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Glycemic index of selected watermelon *(Citrullus lanatus)*

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Abstract

The glycemic index (GI) was conducted on 14 healthy subjects who have consumed 25 g of available carbohydrate portions of glucose (standard food) and four test foods (red-fleshed seedless watermelon, red-fleshed seeded watermelon and yellow-fleshed watermelon, as well as a glass of red-fleshed seedless watermelon juice) in random order after an overnight fast. Red-fleshed seedless watermelon was usually processed as juice than red-fleshed seeded and yellow-fleshed watermelon. Blood glucose was measured at 0, 15, 30, 45, 60, 90 and 120 mins after intake of the foods. Incremental areas under the curve were calculated, whereas the glycemic index was determined by expressing the area under the curve after the test foods, as a percentage of the mean area under the curve after consuming standard food, was carried out. The results showed that the area under the curve for a portion of red-fleshed seedless was 98.17±6.39, red-fleshed seeded (94.10±7.45), yellow-fleshed (92.95±8.73), and a juice of red-fleshed seedless (98.89±6.38) did not have any significant difference (p<0.05). The GI of a portion fruit and the juice of red-fleshed seedless watermelon were 51, while red-fleshed seeded watermelon was 48 and yellow-fleshed watermelon was 47. The study showed red-fleshed watermelon and yellow-fleshed watermelon could be classified as low GI food (GI value below 51) with strong influenced by the fructose content and no significant difference from GI value among red and yellow watermelons. This research will help the experts to explore more based of GI value and also be used as a database reference.

Introduction

Glycemic index (GI) was introduced as a method of grading carbohydrate containing foods based on the blood glucose response after food ingestion and metabolized in a body (Jenkins *et al.*, 1981). Definition of GI expressed as a percentage of the blood glucose response (incremental area under the blood glucose response curve (AUC)) after a portion of food containing 50 g (or 25 g) of the available carbohydrate and prompted by 50 g (or 25 g) of reference food (either glucose or white bread) consumed by the same subject (Wolever *et al.*, 1991). The GI has a wide range from low ranking; medium ranking and high ranking depends on various factors. According to International Organization for Standardization (ISO method 26642:2010), the high GI foods was classified when GI more than 70, whereas GI less than 55 considered as low GI food. Studies have shown that low GI food may increase a lot of goodness to maintain optimum health such as reducing an overall cancer risk (Sieri and Krogh, 2017); cardiovascular risk and disease factor (Ma *et al.*, 2012); and managed and control blood glucose among diabetes patient (Dong *et al.*, 2011).

Fruits are known as the best sources of carbohydrates, vitamins and minerals and considered significant for well-being properties. Nutrition Division of Malaysia also recommends having a lot of fruits portion in diet intake. Nowadays, there was a demand of fruits to eat as a healthy snack (Yen *et al.*, 2015). Watermelon *(Citrullus lanatus)* is easily found at any event in Malaysia. Due to the Malaysian climate as a tropical country, watermelon is not seasonal fruit and available throughout the year to gain reputation as a popular fruit. Malaysian consumption of watermelon was 4.9 kg per capita in 2013 (Department of Statistic Malaysia, 2015) and study by Nurul Izzah *et al.* (2012) also found that watermelon was the third famous fruit of Malaysian. Nevertheless, most studies were fixed on portion of red watermelon and not included yellow watermelon and a juice of watermelon. Hence, this study to determine the GI of selected choices of watermelon (red seedless, red seeded, yellow and juice of red seedless) and the difference of GI value among red and yellow watermelon. This study will be contributed as useful information to educate people about the GI

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and also be used as future database reference.

Materials and methods

Sample collection

Commercial sample of selected watermelons (red-fleshed seedless watermelon (RS), red-fleshed seeded watermelon (RSD) and yellow-fleshed watermelon (Y)) were bought from Pasar Borong Selangor. The matured fruits were chosen according to the Federal Agricultural Marketing Authority (FAMA, 2012) index of maturity. Samples were cleansed by water and wiped to dry the outer layer. Samples were manually cut using utility knife longitudinally from the stem-end to the blossom-end at the central and all outer layer skin, rind and seed were discarded because only edible portion of watermelon was used as a sample.

Nutritional analysis

Analysis of moisture content, crude protein, crude fat and total ash was determined according to AOAC (2000) (No. 925.09; No. 920.152; No. 991.36; No. 940.26). In addition, the available carbohydrate was determined as described by Clegg (1956) using anthrone reagent and glucose anhydrous as a standard. The high performance liquid chromatography (HPLC) method described by Yang *et al*. (2008) was used to determine the content of sugar (glucose, sucrose and fructose). Total dietary fibre (TDF) from the dried homogenate watermelons was analysed based on the methods of Lee *et al*. (1992) using Megazyme the total dietary fibre assay kit.

Study design and sample size

This study design was a cross-over and experimental study. The sample size calculation based on (Wang, 2007) and minimal subjects needed were 13 persons. In this test, 14 persons volunteer (5 females and 9 males) were participated. The ethical clearance approval (Reference Number: FPSK_ Julai(13)06) was approved by UPM human ethic committee on 7th February 2014. The location of subject sampling was at the Faculty of Medicine and Health Sciences, Universiti Putra Malaysia (UPM), while the study was conducted at the Laboratory of Nutrition. All subjects were screened based on certain criteria as mentioned below. All subjects had to sign an information sheet and consent form. Besides, compensation was given after the subjects had completed the study.

The subject was considered eligible to participate in the study if all of the inclusion criteria were met:

- Male or Female aged 18 to 30 years
- Normal BMI (BMI : 18 to 23 kg/m^2)
- Non smoker
- Individual with none of all conditions below

On the other hand, the subject was not considered in the study if he or she met any of the exclusion criteria:

- Individuals with impaired glucose tolerance or with fasting blood glucose level more than 5.4 mmol/L
- Individuals diagnosed with chronic diseases such as diabetes (Type 1 & Type 2), hypercholesterolemia, high blood pressure and disease that could be interfere with glucose metabolism.
- Individual on medication (glucocorticods, anticonvulsant medication, thyroxine) that might affect glucose metabolism.
- Individual with health problems or have a history of an acute medical or surgical within the last 6 months (and others than stated above)

Table 1. Amount of samples and standard food for glycemic index

Samples / Standard	Weight of sample (g)	Available CHO (g)
Glucolin® (Glucose)	32	25
Red-fleshed Seedless	236	25
Red-fleshed Seeded	239	25
Yellow-fleshed	233	25
Red Seedless Juice	236	25

Preparations for standard food and test food

The standard food was 25 g of available glucose (Glucolin®), which was dissolved in 250 ml of water. As for test food, a portion of 25 g available carbohydrate of each watermelon (red-fleshed seeded, red-fleshed seedless, yellow-fleshed watermelons and a glass of juice) had been employed. As for red-fleshed seedless juice, 25 g of available carbohydrate from red-fleshed seedless watermelon was blend using automated juicer machine Panasonic MJ-DJ 31 (Petaling Jaya, Malaysia) without any water and made it around 280 ml per portion. All samples were prepared not more than 3 hours before the analysis was carried out.

Experimental procedure

Subjects were requested to consume 25 g of available carbohydrate of the test food or the standard food on different days in the morning after fasting overnight (10-12 hr). One glass of plain water (250 ml) was provided together with the test food, as well as a standard food. The subjects were given 0 minutes to 15 minutes to finish the food. Standard food was given 3 times in random for each subject on separate days to ensure no changes in subject's glucose response. First finger prick was done after subject arrived as a control. Then, 200 µl of blood sample was taken by a finger prick at 0 minutes before the food was consumed. The blood was continually collected after 15, 30, 45, 60, 90 and 120 minutes after meal. The total blood sample needed for one test food was 1.6 ml. The total finger prick done throughout this study for one subject was 56 finger pricks (7 test x 8 finger prick for one test). The blood samples were drawn into 0.6 ml BD Microtainer® tube (Beckton and Dickinson, NJ, USA) with sodium fluoride as glycolytic inhibitor and centrifuged at 4000 x *g* for 5 mins to obtain plasma. After that, the plasma was collected in a 0.5 ml tube and was stored at -80°C before analysis of glucose using an auto analyser YSI 2300 (YSI Stat Plus, Ohio, USA) was conducted.

Body mass index (BMI), 24 hr diet recall and physical activity measurement

Others measurement were important to support the GI test. Other measurements included body mass index (BMI) measurement, 24 hr diet recall and an assessment of physical activity level. For BMI measurement, weight and height of the subjects were measured and recorded at the beginning of every test. All the subjects recorded the details of food intake the previous day in a 24 hour diet recall form. The calorie intake per subject was calculated using Nutritionist Pro™ diet analysis software (Axxya Systems, Washington, USA).

Physical assessment of subjects was evaluated by using a questionnaire. The subjects were required to fill in self-administered short version of International Physical Activity Questionnaires (iPAQ version 3, 2012) on physical activity level in the last 7 days during each test. All subject completed all questionnaires while waiting for the blood collection throughout the 2 hour test. All answers were calculated using scoring protocol and adhered to the calculation suggested by Patterson (2005).

Glycemic index (GI) calculation

The GI was calculated based on method described by the Standard Australia (2007). The blood glucose responses for every two points of time over 2 hours had been used to calculate the incremental area under the curve (iAUC) for each test meal, including standard food based on standard criteria, ignoring the area beneath the baseline. The GI of each subject was the iAUC after the test meals, which was expressed as a percentage of the mean of iAUC after three glucose tests. The GI for each meal was taken as the average of values from all 14 individuals. The equation of GI is given below:

Table 2. Values of age, BMI, calorie intake and physical activity level of respondents

	Age	BMI	Calorie Intake (kcal)	Physical activity level (MET)
Respondent	21.57±0.1	21.38±0.24	1413±22	1072.67±100

All figures are mean ± SEM among subjects (n=14)

$$\text{Glycemic Index} = \frac{\text{IAUC of 25g available CHO test food}}{\text{IAUC of 25g standard food (glucose)}} \times 100$$

Statistical analysis

Statistical calculation by Statistical Products and Service Solution (SPSS) for Windows, version 20 (IBM, New York, USA) was carried out. The results of glycemic test are expressed as the mean ± standard error means (SEM). The blood glucose values at each point, the iAUC, and the GI values were subjected to analysis of variance (ANOVA), and Pearson correlation. The differences were considered significant if $p<0.05$.

Results and discussion

Glycemic index of watermelon

The characteristic of subjects (n=14; 9 males and 5 females), including age, BMI, calorie intake and physical activity level are described in Table 2. From 24hr diet recall, the calorie intake for all respondents adhered to the recommended of nutrient intake (RNI, 2005) and the physical activity level showed that the respondents had a moderate level of fitness.

The plasma glucose responses after the consumption of glucose and four test foods (red-fleshed seedless, red-fleshed seeded, and yellow-fleshed, as well as juice of red-fleshed seedless) are shown in Figure 1. The calculation of coefficient variation (CV) of the AUC of repeated glucose was 28.8±6% and all GI values (no outlier more than 2SD) were accepted according to ISO methodology. The mean for AUC of glucose was significantly different than all samples ($p<0.05$) and no significant difference was found among test samples (Table 3). After the calculation, the GI of red-fleshed seedless juice (GI = 51±1) and red-fleshed seedless fruit portion (GI = 51±2) had been the highest. Meanwhile, the value of GI for red-fleshed seeded fruit portion was 48±1, whereas the GI value of GI for yellow-fleshed watermelon was 47±2.

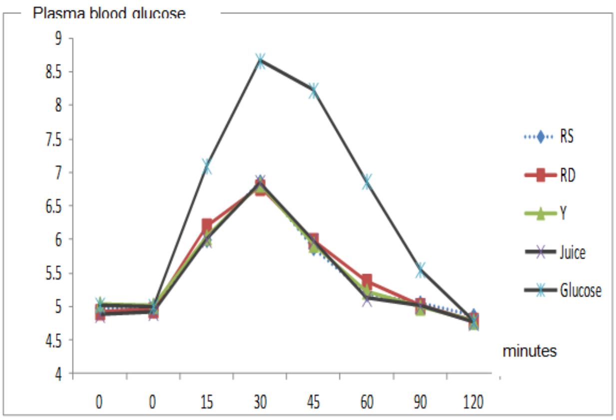

Figure 1. Mean of plasma blood glucose response of standard food and the test foods. (n=14)

Relationship between glycemic index and other nutrients

Watermelon was classified as high sugar fruits (Yativ *et al.,* 2010) and International GI Tables (Foster-Powell *et al.*, 2002) showed GI for red watermelon was 72. However, the GI value obtained in this study was almost similar to those found in previous studies for red watermelon such as 48 (Trinidad *et al.*, 2010), 55 (Roberts *et al.*, 2008) and 54 (Barakatun Nisak *et al.*, 2005).

There are many factors that can affect the value of GI. The component of sugars, processing or treatments of sample and the matrix structure of sample might influence the GI value of food. Other than that, according to Wolever and Miller (1995), the internal factors were fibre content, maturity stage, fat content and presentation of antinutrient. The component of dietary fibre might influence the GI by delaying a digestion and slowly blood glucose response (Wolever *et al.*, 1994; Trinidad *et al.*, 2003; Robert *et al.*, 2008).

Table 4 showed nutrient contents and proportion of sugar for red-fleshed seedless watermelon, red-fleshed seeded watermelon and yellow-fleshed watermelon. From the analysis, a low correlation between total dietary fibre and GI value (r^2 = -0.2721) was determined. Thus, the fibre content of watermelon did not give an impact to blood glucose response and the amount of fat among the samples was too low to influence the GI value.

The stage of ripening and maturity of watermelon depended on moisture and sugar content. All samples were fully matured and have almost similar moisture contents. Although red-fleshed seedless juice was in different physical attire than other test food, there shared similar GI value. Apart, the high moisture found in watermelon due to the watermelon is a soft form of fruits that influenced the physical factor. Furthermore, the processing activity that changed the physical structure of the red-fleshed seedless watermelon from fruit portion to juice portion was minimal. This recent observation had been similar with those reported by Arvidsson-Lenner *et al.* (2004) and Brouns *et a*l. (2005), which concluded that a homogenised structure would give effect to GI value.

Besides, the amounts of monosaccharide and sugar content (glucose and fructose) might contribute to the GI. The relationship between glycemic index and component of sugar (fructose) was investigated. There was a high correlation between fructose content of the fruits and respective GI value (r^2= - 0.91). It explains the respective GI value for red-fleshed seedless watermelon was higher although amount of total sugar for red-fleshed seeded was more than red seedless watermelon. Glucose displayed higher

Table 3. Incremental area under the curve (IAUC) and glycemic index (GI) values of test foods and glucose

Type of watermelon	Area under the curve (mmol x min/L)	Glycemic Index (%)
Red-fleshed seedless	98.17 ± 6.39^a	51 ± 2^a
Red-fleshed seeded	94.10 ± 7.45^a	48 ± 1^a
Yellow-fleshed	92.95 ± 8.73^a	47 ± 2^a
Juice of red seedless	98.89 ± 6.38^a	51 ± 1^a
Glucose	193.83 ± 15.34^b	100^b

Values are mean ± SEM. Means with different letter superscripts are significantly different ($p<0.05$)

Table 4. Moisture contents, proximate, total dietary fiber and sugars parameters of red-fleshed seedless (RS), red-fleshed seeded (RD) and yellow-fleshed (Y) watermelon based on fresh weight basis (g/100g)

Parameters (%)	RS watermelon	RD Watermelon	Y Watermelon
Moisture	89.65 ± 4.29	87.46 ± 2.58	87.03 ± 2.67
Protein	0.76 ± 0.81	0.81 ± 0.07	0.76 ± 0.17
Fat	0.16 ± 0.06	0.18 ± 0.05	0.21± 0.06
Ash	0.36 ± 0.04	0.24 ± 0.07	0.28 ± 0.03
Available CHO	10.6 ± 3.16	10.48 ± 2.65	10.71 ± 2.28
Total CHO	11.41 ± 3.12	11.27 ± 3.03	11.06 ± 1.61
Total Dietary Fiber	0.56 ± 0.04	0.63 ± 0.13	0.54 ± 0.05
Fructose (mg/ml)	46.55 ± 18.83	49.59 ± 10.92	51.43 ± 15.86
Glucose (mg/ml)	31.22 ± 13.63	29.51 ± 14.5	33.91 ± 12.19
Sucrose (mg/ml)	15.93 ± 8.71	34.93 ± 13.55	15.3 ± 5.12
Total sugar (mg/ml)	95.04 ± 25.22	113.78 ± 31.58	100.59 ± 25.45

All figures are mean ± SD of three replications blood glucose response (GI = 99±3) than fructose (GI = 19±2), as glucose can easily enter into the blood circulation, while fructose has to follow another pathway of metabolism before it can change to glucose (Brand-Miller *et al*., 2007). Hence, this finding has exhibited that blood glucose responses for red-fleshed seedless, red-fleshed seeded and yellow-fleshed watermelon were influenced by component of sugar contents consequently by high proportion of fructose that contributed to low GI value among watermelon samples.

Conclusion

Red-fleshed watermelon and yellow-fleshed watermelon had a GI value below 51 with influenced by the fructose content. No significant difference for GI value among red-fleshed and yellow-fleshed watermelons. The glycemic index for red-fleshed and yellow-fleshed watermelons can be classified as low GI food group. Furthermore, this research will help the experts to explore more based of GI value and also be used as a future database reference.

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