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Nutritional and antinutritional profile of newly developed chickpea (*Cicer arietinum* L) varieties

[1]Sharma, S., [1]Yadav, N., [1]Singh, A. and [2]Kumar, R.

[1]Centre of Food Technology, University of Allahabad, Allahabad, Uttar Pradesh, India
[2]Division of Genetics, IARI, New Delhi -110012, India

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Abstract

In this study the variability in nutritional composition, mineral profile, antinutritional factors and *in vitro* starch digestibility of five desi and four kabuli chickpea cultivars were studied. Proximate composition varied significantly ($p<0.05$) among different types of chickpea cultivars. The crude protein content varied from 18 to 31% being higher in kabuli chickpea cultivars than desi chickpea. The iron was the most abundant mineral present in all the cultivars of chickpea (4.6 to 10.5%). In addition appreciable amount of zinc was also present in all nine varieties of chickpea. Among antinutritional factors tannin concentration ranged from 0.07 to 0.22% and trypsin inhibitor's content ranged from 9 to 31 mg/g in both the cultivars of chickpea. *In vitro* starch digestibility was found significantly ($p<0.05$) higher in kabuli chickpea than desi chickpea cultivars. Among the analyzed chickpea cultivars K850 in desi and PUSA 1108, PUSA 1088 and PUSA1053 in kabuli cultivars had good potential as a food crop therefore their cultivation and utilization should be encouraged.

Introduction

Chickpea (*Cicer arietinum* L.) is one of the oldest and most widely consumed legume in the world due to relatively high protein content and wide adaptability as a food grain. It is the second most widely grown legume in the world (FAO, 2008). Chickpeas are good source of protein and carbohydrate. Its protein quality is better than other legumes such as pigeon pea, black gram and green gram (Kaur and Singh, 2005). According to the size, shape and color of the seeds, two types of chickpea are usually acknowledged. Kabuli chickpea is large seeded with salmon white testa, is grown mainly in the Mediterranean area, central Asia and America and Desi chickpea is small seeded with a light brown testa, is cultivated mostly in India and east Africa (Rincon *et al*., 1998).

It is generally accepted that the kabuli type was derived from desi type through mutation followed by conscious selection (Jana and Singh, 1993). Polymorphism has been also reported between Cicer arietinum and its wild genotype *Cicer reticulatum* (Udupa *et al*., 1993). Besides above mentioned factor, many other factors also affect seed quality such as cultivars, cultural practices and locality or environmental conditions (Elshiekh *et al*., 1999). Genetic differences in chemical composition must thus be evaluated while excluding the agro climatic effect. Poor nutritive value of this legume, due to the presence of certain antinutritional factors such as tannins, phytates and trypsin inhibitors has been also reported by some authors earlier (Siddhuraju *et al*., 2000). Trypsin inhibitors and tannins inhibit the digestibility of protein and starch, whereas, Phytic acid reduces the bioavailability of some essential minerals viz. iron and zinc etc. (Rehman and Shah, 2001).

The aim of the present work was to study the nutritional composition, mineral profile, antinutritional compounds (viz. tannin and trypsin inhibitors) and starch digestibility between desi (5 cultivars) and kabuli (4 cultivars) cultivars of chickpea.

Materials and Methods

Raw materials

Nine different cultivars of chickpea (*Cicer arietinum* L.) in their dried state were procured from 'Sardar Vallabhbhai Patel University of Agriculture and Technology', Meerut, India (Table 1). These varieties included five desi (dark brown) types (PUSA-1103, PUSA-362, JG-62, K-850, and JG-74) and four kabuli (white) types (PUSA-1105, PUSA-

*Corresponding author.
Email: *epitome.shruti@gmail.com*

1108, PUSA-1053, and PUSA-1088) of chickpea. The samples were cleaned by hand to remove dirt, grit and broken grains and then packed in air tight plastic containers at room temperature (30±2ºC). Thereafter the seed samples were pulverized using grinder. They were kept in airtight containers for quality analysis purposes.

Proximate composition

Seed samples from different chickpea cultivars were estimated for their moisture, ash and fat content as per standard methods of analysis (AOAC, 1990). Protein content was determined by Lowry's method (1951). Crude fiber was determined in the portion of the moisture and fat free sample that remained after digestion with acid and alkali.

Mineral composition analysis

Mineral analysis was done according to the standard method of analysis AOAC (2005). The mineral contents viz. Iron (Fe) and zinc (Zn) were determined using Atomic Absorption Spectrophotometer (Model No. AAS-700) (Perkin Elmer).

Determination of antinutritional factors

Tannin content

Tannin content in chickpea was determined by Folin-Denis method as described by Sadasivum and Manickam (2005). Color intensity was measured at 700 nm after 30 minutes of incubation period. Standard graph was prepared by using 0-100 µg tannic acid. Tannin content of the samples was calculated as per cent (%) tannic acid from the standard graph.

Trypsin inhibitor content (TI)

Trypsin inhibitor (TI) content was determined according to the method of Kakade *et al*. (1974) as modified by Hammerstrand *et al*. (1981) using BAPNA (N-a-Benzoyl-DL-Arginine p-nitroanilide hydrochloride) as a substrate. Trypsin inhibitor content was determined from the following formula:

$$\text{TI content (mg/g sample)} = \frac{\text{differential absorbance} \times \text{dilution factor}}{0.019 \text{ X } 1.000}$$

Determination of in vitro starch digestibility (IVSD)

In vitro starch digestibility (IVSD) was determined according to the method of Singh *et al*. (1982). 50 mg of ground chickpea sample was taken in a test tube and mixed with 1 ml of 0.2 M phosphate buffer (pH 6.9). To the sample suspension 0.5 ml pancreatic alpha amylase (Sigma,cat. No. 6880, 20 mg enzyme dissolved in 50 ml of the same buffer) was added and incubated at 37°C for 2 h. After the incubation period 2 ml of 3,5 DNS reagent (prepared by dissolving 200 mg crystalline phenol, 1 g 3,5 dinitrosalycylic acid and 50 mg sodium sulphite in 1% NaOH solution) was added immediately. The mixture was heated for 5-15 min in a boiling water bath. After heating 1 ml of 40% K-Na-Tartarate solution was added in the test tubes and allowed to cool at the room temperature (25ºC). Thereafter solution was made up to 25 ml with distilled water and filter prior to measurement of the absorbance at 550 nm. A blank was run simultaneously. A standard curve was prepared using maltose. Values were expressed as mg maltose released/100mg of flour.

Statistical analysis

All results in this study are reported as means of three replications. Results were analyzed with SPSS 7.5 software using one way analysis of variance (ANOVA), followed by Duncan's multiple range test to compare among means. Significance was defined at $p<0.05$.

Results

Proximate analysis

Proximate composition varied significantly ($p \leq 0.05$) among different cultivars of chickpea cultivars. The ash, crude fat, protein and crude fiber contents of flours from desi cultivars of chickpea ranged from 3.2% to 3.9%, 2.6% to 5.6%, 18% to 23% and 3.5% to 5.8%, respectively. In kabuli cultivars the values of the above mentioned parameters ranged from 3.0% to 3.6%, 3.1% to 6.8%, 28% to 31% and 3.8% to 4.1%, respectively (Table 1).

Mineral analysis

The results of the nutritionally valuable minerals viz. Iron (Fe) and zinc (Zn) are presented in Table 2. Significant varietal differences were also found in total content of iron and zinc among chickpea cultivars. Result showed that both Fe and Zn were present in the appreciable amount in desi and kabuli chickpea cultivars. Iron content ranged from 4.6 mg/100g to 10.5 mg/100g. Iron content was found highest in PUSA-1053 (kabuli cultivar) and lowest in JG-74 (desi cultivar). Zinc content ranged from 2.7 mg/100g for JG-62 to 5.8 mg/100g for PUSA-1103.

Tannin content

In present study tannin content ranged from 0.18 to 0.22 g/100g for desi cultivars of chickpea and 0.07 to 0.13 g/100g for kabuli cultivars of chickpea (Table 3).

Trypsin inhibitor content

Data on Trypsin inhibitor (TI) contents in desi and kabuli cultivars of chickpea are presented in Table

Table 1. Nutritional composition of desi and kabuli cultivars of chickpea (Dry weight basis)

Cultivars	Ash (%)	Protein (%)	Fat (%)	Crude fiber** (%)
Desi				
K 850	3.2±0.15^{ab}	23±0.57^{b}	5.6±0.10^{c}	5.8±0.26^{e}
PUSA1103	3.2±0.43^{ab}	18±1.52^{a}	2.6±0.28^{a}	4.4±0.45^{cd}
PUSA362	3.9±0.36^{c}	22±1.52^{b}	4.1±0.28^{b}	5.7±0.20^{e}
JG 62	3.7±0.30^{bc}	22±1.52^{b}	5.3±0.70^{c}	3.5±0.45^{b}
JG 74	3.3±0.26^{abc}	22±1.44^{b}	4.9±0.05^{c}	4.9±0.10^{d}
Kabuli				
PUSA1105	3.5±0.46^{abc}	28±1.00^{c}	6.8±0.35^{d}	3.8±0.10^{a}
PUSA1108	3.6±0.28^{abc}	31±0.57^{d}	5.2±0.25^{c}	3.4±0.17^{b}
PUSA1088	3.7±0.05^{bc}	29±0.57^{cd}	5.0±0.15^{c}	4.1±0.03^{c}
PUSA1053	3.0±0.11^{a}	28±1.00^{c}	3.1±0.40^{a}	3.7±0.02^{b}

*Values are means ± (S.D.) of triplicate analysis. Means followed by same superscript with in a column do not differ significantly (p≤0.05).
**Crude fiber values are expressed as fat free basis.

Table 2. Mineral composition of desi and kabuli biotypes of chickpea (dry weight basis)

Cultivars	Iron (mg/100g)	Zinc (mg/100g)
Desi		
K 850	8.6±0.20^{e}	5.3±0.32^{e}
PUSA1103	7.4± 0.20^{d}	5.8±0.10^{fg}
PUSA362	5.8±0.17^{c}	2.8±0.20^{b}
JG 62	5.1±0.26^{b}	2.7±0.26^{b}
JG 74	4.6± 0.20^{a}	3.6±0.32^{c}
Kabuli		
PUSA1105	10.3±0.20^{f}	5.5±0.20^{ef}
PUSA1108	6.1± 0.20^{c}	4.5 ±0.20^{d}
PUSA1088	7.1±0.22^{d}	2.2±0.26^{a}
PUSA1053	10.5±0.45^{f}	6.2±0.26^{g}

Values are means ± (S.D.) of triplicate analysis. Means followed by same superscript with in a column do not differ significantly (p≤0.01).

Table 3. Antinutritional factor contents of desi and kabuli biotypes of chickpea (mean ± SD, n=3; dry weight basis)

Cultivars	Tannin (%)	Trypsin inhibitor's content (mg/g)
Desi		
K 850	0.21±0.02^{c}	24±1.52^{e}
PUSA1103	0.19±0.01^{c}	22±2.64^{de}
PUSA362	0.20±0.01^{c}	31±3.00^{cd}
JG 62	0.22±0.02^{c}	17±2.64^{bc}
JG 74	0.18±0.03^{c}	25±2.08^{e}
Kabuli		
PUSA1105	0.08±0.01^{a}	12±3.78^{ab}
PUSA1108	0.13±0.03^{b}	16±1.52^{bc}
PUSA1088	0.12±0.03^{b}	11±2.08^{a}
PUSA1053	0.07±0.01^{a}	09±1.52^{a}

Means followed by same superscript with in a column do not differ significantly (p≤0.05).

3. The concentration of TI in desi cultivars ranged between 17 to 31 mg/g of sample, while in Kabuli cultivars it ranged between 09 to 16 mg/g of sample.

In vitro starch digestibility (IVSD)

Data on starch digestibility is presented in Figure 1. In present study *in vitro* starch digestibility of desi and kabuli cultivars of chickpea varied significantly. Among chickpea cultivars (desi and kabuli) in vitro starch digestibility in terms of maltose released

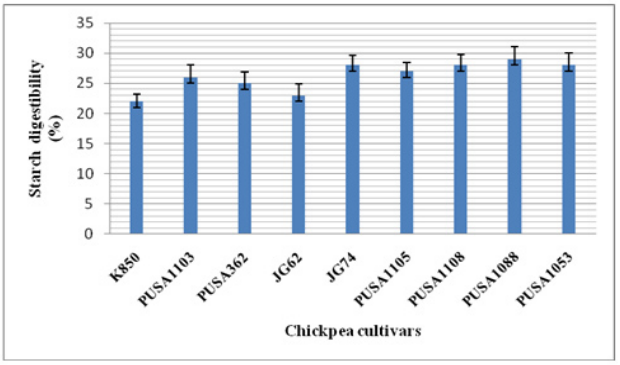

Figure 1. Starch digestibility of desi and kabuli chickpea cultivars (dry weight basis). Values are expressed as means ± S.D of triplicate analysis

ranged between 22 to 29 mg maltose released/100mg of flour and found lowest in K850 and highest in PUSA 1088.

Discussion

Results of the proximate composition are in line with Milan-Carillo *et al*. (2000) who have reported mean values for protein, lipid and ash content of 22.5, 5.01 and 2.98%, respectively for desi chickpea cultivars. Protein content are also in agreement with Singh and Jambunathan (1981) who compared 8 desi and 7 kabuli chickpea cultivars and found higher crude protein content for kabuli types (241 g/kg) than desi type (217g/kg). On the other part, Saini and Knights (1984) found no difference in crude protein content on comparing 7 varieties of desi with 7 varieties of kabuli chickpea cultivars. In addition to genetic differences, difference in crude protein content has been reported to depend on geographical origin of seed, although the contribution of location and season in the genotypic expression of protein content is generally small.

Nevertheless, crude fat content does not qualify these chickpea cultivars as an oil rich legume, especially when compared with groundnuts and soybeans (Vadivel *et al*., 2011). Results showed that kabuli cultivars had significantly higher fat content than desi cultivars which was in agreement with Rincon *et al*. (1998). Jana and Singh (1993) have studied geographical divergence in crude fat content and indicated that kabuli chickpea in Mediterranean basin is characterized by the high amount of the protein content so naturally they will have low fat content. So genetic selection in order to obtain higher protein content may explain the relative decreased of the fat content (Rincon *et al*., 1998). In our study desi types showed low fat levels while kabuli types displayed comparatively higher fat content. Among the nine cultivars of chickpea K-850 (desi cultivar) had the

highest crude fiber content (5.8 %) in comparison to other chickpea cultivars. The difference in proximate composition between flours from two different cultivars of chickpea viz. desi and kabuli in our study could be due to the inherited differences.

Significant varietal differences were also found in total content of iron and zinc among chickpea cultivars. Result showed that both Fe and Zn were present in the appreciable amount in desi and kabuli chickpea cultivars. Findings of this study corresponds to Singh and Jambunathan (1981) who examined mineral and trace element composition in eight desi and seven kabuli chickpea cultivars and found that iron content ranged between 2.3 to 4.1 mg/100g for desi and 8.2 to 16.8 mg/100g for kabuli chickpea cultivars. Similarly they also reported that zinc content ranged between 3.3 to 4.2 mg/100g for desi and 3.3 to 5.4 mg/100g for kabuli chickpea cultivars. Zia-Ul-Haq (2007) reported that iron and zinc content of four desi chickpea cultivars were ranged from 2.4 to 4.1% and 3.5 to 6.0%, respectively. These results revealed that both desi and kabuli cultivars of chickpea provide a sufficient amount of minerals to meet the human mineral requirement. Now a days Iron deficiency anemia is widely prevalent among pregnant women and young children in India and other developing countries. Therefore, selection of variety PUSA-1053 for cultivation can be the option to improve the mineral status of the vulnerable group.

Although legumes are important source of dietary protein and starch for human but their acceptability and utilization has been limited due to some antinutritional substances such as trypsin inhibitors, phytate, tannins etc. Tannins are polymeric flavonoids that comprise a small part of the broad and diverse group of phenolics compounds produced by plants as secondary metabolites (Diaz *et al*., 2010). Tannins have been reported to inhibit the digestive enzymes and there by lower the digestibility of most important nutrients especially protein and starch (Khattab and Arntfield, 2009). In the present study tannin content was found significantly ($p<0.05$) higher in desi chickpea than kabuli, because most of the tannins present in the colored seed coat of the legumes. Tannins also inhibit the utilization of nutrients through astringency and enzyme inhibition. As phenolics tannins are water soluble, they may be eliminated by thermal and hydrothermal processing treatments.

Trypsin inhibitors are widespread antinutritional substance which blocks trypsin activity and thereby reducing digestibility of protein. In this study TI content varied significantly ($p<0.05$) among desi and kabuli cultivars. It is very difficult to compare enzyme inhibitory potential of legumes, as reported by different authors, primarily, because of the difference in the methods and units used, Smith *et al*. (1980) reported trypsin inhibitor values (using the same method as used in this work) 16.6 mg/g to 30 mg/g of raw soybeans meal. Results also found in agreement with Kansal *et al*., 2008 who reported trypsin inhibitor content of 8.71 to 39.47 mg/g in ten desi chickpea cultivars. Kocchar *et al*., (1998) also reported TI content of 4.6 to 13.3 mg/g seeds of Vigna unguiculata. Abedowale *et al*. (2005) reported the values between 18 to 26 mg/g for 6 different species of *Mucuna* L.

Data on starch digestibility is presented in Figure 1. Starch digestibility in pulses is the result of the combined action of number of chemical and physical factors, such as size, composition and structure of the starch granules. Our results showed parity with Khatoon and Prakash (2006) who reported almost similar values of *in vitro* starch digestibility for green gram (9.5mg maltose released/100mg of flour) and horse gram (7.0 mg maltose released/100mg of flour). Various factor such as amylase inhibitors, phytate, etc. are supposed to affect the starch digestibility in legumes (Yadav and Khetarpaul, 1998). Reduced digestibility of starch lowers glucose release into the blood stream, which is beneficial for diabetic patients (Zia-Ul-Haq *et al*., 2007). However, digestibility of starch can be improved through heat treatments e.g. cooking, roasting and autoclaving.

Conclusion

The findings of this study demonstrate that the analyzed nine chickpea cultivars are a good source of protein. *In vitro* starch digestibility is higher than most common legumes. The presence of antinutritional factors identified in this paper should not pose a problem to humans if the seeds are properly processed. In view of the overall nutrient and proximate composition analysis, these desi and kabuli chickpea cultivars can be an economic and alternative protein source that could alleviate protein malnutrition in developing countries and improve overall nutritional status of functional food in the developed countries. More agronomic studies should be done on this legume with a view to cultivating it.

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