



Comparative Analysis of Nutritional Composition and Effect of Dietary Fiber Extracts of Chickpea and Bengal Gram on Blood Glucose and Cholesterol Levels of Male Induced Diabetic and Hypercholesterolemic Rats

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ABSTRACT

Albino Sprague-Dawley rats (n=42) were randomly divided into seven groups, each of six rats. One group comprised of normal rats fed on basic purified diet AIN-76-A, three groups were alloxan induced diabetic rats which were fed basic purified diet AIN-76-A, Kabuli chickpea HFD and Bengal gram HFD and were monitored for random blood glucose and weight for a period of six weeks. Another three groups of rats with induced hypercholesterolemia were fed the same diet as the diabetic rats and were monitored for total serum cholesterol. It was found that both Bengal gram and chickpea had a significant effect on lowering glucose level and total cholesterol of these experimental animals. Bengal gram however, proved to have a significantly (p<0.05) greater effect on lowering of these parameters.

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Authors' Contributions

AH designed the study, executed the experimental work and wrote the article. S K supervised in writing the article.

Key words

High fiber diet, Random blood sugar, Total serum cholesterol.

INTRODUCTION

Chickpea (*Cicer arietinum*) belongs to the Fabaceae family and subfamily Faboidea (Deshpande, 1992). It is an important part of diet in Asia, Central and South America (Nestares *et al.*, 1996). There are two popular varieties in Pakistan named Kabuli Chana (chickpea) and Desi Chana (Bengal gram) (Khan, 1990; Muehlbauer and Kaiser, 2012). Both varieties of chickpea and Bengal gram seeds are grown mainly in the Mediterranean area, the Near East, Central Asia and America. Both *Kabuli Chana* and *Desi Chana* variety is used invariably both in Pakistan and India (Zia-Ul-Haq *et al.*, 2007).

Chickpea is the most widely used variety which absorbs more water, whereas Bengal gram has a thick seed coat and takes longer to be cooked (Khan *et al.*, 1995). In the Indo-Pak Subcontinent the de-hulled flour of chickpeas (basen) is used in making bread and a wide array of snacks (Singh, 1988). It renders unique range of taste to various local cuisines and is also packed with nutrients (carbohydrates, minerals) to the staple dish which ensures a balanced diet and also has adequate amount of dietary

fiber in it as well (Zia-Ul-Haq *et al.*, 2007). Studies have shown that chickpea is beneficial for heart burns, skin diseases, blood disorders, biliousness, liver, spleen and bronchitis (Sastry and Kavathekar, 1990).

The epidemiological prevalence of the non-communicable metabolic disorders such as hypercholesterolemia leading to cardiovascular diseases (CVDs) and of diabetes mellitus has risen tremendously in developing countries like Pakistan due to excessive consumption of junk food (Kapoor and Anand, 2002). Various substances have been tested for hypoglycemic and hypocholesterolemic effects (Abdel-Sattar *et al.*, 2011), however, there is a need to focus on health benefits of foods such as chickpea and Bengal gram which are consumed by the local population. The present study compares the effect of dietary fiber extracts of chickpea and Bengal gram on blood glucose and cholesterol level of induced diabetic and hypercholesterolemic rats.

MATERIALS AND METHODS

Chickpea and Bengal gram seeds were purchased from The National Seed Council (NSC) chickpea and Bengal gram were ground into flour.

Chemical evaluation of feed additives

The nutritional composition of the raw ground samples

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of chickpea and Bengal gram samples was determined as per (AOAC, 2005). The results were submitted to analysis of variance (ANOVA) test.

Animal diet

Three types of rats feeds were prepared and the basic purified diet (AIN-76A) was used for the two control groups (Reeves *et al.*, 1993). The chickpea high fibre diet (HFD) and Bengal gram HFD contained more fiber content as compared to the amount recommended in the protocol for the preparation of rat and rodent diets. To prepare HFD, the chickpea and Bengal gram were coarsely ground to separate the outer crust by sieving through sieve of 200-300 pore size. The ingredients were mixed in a room with defused light and diet pellets were made and oven dried at a very low heat. These pellets were then stored in plastic containers with air tight fitted lids in a cool and dark place until further use.

Experimental design

A total of 48 male albino rats 6 to 18 months, weighing between 200g to 250g were procured from the Animal House of PCSIR Laboratories. Animals were housed in a temperature (20 to 23°C) and humidity (approximately 50%). To measure the exact amount of food consumption, the rats were housed in individual stainless steel hanging wire-mesh cages, with food and tap water provided according to the need. The amount of feed consumed by the rats was determined by weighing feed in grams before putting it in the feed hoppers. Later the remaining feed was weighed once again to determine the exact amount of feed consumed by each rat in the individual cage (Jackson *et al.*, 1994). The average feed intake was 15g/day/rat. The rats were divided into 8 groups with 6 rats in each group.

The groups I and V fed on basic purified diet (AIN-76-A), group II, III and IV which comprised alloxan induced diabetic rats were fed on AIN-76-A (group II), on chickpea HFD (group III), and on Bengal gram HFD (group IV), group VI-VIII which were induced hypercholesterolemic rats were fed on AIN-76-A (group VI) on chickpea HFD (group VII) and on Bengal gram HFD (group VIII).

Induction of diabetes and hypercholesterolemia

Diabetes was induced in three groups with ALX monohydrate (Sigma-Aldrich Company) after 12 h of fasting (Ashok-Kumar *et al.*, 2010). The animals were administered an IV dose of Alloxan monohydrate dissolved (0.9% saline solution) at 70mg/kg (Orsollic *et al.*, 2011; Bilal *et al.*, 2014), intravenously at the coccygeal lateral vein of the rat (Thorington, 1966; Young and Dawson, 1981). Immediately after ALX monohydrate injection the rats were given glucose diluted in water to prevent

hypoglycemia. Blood glucose level was determined at 0 h and 10 h after ALX injection ascertain the glucose level, which should be <200mg/dl.

The procedure for induction of hypercholesterolemia has already been described elsewhere (Bilal *et al.*, 2014).

Estimation of glucose and cholesterol

Blood glucose estimation was done by One Touch Ultra 2 Glucometer of OneTouch Verio IQ, U.S.A. (Bilal *et al.*, 2014). The cholesterol level was estimated by using Richmond (1973) method.

Statistical analysis

These results were analyzed by one-way analysis of variance (ANOVA), followed by LSD to evaluate the significance of the difference between the mean value of the measured parameters in the respective test and control groups. A significant change was considered acceptable at $p < 0.05$.

RESULTS AND DISCUSSION

Table I shows the proximate analysis of chickpea and Bengal gram. Chickpea had a significantly ($p < 0.05$) higher moisture, protein and fat content compared to Bengal gram. Bengal grams on the other hand had higher ash content, dietary fibre, lignin and cellulose compared to chickpea.

Table I.- Proximate analysis of indigenous chickpea and Bengal gram (Mean±SD).

| Components percentage | Chickpea (KC-98) | Bengal gram (Desi Chana-Karak-1) |
|-----------------------|------------------|----------------------------------|
| Moisture | 10.32±0.59 | 8.16±0.51 |
| Ash | 2.70±0.18 | 3.24±0.30 |
| Protein | 26.10±0.25 | 24.32±0.50 |
| Fat | 5.16±0.17 | 2.52±0.40 |
| Dietary fiber | 4.72±0.14a | 9.33±0.44 |
| Lignin | 1.79±0.16 | 2.21±0.22 |
| Cellulose | 2.19±0.17 | 3.00±0.29 |

The mean value of various nutrients in chickpea and Bengal gram were as following, protein (27.3 vs. 25.4 %), fat (5.1 vs. 3.6%) carbohydrate (54.10 vs. 47.00%), crude fiber (2.7 vs. 10.9%), ash (2.9 vs. 4.5%), cellulose (1.3 vs. 1.5%) and lignin (1.9 vs. 2.0%). Several studies have also reported that chickpea and Bengal gram are rich in carbohydrates; however, the chickpea variety can have up to 59% of carbohydrates, whereas the Bengal gram can have about 38% of carbohydrates. The protein ranges from 18-25% in both the varieties, depending on their

Table II.- Blood glucose level of normal and alloxan-induced albino Sprague Dewley rats fed on BPD AIN-76, Kabuli Chickpea HFD and Bengal gram HFD.

| | Blood glucose Level (mg/dl) After Induction of diabetes on different weeks | | | | | | |
|--|--|-----------------|-----------------|-----------------|-----------------|-----------------|------------------|
| | Zero | 1 st | 2 nd | 3 rd | 4 th | 5 th | 6 th |
| 1. Normal on BPD_AIN-76 | 101.50±12.00 | 99.33±14.58 | 108.33±10.74 | 102.66±15.27 | 106.66±11.94 | 106.50±8.93 | 99.66±13.35 |
| 2. Induced diabetic on BPD AIN-76 | 100.66±15.55a | 264.33±19.52b | 253.66±46.66c | 242.16±25.49d | 262.16±14.83eh | 260.00±10.52fh | 267.00±14.1gh |
| 3. Induced diabetic on Chickpea HFD | 99.66±12.46a | 252.33±15.95b | 237.16±16.65ci | 220.83±14.20dhj | 219.33±17.00ej | 220.66±7.33fij | 218.16±11.66ghij |
| 4. Induced diabetic on Bengal gram HFD | 102.16±13.74a | 259.50±8.16b | 225.81±17.06c | 201.50±11.99dhj | 195.16±9.02ehj | 199.33±6.37fij | 197.83±10.62gij |

Mean values followed by different letter in a rows are significantly different at $\alpha=0.05$ BPD, basic protein diet, HFD, high fibre diet.

Table III.- Blood glucose level of normal and alloxan-induced albino Sprague Dewley rats fed on BPD AIN-76, Kabuli Chickpea HFD and Bengal gram HFD.

| | Blood glucose Level (mg/dl) After Induction of diabetes on different weeks | | | | | | |
|--|--|-----------------|-----------------|-----------------|-----------------|-----------------|------------------|
| | Zero | 1 st | 2 nd | 3 rd | 4 th | 5 th | 6 th |
| 1. Normal on BPD_AIN-76 | 101.50±12.00 | 99.33±14.58 | 108.33±10.74 | 102.66±15.27 | 106.66±11.94 | 106.50±8.93 | 99.66±13.35 |
| 2. Induced diabetic on BPD AIN-76 | 100.66±15.55a | 264.33±19.52b | 253.66±46.66c | 242.16±25.49d | 262.16±14.83eh | 260.00±10.52fh | 267.00±14.1gh |
| 3. Induced diabetic on Chickpea HFD | 99.66±12.46a | 252.33±15.95b | 237.16±16.65ci | 220.83±14.20dhj | 219.33±17.00ej | 220.66±7.33fij | 218.16±11.66ghij |
| 4. Induced diabetic on Bengal gram HFD | 102.16±13.74a | 259.50±8.16b | 225.81±17.06c | 201.50±11.99dhj | 195.16±9.02ehj | 199.33±6.37fij | 197.83±10.62gij |

Mean values followed by different letter in a rows are significantly different at $\alpha=0.05$ BPD, basic protein diet, HFD, high fibre diet.

subtype (Zia-Ul-Haq *et al.*, 2007). The fat content is higher in the chickpea being 4-5% as compared to the Bengal gram (2.0-2.5%). However the dietary fiber content is higher in Bengal gram 14-15% as compared to 3-4% of chickpea variety (Fernandez and Berry, 1988; Hulse, 1991; Maheri-Sis *et al.*, 2010).

Table II shows Blood glucose level of all the three treatment groups. Control group fed on BPD AIN-76 showed no significant change in the blood glucose level over the entire period of the experiment. The diabetic group fed on BPD AIN-76 showed a significant increase in the blood glucose level (from 100.66±15.55 mg/dl to 264.33±19.52 mg/dl) during the first week. This increase in the blood glucose level remained on the higher side almost throughout the observation period of 6 weeks.

The blood glucose level of diabetic group fed on chickpea HFD after the induction of diabetes showed a great increase in the blood glucose level (from 99.66±12.46 mg/dl to 252.33±15.95 mg/dl) during first week compared to zero week. After this increase recorded during the 2nd and 3rd week there was a significant decrease in blood glucose level which was maintained till the end of experiment.

The diabetic group fed on Bengal gram HFD also showed the significant increase in the blood glucose level during the 1st week after the induction of diabetes (102.16±13.74 to 259.50±8.16). Later it decreased during the 2nd week and 3rd weeks (225.81±17.06 to 201.50±11.99), and then remained unaltered during the rest of experimental period.

One Way ANOVA paired comparison post-hoc LSD test results showed that there was no significant difference ($p < 0.05$) in the random blood glucose level (RBGL) of normal and all other experimental groups at week 0. One week after the induction of diabetes however, the RBGL of the normal group fed on BPD AIN-76 was significantly lower than the rest of the groups and it continued to be the lowest for the rest of the period. However the RBGL of group fed on Bengal gram remained significantly ($p < 0.05$) lower than that of the diabetic group fed on BPD AIN-76 and the diabetic group fed on chickpea HFD throughout the experimental period.

The results of the present study suggest that both types of the chickpeas have a blood glucose lowering effect in the diabetic rats. Bengal gram HFD lowered the blood glucose level significantly ($p < 0.05$) more as compared to the chickpea HFD. Tiwari *et al.* (2013) showed that both chickpea and Bengal gram sprouts are beneficial in diabetic rats. The seeds and sprouts of chickpea and Bengal gram proved to mitigate starch-induced postprandial glycemic spikes in rats. Pittway *et al.* (2008) have shown relationship between the consumption of different varieties of chickpea on plasma glucose, insulin, and triglyceride

concentrations.

A similar study conducted by Nestel *et al.* (2004) on the effect of a single meal of chickpea on plasma glucose, insulin and triglyceride levels showed that after the chickpea meal plasma insulin and HOMA were significantly lower ($P < 0.05$) as compared to the regular meal. However, this study failed to illustrate the long-term, significant differences in plasma glucose, insulin, or HOMA either in the fasting state or after a glucose load. Haq *et al.* (2007) and Amjad *et al.* (2006) also observed lower insulinemic responses with single meals of fiber-rich foods yet no clear benefit when similar foods were consumed over several weeks.

Blood cholesterol

Table III shows the effect of the same high fiber diets on serum total cholesterol (STC) levels. The control group fed on BPD AIN-76 showed no change in blood cholesterol level throughout the period of six weeks. Hypercholesterolemic rats fed on BPD AIN-76; however, showed no significant change in STC during the first two weeks, but decreased significantly during the remaining experimental period.

The hypercholesterolemic rats fed on chickpea HFD showed significant increase in blood STC during the 1st week compared to the week zero. A significant decrease was recorded in 2nd week which was maintained during the subsequent experimental period. A slight increase in STC level observed in the last week was non significant.

The hypercholesterolemic group fed on Bengal gram showed significant increase in blood STC during 1st week compared to the week zero. During the 2nd and subsequent week the blood STC level decreased.

CONCLUSIONS

The nutritional evaluation of Kabuli chickpea and Bengal gram showed that former had higher moisture, protein and fat content, while the later had higher dietary fiber, cellulose and lignin contents.

Both the chickpea and Bengal gram high fiber diet decreased the blood glucose level of the alloxan-induced diabetic rats during six weeks of experiment compared to the diabetic rats basic purified diet AIN-76. However, the Bengal gram high fiber diet reduced the random blood glucose level of the diabetic rats significantly more than the chickpea high fiber diet. Similarly when the same high fiber diets were given to the rats with induced hypercholesterolemia it was noticed that the Bengal gram high fiber diet reduced the serum cholesterol level more than the chickpea HFD.

Statement of conflict of interest

The Authors declare no conflict of interest.

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