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Saloni Chauhan
Department of Food and Nutrition, Rajendra Prasad Central Agricultural University, Pusa, Samastipur, Bihar, India

Corresponding Author:
Saloni Chauhan
Department of Food and Nutrition, Rajendra Prasad Central Agricultural University, Pusa, Samastipur, Bihar, India

Impact of processing on glycemic index of Bengal gram flour before and after processing through *in vitro* condition

Saloni Chauhan

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Abstract
The consumption of pulses is associated with many health benefits. Currently, there is a growing body of evidence that suggests the regular consumption of bengal gram, along with other pulses, is associated with reduced risk for cardiovascular disease and type 2 diabetes, as well as improved glycemic control and lowering of blood pressure The current study was undertaken to determine the Impact of processing on glycemic index of bengal gram flour before and after processing through *in vitro* condition. In this study, results revealed that the freshly harvested bengal gram i.e. raw bengal gram had been taken as control sample and after processing like soaking had been taken as treated sample. It was revealed that bengal gram control flour sample was 15.8 GI and treated bengal gram sample was 11.51 GI. The lower level of glycemic index in bengal gram after treatment is recommended for persons with Diabetes, obesity and cardiovascular diseases.

Keywords: Glycemic index (GI), processing, *in vitro* method, bengal gram

Introduction
Glycemic index has proven to be a more useful nutritional concept than is the chemical classification of carbohydrate (as simple or complex, as sugars or starches, or as available or unavailable), permitting new insights into the relation between the physiologic effects of carbohydrate-rich foods and health. Several prospective observational studies have shown that the chronic consumption of a diet with a high glycemic load (GI dietary carbohydrate content) is independently associated with an increased risk of developing type 2 diabetes, cardiovascular disease, and certain cancers.

Glycemic index is a measure of the effect of carbohydrates on blood sugar levels. Carbohydrates that break down quickly during digestion releasing glucose rapidly into the bloodstream, have a high GI; Carbohydrates that break down more slowly, releasing glucose more gradually into the blood stream, have a low GI. For most people, foods with a low GI have significant health benefits. These factors include physical entrapment, rate of digestion, food form (physical forms, particle size), type of preparation (processing and cooking method), nature of starch (amylose or amylopectin), amount and presence of fibre, fat and protein and the presence of organic acids.

A popular application of GI is for body weight management. A low-GI diet is thought to promote weight loss through reduced food intake, reduced fat storage, and increased fat oxidation. Physiological and metabolic advantages observed from consuming low GI foods are due to reduced rate of carbohydrate absorption in the small intestine. The metabolic advantages include; lower postprandial glucose rise; reduced daily insulin levels, flatter gastric inhibitory polypeptide response decreased 24 hours uninary C-peptide output, prolonged suppression of plasma free fatty acids; reduced urinary catecholamine cholesterol levels, reduced hepatic cholesterol synthesis, decreased serum uric acid levels and increased urinary uric acid excretion.

Pulses are one of the main components of Indian diets. India has been growing 12 different pulse crops. Indian pulses mainly include chickpea, pigeon pea, moong bean, black gram, lentil, field pea, bengal gram etc. They are the excellent source of proteins, carbohydrates and minerals.

Pulses also act as the main source of protein in a diet of vegetarian Indian population, which supply almost all essential amino acids. Legumes are nutritionally important but their use in diet is recommended to be limited owing to presence of antinutritional factors. Starch is the major carbohydrate in pulse seeds, which accounts for 22–45% of the dry matter2. Legumes consumption is generally recommended in diabetic condition, as they have low glycemic index (GI). The productivity of pulse is increasing in India day by day, but the intake of pulse is decreasing. Hence the risk factors are also increasing day by day. So, considering all these points in mind it had been undertaken to study the impact of processing on glycemic index of bengal gram under *in vitro* condition.

Materials

Selection of the raw material

The most common varieties of bengal gram was selected for the study.

Procurement of quality protein maize grains and bengal gram

For the study, freshly harvested bengal gram was collected from farmers of Bisanpur, Dighra, (District – Samastipur) in one lot. For the study, 5 kg bengal gram were procured. The collected grains were cleaned by isolating damaged and unhealthy seeds and also by removing foreign matter. In case of bengal gram, the lot was divided into two portion for processing. Each portion was subdivided for processing into triplicate.

Processing of pulse grains.

For the study, out of two sets (in triplicate), one set was kept as such as control (in triplicate). The other sets (each in triplicate) were kept for processing. The processing methods applied were soaked for overnight (8 to 10 hours) then dried in sunlight and then ground into flour.

Preparation of the sample

After the application of processing methods, the control as well as processed pulse grains (all together 6 replicates) were subdivided into two sets. Then, whole bengal gram were converted into flour with the help of grinder.

Methods

In vitro determination of glycemic index

Procedure

In the first phase, samples were mixed with an equal weight of water, homogenized and incubated with α-amylase (185 U/g available carbohydrates) at 37 °C for 15 min in a shaking incubator, in order to mimic the oral digestion. Subsequently, the pH was adjusted to 2.5 with 0.1 M HCl. In cases that a food's homogenate volume was less than 2 ml, water (pH adjusted to 2.5 with 0.1 M HCl) was added up to 2 ml and transferred in duplicates to wells in a six-well plate. In each well plate, 0.1 ml of pepsin (porcine pepsin preparation, suspended in 4 g/100 mL in 0.1 M HCl,) was added and the plates were placed on a shaking incubator at 37 °C for 2 h, simulating the gastric phase of human digestion. After 2 h, a cylindrical insert, with a piece of dialysis membrane fastened to one end with an elastic band was placed in each well plate. Each ring was filled with 2 ml 0.1 M PIPES buffer pH 6.5 (piperazine-1, 4-bis (2-ethane-sulfonic acid) disodium salt, simulating the gradual increase of pH in the human small intestine. The plates were incubated for another 30 min, shaking at 37 °C.

The second phase of the *in vitro* digestion started after the end of this incubation period and lasted 120 min. An aliquot (0.2 ml) from the dialysate was taken (t = 0 min). Subsequently, the insert was carefully removed and 10 µL of amyloglucosidase (3260 U/mL amyloglucosidase) and 0.5 ml of a pancreatin–bile salt mixture (0.2 g porcine pancreatin from porcine pancreas, and 1.2 g bile extract, suspended in 100 mL 0.1 M $NaHCO_3$) was added on to each digested sample. The cylindrical insert was placed back and the incubation continued in a shaking incubator for 2 h, taking aliquots (0.2 ml) every 30 min from the dialysate for the determination of glucose (t = 30 min, t = 60 min, t = 90 min, t = 120 min, where t = 0 min is set at the start of the second phase of the *in vitro* digestion procedure). The digested samples (0.2 ml aliquots) were mixed immediately with 0.8 ml ethanol in a microcentrifuge tube and 30 min later the tubes were centrifuged for 10 min at 5000 rpm at 20 °C to clarify the ethanol supernatant fraction before analysis of sugars. Dialyzable glucose, *i.e.*, the concentration of glucose in the soluble and low molecular weight fraction of the digest, was tested as an index for the prediction of glycemic response. Glucose determination was performed spectrophotometrically using the dinitrosalicylic method at 562 nm.

DNS Method

Procedure

1. Add 3ml of DNS reagent to 3 ml of glucose sample in a tightly capped test tube. (To avoid the loss of liquid due to evaporation cover, the test tube with a piece of paraffin film if plain test tube is used.)
2. Heat the mixture at 90 ºC for 5-15 minutes to develop the red brown color
3. Add 1ml of a 40% potassium sodium tartrate (Rochelle salt) solution to stabilize the color.
4. After cooling to room temperature in a cold water bath, record the absorbance with a spectrophotometer at 575nm.

Data analysis of glycemic index

The data obtained upon determination of quality parameter of maize grains had been analyses for statistical implication by using standard deviation and paired 't' test to find out the Impact of processing on glycemic index of quality protein maize based flour (Snedecor and Cochran, 1989) [8].

Results and Discussion

Glycemic index of bengal gram flour before and after processing

The glycemic index of whole bengal gram was determined. The data obtained on glycemic index in flour and changes in glycemic index after application of processing methods have been presented in Table.1 illustrated through Fig 1.

The freshly harvested bengal gram i.e. raw bengal gram had been taken as control sample and after processing like soaking had been taken as treated sample. It was revealed from the table that bengal gram control flour sample was 15.8 GI and treated bengal gram sample was 11.51 GI.

It can be observed in Table 1 the value of GI in control sample was highest (15.8) followed by treated bengal gram sample (11.51).

Table 1: Glycemic index of whole bengal gram before and after processing *(in-vitro)*

Bengal gram flour sample	Glycemic index of bengal gram (mean ± S.D)
Control (A)	15.8 ±0.41
Processed (B)	11.51± 0.43
't' value flour among sample A×B	17.05 **

Each value is the mean of six observations
NS Not significant
*Significant at 5% level of probability
**significant at 1% level of probability

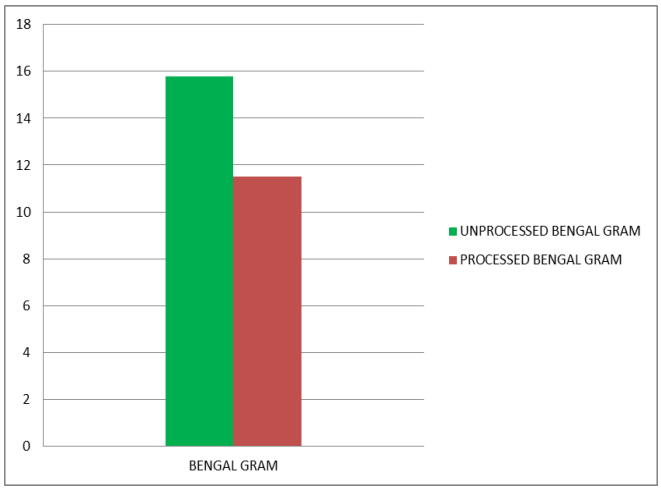

Fig. 1: Glycemic index of whole bengal gram before and after processing *(in-vitro)*

The statistical analysis clearly showed that the GI of bengal gram control samples was significantly higher than the treated bengal gram sample ('t' value 11.51) at 1% level of probability.

Conclusion

The GI found in control bengal gram flour sample was highest (15.8) followed by treated bengal gram (11.51). Hence, low glycemic index bengal gram based food mixes is recommended in stress condition such as obesity, diabetes, heart diseases etc. and also high glycemic index bengal gram flour based food mixes is recommended in case of malnutrition, given after exercise for extra energy, sports person, lactating and pregnant women for extra energy to fulfill their requirement.

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