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Glycemic and Insulinemic Responses of Multi-Grain Types of Bread Marketed in Jordan

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Abstract

Introduction: Bread is the staple food in Jordan and there are plenty of bread types advertised there as healthy and multi-grain breads. These breads have diverse health and nutritional effects. This research aimed at studying the glycemic and insulinemic responses of 6 types of multi-grain breads marketed in Jordan, in addition to the local white bread, and verifying the nutrient and health claims of these breads.

Methods: The glycemic and insulinemic responses were obtained in 10 participants (5 males and 5 females) according to the standard method. Six types of test breads (TB1 – TB6) were tested against white bread (WB) as a reference.

Results: Among all the 6 tested breads and the local white bread, the lowest GI bread (TB6) has the lowest energy density and percentages of carbohydrate and fat, while having the highest percentages of protein, ash, fiber, and moisture. On the other hand, the highest GI bread (TB5) has only the lowest moisture percentage and the highest carbohydrate percentage. **Conclusion:** In this study, 3 types of the tested bread were of low GI and the other 3 types were of medium GI. The insulin response was found to parallel the glucose response for all studied breads. The peak of insulin and glucose responses were at 30-min.



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Keywords

Bread; Glycemic; Index; Insulin; Jordan; Multi-Grain; Response.

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Introduction

Carbohydrates, the major part of the diet, are categorized according to the chemical composition into simple sugars and complex carbohydrates. Nevertheless, carbohydrate effects on health are better classified according to postprandial insulin secretion and glycemia.^{1,2}

The glycemic index (GI) concept has a standard definition which was proposed in 1980s by Jenkins *et al.*,^{3,4,5} The GI standardizes the glycemic response of carbohydrate-containing foods and accounts for the variability among individuals by taking the average results of not less than 10 persons.⁶

Glucose was the first standard used in the determination of Gl.^{3,7} At present, local white bread is also used as a standard. Bread is preferred as the standard instead of glucose because bread follows the physiological digestion and absorption in the human gastrointestinal tract as other foods,⁸ whereas glucose has a high osmolarity^{3,9} and can cause nausea in some individuals. Thus, the Gl values can be determined with either glucose or local white bread and interconverted with a ratio of 1.4.¹⁰

The glycemic response and insulinemic response to foods are dependent on many factors; including the sugar type, arrangements of molecular, starch granule size, other components of the food such as fiber, fat, moisture, protein, amylose to amylopectin ratio in addition to external determinants as processing methods and the total food consumed. From the functional point of view, carbohydrate is classified regarding its postprandial glycemic effect, which depends principally on its intestinal digestion and absorption.¹¹

The insulin response to foods also depends on food composition but may or may not be parallel the glucose response.¹² Hence, Insulin Index (II) is determined by the same way of GI determination. This index is of significant importance in cases of insulin resistance or insulin-deficiency.¹¹

Low glycemic index or glycemic load (GL) foods help in normalizing the fasting glycemia, improving the concentration of glycated hemoglobin, and enhancing sensitivity of insulin in diabetics and nondiabetics.^{13,14} Low GI foods may decrease blood cholesterol and the risk of having coronary heart disease.^{6,15} In addition, meta-analysis studies support the hypothesis that high glycemic index foods contribute to the development of many diseases.¹⁶

Food GI and GL were not correlated with increased development of risk of overweight/obesity in a cohort of Mediterranean young adults who had low average BMI and ingested plenty of vegetables and fruits. On the other hand, it was found that high white bread consumption was a risk factor for overweight/obesity in that population.¹⁷

In Mediterranean countries, cereals have been used in many ways, nevertheless, they are mainly used in refined forms. In the Arab Middle East countries, wheat bread is the traditional and most commonly consumed bread.^{18,19} In Jordan, wheat and rice are the staple foods. Khobez (pita bread), a leavened flat wheat bread, is consumed daily with most meals.²⁰ In Jordan there is an increasing interest in determining the glycemic effect of local foods. Abu Rajab et al.,²¹ have studied the glycemic and insulinemic responses of different types of Jordanian honey. In the same context, Takruri & Alkurd¹⁹ have studied the glycemic index of a "healthy bread -Biobreadcomposed of whole wheat and rye flours, in addition to linseed and soy powders, wheat bran, fennel, anise and caraway seeds, and dried soaked wheat.

The objectives of this study are:

- Studying the nutrient makeup and health properties of 6 types of multi-grain breads marketed in Jordan and comparing them with the local white bread.
- Verifying the nutrient and health claims of these breads.
- Determining the glycemic and insulinemic responses to these 6 types of bread in comparison with local white bread.

Materials and Methods Subject Selection

After signing a consent to adhere to the experiment, clinically 10 healthy (non-diabetic) adult subjects (5 males and 5 females) were recruited. Subjects on any medication were excluded. Other study

inclusion criteria included: readiness to fast for not less than 10 hours before each of 8 study visits, and readiness to perform 5 venipunctures during a two-hour period, on 8 separate occasions over two weeks. The fasting began at \leq 23:00 hours the night preceding the test date, and the day of testing always started at 09:00 hours. Each participant was as his/ her own control. Vigorous activity was not allowed for a whole day before each study visit. A baseline venipuncture blood glucose measurement was performed immediately prior to bread administration.

Bread Selection

Six types of bread claimed by the manufacturers to be healthy, in addition to the local white bread, were selected for the experiment. The breads were purchased fresh from different bakeries and markets in Amman Governorate and fed to the participants between 9:00 to 10:00 hours in every experimental day.

Ethics committee approval was obtained from the

Jordan Center for Pharmaceutical Research in 2018.

Experimental Design

After fasting overnight, participants ingested an equivalent amount of 50 g carbohydrate of each test bread (once) and white bread (twice) as a reference food on different days.⁹ Ten minutes were allowed for participant to ingest the total bread portion with 250 ml of water during each two-hour sampling period.

Blood Sampling and Analysis

Blood was obtained from the non-dominate venipuncture to measure the serum glucose and insulin levels. Samples of blood were drawn at fasting (0 min) and at 30, 60, 90, and 120 min intervals after the consumption of white bread (WB) and test breads (TBs). The same investigator performed all blood glucose measurements. The area under the curve of each bread was calculated for each participant. The simple application of the trapezoid rule was used to calculate the net incremental area under the curve (IAUC) to increments of all blood glucose. The glycemic and insulinemic IAUCs, as well as the peak glycemic and insulinemic responses of TBs were compared with those of WB. Differences were considered significant a p-value of <0.05 using twotailed testing.11,19,21

Chemical Analysis

Proximate analysis for the tested breads was performed to determine their average contents of moisture, protein, fat, carbohydrate, fiber and ash. Each bread was analyzed in triplicates.

Calculation of Glycemic Index

In this study, the GI was calculated according to Brouns *et al.*, using the method of IAUC.9 This method uses the mean of the ratios method (f:r), where f is an individual participant's IAUC after ingesting the test bread and r is the IAUC for the same participant after ingesting the standard white bread. The individual values of f:r were averaged for all participant to calculate the GI for the test breads.²² In each participant, the GI (%) was calculated by using this formula: GI = (IAUCTB / IAUCWB) X 100%, where

- IAUC_{TB} Incremental Area Under the blood glucose response Curve for the tested bread
- IAUC_{WB} Incremental Area Under the blood glucose response Curve for the white bread

The GI for each tested bread was calculated as the mean of the average GIs for the 10 subjects.

Results

Participants

As shown in Table (1), the mean \pm SEM age (year) of the 10 participants was 28.7 ± 1.7 (range: 22-36). The mean (\pm SEM) body mass index (BMI in kg/m²) of the participants was 23.9 ± 0.9 (range: 18.9-27.1). When comparing male and female characteristics, there were no significant differences between their mean age, BMI, waist/hip ratio, and body fat mass indices. On the other hand, there were significant differences (p<0.05) in their mean height, weight, and % body fat indices.

Glycemic Index (GI)

Table 2 shows the results of the GI experiment. The fasting blood glucose values of the 10 subjects for the six tested breads (TBs) and the reference local white bread (WB) ranged between 68 and 95 mg/dl (3.78 and 5.28 mmol/L). Additionally, the 2-hr blood glucose level of the subjects for the 7 breads ranged between 70 and 103 mg/dl (3.89 to 5.72 mmol/L).

In the Table, the GI of TB5 was significantly (p<0.05) higher than the GIs of TB2, TB4, and TB6. There was no significant difference (p<0.05) between TB1 and

the other 5 types of bread. On the other hand, the GIs of TB4 and TB6 were significantly (p<0.05) lower than that of TB3 and TB5.

Sex	Age (yrs)	Height (cm)	Weight (kg)	BMI (kg/m²)	Waist/hip ratio	Body fat mass (kg)	%body fat
F1	34	158	66.3	26.6	0.96	26.2	39.5
F2	33	165.5	60.4	22.1	0.89	19.7	32.6
F3	30	166	70.7	25.7	0.95	29.8	42.2
F4	24	151	43.1	18.9	0.82	10.0	23.2
F5	31	158.5	51.4	20.5	0.86	15.4	29.9
Sub Mean	30.4 ± 2.0^{a}	159.8±3.1ª	58.38 ± 5.6^{a}	22.76±1.7ª	0.90 ± 0.06^{a}	20.2 ± 3.6^{a}	33.5 ± 3.4^{a}
F±SEM							
M1	22	167	75.6	27.1	0.88	22.4	29.7
M2	23	176	77.3	25.0	0.97	20.5	26.5
МЗ	36	160	61.8	24.1	0.88	13.6	22.1
M4	31	184	91.7	27.1	0.93	22.5	24.6
M5	23.0	182	71.6	21.6	0.77	13.3	18.5
Sub Mean M±SEM	27±3.1ª	173.8±5.1⁵	75.6±5.4 ^b	25.0±1.2ª	0.89±0.08ª	18.5±2.1ª	24.3±1.9 ^b
F and M Mean±SEM	28.7±1.7	166.8±3.4	77.0±4.4	23.9±0.9	0.89±0.06	19.3±2.0	28.9±2.4

Table 1: Characteristics of the female (F) and male (M) participants*

*Submeans having the same letter in the same column indicate no significant differences (p<0.005).

	Bread type	Bread characteristics	GI (compared to WB)	GI (compared to glucose)**
TB1	Brown, healthy	Whole wheat flour Claim: enhanced and healthy	87.15±16.84 ^{abc}	61.0
TB2	Oat, sugar-free	Wheat flour + oat	61.00±16.04 ^{bc}	42.7
TB3	Multi-grain	Flour of whole wheat + barley, millet, corn, soybean, etc.	94.76±12.64 ^{ab}	66.3
TB4	Multi-grain	Whole wheat flour + different grains	58.94±13.19°	41.3
TB5	Barley	Whole wheat flour + barley flour	97.55±12.06ª	68.3
TB6	High fiber, high protein, and low carbohydrate	Claim: 36% fiber, 80% less carbohydrate Per 100 g: 34.2 g protein, 36.3 fiber, 2.9 carbohydrate	54.87±13.26°	38.4
WB	White bread	Mowahad flour (73-78% extraction)	100.00	70.0

Table 2: Some characteristics of the test breads and their GIs (Mean±SEM)*

*Means having a different letter indicate highly significant differences (p<0.0001).

**To convert to a glucose standard, multiply by 0.7.

	Bread types	% fat	% protein	%СНО	% ash	% fiber	%moist.	GI (compared to white bread)	Energy density) (kcal/100 g)
TB1	Brown, healthy	0.27±0.03℃	10.58±0.49e	52.16±0.94°	1.95±0.09e	0.96±0.04°	34.08±0.51 ^b	87.15±16.84ªbc	253.36 ^d
TB2	Oat, sugar-free	1.08±0.08 ^b	12.80±0.06°	61.14±0.23ª	2.63±0.02°	1.78±0.09 ^b	20.57±0.209	61.00±16.04 ^{bc}	306.01ª
TB3	Multi-grain	3.16±0.38ª	12.82±0.54°	56.32±1.11 ^b	2.18±0.06₫	0.62±0.07	24.89±0.54	94.76±12.64 ^{ab}	305.04ª
TB4	Multi-grain	3.26±0.33ª	14.96±0.09 ^b	50.10±0.28 ^d	2.80±0.07⁵	0.88±0.11°	28.01±0.25°	58.94±13.19°	289.55°
TB5	Barley	1.19±0.06⁵	11.43±0.24 ^d	60.76±0.39ª	2.65±0.06°	2.1±0.17a	21.86±0.25f	97.55±12.06ª	299.51 ^b
TB6	High fiber, high	0.60±0.03°	33.51 ± 0.44^{a}	19.91±0.06e	2.96±0.02ª	2.02±0.20ª	41.01 ± 0.46^{a}	54.87±13.26°	219.05 ^e
	protein, and low								
	carbohydrate								
WB	White bread	0.42±0.05°	10.32±0.73 [®]	60.87±0.46ª	0.99±0.03 ^f	0.42±0.04	26.98±0.21 ^d	100	288.58c
	Mean±SEM	1.43±0.26	15.20±1.70	51.61±3.04	2.31±0.14	1.25±0.15	28.20±1.49		280.16±6.70
*Mea	ins having a differ	ent letter in t	he same colur	nn indicate hig	thly significar	it differences	(p<0.0001).		

Table 3: The proximate analysis (mean±SEM) on wet-matter basis of the test breads'

Table 2 shows the nutrition and health claims, while Table 3 presents the nutrient composition of these breads. There were significant (p<0.05) differences in the energy density (kcal/100 g) among TB1, TB5, TB6, and WB (Table 3). Despite that there was no significant difference between TB2 and TB3, there was a significant difference (p<0.05) between these two breads and the other 5 types of bread. In addition, the energy density of TB6 (219.05) was significantly (p<0.05) the lowest among all breads. When compared to white bread, the highest GI breads, TB5 (barley bread), TB3 (multi-grain bread), and TB1 (brown, healthy bread): all had significantly (p<0.05) higher fiber content; two of them (TB3 and TB5) had significantly (p<0.05) higher while TB1 had significantly (p<0.05) lower energy density (kcal/100 g) in comparison with the mean of all breads (280.16) and that of WB (288.58).

Insulin Response

In the present study, the insulin response (µU/ml; mean±SEM) to equal guantities of carbohydrate (50 g) in test breads (TBs) was compared with that of white bread (WB) up to 2 hours (Table 4). The peak insulin response (41.14±4.84) was at 30 min for all breads, including WB, and continued over the baseline (7.76 ±0.71) even after 120 min (14.65±3.14) for all these breads. The means of insulin levels of the 6 tested breads and white bread were significantly (p<0.05) different at all of the 5 times of measurement. The mean insulin responses of all of breads, including WB, were almost equal at 0, 60, 90, and 120 min. While the mean insulin response at 30 min was significantly (p<0.05) higher for the 6 test breads (43.03 ± 3.94) in comparison to that of WB (35.45 \pm 4.41). The duration of postprandial insulin level for all breads, including WB, to come back to fasting level required longer than 120 min.

As seen in Table 4, there were no great differences concerning the mean glucose levels between all the tested breads and white bread at 0-, 30-, 60-, 90-, 120-min. The mean insulin levels of the tested breads are higher than that of white bread at 0-, 30-, 60-, and 90- min. The insulin concentration (μ U/mI) of the tested breads at 120-min was 14.35±2.90 which is lower than that of WB (15.54±5.00).

Bread	fastin	D	30 m	-	60 m		90 m		120 n	-
types	glucose	insulin	glucose	insulin	glucose	insulin	glucose	insulin	glucose	insulin
TB1	85.6±1.74	8.44±0.93	92.8±4.72	36.77±6.11	87.63±4.25	25.93±3.83	86.58±5.17	29.68±6.54	85.1±2.87	16.13±3.87
TB2	81.4±2.13	7.93±2.06	96.5±4.49	43.02±6.87	82.0±3.33	31.99±6.99	80.15±3.59	22.49±4.94	81.7±1.10	11.73±2.03
TB3	84.0±1.61	8.75±1.22	99.48±4.07	44.84±7.60	87.9±3.17	37.15±4.43	84.2±4.47	20.02±4.07	87.8±2.52	12.10±2.06
TB4	87.4±1.64	7.32±1.28	91.59±4.75	44.311±9.49	81.9±5.38	33.96±5.69	86.8±2.82	25.69±6.04	89.5±1.96	17.66±3.63
TB5	86.9±1.35	8.14±0.98	103.1±2.92	39.32±3.23	86.3±5.80	27.54±3.13	92.19±2.41	23.85±4.72	84.6±2.28	11.15±1.74
TB6	81.7±1.49	7.77±1.43	88.53±2.58	44.23±6.17	82.17±2.82	29.98±4.09	83.12±2.04	22.71±3.54	83.6±1.65	16.01±2.19
Mean WBV*	81.8±1.3	6.88±0.73	93.85±2.58	35.45±4.41	84.15±3.02	29.62±3.47	86.23±2.40	21.34±3.36	85.45±1.78	15.53 ± 3.23
Mean TBs*	84.5±0.72	8.06±0.54	95.33±1.69	42.08±2.79	84.65±1.71	31.09±1.96	85.51±1.51	24.07±2.02	85.38±0.9	14.13±1.11
Mean TBs+WB*	⁺ 83.83±0.64 ^b	° 7.76±0.44 ^E	94.96±1.42ª	40.42±2.31 ^A	84.53±1.48 ^b	30.72±1.70 ^B	85.69±1.27 ^b	23.39±1.73°	85.4±0.8⁵	14.48±1.15 ^D
*Mean±SEM										

Table 4: Blood glucose (mg/dl) and insulin (µU/ml) levels* for breads

Table 5: Correlation values and degree of significance between blood glucose levels and insulin levels for breads* TB2 Bread types TB1 TB2 *The overall correlation between blood glucose levels and insulin levels for all breads is 0.53 (p<.001).</p>

0.0001

0.004 0.40

0.0001 0.61

0.004 0.40

0.0001 0.56

0.0001 0.55

0.0001 0.60

Significant degree Correlation value

0.65

МВ

TB6

TB5

TB4

TB3

Table 5 shows that the overall correlation between blood glucose levels and insulin levels for all breads is 0.53 which indicates a highly significant difference (0.001).

Discussion **Subjects**

The results indicate that both male and female subjects of this study are mutually matched. The significant differences in height, weight, and % body fat are expected according to the different natures of males and females.

Glycemic Index (GI)

The nondiabetic fasting normoglycemia range is defined to be <100 mg/dl (<5.6 mmol/L) and <140 mg/dl (<7.8 mmol/L) 2 hours after eating.^{23,24} The results of fasting and 2-hr postprandial blood glucose levels of all subjects for the 7 breads in the present were within the normal limits.

Table 2 shows that all the tested breads had GIs lower than that of the local white bread. This indicates that the tested breads have ingredients, preparation methods, or other characteristics that lower their GIs. As it is well known, all these breads are produced by baking and preparation methods that are almost similar. Therefore, it is expected that the major differences among these breads may be due to their ingredient composition. The ingredient composition can be inferred from the breads' names and their nutrition and health claims (Table 2).

As seen in Table 2, bread TB6 had the lowest GI (54.87±13.26). This bread has a special ingredient formula of high protein and fiber and low carbohydrate which might be responsible for its lowest GI value. Two of the tested breads, TB3 and TB4, are both claimed as multi-grain. These two breads had significantly (p<0.05) different GIs: TB4 had the second-lowest value (58.94±13.19), while TB3 had the second-highest value (94.76±12.64). This may indicate that the ingredients do not always reflect the bread's GI value. It also seems that other characteristics of the bread affect their GI. The highest GI (97.55±12.06) was that of TB5, a barley bread. Also, TB1, a brown healthy bread, had a GI of 87.15±16.84; it occupied the third-highest value. TB2, an oat-containing, and sugar-free bread had a relatively low GI (the third-lowest GI value; 61.00±16.04).

When corrected for glucose as a reference food (as shown in Table 2), TB2, TB4, and TB6 are considered low GI foods (<55). The other 3 breads, TB1, TB3, and TB5, are considered medium GI foods (55 -<70) Atkinson *et al.*,²⁵ stated that breads, including whole grain, were among the high and low GI classifications. It was also noted that, although some types of whole-grain bread have a low GI, most breads are of medium GI.²⁶

The lowest GI breads: TB6 (claimed as high fiber, high protein, and low carbohydrate) and TB4 (multigrain bread, made from whole wheat flour and different grains) (Tables 3 and 4) have 2 common characteristics in comparison with the other 4 breads: the lower significant (p<0.05) carbohydrate and the higher significant (p<0.05) protein contents. In comparison to white bread (WB), TB4 and TB6 breads have significantly (p<0.05) higher moisture, fiber, ash, and protein contents but significantly (p<0.05) lower carbohydrate content.

Insulin Response

As mentioned earlier, the blood sugar response is affected by many food internal and external factors.²⁷ Thus, considering that high starch foods as efficient in reducing glycemic and insulinemic responses may not be practically true.²⁸

The metabolic control is effected by both the glucose and insulin levels postprandially; therefore, studying their independent effects on glycemic control is important.¹¹ In this study, the insulin response was found to parallel the glucose response for all studied breads, which agrees with the findings of.²⁹ The peak of insulin and glucose responses were at 30-min, which is in agreement with Febbraio *et al.*,³⁰ and Wee *et al.*,³¹

It is concluded from this study that 3 types of the tested bread are of low GI and the other 3 types are of medium GI.

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Conflict of interest

The authors declare no competing interests.

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