Full Length Research Paper

Nutritional evaluation of *kabuli* and *desi* type chickpeas (*cicer arietinum* L.) for ruminants using *in vitro* gas production technique

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Accepted 8 July 2008

The aim of the present study was to determine the chemical composition and estimation of nutritive value of *kabuli* and *desi* type chickpeas using *in vitro* gas production technique in sheep. The samples were collected from East Azerbaijan, Iran, pea packaging and processing factories. The feed samples (200 mg from each) were incubated with rumen liquor taken from three fistulated rams at 2, 4, 6, 8, 12, 24, 48, 72 and 96 h. The results showed that neutral detergent fiber (NDF) and crude fiber (CF) in *desi* type were significantly higher than that of *kabuli* (p < 0.01) while crude protein (CP), non fibrous carbohydrates (NFC) and soluble sugars in *kabuli* were significantly greater than that of *desi* (p < 0.05). Total tannins in *desi* type were higher than *kabuli* chickpeas (p < 0.05). There were no significant differences between dry matter (DM), organic matter (OM), ether extract (EE), starch and total phenolic compounds (TPC) content of the two experimental chickpea types. There were significant differences in organic matter digestibility (OMD), short chain fatty acids (SCFA) and metabolizable energy (ME) contents of the two chickpea types (p < 0.05). Gas productions at 24 h for *kabuli* type was higher than that of *desi* for ruminants.

Key words: Chickpea, gas production, metabolizable energy, nutritive value, sheep.

INTRODUCTION

Legume grains comprise an important part of the human diet in developing countries in tropical and subtropical areas, where their nutritional contribution is of paramount importance as a large segment of the populations in these areas have limited access to food of animal origin (Bressani, 1975; Ramalho Ribeiro and Portugal Melo, 1990). Chickpea (*Cicer arietinum* L.) is a major food legume in Southern Europe, North Africa, India and Middle East countries (Iqbal et al., 2006; Viveros et al., 2001). It is cultivated mainly in Algeria, Ethiopia, Iran, India Mexico, Morocco, Myanmar, Pakistan, Spain, Syria, Tanzania, Tunisia and Turkey (Naghavi and Jahansouz, 2005). There are two main types of chickpea, distinguished by seed size, shape and color: one produces relatively small seeds with an angular shape, dark color and called *desi*, the other produces large, rounded, cream color seeds and is called *kabuli* (Naghavi and Jahansouz, 2005; lqbal et al., 2006). *Kabuli* chickpea seeds are grown in temperate regions, whereas the *desi* type is grown in the semi-arid tropics (Naghavi and Jahansouz, 2005; lqbal et al., 2006). Both types of chickpea are grown in East Azerbaijan, Iran (Maheri-Sis et al., 2007).

Although most chickpeas are produced for human consumption, they provide the livestock industry with an alternative protein and energy feedstuff (Christodoulou, 2005). During the last decades there has been an increase in interest in their role in animal diets (Dixon and Hosking, 1992) due to ban of animal origin proteins and dissemination of using genetically modified organism (GMO) products (Lanza et al., 2003). Chickpea seed contains 29% protein, 59% carbohydrate, 3% fiber, 5% oil

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Figure 1. Iranian *desi* type chickpea.



Figure 2. Iranian kabuli type chickpea.

and 4% ash. Chickpea protein is rich in lysine and arginine but most deficient in sulphur-containing amino acids methionine and cystine (Iqbal et al., 2006). Chickpea is also a good source of absorbable Ca, P, Mg, Fe and K (Chavan et al., 1989; Christodoulou, 2005).

As the chemical composition of crops varies with crop cultivars, soil and climatic conditions of the area (Iqbal et al., 2006), it is imperative to study the chemical composition and nutritive value of the crops such as legumes cultivars grown in Iran.

Several methods such as *in vivo*, *in situ* and *in vitro* techniques have been used in order to evaluate the nutritive value of feedstuffs (Maheri-Sis et al., 2007). The in vitro gas production technique has proved to be a potentially useful technique for feed evaluation (Menke and Steingass, 1988; Getachew et al., 2004), as it is capable of measuring rate and extent of nutrient degradation. In addition, *in vitro* gas production technique provide less expensive, easy to determine (Getachew et al., 2004)) and suitable for use in developing countries (Chumpawadee et al., 2005; Maheri-Sis et al., 2007). This method also predicts feed intake, digestibility, microbial nitrogen supply, amount of short chain fatty acids, carbon dioxides and metabolizable energy of feed for ruminants (Babayemi, 2007).

Information on the nutritional value of chickpeas for ruminants especially in Iran is limited (Maheri-Sis et al., 2007). Thus, the objectives of this study were to determine the chemical composition and estimating nutritive value of *kabuli* and *desi* types of chickpea for ruminants using *in vitro* gas production technique.

MATERIALS AND METHODS

Animals and feeds

Three fistulated rams (1.5 years old, about 55 kg weights) were used for rumen liquor collection for application in gas production

technique. The experimental samples including *desi* (black coat; Figure 1) and *kabuli* (cream color; Figure 2) type chickpeas were collected from chickpea packaging and processing units in East Azerbaijan, Iran. The collected samples were mixed and milled through a 1 mm sieve in animal nutrition laboratory of Animal science research institute, Karaj, Iran.

Chemical analysis

Collected samples were milled through a 1 mm sieve for chemical analysis and gas production procedure. Dry matter (DM) was determined by drying the samples at 105° C overnight and ash by igniting the samples in muffle furnace at 525° C for 8 h. Nitrogen (N) content was measured by the Kjeldahl method. Crude protein (CP) was calculated as N x 6.25 (AOAC, 1990). Neutral detergent fiber (NDF) was determined by procedures outlined by Goering and Van Soest (1970) with modifications described by Van Soest et al. (1991); sulfite was obtained from NDF analysis. Starch content was determined by the method of MacRea and Armstrong (1968). Total phenolic compounds and tannin contents measured through techniques outlined by Khazaal et al. (1996).

In vitro gas production

Rumen fluid was obtained from three fistulated rams fed twice daily at the maintenance level with a diet containing alfalfa hay (60%) and concentrate (40%). The samples were incubated *in vitro* with the rumen fluid in calibrated glass syringes following the procedures of Menke et al. (1979). The 200 mg samples were weighed in triplicate into calibrated glass syringes of 100 ml. The syringes were prewarmed at 39°C before the injection of 30 ml rumen fluid-buffer mixture into each syringe followed by incubation in a water bath at 39°C. Readings of gas production were recorded before incubation (0) and 2, 4, 6, 8, 12, 24, 48, 72 and 96 h after incubation. Total gas values were corrected for blank incubation. Cumulative gas production data were fitted to the model of Orskov and McDonald (1979)

 $Y = a + b (1 - e^{-ct})$

Where: a = the gas production from the immediately soluble fraction (ml); b = the gas production from the insoluble fraction (ml); c = the

Constituents	kabuli	desi	Difference
Dry matter (DM)	92.08	91.17	NS
	%DM basis		
Organic matter (OM)	97.84	97.15	NS
Crude protein (CP)	24.63	22.76	*
Crude fibre (CF)	6.49	9.94	**
Neutral detergent fiber (NDF)	16.70	20.47	**
Ether extract (EE)	7.38	7.11	NS
Total tannin	0.09	0.125	*
Total Phenolic compounds (TPC)	0.270	0.265	NS
Non fibrous carbohydrate (NFC)	49.13	46.81	*
Starch	39.12	38.48	NS
Soluble sugars	8.43	7.53	*

Table 1. Chemical composition of kabuli and desi types of chickpea on dry matter basis (%).

NS: Non Significant; *: p < 0.05; **: p < 0.01

gas production rate constant for the insoluble fraction (ml/h); a + b = potential gas production (ml); t = incubation time (h); Y = gas produced at time t.

The non fibrous carbohydrates (NFC), short chain fatty acids (SCFA), organic matter digestibility (OMD) and metabilizable energy (ME) values in experimental chickpea types were calculated using equations as below:

NFC = 100 - (NDF + CP + EE + Ash) (NRC, 2001)

SCFA = 0.0222 Gas - 0.00425 (Makkar, 2005)

OMD = 0.9991 Gas + 0.0595 CP + 0.0181 CA + 9 (Menke and Steingass, 1988)

ME = 0.157 Gas + 0.0084 CP + 0.022 EE - 0.0081 CA + 1.06 (Menke and Steingass, 1988)

Where: Gas is gas production at 24 h incubation (ml/200 mg DM); a, b, c are gas production parameters described by Orskov and McDonald (1979) and NDF, CP, EE and CA are neutral detergent fiber, crude protein, ether extract and crude ash (% DM), respectively.

Statistical analysis

All of the data were analyzed by using software of SPSS (2002) and means of two sample groups were separated by independent-samples t-test (Steel and Torrie, 1980). All data obtained from three replicates (n = 3).

RESULTS AND DISCUSSION

Chemical composition of the two chickpea types are presented in Table 1. The neutral detergent fiber (NDF) and crude fiber (CF) in *desi* type were significantly higher than that of *kabuli* (p < 0.01) while crude protein (CP), non fibrous carbohydrates (NFC) and soluble sugars in *kabuli* were significantly greater than that of *desi* (p < 0.05). The NDF content of experimented Iranian *desi* was lower than Australian (20.47% vs. 25.19) but higher than that of Canadian (20.47 vs. 12.8%) *desi* type chickpeas.

While the NDF content of Iranian kabuli type was higher than those of Australian and Canadian (16.70 vs. 11.9 and 5%, respectively) kabuli types of chickpea (Wang and Daun, 2004). The NDF content of kabuli and desi types, were in agreement with several researches (Cordesse, 1990; Ramalho Ribeiro and Portugal Melo, 1990; Saskatchewan Pulse Growers, 2000). The CF content of kabuli and desi chickpeas in our study were higher than those of reported by Ramalho Ribeiro and Portugal Melo (1990), Saskatchewan Pulse Growers (2000) Saleh and El-Adawy (2006) and in line with Dixon and Hosking (1992) and Viveros et al. (2001). The CP content of *desi* and *kabuli* types were lower than reported by Viveros et al. (2001), higher than reports of Saskatchewan Pulse Growers (2000) and in line with Ramalho Ribeiro and Portugal Melo (1990), Wang and Daun (2004), and Igbal et al. (2006). The NFC content of experimental chickpea types was lower than that of reported from Greece (Christodoulou et al., 2005). The soluble sugars contents were in agreement with Ramalho Ribeiro and Portugal Melo (1990), and Wang and Daun (2004).

Total tannins in Iranan *desi* type were higher than *kabuli* chickpeas (p < 0.05), while in the study of Viveros et al. (2001), in Spain, tannin contents of *kabuli* was higher than that of *desi* types. Total tannins content of chickpea types in our study were in line with Australian chickpeas (Wang and Daun, 2004) and lower than that of El-Niely (2007), findings.

There were no significant differences between dry matters (DM), organic matter (OM), ether extract (EE), starch and total phenolic compounds (TPC) content of two experimental chickpea types. The starch content of experimented chickpea types was lower than Ramalho Ribeiro and Portugal Melo (1990), higher than Viveros et al. (2001), and in agreement with Saleh and El-Adawy (2006) and Canadian chickpea types (Wang and Daun, 2004). The OM content were in line with several studies

Items	kabuli	desi	Difference
Gas volume at 24 h (ml/200 mg DM)	78.66	73.96	*
a(ml)	-15.64	-16.11	NS
b(ml)	107.30	104.35	*
c(ml / h)	0.074	0.071	*
OMD (%)	89.09	84.29	*
SCFA (mmol)	1.742	1.638	*
ME (MJ / kg DM)	13.76	13.00	*

Table 2. Gas production parameters, dry matter digestibility (DMD), organic matter digestibility (OMD), short chain fatty acids (SCFA) and metabolizable energy (ME) contents of *kabuli* and *desi* types of chickpea.

a = Gas production from the immediately soluble fraction (ml); b = gas production from the insoluble fraction (ml); c = gas production rate constant for the insoluble fraction (b); NS: non significant; *: p < 0.05.

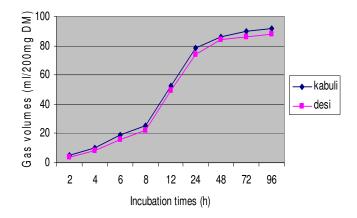


Figure 3. Gas production in *kabuli* and *desi* type chickpea at difference incubation times.

1992; Saskatchewan (Dixon and Hosking, Pulse Growers, 2000; Viveros et al., 2001; Wang and Daun, 2004; Christodoulou et al., 2005) and higher than Ramalho Ribeiro and Portugal Melo (1990), Cordesse (1990), and Igbal et al. (2006). The wide range of variation in chemical composition of chickpea can be due to different crop cultivars, seed source, growing conditions (and geographic, seasonal variations, climatic conditions and soil characteristics), type of seed, extent of foreign materials and impurities (Shridhar et al., 2005; lobal et al., 2006; Maheri-Sis et al., 2007), Different chemical composition leads to different nutritive value, because chemical composition is one of the most important indices of nutritive value of feeds. Variation in chemical components of feeds such as starch, NFC, OM, CP NDF and soluble sugars contents can result in variation of in vitro gas production extent (Getachew et al., 2004). Gas production volumes (ml/200 mg DM) in different incubation times (Figure 3), gas production parameters (a, b, c) and calculated amounts of SCFA, OMD and ME of kabuli and desi type chickpeas are presented in Table 2.

The gas volumes for *kabuli* in different incubation times were higher than that of *desi* type. Gas volume at 24 h incubation (for 200 mg dry samples), soluble fraction (a), insoluble but fermentable fraction (b), for kabuli was 78.66, -15.64 and 107.30 and for *desi* were 73.96, -16.11 and 104.35 ml, respectively. The negative (a) value for both chickpea types due to delay in onset of fermentation and microbial attachment was in agreement with Chumpawadee et al. (2005). Rate of gas production expressed in ml/h in kabuli (0.074) was significantly (P < 0.05) greater than desi (0.071). The gas volume after 24 h incubation in current study was higher than reported by Ramalho Ribeiro and Portugal Melo (1990). Different gas production in these studies can be due to different chemical constituents of chickpeas, animal types and breeds and quality of inoculums source (Menke et al., 1979; Getachew et al., 2004). There was a positive correlation between NFC content of feeds and gas production, but feed CP, NH₃-N and NDF levels were negatively correlated with gas production (Getachew et al., 2004; Maheri-Sis et al., 2007).

The ME, SCFA and OMD of kabuli were significantly higher than that of desi (P< 0.05). The ME content of kabuli and desi in this experiment were 13.76 and 13.00 MJ/Kg DM, respectively, and were higher than reported by Hawthorne (2006) for Australian varieties (12.1 MJ/ Kg DM) and almost in line with range of Mediterranean chickpeas (11.8 - 13.2 MJ/ Kg DM) reported by Ramalho Ribeiro and Portugal Melo (1990) and Dixon and Hosking (1992). The ME (MJ/Kg DM), SCFA (mmol) and OMD (%) for kabuli culled chickpea were 13.26, 1.70 and 85.76, respectively (Maheri-Sis et al., 2007). High energy content of kabuli than desi chickpeas also previously has been reported by Saskatchewan Pulse Growers, (2000), Viveros et al. (2001). The reason for energy content of kabuli being higher than desi can be due to difference in chemical composition (especially soluble carbohydrates, CP, NFC and NDF) and volume of gas production (Menke and Steingass, 1988; Getachew et al., 2004). The OMD of chickpea varieties (79.7 - 88.8%) reported

by Ramalho Ribeiro and Portugal Melo (1990) confirm our findings on chickpea types (84.29 - 89.09%). The SCFA content of *kabuli* type (1.742 mmol) was significantly higher than that of *desi* (1.638 mmol) chickpeas (P< 0.05). Blummel et al. (1999) reported that the gas volume in the bicarbonate buffered Hohenheim *in vitro* gas production test reflect SCFA production very closely. Gas volumes were produced quantitatively and qualitatively as a result of SCFA production (the amount of fermentative CO₂ and CH₄ could be accurately calculated from the amount and proportion of acetate, propionate and butyrate present in the incubation medium). Thus increasing amount of SCFA was lead to increase in gas production which is resulted in high digestibility and energetic value.

Conclusion

Overall, the nutritive value (chemical composition, gas production characteristics, organic matter digestibility and metabolizable energy content) of *kabuli* type chickpeas were better than that of *desi*. However, both types of chickpea can be used as potential energy and protein sources in ruminant nutrition.

ACKNOWLEDGMENTS

This research was carried out as Ph.D. thesis in animal sciences in Islamic Azad University – Sciences and Research branch. The authors are grateful to the laboratories of Animal Science Research Institute, Karaj, Islamic Azad University- Shabestar Branch and Sciences and Research Branch.

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