

# GLYCEMIC AND INSULINEMIC RESPONSES TO SIX CEREAL PRODUCTS IN HEALTHY ADULTS

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## SUMMARY:

*Glycemic index (GI) and insulin index (II) scores of six cereal-based foods (biscuits and shortbreads) were determined and the relationship between the glycemic and insulin responses of the foods was examined. The study was conducted using recommended by FAO/WHO methodology using glucose as a standard food. We obtained a range of GI scores from 49 to 68% and II scores from 51 to 75%. The test foods' blood insulin responses were in parallel to their glycemic responses although in every case the level of II was higher than the level of GI. The foods' II scores were closely related to their GI scores ( $r=0.80$ ).*

**Key words:** glycemic index, insulin index, biscuits, shortbreads

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## INTRODUCTION

The glycemic index (GI) is a ranking of foods according to the extent to which they raise blood sugar levels after eating. Foods with a high GI are those which are rapidly digested and absorbed and result in marked fluctuations in blood sugar levels. Starchy high-fiber low-GI foods, by virtue of their slow digestion and absorption, produce gradual rises in blood sugar and insulin levels and have proven benefits for health. Recent studies indicate that the risks of chronic degenerative diseases such as type 2 diabetes, obesity, coronary heart disease and colon cancer are strongly related to the GI of the overall diet (1–13). The World Health Organization (WHO) and Food and Agriculture Organization (FAO) recently recommended that people in industrialized countries should base their diets on low-GI foods in order to prevent the most common diseases of affluence, such as coronary heart disease, diabetes and obesity (14). The usefulness of the idea of glycemic index is still under discussion, but many facts support the thesis, that the glycemic index value should be taken into account in rational food choice (15–18)

Terms such as complex carbohydrates and sugars, which commonly appear on food labels, are now recognized as having little nutritional or physiological significance. The WHO/FAO recommend that these terms be removed and replaced with the total carbohydrate content of the food and its GI value (14).

Glycemic index of over 500 high-carbohydrate foods was measured so far based on over 80 studies and international tables of GI were published (19).

Cakes and shortbreads are high carbohydrate products of cereal origin. They are widely consumed in many countries, also in the countries of Central and Eastern Europe. There is a variety of products from that group of food products on the market. Up to now only in a few studies glycemic index of cakes and shortbreads was measured.

The aim of the study was to determine the glycemic index

scores of six cereal-based foods. Insulinic response and insulin index was also measured to examine the relationship between the glycemic and insulin responses of the foods.

## MATERIAL AND METHODS

### Subjects

Twelve healthy, non-smoking subjects (6 males, 6 females) voluntarily participated in this study. The mean  $\pm$  SD age of the subjects was  $23.75 \pm 7.7$  years (range: 18 – 38 years), and the mean  $\pm$  SD body mass index value was  $22.5 \pm 1.8$  kg/m<sup>2</sup> (range: 19.0 – 24.6 kg/m<sup>2</sup>). The subjects were recruited from the staff and student population of National Food and Nutrition Institute and District Hospital in Warsaw. All the subjects were characterised by normal blood and urine biochemical values, normal blood pressure and heart rate and normal dietary and physical activity habits. No any chronic disease was diagnosed and treated.

The approval of Bioethics Committee of Institute of Food and Nutrition was obtained. Each person signed an approval for including into the study.

### Test Foods

Six foods were tested in this study (biscuits and shortbreads) and each was identified by a separate code. The investigators did not know the ingredients or recipes for the products. The macronutrient contents of the test products are listed in Table 1. The test portions contained 50 g of digestible carbohydrate.

Products A, B, D and E were standard commercially available products and were fit for human consumption. While products C and F were currently being developed for commercial production their main components such as flour, sugar, cacao, fruits and vegetable fat were safe for human consumption. Food technologies used for these products were those commonly used in biscuit manufacturing.

**Table 1.** The nutritional composition of the test products per 100 g and the portion sizes required for providing 50 g of available carbohydrate

Products (Identification code)	A (R9P70)	B (R9P103)	C (R9P102)	D (R9P68)	E (R9P69)	F (R9P92)
Moisture (g/100g)	1.1	2.7	3.6	4.1	3.4	4.4
Protein (g/100g)	7.1	6.5	6.5	6.3	6.5	7.7
Fat (g/100g)	11.7	18.1	16.7	16.8	18.2	14.1
Total carbohydrate (g/100g)	75.7	65.7	63.5	62.3	63.1	64.7
Test portion size (g) providing 50 g of available carbohydrate	62.0	72.2	74.4	75.7	74.6	72.4

## METHODS

This study was conducted using internationally recognised GI methodology. Short-term (2 hours) postprandial glycemic and insulinemic effects of six cereal-based foods was measured, in relation to the effects produced by pure glucose sugar (the reference food). The reference food was tested on three separate occasions, and the six cereal-based foods were each tested once only. The interval between individual measurements was stated as at least two days. Each measurement started early in the morning after nocturnal fasting period. The reference food and the six test foods were fed to the subjects in equal-carbohydrate portions containing 50 g of available carbohydrate with 250 ml of Evian mineral water. Each subject consumed glucose on the first, fifth and ninth test sessions (first, middle and last) and the six test foods were presented to the subjects in a random, counter-balanced order, in between the reference tests.

Plasma glucose and insulin concentrations were determined in venous (not arterialized) blood.

Plasma glucose concentrations were measured in duplicate using a Dimension®RXL automatic spectrophotometric centrifugal analyzer (Dade Behring, Marburg, Germany) employing the glucose hexokinase /glucose-6-phosphate dehydrogenase enzymatic assay.

Plasma insulin concentrations were measured in duplicate and the insulin assays were performed at a local hospital laboratory, which is authorized to conduct biochemical hormone assays requiring radioactive materials. INS-RIA-PROP kit containing polyclonal anti-insulin antibodies (produced by Isotope Centre POLATOM in Swierk) was used. It is a radioimmunologic method RIA (marked <sup>125</sup>I pork insulin). Free hormone is separated from the hormone-antibody complex by solid phase method. Polyester tubes (produced by firm CIS) coated by polyclonal anti-insulin antibody were employed.

### Calculating Glycemic Index (GI) and Insulin index (II) Scores

All of the subjects' relevant characteristics and biochemical results were tabulated on data spreadsheets using Microsoft Excel 1998 software.

The calculation of GI and II scores was determined using approved by FAO/WHO protocol (14).

The average value of the two plasma glucose levels (duplicate measures were performed on each blood sample) obtained for each blood sample was used as the final plasma glucose value for the calculation of GI scores. Similarly, the average value of

the two plasma insulin levels recorded for each blood sample was used for the calculation of the II scores. For each subject, the incremental area under the two-hour plasma glucose response curve (AUC) for each test food and reference food was calculated using the trapezoidal rule and truncated at the baseline value (zero). For both GI and II scores, the baseline value was the average concentration of the – 5 min and 0 min fasting blood samples. AUC values allow the comparison of the integrated effects of the test foods over a fixed time period. Any negative area under a response curve was ignored. For each subject, a GI score was calculated for each food by dividing the plasma glucose AUC value for this test food by the average plasma glucose AUC value for the reference food and multiplying by 100 to obtain a percentage score.

II scores were calculated using the same equation as above, substituting insulin AUC values for the glucose AUC values.

### Statistical Analyses

Statistical analyses were performed using Statistica 5 software. The descriptive statistics included the coefficient of variation (CV), which was calculated by dividing the standard deviation (SD) by the mean and multiplied by 100 to obtain a percentage value [ $CV = (SD/mean) \times 100\%$ ].

For normally distributed data, repeated-measures analysis of variance (ANOVA) was used to determine whether any significant test food effects existed and the Fisher PLSD repeated measures test was used as a post-hoc test to identify the significant differences. If some data were not normally distributed, the Friedman test was intended to use as an alternative to two-way analysis of variance.

Linear regression analysis was used to examine the associations between postprandial glucose and insulin responses and the nutrient composition of the test foods.

## RESULTS

### Blood Glucose Responses to The test Foods

For one subject the AUC value for standard food (glucose) was extremely low. That was the reason that the results of GI for that subject were very dispersed [from 38 to 187(!)]. We excluded results obtained by this subject from further analysis. So we calculated the results only of 11 remaining subjects.

The average plasma glucose concentrations for each test food are listed in Table 2. The average two-hour incremental plasma glucose response curves for the test foods are shown in Fig. 1,

**Table 2.** The mean  $\pm$  SEM (Standard Error of the Mean) absolute plasma glucose concentrations for the seven blood samples (mmol/l) collected over two hours for the test foods and the mean incremental areas under the two-hour plasma glucose response curves (AUC) ( $n = 11$ ). The results for the glucose (reference food) are the mean values from three separate repeated tests for this food for each subject

Time (min)	Glucose reference food	Product A	Product B	Product C	Product D	Product E	Product F
0	4.17 $\pm$ 0.08	3.87 $\pm$ 0.11	3.94 $\pm$ 0.09	4.04 $\pm$ 0.14	4.08 $\pm$ 0.13	4.03 $\pm$ 0.12	4.12 $\pm$ 0.09
15	5.32 $\pm$ 0.11	4.19 $\pm$ 0.12	4.32 $\pm$ 0.21	4.17 $\pm$ 0.16	4.60 $\pm$ 0.17	4.32 $\pm$ 0.18	4.58 $\pm$ 0.17
30	6.78 $\pm$ 0.31	5.50 $\pm$ 0.19	5.45 $\pm$ 0.25	5.72 $\pm$ 0.22	5.62 $\pm$ 0.30	5.55 $\pm$ 0.25	5.67 $\pm$ 0.20
45	6.72 $\pm$ 0.47	5.54 $\pm$ 0.28	5.07 $\pm$ 0.35	5.91 $\pm$ 0.34	5.67 $\pm$ 0.32	5.16 $\pm$ 0.35	5.36 $\pm$ 0.38
60	6.03 $\pm$ 0.49	4.71 $\pm$ 0.23	4.43 $\pm$ 0.28	4.92 $\pm$ 0.31	4.78 $\pm$ 0.36	4.69 $\pm$ 0.32	4.73 $\pm$ 0.25
90	4.64 $\pm$ 0.35	4.02 $\pm$ 0.22	3.98 $\pm$ 0.20	4.34 $\pm$ 0.22	4.26 $\pm$ 0.23	4.03 $\pm$ 0.26	3.95 $\pm$ 0.15
120	3.67 $\pm$ 0.27	4.32 $\pm$ 0.15	4.43 $\pm$ 0.19	4.42 $\pm$ 0.19	4.35 $\pm$ 0.19	4.32 $\pm$ 0.18	4.13 $\pm$ 0.10
AUC	159.8 $\pm$ 28.1	95.0 $\pm$ 14.9	72.1 $\pm$ 14.4	98.4 $\pm$ 21.7	88.3 $\pm$ 16.4	77.2 $\pm$ 18.3	67.6 $\pm$ 11.5

illustrated as the change in plasma glucose from the fasting baseline value (i.e. actual plasma glucose concentration minus the fasting plasma glucose concentration).

The glucose sugar (reference food) produced the highest peak plasma glucose level at 30 minutes and the largest overall blood glucose response. Among the six test foods, product B (followed by product F) produced the lowest peak plasma glucose level at 30 and 45 minutes. Plasma glucose levels following the consumption of the glucose sugar were higher than those produced by the six test foods at each time point during the first 90 minutes. The average plasma glucose levels for the glucose sugar and the six test foods decreased between 60 and 120 minutes, although the foods differ in the rate at which plasma glucose declined. The glucose sugar was associated with the most rapid decline in plasma glucose during the last hour.

### Blood Insulin Responses to the Test Foods

Similarly to the results of GI – the value of AUC for standard food (glucose) was very low for one (the same) subject [the results of II of tested products in that case varied from 47 to 208(!)]. We excluded this subject from further calculation of the results.

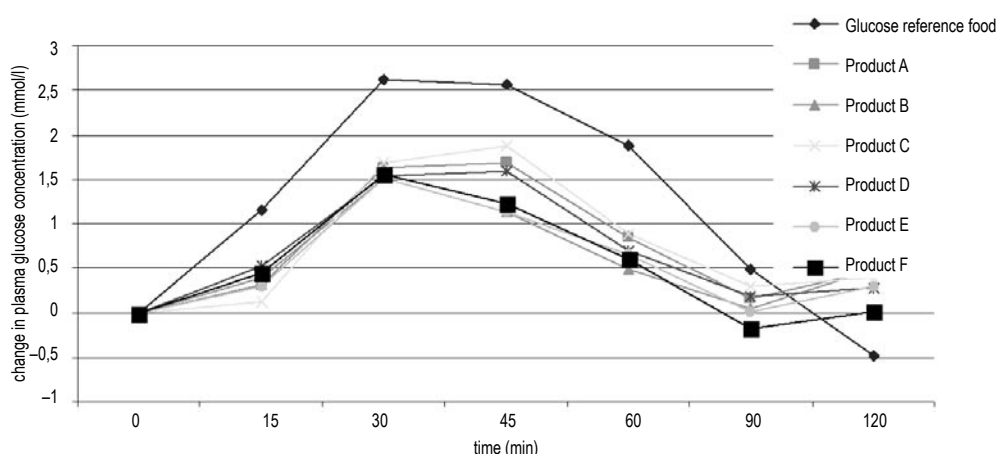
The average plasma insulin concentrations for each test food are listed in Table 3. The average two-hour incremental plasma insulin response curves for the test foods are shown in Fig. 2,

illustrating the change in plasma insulin from the fasting baseline value (i.e. actual plasma insulin concentration minus the fasting plasma insulin concentration).

The glucose sugar (reference food) produced the highest peak plasma insulin level at 45 minutes and the largest overall plasma insulin response. The average insulin responses to the test foods did not follow the same rank order as the plasma glucose responses, as confirmed by the insulin index scores presented in the next section. Among the six test foods, product F produced the lowest peak plasma insulin level at 30 minutes and product B at 45 and 60 minutes. Plasma insulin levels following the consumption of the glucose sugar were greater than those produced by the six test foods at each time point up until 120 minutes. Similar to the plasma glucose responses, plasma insulin responses rose during the first 30 minutes of food consumption and then decreased at varying rates over the rest of the experimental period in the case of product B and E. Rest of the products gave the rise of plasma insulin response during the first 45 minutes. Plasma insulin levels were close to fasting values by the end of the 120-minute period for all of the test foods.

### The test Foods' GI and II Scores

The average plasma glucose and insulin response curves indicated that the test foods varied in their glycemic and insulinemic effects.



**Fig. 1.** The average two-hour plasma glucose response curves depicted as change in plasma glucose concentration from the fasting baseline concentration ( $n = 11$ ).

**Table 3.** The mean  $\pm$  SEM absolute plasma insulin concentrations (pmol/l) for the seven blood samples collected over two hours for the test foods and the mean incremental areas under the two-hour plasma insulin response curves (AUC) ( $n = 11$ ). The results for the glucose (reference food) are the mean values from three repeated tests for this food for each subject

Time (min)	Glucose reference food	Product A	Product B	Product C	Product D	Product E	Product F
0	8.18 $\pm$ 0.89	7.20 $\pm$ 0.92	6.97 $\pm$ 0.37	7.50 $\pm$ 1.04	8.57 $\pm$ 1.20	8.12 $\pm$ 1.13	7.14 $\pm$ 0.97
15	24.53 $\pm$ 2.80	12.50 $\pm$ 1.96	16.94 $\pm$ 3.59	12.60 $\pm$ 1.81	18.79 $\pm$ 387	13.06 $\pm$ 179	15.88 $\pm$ 281
30	50.84 $\pm$ 5.83	40.50 $\pm$ 5.90	39.86 $\pm$ 5.65	45.25 $\pm$ 7.37	42.39 $\pm$ 934	47.30 $\pm$ 847	37.40 $\pm$ 673
45	55.20 $\pm$ 7.12	50.28 $\pm$ 6.28	33.77 $\pm$ 4.43	49.09 $\pm$ 8.93	50.25 $\pm$ 962	46.10 $\pm$ 611	41.42 $\pm$ 537
60	48.52 $\pm$ 7.55	34.11 $\pm$ 7.85	22.22 $\pm$ 3.86	33.73 $\pm$ 6.75	33.33 $\pm$ 598	32.04 $\pm$ 568	29.05 $\pm$ 440
90	29.22 $\pm$ 7.39	22.15 $\pm$ 4.93	12.91 $\pm$ 2.15	18.59 $\pm$ 5.28	17.45 $\pm$ 453	17.12 $\pm$ 441	14.35 $\pm$ 323
120	13.13 $\pm$ 4.30	14.18 $\pm$ 3.36	10.42 $\pm$ 1.82	12.92 $\pm$ 2.73	13.11 $\pm$ 293	11.81 $\pm$ 319	10.15 $\pm$ 202
AUC	3212.4 $\pm$ 529.0	2390.0 $\pm$ 474.1	1620.4 $\pm$ 287.5	2274.2 $\pm$ 464.5	2192.7 $\pm$ 455.1	2096.3 $\pm$ 376.0	1859.5 $\pm$ 314.0

The extent of the variation among the test foods' total two-hour blood glucose and insulin responses is more clearly reflected by their GI and II scores. The mean  $\pm$  SEM GI scores for the test foods and the range, median value and coefficient of variation (CV) of the GI scores for each product are listed in Table 4. The mean  $\pm$  SEM II scores for the test foods and the range, median value and coefficient of variation (CV) of the II scores for each product are listed in Table 5. The mean GI and II scores for the test foods are illustrated in Fig. 3.

#### Significant Differences among the Foods' GI and II Scores

Repeated-measures analysis of variance (ANOVA) indicated that significant test food effects existed for both the GI and II scores. Both the GI and II data were normally distributed. Therefore, the Fisher PLSD test for repeated measures was used as a post-hoc test to identify the significant differences among the test foods' GI and II scores.

*Significant differences among the mean GI scores:* The mean GI score of the glucose sugar (reference food) was significantly greater than the mean GI scores of all of the six test foods ( $p < 0.001$

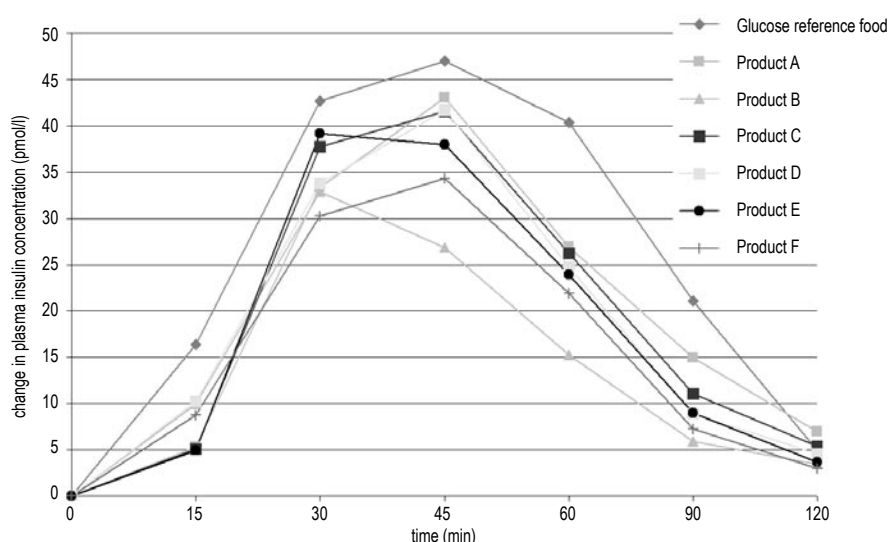
in case of products B-F and  $p < 0.01$  in case of product A). No significant differences were found between the tested products.

*Significant differences among the mean II scores:* The mean II score of the glucose sugar (reference food) was significantly greater than the mean II scores of all of the 6 test foods ( $p < 0.001$  in case of products B-F and  $p < 0.01$  in case of product A). No significant differences were found between the tested products.

*The relationship between the GI and II scores:* As expected, the mean GI and II scores for the six test foods were significantly associated ( $r = 0.80$ ,  $n = 6$ ,  $p < 0.05$ ) (Fig. 4).

## DISCUSSION

The term of glycemic index was established by David Jenkins in 1981 (20). He began to investigate a battery of foods to healthy individuals and ranking the foods' glycemic indexes on a 100-point scale, where 100 was pure glucose. The higher the GI values, the more rapidly carbohydrates turn into blood sugar.



**Fig. 2.** The average two-hour plasma insulin response curves depicted as the change in plasma insulin concentration from the fasting baseline concentration ( $n = 11$ ).

**Table 4.** The mean  $\pm$  SEM GI scores and the range, median value and CV of the GI scores for the test foods ( $n = 11$ )

Food	GI score (%)	Minimum score (%)	Maximum score (%)	Median score (%)	CV (%)
Product A	67.5 $\pm$ 10.9	25.1	138.2	52.0	53.6
Product B	49.3 $\pm$ 7.5	5.6	87.3	43.1	50.3
Product C	56.9 $\pm$ 8.4	10.6	110.6	60.4	48.9
Product D	57.0 $\pm$ 5.9	16.6	85.5	56.7	34.5
Product E	51.0 $\pm$ 9.3	7.7	116.6	47.0	60.3
Product F	48.9 $\pm$ 6.4	13.9	88.5	49.8	43.7
Glucose	100 $\pm$ 0	100.0	100.0	100.0	0.0

**Table 5.** The mean  $\pm$  SEM II scores and the range, median value and CV of the II scores for the test foods ( $n = 11$ )

Food	II score (%)	Minimum score (%)	Maximum score (%)	Median score (%)	CV (%)
Product A	75.3 $\pm$ 10.3	30.0	143.9	62.7	47.0
Product B	50.6 $\pm$ 6.2	23.7	94.2	48.5	40.8
Product C	68.5 $\pm$ 6.1	39.5	107.0	69.0	29.4
Product D	65.5 $\pm$ 6.9	34.1	96.0	64.4	34.9
Product E	66.9 $\pm$ 8.0	35.3	124.6	61.3	39.8
Product F	61.5 $\pm$ 5.3	35.4	84.8	61.4	28.4
Glucose	100 $\pm$ 0	100.0	100.0	100.0	0.0

Due to differences in body weight and metabolism, blood glucose and insulin responses to the same food can vary between different people. The use of the reference food to calculate GI and II scores reduces the effect of the natural differences between

the subjects, so the GI and II scores for the same food vary less between the subjects than their AUC values for this food.

During the 80-es and 90-es the procedure of measurement GI was defined precisely (21, 22). In 1997 FAO and WHO elaborated the standard of measuring GI (14). Some crucial points of standard procedure are:

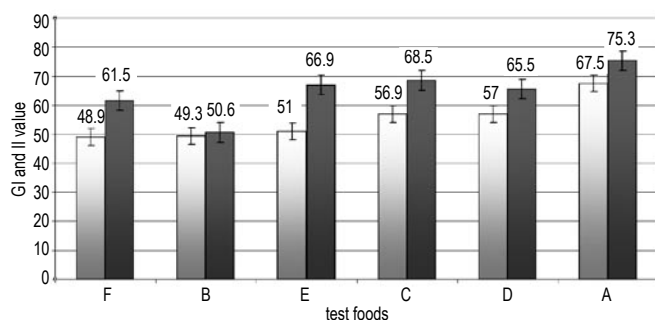
- measuring 50 g of available (except fiber) carbohydrates,
- using standard food to compare the effect (50 g of glucose or white bread is used – GI of white bread is 70 in comparison to glucose),
- repeating three times measurement of standard food effect,
- using the same subjects (usually 10 – 12 persons) for all tested products,
- calculation incremental area under the curve during 2 hours testing.

GI do not relay to the number of calories or portion size.

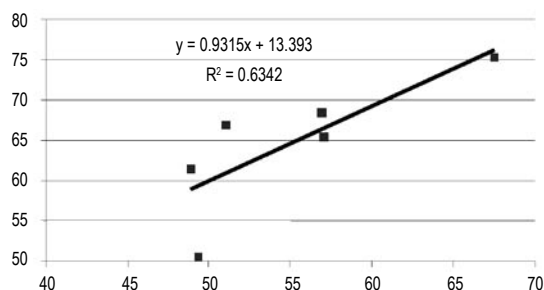
Meals containing low GI foods reduce both postprandial blood glucose and insulin responses. Diets based on low GI carbohydrates improve serum cholesterol and triacylglycerol levels and the ratio of LDL do HDL (2, 4, 23). Animal studies suggest that low GI diet delays the onset of insulin resistance. Many epidemiological studies indicate that low GI diet is associated with reduced risk of developing non-insulin diabetes, ischemic heart disease, obesity and colon cancer (2, 3, 6, 8, 9, 12, 17, 18). In patients with diabetes low GI diet improves the control of glycemia (5,7,10).

Foods are ranked as low GI when their glycemic index is below 50, medium GI when it is between 50 and 70, and high GI when it exceeds 70.

Many factors are contributing to the value of GI. Among them we should mention the nature of monosaccharide components, nature of the starch, non-carbohydrate food components



**Fig. 3.** The average GI and II scores for the test foods (mean  $\pm$  SEM,  $n = 11$ ).



**Fig. 4.** The relationship between the mean GI and II scores ( $r = 0.80$ ,  $n = 6$ ,  $p < 0.05$ ).

(for example dietary fiber) and cooking or food processing (for example food form, cellular structure, degree of starch gelatinization) (24, 25, 26). Foods particularly sensitive to processing include potatoes, rice and bananas. Important factor is also particle size. The GI of wheat, maize and oats increases from whole grains (lowest GI), cracked grains, coarse flour to fine flour (highest GI) (27).

Lately, the studies of insulin response to the carbohydrate foods measured by insulin index (II), were started. The protocol of II testing is adequate to the GI measurement (14, 28). Usually, a high correlation between GI and II score was found. Although bakery products, which are high in fat and refined carbohydrates, protein-rich foods and fermented milk products give disproportional higher insulin responses than glycemic responses (29). It is suspected that non carbohydrate components of those products (proteins or organic acids) are responsible for that effect (23, 29). The degree of the gelatinisation should also be taken into account (25, 30).

The aim of the present study was to determine GI and II of 6 bakery foods (biscuits and shortbreads). We obtained a range of GI scores (49–68%), covering the range of low and intermediate GI foods. The test foods' blood insulin responses were in parallel to their glycemic responses although in every case the level of II was higher than the level of GI. Therefore, the foods' II scores were closely related to their GI scores ( $r=0.80$ ), although the correlation was not perfect. In another study measuring glycemic index of several processed cereal foods similar results were obtained (30).

Up to now, the GI of several high-carbohydrate foods have never been tested (for example celery or tomatoes). But it seems that the idea of glycemic index is still emerging, considering the increasing number of studies in this area year by year. FAO and WHO recommends that the "glycemic index should be used to compare foods of similar composition within food groups" and "both glycemic index and food composition must be considered" when choosing carbohydrate-containing foods (14). In some countries even now GI score is put on the label of the product. The new dietary guidelines recommend the consumption of low glycemic foods (31). So the concept of glycemic index foster much interest among the scientists, physicians, dietitians, food industry and last, but not least, among all the population of consumers.

## CONCLUSIONS

- The six cereal products tested produced a range of blood glucose responses and a range of GI scores (49 – 68%), covering the range of low and intermediate GI foods.
- The test foods' blood insulin responses were in parallel to their glycemic responses although in every case the level of II was higher than the level of GI. Therefore, the foods' II scores were closely related to their GI scores, although the correlation was not perfect.
- The glycemic index concept is an emerging issue and should be further intensively investigated.

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