Cola (light, Zero or plus)
Drink moderately 	- Soya Milk - Almond Milk - Cow Milk (Skim, 1%, 2%, whole) - Chocolate Milk - Tea with milk - Lemon Tea (no added sugar) - White Coffee (Cappuccino, Macchiato, Latte, Mocha) - Pure coconut water - Tomato juice - Smoothies (made from milk and fruit) - Fruit Juices (eg. Apple juice, carrot juice, orange juice, tomato juice, etc.) - Yakult (normal/ light) - Sports drinks (no added sugar)
N – O, NO! 	Say NO to ALL non-listed drinks … Just 5 days!!! - Regular soft drinks - ALL sugary drinks - Bubble Tea with sugar - alcoholic drinks

Drink list during high glycaemic index intervention

Drink list (Date: _____)

Thirsty? No worries! Here's what you can drink! :P
Forbidden fruits tastes so much sweeter…??? <- That's NOT the truth!!!
When you have so many options here, why you have to seek those are not on the list!?

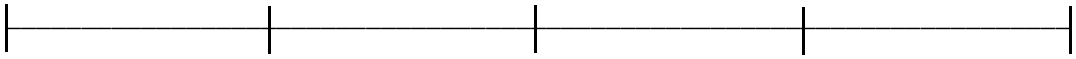
	Beverages
YES! Enjoy~ ☺ 	Drinks below; **NO added sugar** - **Plain water [your best friend to quench your thirst!!!]** - Sparkling (mineral) Water - Tea (Black, White, Green, buckwheat, corn silk, etc.) - Black Coffee (Long black, Espresso, Americano) - Lemon water - Coca-Cola (light, Zero or plus)
Think again… 	- Rice Milk - Sports drinks; if necessary - Energy Drink (Lucozade) - Hong Kong Style Milk Tea (condensed milk) - watermelon juices
N – O, NO! 	Say NO to ALL non-listed drinks … Just 5 days!!! - Regular soft drinks - ALL sugary drinks - Bubble Tea with sugar - Cow's milk/ soya milk/ Almond milk - alcoholic drinks

Online Supplementary Appendix 3 – Likert scale for food preference

Code: _____

After the first 5- day period (_____):

How much do you like your food in these 5 days?

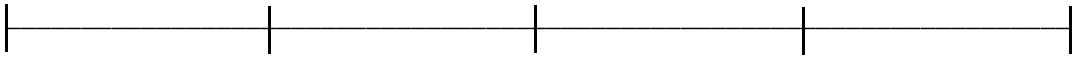


1 2 3 4 5

Not at all Very much

After the second 5-day period (_____):

How much do you like your food in these 5 days?



1 2 3 4 5

Not at all Very much

Online Supplementary Appendix 4

Adjustment of FreeStyle Libre – Flash Glucose Monitoring System (FSL-FGMS) readings

The glucose readings measured by the interstitial fluid through FSL-FGMS system have been proven to reliably reflect changes in glucose levels [1-3]. Adjustment was unnecessary and was not performed in the incremental area-under-curve (iAUC) and maximum peak glucose rise (MPGR) analyses as it may change the shape of the curve [1]. However, the average absolute blood glucose level tested by finger-prick blood glucose test was higher than the glucose readings indicated by FSL-FGMS sensor (mean difference= +0.6, ± 0.9). This suggests the glucose readings from FSL-FGMS may not be a true representation of the absolute blood glucose level, affecting the outcomes related to absolute readings. Therefore, adjustment was performed on the glucose readings data for postprandial glucose (PPG) analysis, using the gender-specific equations developed by our group [1].

Handling of missing data

The methods of pairwise deletion and listwise deletion were used to handle missing data in this study. Pairwise deletion means that when there was missing data or error in one meal, the data of the paired meal was also excluded (e.g. missing data in LGI day 1 breakfast means the data in HGI day 1 breakfast was also excluded). The advantage of this method is to minimize the sample size loss. On the other hand, listwise deletion was used when there was missing data or error in one meal, the data on that day (5 meals in total) and the paired day was excluded (e.g. missing data in LGI day 1 breakfast means the data of LGI day 1 and HGI day 1, from breakfast to dinner were also excluded). It was considered as a valid and conservative way to handle the data when

there was uncertainty. For example, when there was missing peak (<5 peaks in a day) while all five meals were consumed, the combined meal effect (when the mealtime between two meals being too close) would result in only one peak observed for two meals. However, the disadvantage is great loss of sample size. The results of PPG and MPGR analyzed based on pairwise deletion and listwise deletion were compared and the number of pairs of meals (n_m) was set as the unit for PPG and MPGR analysis. The participant flow for each analysis was shown in **Online Supplementary Appendix 5**.

Missing data were classified as missing completely at random (MCAR) when there was no specific pattern observed between LGI and HGI diets. The sources of missing data includes sensor error (SE), reflected by abnormally low glucose level (below 4 mmol/L in most of the time during the day) on the last day of the intervention; food-related issue (F) when the whole provided meal was not eaten because of food spoilage; sensor-scanner issue (S) when there were missing data in a period of time during the day with meals consumed because of insufficient scanning by the participant and peaks observation issues (P) when there was a missing peak of a particular meal.

Because of the use of paired-*t*-test, the data were considered as a pair instead of single data points. Number of pairs of days (n_d) was set as the unit for iAUC analysis, and pairwise deletion was used to handle the missing data. When there was missing data or error in one particular day, the data of the paired day was also excluded (i.e. missing data in LGI day 1 means the data in HGI day 1 was also excluded). Although last observation carried forward (LOCF) is one of the ways to replace the longitudinal missing data [4], the missing data observed in the participants violate the assumptions

that there was no food consumed during that period of time. Hence, LOCF is not an appropriate approach to handle the missing data

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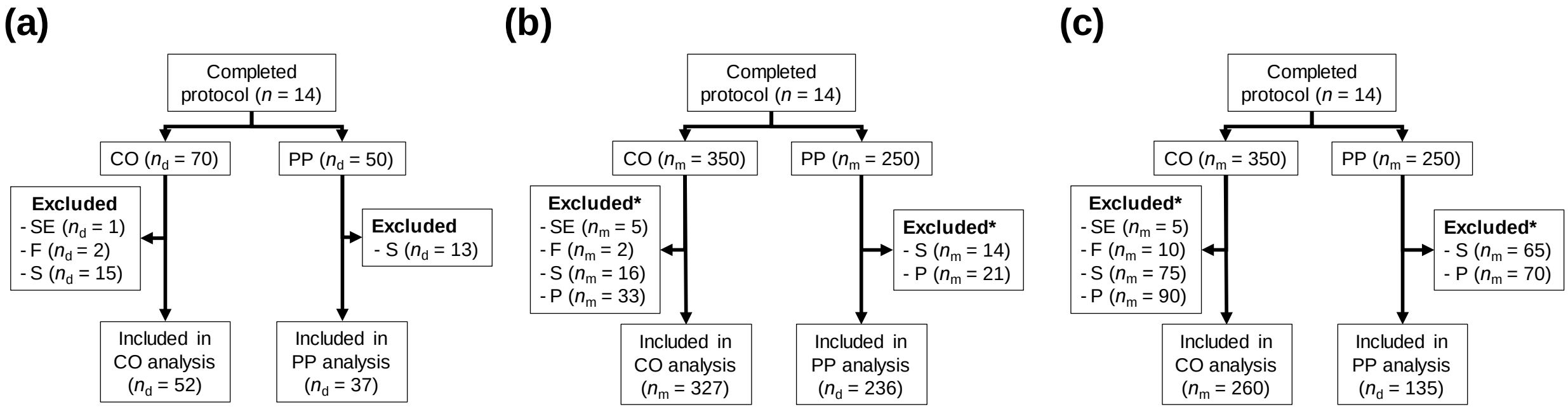
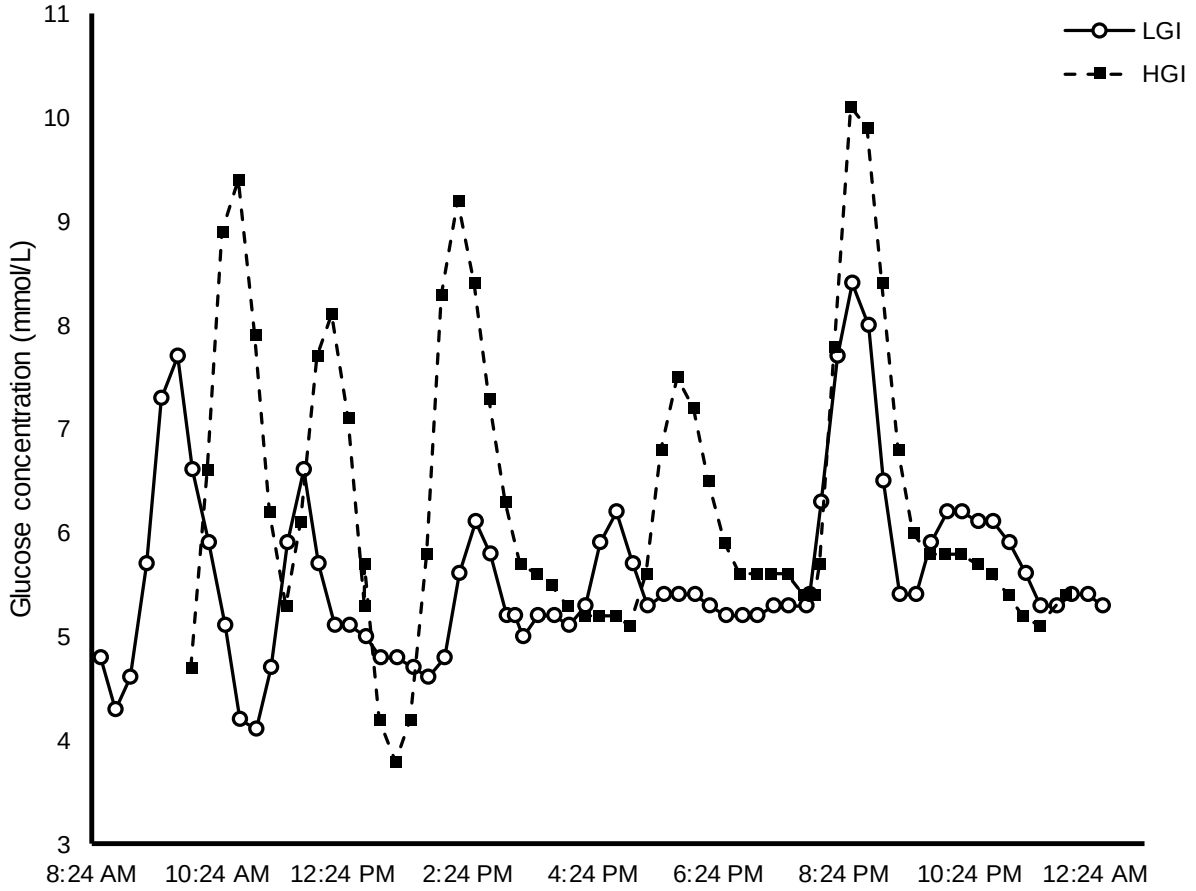


Online Supplementary Appendix 5 – Schematic diagram illustrating how missing data were handled in: (a) iAUC analysis; (b) PPG and MPGR analyses with the use of pairwise deletion, and (c) PPG and MPGR analyses with the use of listwise deletion. * reasons for exclusion were not mutually exclusive. CO, completers only; F, Food-related issue; iAUC, incremental area under curve; MPGR, maximum postprandial glucose rise; n_d, number of pairs of days; n_m, number of pairs of meals; P, peaks observation issue; PP, per-protocol; PPG, peak postprandial glucose level; S, sensor-scanning issue; SE, sensor error.



Online Supplementary Appendix 6 – sample daylong glycaemic profile of a participant when following the LGI and the HGI diet. LGI, low glycaemic index. HGI, high glycaemic index.