



Does type of bread ingested for breakfast contribute to lowering of glycaemic index?

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HIGHLIGHTS

- Glycaemic index of mixed-food meal is a tool to estimate effects on glucoregulation.
- Glycaemic response to wholemeal bread is slower compared to white bread.
- Viscous β -glucans in a meal might reduce postprandial blood glucose.
- Glycaemic index shows high inter-volunteer variability.
- Breakfast defined by state authorities affects schoolchildren nutritional behaviour.

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ABSTRACT

Recent interest has focused on the use of high-fibre foods as potential ingredients for lowering the glycaemic index of most carbohydrate-based meals. The objective of this study was to examine glycaemic responses to two defined breakfasts. The reference breakfast and the test breakfast differed only in the type of bread (white vs. wholemeal), and were administered three times to volunteers (2 men, 8 women). Capillary blood glucose was monitored in the fasting state 15, 30, 45, 60, 75, 90, 105 and 120 min after breakfast consumption. The incremental areas under the glucose response curves were calculated for each type of breakfast and compared with those of glucose to determine the glycaemic indices. The glycaemic index of the reference breakfast was 26.6 (\pm 6.2), with that of the test breakfast significantly lower at 18.1 (\pm 6.0). The mixed-food test breakfast with wholemeal bread was rich in soluble fibre (β -glucans) for a lower glycaemic load than the white bread, and provided a lower glycaemic response in the volunteers. The inter-volunteer variability in glycaemic index was large, with some showing lower responses to the test breakfast (glycaemic index, 12.4–21.1) and some showing high responses to the test breakfast (glycaemic index, 25.9–27.4).

1. Introduction

The glycaemic index is a measure of the effect that foods have on blood glucose [18]. Factors recognized to raise the glycaemic index include high glucose or starch content of food, and low concentrations of food components that will inhibit digestion or absorption. Conversely, foods that have lower glycaemic index are often rich in fructose, galactose, fibre (e.g., viscous fibre, fructan type of fermentable fibre), fat and/or protein glucose [18]. The study of the effects of diets with high glycaemic indices and glycaemic loads on postprandial blood glucose is of interest mainly due to the effects they can have on the health of the consumer, and particularly on the incidence of diabetes mellitus and the cancer risk, through effects on insulin-like growth

factor [4,18,24,30].

Some parts of meals have high levels of carbohydrates (e.g., pasta, bread, honey), and so the determination of the glycaemic index of meals is an important criterion when evaluating the nutritional and physiological advantages of mixed-food meals [22,23]. Indeed, a recent initiative of the Ministry of Agriculture, Forestry and Food, the Ministry of Education, Science and Sport, and the Ministry of Health defined a Slovene breakfast for schoolchildren [25]. Once a year, usually in November, the kids get this breakfast in educational institutions, and consist of bread, butter, honey, milk and apples. The main parts of this breakfast include carbohydrate foods, such as bread and honey. Therefore, from a nutritional point of view, it would be desirable for this meal to contain more dietary fibre, as foods formulated with

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soluble dietary fibre have been shown to lower serum cholesterol levels and postprandial blood glucose and insulin responses [19]. A shortage of foods with low glycaemic index in mixed-food meals also has disadvantages, and many common starchy staple foods, such as bread products [6,15], while that for white bread is in the range of 59–89 [2].

For this reason, interest has focused on the use of high-fibre foods as potential ingredients for lowering the glycaemic index of most carbohydrate-based meals. Jenkins et al. [16] reported that bread made from 100% wholemeal (wheat) flour produced glycaemic responses that were not significantly different from those for white bread (glycaemic index, 92 vs. 100, respectively). However, it can be expected that by replacing white bread with wholemeal bread, the glycaemic response in terms of sugar release kinetics will be changed, mainly due to the increased solubility of the fibre. The presence of the viscous β -glucans fibre will help to reduce the postprandial rise in blood glucose concentration [17]. Therefore, the glycaemic index in meals with wholemeal bread instead of white bread should be lower or unchanged, and a flattening of the glucose response should be observed.

The aim of the present study was to determine whether additional dietary fibre (i.e., wholemeal bread) in complex meals (i.e., breakfast) contributes to lowering of the glycaemic index, which will be relatively high, mainly due to the inclusion of honey.

2. Materials and methods

2.1. Subject

Ten volunteers (2 men, 8 women; mean age [\pm SD], 41.3 ± 10.5 years, range 25–61 years; mean body mass index [\pm SD], 23.0 ± 3.1 kg/m², range 18.1–27.4 kg/m²) with no symptoms or history of severe gastrointestinal disease or diabetes mellitus were recruited from the staff of the Department of Laboratory Diagnosis of General Hospital Jesenice, and the Department of Food Science and Technology of the Biotechnical Faculty, University of Ljubljana, Slovenia. The venous blood baseline parameters were determined for all volunteers, in terms of content of glucose, glycated haemoglobin, cholesterol, triglycerides, high-density lipoprotein, low-density lipoprotein, creatinine, glomerular filtration rate, and hepatograms for direct and total bilirubin, aspartate, alanine transaminase, gamma-glutamyl transferase and alkaline phosphatase. The volunteers were only included if these parameters were in the normal reference range at baseline, to represent normal, healthy subjects. The main exclusion criterion was for baseline glycated haemoglobin not within the normal reference range.

Data collection was carried out in accordance with the Helsinki Declaration, at the Department of Food Science and Technology, Biotechnical Faculty, University of Ljubljana, Slovenia.

2.2. Study design

The study was carried out according to Goñi et al. [13], with some modifications. The volunteers were examined between 07:00 h and 09:15 h after a 12-h fast. If they were thirsty, they were allowed to drink half of a glass of water (at most) on the morning of the study days. Alcohol intake and intensive physical activity were not permitted the day before the study days. Tobacco use was prohibited from 10 h before the tests and during the tests.

All of the volunteers ingested the different breakfasts at 07:00 h over two consecutive weeks: for the first week, the reference breakfast with white bread (from wheat flour) for three consecutive days; for the second week, the test breakfast with wholemeal bread (from whole grain wheat flour) for three consecutive days. For the third week, they ingested 50 g glucose dissolved in 2 dL water for three consecutive days. The breakfasts had to be ingested within 10 min.

Blood glucose concentrations were measured in capillary whole blood obtained by a finger prick (Accu-Chek Advantage System, Roche

Diagnostics Limited, Lewes, UK), in the fasting state and 15, 30, 45, 60, 75, 90, 105 and 120 min after the consumption of the breakfast or the glucose.

Bread was the only solid component of these breakfasts, and for its analysis, samples were homogenised in a blender (Grindomix 200, Retch, Germany). The other components of the breakfasts (i.e., honey, butter, milk) were used separately in the chemical analyses, which were carried out in duplicate. The following parameters were measured and analysed for each of the breakfast samples: content of water, crude protein, crude fat, carbohydrate, available carbohydrate (that providing carbohydrate for metabolism; 'glycaemic'), crude fibre, β -glucans and ash, with energy values also calculated.

2.3. Breakfasts

The reference breakfast had the following composition: two slices of white bread (50 g), butter (16.7 g), honey (20 g) and milk (2 dL, 3.5% fat). The test breakfast was similar to the reference breakfast, except the white bread was replaced by wholemeal bread (50 g). The volunteers began the meal by eating the slices of bread, which were spread with butter and honey, while drinking the milk.

The white and wholemeal bread (Žito d.o.o., Slovenia), butter (Meggle AG, Germany), wildflower honey (Medex d.o.o., Slovenia), and Alpen milk with 3.5% milk fat (Alpsko mleko, Ljubljanske mlekarne d.o.o., Slovenia) were purchased from a local supermarket.

2.4. Methods

2.4.1. Chemical composition

The water contents of the breakfast components were determined by drying samples in an oven (Kambič) at 105 °C (AOAC 950.46) [1]. The total protein content (crude protein, $N \times 6.25$) was determined by the Kjeldahl method (AOAC 928.08) [1]. The crude fat content was determined by extracting the total lipids using hot petroleum ether as solvent (AOAC 935.38, Fat (crude) in bread) [1]. The ash content was determined by mineralisation of the samples at 550 °C (AOAC 920.153) [1]. The crude (dietary) fibre content was determined by AOAC method 985.29 (Total Dietary Fibre in Foods) [1], and β -glucans by AOAC method 995.16 (β -D-Glucans in barley and oats) [1]. Total carbohydrate was calculated from the difference, as $[100 - (\text{crude protein} + \text{crude fat} + \text{ash} + \text{crude fibre})]$ [14]. To calculate the available carbohydrate by difference, the amount of dietary fibre was analysed and subtracted from the total carbohydrate [7]. Data from the chemical analyses are expressed on a wet matter basis.

2.4.2. Estimated glycaemic index

The incremental areas under the blood glucose response curves (IAUCs) were calculated geometrically using the trapezoid rule, ignoring the area below the fasting baseline. For each experimental group of breakfasts, the IAUCs are expressed as percentages of the mean IAUC of the iso-carbohydrate reference food glucose (tested three times), as consumed by the same volunteer [32]. The glycaemic index of each meal was then calculated as the mean value across all of the volunteers who consumed that meal.

2.4.3. Glycaemic indices of the meals

The meal glycaemic indices were determined by calculating the weighted glycaemic indices of the components of each meal [11]. The glycaemic indices for these components were obtained from the international tables of glycaemic index and glycaemic load [2]. Each food had a number of glycaemic indices indicated, and the selection was made hierarchically in order of preference, from the mean value, and otherwise from the closest match. Equation (1) was used for the calculation of the meal glycaemic index (GI) from the individual food items:

Meal

$$GI = \frac{\Sigma(GI_{\text{bread}} \times \text{availCHO}_{\text{bread}} + GI_{\text{honey}} \times \text{availCHO}_{\text{honey}} + GI_{\text{milk}} \times \text{availCHO}_{\text{milk}})}{\text{total avail CHO}} \quad (1)$$

where avail CHO is the available carbohydrate of the given source (in grammes). The formula to calculate the meal glycaemic load (GL) was:

$$\text{Meal GL} = (GI \times \text{avail CHO}_{\text{meal}}) / 100 \quad (2)$$

2.4.4. Data analysis

The experimental data were evaluated statistically using the IBM SPSS Statistics 20 programme. The data were tested for normal distribution by Shapiro-Wilk tests. The differences in glycaemic indices between the reference and test breakfasts were tested using paired *t*-tests, with equal variance of the breakfasts assumed (Levene's test, $P = 0.316$), and for the determination whether there were any statistically significant differences in glycaemic indices between volunteers one-way ANOVA was used. On the basis of the post hoc test (Least Significant Difference) used on glycaemic indices for the individual volunteers 120 min after test breakfast consumption two groups of volunteers was formed, those with the highest values of GI and all others. The effects on the glycaemic index were evaluated according to breakfast type (reference vs. test breakfast; white vs. wholemeal bread), metabolic type of the volunteer (high: glycaemic index, 25.9–27.4; low: glycaemic index, 12.4–21.1), breakfast \times metabolic type interaction, and repetition/volunteer (1–10), using the general linear model procedure. Means were calculated for the experimental groups using the least-squares means procedure, and were compared at the 5% probability level.

3. Results and discussion

3.1. Breakfasts

The nutritional information for the reference and test breakfasts is given in Table 1. Compared to the reference breakfast, the test breakfast had an 11% lower carbohydrate content (available CHO, 9% lower), 2.7-times higher dietary fibre content, almost the same crude fat content, and 14% higher crude protein content. Among the nutrients contained in the food, carbohydrate has the strongest activity for raising postprandial blood glucose levels, and its amount, rather than its type, was directly associated with postprandial blood glucose levels [20]. Furthermore, the available carbohydrate, as that providing carbohydrate for metabolism (i.e., 'glycaemic'), and the crude fibre, in particular, are of greatest interest in terms of blood glucose levels. The available carbohydrate is defined as 'starch and soluble sugars', and the 'unavailable' carbohydrate (i.e., non-glycaemic) as 'mainly hemicellulose and fibre (cellulose)' [8,11].

Table 1

Nutritional composition of the reference and test breakfast.

Breakfast type	Component	Constituent [g kg ⁻¹]							Ash [g kg ⁻¹]	Energy [kcal]	Mass [g]	
		Water	Crude protein	Crude fat	Carbohydrate	Available carbohydrate	Crude dietary fibre	β -Glucans				
Reference	Butter	157.4	7.0	828.1	6.0	6.0	–	–	1.5	738	3086	16.5
	Honey	163.2	3.0	0	832.0	832.0	–	–	1.8	339	1420	20
	Milk	875.7	34.1	35.0	48.2	48.2	–	–	7.0	64	269	200
	White bread	365.4	92.0	41.0	481.8	476.3	5.5	< 0.1	19.8	269	1127	50
Test	Butter	157.4	7.0	828.1	6.0	6.0	–	–	1.5	738	3086	16.5
	Honey	163.2	3.0	0	832.0	832.0	–	–	1.8	339	1420	20
	Milk	875.7	34.1	35.0	48.2	48.2	–	–	7.0	64	269	200
	Wholemeal Bread	392.4	105.0	42.0	438.6	423.5	15.1	3.2	22.0	258	1080	50

- Content not determined.

Table 2

Glycaemic indices for the individual volunteers 120 min after the reference and test breakfast consumption.

Volunteer	Glycaemic index	
	Reference breakfast	Test breakfast
9	22.4 \pm 2.4	27.4 \pm 0.6 ^a
6	27.4 \pm 3.7	25.9 \pm 3.1 ^{ba}
3	24.8 \pm 5.5	21.1 \pm 2.7 ^{bc}
10	24.9 \pm 8.9	20.0 \pm 5.7 ^{bc}
5	27.5 \pm 5.4	18.8 \pm 5.7 ^{cdc}
4	28.3 \pm 11.6	15.8 \pm 2.6 ^{cdc}
8	27.9 \pm 2.9	13.5 \pm 2.6 ^{ed}
2	27.5 \pm 10.1	13.2 \pm 0.7 ^{ed}
7	27.8 \pm 4.8	13.2 \pm 4.4 ^{ed}
1	27.5 \pm 8.7	12.4 \pm 3.6 ^e
<i>P</i> value	0.988	\leq 0.001

Data presented as means \pm standard deviation, in descending order of the test breakfast. Data with different superscript letters within a column differ significantly ($P < 0.05$).

3.2. Calculated glycaemic indices

The glycaemic indices of the meals were calculated from the tabulated glycaemic indices of the individual food items of which each meal was composed. The reference breakfast had a calculated glycaemic index of 41, and test breakfast, of 39.

The measured glycaemic indices of the meals were 35% and 53% lower than the corresponding values calculated from the tables of glycaemic indices of the individual food items. These data are in good agreement with findings of other studies [10,12], with the range of the measured values generally a little lower, from 20% to 50%. It has been suggested that the interactions among the food items within the human gastrointestinal tract often slow the rate of glucose absorption (thereby reducing the measured glycaemic index of a mixed-food meal) [10,27].

3.3. Glycaemic indices of the volunteers

The distribution of the glycaemic indices across the volunteers was analysed using Shapiro-Wilk tests (IBM SPSS Statistics 20) for normal distribution of parameters. These tests showed normal distribution of the data ($n = 60$, $P = 0.315$): median, 23.3; mean, 22.4 (minimum, 8.6; maximum, 38.5; standard error of mean, 0.96; standard deviation, 7.4).

The glycaemic indices determined from the measured postprandial changes in plasma glucose concentrations that followed the consumption of an equal amount of breakfast compared with the changes after the consumption of 50 g of glucose (in both cases, measured during the first 120 min post consumption) were significantly different. Compared to the glycaemic index of the reference breakfast (26.6 \pm 6.2), that for the test breakfast (18.1 \pm 6.0) was significantly reduced for a mean difference of 8.4 ($n = 30$; standard error, 1.7; lower confidence level,

Table 3Evaluation of the effects of the *Breakfast* × *Metabolic type* interaction on the glycaemic indices in capillary blood of the volunteers 120 min after consumption.

Volunteer	Glycaemic	n	Glycaemic index		Significance
metabolic type	index range		Reference breakfast	Test breakfast	breakfast × metabolic type
High	25.9–27.4	2	24.9 ± 2.2 ^a	26.7 ± 2.2 ^a	< 0.001
Low	12.4–21.1	8	27.0 ± 1.1 ^a	16.0 ± 1.1 ^b	

Data are least squares means ± standard error; volunteer metabolic type defined according the test breakfast glycaemic index; n – number of volunteers.

5.0; upper confidence level, 11.9; 2-tailed sigma, $P < 0.001$).

At closer inspection, low glycaemic indices of food items (e.g., butter) produce smaller postprandial increases in plasma glucose concentrations, while high glycaemic indices of food items (e.g., honey, bread) produce larger postprandial increases in plasma glucose concentrations. As can be seen from the data in Table 1, the main differences between the reference and test breakfasts were the content of available carbohydrate, and especially dietary fibre and β -glucans, due to the different breads included, as white or wholemeal. The approximately 50% higher dietary fibre content in the test breakfast in comparison with the reference breakfast will have changed the glycaemic response of these volunteers in terms of the sugar release kinetics, which will mainly be due to the increased solubility of the fibre, as reported in previous studies [13,17,28]. The presence, in particular, of the viscous β -glucans fibre (the main component of soluble fibre) helps to reduce the postprandial rise in blood glucose [9,13,28]. The present data confirm these findings; in comparison to the reference meal β -glucans ($< 0.1 \text{ g kg}^{-1}$), with 3.2 g kg^{-1} β -glucans defined for the test meal with wholemeal bread. Attenuation of postprandial blood glucose levels by β -glucans is associated with their high viscosity in solution at low concentrations, and their molecular weights and concentrations. Consequently, the increase in luminal/intestinal viscosity will impair the rate of starch digestion and the absorption of glucose, due to reduction of pancreatic amylase activity and of the access of the released sugars to the gut wall [13,28,33]. As well as β -glucans, some whole grain or minimally processed grain foods (e.g., steel-cut oats, quinoa, pumpernickel bread) also reduce postprandial glycaemia, although the majority of whole grain foods do not [3]. The main factor that might be responsible for these altered glycaemic responses could go beyond the chemical composition of the foods, as reduced rates of digestion and small intestinal absorption will also delay gastric emptying [27,28] when the wholemeal bread was consumed.

It has been reported that different honeys have varying fructose contents and fructose/glucose ratios [5,21]. Honey with lower glycaemic index, such as for acacia honey (glycaemic index, 32) but with comparatively high concentrations of fructose ($392.0 \pm 31.1 \text{ g kg}^{-1}$), can also lower the postprandial rise in blood glucose. However, in the present study, both experimental groups used wildflower (multiflower) honey ($352.5 \pm 23.4 \text{ g kg}^{-1}$ fructose) that had a medium/high glycaemic index, which would thus have equally affected the glycaemic indices of both breakfasts. According to the international table of glycaemic indices and glycaemic loads [1], wildflower honey has medium (55–69) or high (70+) glycaemic indices [31]. Of note here, the wildflower honey used in these breakfasts could be replaced with acacia, spruce or chestnut honeys, as these would further reduce the glycaemic indices.

Straightforward comparisons of the data in the present study with the literature are not easy. Ohlsson et al. [26] examined different recommended diabetes diets with equivalent energy contents. Their reference breakfast was composed of low-fat flavoured yoghurt, one slice of rye bread, butter spread, hard cheese, one glass of orange juice, and one cup of either coffee or tea. This breakfast and the reference breakfast in the present study had similar glycaemic loads, as 34 versus 36. It is interesting that replacement of white bread with wholemeal bread lowered the breakfast glycaemic load here from 36 to 24. When the glycaemic loads were obtained from the calculated glycaemic index,

these were 56 and 51 for the reference and test breakfasts, respectively.

3.4. Variability in glycaemic indices between volunteers

There was relatively high coefficient of variability for the glycaemic indices in the present study (35%). Table 2 shows glycaemic indices for the individual volunteers 120 min after the reference and test breakfast consumption; significant differences between volunteers were seen after test breakfast consumption.

Glycaemic index data can be divided into the assessment of two metabolic profiles of volunteers, one with lower responses to the test meal (glycaemic index, 12.4–21.1; 8 volunteers) and the remaining volunteers with high responses (glycaemic index, 25.9–27.4; 2 volunteers; in Table 3 marked with superscript letter 'a'). The reasons for these post-consumption changes in plasma glucose concentrations in these volunteers could be the consequence of several physiological processes, such as carbohydrate hydrolysis and absorption in the small intestine, and tissue glucose uptake. These processes are affected by the inherent chemical characteristics of the food items as well as inter-volunteer variability in the alimentary canal, gastrointestinal and pancreatic efficiencies [9,12].

Although for this experiment consumption of the test breakfast reduced the glucose responses from the reference breakfast, the problem of inter-volunteer variability remains. Table 2 shows that for two of the volunteers, the postprandial blood glucose response was not affected by the change in the glycaemic indices of the meals, nor by the ingested carbohydrate levels, where the available carbohydrate and energy in the test and reference breakfasts were comparable (carbohydrate: 168.6 vs. 176.2 g kg^{-1} ; energy: 1580 vs. 1560 kcal kg^{-1}). The postprandial blood glucose responses of the volunteers after consumption of the test and reference breakfasts varied a little, with increases in some cases, and decreases in others. Fig. 1 illustrates these changes in capillary blood glucose responses in volunteers, divided according the test breakfast glycaemic index into groups with high and low glycaemic index range.

The postprandial capillary blood glucose levels were higher after the reference breakfast than for the test breakfast, and the inclusion of the wholemeal bread flatten the sharp capillary blood glucose peak (slow down glycaemic response) observed 30–60 min after the white bread consumption (Fig. 1b). This helped to maintain moderate level of capillary blood glucose until 120 min after consumption. A low glycaemic index (16.0 ± 6.4) was obtained here.

4. Conclusions

There is the need to improve the amount and quality of information on glycaemic indices that are made available to the general public and to health professionals, as has been expressed on different occasions [21,29]. Furthermore, previous studies have indicated that glycaemic indices given in the international tables are often not good predictors of the equivalent measured glycaemic indices, for various reasons, including for the differences between countries for methodologies and cooking methods [29,31], with the remaining need for local information.

The present study contributes to clarification of the regulation of glucose intake from individual foods when consumed in the context of mixed meals. Here, substitution of foods in a meal with high glycaemic

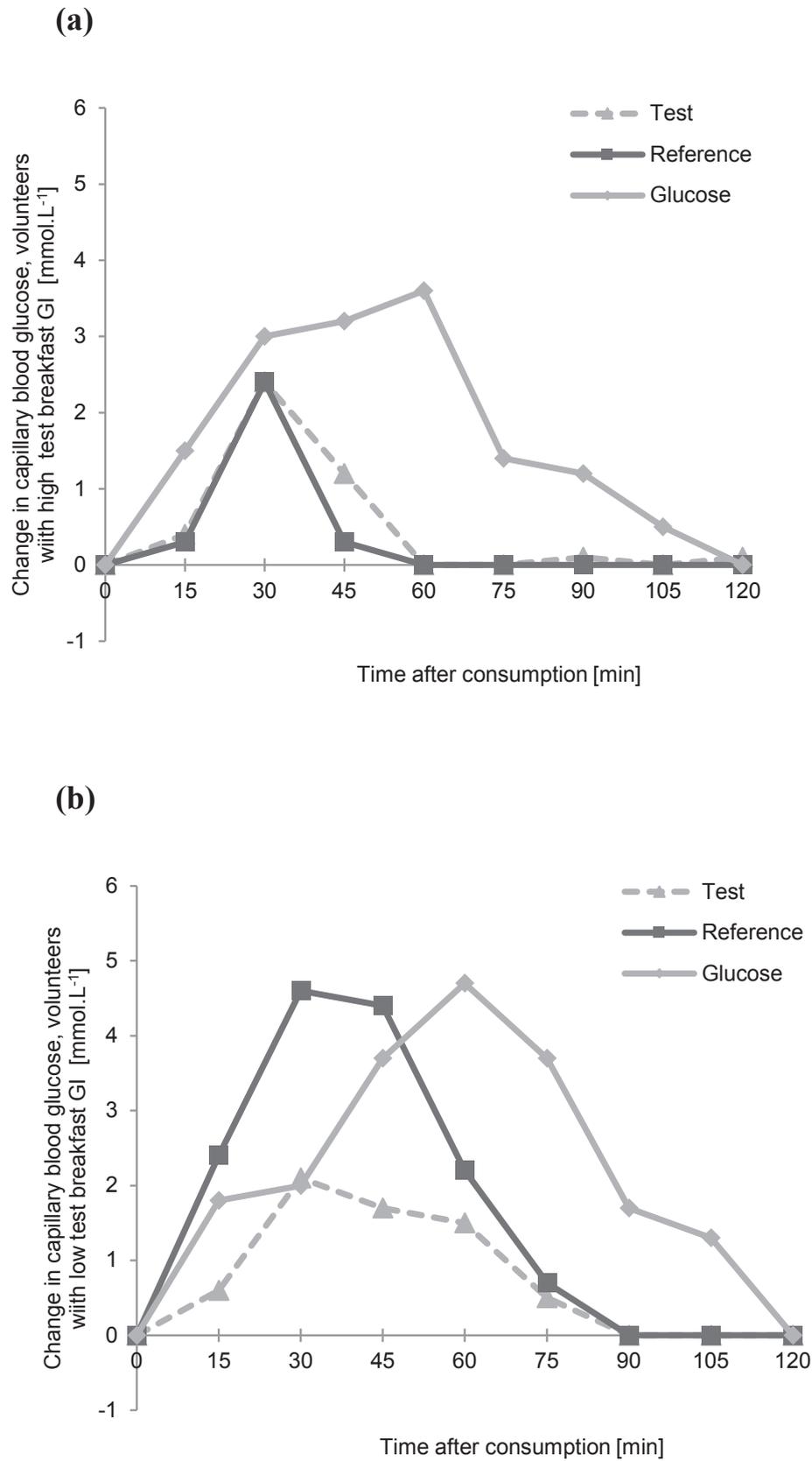


Fig. 1. Changes over 120 min for the capillary blood glucose responses (normalised to baseline) in two volunteers, divided according the test breakfast glycaemic index into volunteer with high (a) and volunteer with low glycaemic index range (b). Available carbohydrate consumed: reference breakfast, 47.6 g; test breakfast, 42.3; and glucose, 50 g.

indices for those with low glycaemic indices within the same category (e.g., as here, from white bread to wholemeal bread) shows significant contributions to reduced glycaemic indices of the meal, even with the inclusion of high glycaemic index components in the meals (e.g., as here, honey). Alternatively, inclusion of foods with higher soluble fibre in mixed-food meals will reduce the capacity of this food to increase glycaemia levels after meals. In particular here, the glycaemic index of breakfasts for schoolchildren might have long-term relevance in the diet of children and adolescents as nutritional behaviours are shaped during childhood and adolescence. We have shown here that the selection of carbohydrates from sources that are rich in dietary fibre (e.g., as here, from white bread to wholemeal bread) can indeed confer benefits for overall nutrient adequacy. Generally, efforts to reduce the dietary glycaemic index in populations should best be targeted to energy-dense starchy foods, such as sugar as glucose, and whole grain products with high glycaemic indices, as these provide considerable contributions to the total dietary glycaemic load.

Conflicts of interest

The authors declare that they do not have any conflict of interest.

Ethical statements

This study was approved by the Medical Ethics Committee of the General Hospital Jesenice (N° 0307–121/2018:2). Written informed consent was obtained from all study participants.

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