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Research Article

Quality evaluation of fermented chickpea (*Cicer arietinum*)

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ABSTRACT

Chickpea (*Cicer arietinum* L.) is a most important legume crop that cultivated and utilized world-wide and good source of protein. The current study was designed to develop fermented chickpea dip and analyzed fermented chickpea dip qualitatively. For this, proximate analysis of chickpea were performed to examine its nutritional quality. Then fermentation of chickpea was done by using lactic acid bacteria. For minerals and heavy metals analysis, assessment of fermented chickpea dip was conducted by using atomic absorption and flame photometer. Proximate composition of chickpea indicated that it contained 11.08±0.14% moisture, 19.05±1.24% crude protein, 6.65±0.15% crude fat, 3.67±0.44% ash and 6.06±0.19% crude fiber. Physicochemical evaluation results for fermented chickpea dip showed that 67.13±0.18% moisture, 26.33±0.22% crude protein, 11.70±0.05% crude fat, 2.67±0.05% ash and 5.11±0.03% crude fiber. Organoleptic analysis of fermented chickpea dip indicated that flavor, texture and aroma was ranked as acceptable by the evaluation panel whereas color and taste declined with the passage of time. Mineral profile of fermented chickpea dip contained 120.88±0.15mg/100g sodium (Na), 173.40±0.18mg/100g magnesium (Mg), 879.72±0.16mg/100g potassium (K), 139.79±0.14mg calcium (Ca), 5.36±0.01mg/100g iron (Fe), 1.15±0.02mg/100g copper (Cu), 3.84±0.02mg/100g zinc (Zn).

Keywords: chickpea dip, organoleptic analysis, physicochemical attributes, lactic acid.

1. INTRODUCTION

Chickpea (*Cicer arietinum* L.) is one of the oldest legume owing to good protein and wide versatility as a food grain. It is the second most commonly grown legume of the world (FAO, 2003). Cultivated chickpea consist of two different varieties, Desi and Kabuli. Desi (*microsperma*) variety contains anthocyanin stain on stems, pink flowers, and a colored and thick seed coat (Frimpong *et al.*, 2009). Kabuli variety is mostly cultivated in North Africa, West Asia, North America and Europe. The local seed varieties are black, brown or green in color and are grown as leguminous crop (Iliadis, 2001).

Chickpea is a rich source of protein (varies from 12.6 to 31.4%) and carbohydrates, which collectively can contribute about 8.78% of total dry seed mass. The protein present in chickpea is rich in arginine but lowering amount of Sulphur-containing amino acids methionine and cysteine (Iqbal *et al.*, 2006). The percentage of starch in chickpea is varying from 40% to 48%. Kabuli variety of chickpea contains soluble sugars in excess amount (Harsha *et al.*, 2014).

The chickpea contains sufficient vitamins like vitamin A, niacin, thiamine and folate.

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The poor consumer that lives on vegetarian diet needs 6% protein which is present in chickpea. The amount of fiber is different in both varieties of chickpea kabuli and desi (Lev-Yadun *et al.*, 2000).

The chickpea used as a component of our regular diet and aids in reduction of blood pressure because it contains β -sitosterol, linoleic acid and phytosterol. The oil of chickpea seed consists of tocopherols, sterols and tocotrienols. These phytosterols are explained to confirm the anti-bacterial, anti-inflammatory, anti-fungal and anti-ulcerative properties which can help in the reduction of cholesterol level, cardiovascular and cancer disease (Moreau *et al.*, 2002)

Along with high nutritional profile and various health benefits, chickpea contains anti-nutritional factors. Anti-nutritional factors include trypsin, chymotrypsin inhibitors, phytic acid/phytate, flavonoids, phyto hemagglutinins (lectins), α -amylase inhibitors, phenolics, tannins, saponins, certain goitrogens and oxalic acid. These anti-nutritional factors reduce the protein availability and digestibility by making the bonds with protein and other minerals (Jukanti *et al.*, 2012). So, it is necessary to breakdown or reduce the level of anti-nutritional factors to enhance the protein availability and digestibility.

The use of different processing methods such as fermentation, microwave cooking, boiling, germination, soaking and autoclaving proves effective to reduce anti nutritional factors. By use of different treatment there is a significant reduction in protease inhibitors and no hema-glutinating activity in chickpeas (El-Adawy *et al.*, 2002).

Fermentation is a best process of biodegradation to increase the nutritive profile of pulses by enhancing the level of essential nutrients and decreasing the level of anti-nutritional factors. It ultimately contributes towards the improvement in the

in-vitro digestibility of pulses (Granito *et al.*, 2009). When fermentation occurs in cereals different volatile compounds are formed, which are responsible for a complex blend of flavors in products. Due to the presence of acetic acids and butyric acids in cereals-based products, aroma is produced, making fermented cereal-based products more appetizing (Blandino *et al.*, 2003).

2. MATERIALS & METHODS

2.1. Procurement of material

The chemicals were made available in the laboratory. Chickpea was provided by Institute of Plant Breeding and Biotechnology, MNS-University of Agriculture, Multan, Pakistan. The equipment used for this study was provided by the Central lab, MNS-University of Agriculture, Multan, Pakistan.

2.2. Nutritional composition of chickpea

The percentage composition of chickpea was determined for crude fat, crude protein, ash, crude fiber and moisture contents according to methods of AOAC (2000).

2.3. Moisture analysis

Moisture contents of chickpea flour determined in hot air oven according to the procedure followed by AACC (2000). About 5g sample of chickpea flour was weighed in already weighed china dish and then placed in drying oven at 105°C for 24 hours. After drying, samples were again weighed and noted the final reading which was a percentage of actual moisture calculated by using the equation shown below.

$$\text{Moisture \%} = \frac{\text{weight of actual sample} - \text{weight of dried sample}}{\text{weight of actual sample}} \times 100$$

2.4. Ash

Ash percentage of chickpea flour was evaluated by the process as defined in AACC (2000). About 10 g sample of chickpea flour was weighed in crucible and then put into the muffle furnace at 550°C for 5 hrs. After 5 hrs the flour was converted into grayish white residue. The percentage of ash was estimated

by the equation given below. The percentage of ash was estimated by the equation given below.

$$\text{Ash \%} = \frac{\text{weight of grayish residues}}{\text{weight of the actual sample}} \times 100$$

2.5. Crude protein

Protein contents of chickpea was estimated through Kjeldhal apparatus by adopting the method of AOCC (2000). About 5g of sample of chickpea was added in digestion flask. The digestion process was carried for 4-5 hours in the digestion tube into the digestion unit for 4-5 hours and when green color residues are starts to develop in digestion tube then cool the sample and then put into the 100ml volumetric flask. Then neutralize the mixture with almost 70ml of sodium hydroxide solution (NaOH) for purpose of ammonia gas releasing. Then this neutralized solution of sample was distilled in Kjeldahl's distillation apparatus. Then titration process of ammonia was started with 0.1N solution of sulphuric acid until the purple color was appeared. The percentage of crude protein of chickpea was expressed by following equation.

$$\text{Nitrogen(\%)} = \frac{\text{Titrate value} \times \text{Normality of sulphuric acid} \times 0.0014 \times \text{Dilution}}{\text{Weight of sample} \times \text{volume}} \times 100$$

2.6. Crude fat

Fat contents of chickpea was subjected through Soxhlet apparatus regarding the protocol of which is described by AOAC (2000). 5g sample of chickpea was properly weighed into extraction thimble which was clean on weighing balance and then placed into a Soxhlet apparatus. Petroleum ether used for the extraction of fat and extraction process was carried out for about 5-6 hours. After evaporation of solvent flask was completely dried at 105°C in the hot air oven. After completion of drying sample containing flask was properly cooled in desiccator and then amount of fat is measured. The crude fat was calculated as a % of the dry mass of the chickpea used.

$$\text{Crude fat (\%)} = \frac{W1 - W2}{W} \times 100$$

2.7. Crude fiber

Crude fiber of chickpea was estimated through the procedure of AOCC (2000). In this method weighed 5g of fat free chickpea sample was taken and then digested with 200 mL boiling solution of 1.25% H₂SO₄. After digestion, sample was filtered and washed with hot water. This washing process was repeated three times. After digestion, sample was placed in the muffle furnace at 600°C to complete the churning process. After the completion of churning process, sample was placed into the desiccators for controlling the moisture and then sample was again weighed.

$$\text{Crude Fiber(\%)} = \frac{W2 - W1}{W}$$

2.8. Mineral profile

In chickpea, minerals were estimated by following procedure of AOAC (2006). Weighed 5g and then placed into a conical flask. After pouring the sample in conical flask then 5ml HClO₄ and 10ml HNO₃ solutions were added into the sample and put the flask on heating chamber for maintaining the temperature of 180°C. Heating was stopped when 2-3ml solution was left. After completion of heating process then sample was dilute up to 25ml and run it on flame photometer. Same digested material can be used for running on atomic absorption spectrophotometer. Only K and Na were measured through Flame photometer while Ca and Mg were determined through Atomic absorption spectrophotometer.

2.9. Lactic acid bacterial treatment of chickpea

The pulses sample were inoculated with different cultures *Lactobacillus bulgaricus* and *Lactobacillus acidophilus* by using 2% inoculum size (10⁸ cfu/mL) and incubated temperature was 37±2°C. The inoculum was prepared by inoculation of skim milk with the selected strains and subsequent incubation at 37±2°C for 24 hours. The fermented samples were dried in hot air oven at 40-50°C and dried samples were grounded and stored in were drawn at 0, 2, 4, 6, 8 and 10 hours of fermentation. The distilled water in replacement of culture was used as a control

treatment. Selected pulses will be treated with lactic acid bacteria at 37°C for 24 hrs. Anaerobic conditions will be continued by leaving the conduct experiment within a rectangular jar (Yousif *et al.*, 2000).

2.10. Product development

Fermented chickpea dip developed by following the fermentation process documented by Singh (2012). Flow diagram of fermented chickpea dip are given in Fig 1.

2.11. Product analysis

2.11.1. Physico-chemical analysis

The prepared fermented chickpea was stored at 4-6°C and analyzed for physico-chemical parameters like, moisture, ash, crude fiber and crude protein by following the method of AOAC (2000).

2.11.2. pH

By the help of digital pH meter, the pH of fermented chickpea was determined (InoLab 720, Germany) according to AOAC (2006).

2.11.3. Acidity

The acidity of fermented chickpea was determined adopting the protocol of AOAC (2006). The samples were titrated against 0.1N sodium hydroxide solution until pink color is appeared.

2.11.4. Mineral profile

Mineral contents of fermented chickpea were determined through Atomic Absorption Spectrophotometer and Flame Photometer (AOAC, 2006).

2.11.5. Sensory evaluation

Suitability and consumer acceptability of fermented chickpea was determined by using Hedonic Rating Scale with 6 sensory parameters, (aroma, appearance, body texture, taste, overall acceptability and mouth feel) through a panel of assessors (Khattab *et al.*, 2009).

2.11.6. Statistical analysis

The obtained data from research was subjected to analysis of variance (ANOVA) by using STATISTIX (Version 8.1) software as prescribed by Steel *et al.*, (1997).

3. RESULTS AND DISCUSSION

3.1. Proximate study of chickpea

The proximate composition of chickpea showed that it contained 11.08±0.14%, 6.65±0.15%, 6.06±0.19%, 3.67±0.44% and 19.05±1.24% of moisture, crude fat, crude fiber, ash and crude protein, respectively (Table 1). The results further showed high protein contents (19.05±1.24%) in the chickpea which was an indicator to be a valuable and cheapest source of protein to overcome the protein deficiency and malnutrition. The present results are well agreed with the findings of Muhammad *et al.* (2007) during working on comparison of proximate composition of some cultivars of chickpea (*Cicer arietinum* L.). They were found that protein contents 12.72% to 19.46%, crude fiber 7.1 %.

Table 1: Physico-chemical analysis of chickpea

Parameters	Values (%)
Moisture	11.08±0.14
Fat	6.65±0.15
Fiber	6.06±0.19
Ash	3.67±0.44
Protein	19.05 ±1.24
Total plate count	6 ±1.25 CFU/mg
Total phenolic contents	40.92± 0.11mg/kg

Mean±S.E

3.2. Mineral profile of chickpea

The mineral composition of raw chickpea seeds showed that it contained 122.00±2.04mg/100g, 161.00±8.73mg/100g, 1246.40±13.86mg/100g, 149.40±9.46mg/100g, 5.72±0.38mg/100g, 1.14±0.09mg/100g and 3.11±0.27mg/100g of Na, Mg, K, Ca, Fe, Cu and Zn respectively (Table 2). The results further showed that the chickpea contained high quantity mineral contents especially K, Ca and Fe which ultimately defined its potential nutritional benefits. The present result of mineral contents are closely in resemblance to the result of Kerem *et al.* (2016) during work on

the nutritional value and health benefits of chickpeas they found that Calcium 57 mg/100g, Iron 4.31mg/100g, Magnesium 79mg/100mg, Potassium 718mg/100g, Sodium 24mg/100g, Zinc 2.76mg/100g, Copper 0.656mg/100g.

Table 2: Mineral profile of raw chickpea

Parameters	mg/100g
Na	122.00±2.04
Mg	161.00±8.73
P	419.40±10.74
K	1246.40±13.86
Ca	149.40±9.46
Mn	3.36±0.31
Fe	5.72±0.38
Cu	1.14±0.09
Zn	3.11±0.27

Mean±S.E

3.3. Product development

Chickpea dip is chickpeas-based product made with mashed chickpeas, salt, garlic, and little amount of sesame paste. From past few year chickpea dip consumption increased rapidly day by day. Due to many health benefits and higher amounts in protein and lower in fat chickpea dip is better from other spreads or dip (Browne, 2011).

3.4. Proximate study of chickpea dip

The proximate composition of chickpea dip showed that it contained 63.46±0.18 to 67.92±0.12 moisture, 3.34±0.02 to 2.97±0.05 ash, 22.05±0.32 to 23.06±0.01 crude protein, 12.07±0.01 to 11.70±0.03 crude fat, 5.62±0.03 to 5.25±0.03 crude fiber, 5.18±0.03 to 4.75±0.03 pH and 0.5±0.02 to 0.59±0.01 acidity from zero days of storage to 10th day of storage (Table 3a). While control (non-fermented) group of chickpea dip contain 62.44±0.04, 3.62±0.04, 18.87±0.17, 12.12±0.01, 5.78±0.04, 5.26±0.04, 0.48±0.01 moisture, ash, crude protein, crude fat, crude fiber, pH and acidity, respectively. While fermented group of chickpea dip contain 67.13±0.18, 2.67±0.05, 26.33±0.22, 11.70±0.05, 5.11±0.03, 4.66±0.04, 0.62±0.01 moisture, ash, crude

protein, crude fat, crude fiber, pH and acidity, respectively (Table 3b).

Table 3a: Physico-chemical analysis of fermented chickpea dip

Parameter	% Value
Moisture	67.13±0.18
Fat	11.7±0.05
Fiber	5.11±0.09
Ash	2.67±0.05
Protein	26.33±0.22

Mean±S.E

Table 3b: Physico-chemical analysis of non-fermented chickpea dip

Parameter	% Value
Moisture	62.44±0.047
Fat	12.14±0.35
Fiber	5.89±0.16
Ash	3.62±0.04
Protein	18.78±0.17

Mean±S.E

According to Blandino *et al.*, (2003) chickpea fermented dip showed higher increase in protein and ash. Fermentation leads to production of acids and probable bacteriocin that prevent growth of microorganism hence increasing shelf life of fermented products Blandino *et al.*, (2003). According Rachwarosiak *et al.*, (2015) the crude fat of the raw sample was 19.05 ±1.24%. There was significant decrease in the fat content of the fermented sample from 12.12±0.01% to 11.70±0.05 %. According to Rachwarosiak *et al.*, (2015) the crude fiber of unprocessed sample was 5.78±0.04% and the fermented sample had the lowest crude fiber of 5.11±0.03%.

According to Blandino *et al.*, (2003) the moisture content of fermented chickpea dip ranged from 62.44±0.04 to 67.13±0.18 respectively. The moisture content of the fermented chickpea dip was significantly different ($p < 0.05$) from each other. The lower moisture content of all the samples tested showed that they will have better keeping quality.

3.5. Mineral contents of fermented chickpea dip

The mineral contents of chickpea dip overall mean showed that it contained 116.4±0.08 to 118.4±0.13 Na, 164.0±0.10 to 164.4±0.22 Mg, 874.4±0.14 to 872.8±0.28 K, 136.0±0.15 to 137.2±0.12 Ca, 4.7±0.01 to 4.9±0.01 Fe, 1.0±0.01 to 1.1±0.01 Cu and 3.5±0.01 to 3.7±0.01 Zn from zero days of storage to 10th day of storage (Table 4a). While control

(non-fermented) group of chickpea dip contain 113.68±0.09, 155.21±0.13, 867.21±0.28, 133.37±0.12, 3.94±0.04, 0.95±0.01, 3.33±0.01 Na, Mg, K, Ca, Fe, Cu and Zn (Table 4b). Fermented group of chickpea dip contain 120.88±0.15, 173.40±0.18, 879.72±0.16, 139.79±0.14, 5.36±0.01, 1.15±0.02, 3.84±0.02 Na, Mg, K, Ca, Fe, Cu and Zn, respectively (Table 4c).

Table 4a: Overall mean value for mineral profile of chickpea dip

Minerals	Time period					
	0 day	2 nd day	4 th day	6 th day	8 th day	10 th day
Na	116.4±0.08	116.7±0.15	117.1±0.12	117.3±0.15	118.0±0.10	118.4±0.13
K	874.4±0.14	873.8±0.18	873.6±0.19	873.3±0.23	872.9±0.29	872.8±0.28
Mg	164.0±0.10	164.2±0.12	164.4±0.09	164.4±0.17	164.4±0.21	164.4±0.22
Ca	136.0±0.15	136.2±0.09	136.5±0.16	136.7±0.15	137.0±0.12	137.2±0.12
Fe	4.7±0.01	4.5±0.03	4.5±0.07	4.6±0.03	4.8±0.01	4.9±0.01
Cu	1.0±0.01	1.0±0.01	1.0±0.01	1.1±0.01	1.1±0.01	1.1±0.01
Zn	3.5±0.01	3.5±0.01	3.6±0.01	3.6±0.01	3.6±0.01	3.7±0.01

Mean±S.E

Table 4b: Mineral profile of non-fermented chickpea dip

Minerals	Time period					
	0 day	2 nd day	4 th day	6 th day	8 th day	10 th day
Na	114.4±0.07	113.9±0.06	113.8±0.12	113.6±0.12	113.3±0.09	113.2±0.09
K	870.2±0.21	868.5±0.18	867.6±0.23	866.6±0.31	865.5±0.44	864.9±0.41
Mg	156.4±0.12	156.0±0.12	155.7±0.07	155.2±0.12	154.3±0.18	153.5±0.18
Ca	133.4±0.12	133.3±0.06	133.4±0.12	133.3±0.18	133.4±0.12	133.4±0.12
Fe	4.1±0.01	3.8±0.06	3.6±0.12	3.8±0.03	4.1±0.01	4.2±0.01
Cu	1.0±0.01	1.0±0.01	1.0±0.01	1.0±0.01	0.9±0.01	0.9±0.00
Zn	3.3±0.00	3.3±0.00	3.3±0.01	3.3±0.01	3.3±0.01	3.3±0.01

Mean±S.E

Table 4c: Mineral profile of fermented chickpea dip

Minerals	Time period					
	0 day	2 nd day	4 th day	6 th day	8 th day	10 th day
Na	118.4±0.09	119.4±0.23	120.4±0.12	121.0±0.19	122.6±0.12	123.5±0.18
K	878.5±0.17	879.1±0.18	879.7±0.15	880.0±0.15	880.3±0.15	880.7±0.15
Mg	171.6±0.09	172.4±0.12	173.0±0.12	173.6±0.23	174.5±0.23	175.3±0.26
Ca	138.5±0.18	139.0±0.12	139.6±0.21	140.0±0.12	140.6±0.12	141.0±0.12
Fe	5.2±0.01	5.2±0.01	5.3±0.02	5.4±0.02	5.5±0.01	5.6±0.01
Cu	1.1±0.01	1.1±0.02	1.1±0.02	1.2±0.02	1.2±0.02	1.3±0.02
Zn	3.7±0.02	3.8±0.01	3.8±0.02	3.9±0.02	3.9±0.02	4.0±0.02

Mean±S.E

3.6. Sensory study of chickpea dip

The sensory study of chickpea dip overall mean showed that it contained 8.75±0.17 to 6.33±0.33 color, 8.85±0.12 to 7.42±0.31 taste, 8.58±0.23 to 6.17±0.17 aroma, 8.75±0.17 to 6.50±0.17 texture, 8.75±0.17 to 7.67±0.17 consistency, 8.4±0.05 to 8.22±0.03 overall acceptability from zero days of storage to 10th days of storage (Table 5a). While control (non-fermented) group of

chickpea dip contain 7.61±0.30, 8.40±0.20, 7.67±0.25, 7.89±0.19, 8.22±0.17, 8.32±0.04 color, taste, aroma, texture, consistency and overall acceptability (Table 5b). Fermented group of chickpea dip contain 7.56±0.24, 7.58±0.44, 7.31±0.16, 7.64±0.21, 8.31±0.19, 8.27±0.03 color, taste, aroma, texture, consistency and overall acceptability, respectively (Table 5c).

Table 5a: Overall mean for sensory evaluation of chickpea dip

Sensorial Attributes	Time period					
	0 day	2 nd day	4 th day	6 th day	8 th day	10 th day
Color	8.75±0.17	8.58±0.23	7.75±0.30	7.25±0.25	6.85±0.33	6.33±0.33
Taste	8.85±0.12	8.68±0.19	8.08±0.33	7.50±0.55	7.42±0.42	7.42±0.31
Aroma	8.58±0.23	8.25±0.25	7.92±0.17	7.42±0.17	6.58±0.25	6.17±0.17
Texture	8.75±0.17	8.33±0.17	8.08±0.23	7.67±0.23	7.25±0.17	6.50±0.17
Consistency	8.75±0.17	8.75±0.17	8.33±0.17	8.25±0.17	7.83±0.23	7.67±0.17
Overall Acceptability	8.4±0.05	8.35±0.03	8.33±0.06	8.25±0.03	8.22±0.02	8.22±0.03

Mean±S.E

Table 5b: Sensory evaluation of non-fermented chickpea dip

Sensorial Attributes	Time period					
	0 day	2 nd day	4 th day	6 th day	8 th day	10 th day
Color	8.83±0.17	8.67±0.17	7.67±0.44	7.33±0.44	6.83±0.33	6.33±0.33
Taste	8.87±0.07	8.87±0.09	8.17±0.17	7.83±0.44	8.17±0.17	8.50±0.29
Aroma	8.67±0.17	8.33±0.33	8.17±0.17	7.67±0.17	6.83±0.33	6.33±0.33
Texture	8.83±0.17	8.5±0.29	8.17±0.17	7.83±0.17	7.33±0.17	6.67±0.17
Consistency	8.67±0.17	8.83±0.17	8.33±0.17	8.17±0.17	7.67±0.17	7.67±0.17
Overall Acceptability	8.43±0.07	8.47±0.03	8.37±0.07	8.23±0.03	8.23±0.03	8.17±0.03

Mean±S.E

Table 5c: Sensory evaluation of fermented chickpea dip

Sensorial Attributes	Time period					
	0 day	2 nd day	4 th day	6 th day	8 th day	10 th day
Color	8.67±0.17	8.50±0.29	7.83±0.17	7.17±0.17	6.87±0.32	6.33±0.33
Taste	8.83±0.17	8.5±0.29	8.00±0.50	7.17±0.67	6.67±0.67	6.33±0.33
Aroma	8.5±0.29	8.17±0.17	7.67±0.17	7.17±0.17	6.33±0.17	6.00±0.00
Texture	8.67±0.17	8.17±0.17	8.00±0.29	7.50±0.29	7.17±0.17	6.33±0.17
Consistency	8.83±0.17	8.67±0.17	8.33±0.17	8.33±0.17	8±0.29	7.67±0.17
Overall Acceptability	8.37±0.03	8.23±0.03	8.3±0.06	8.27±0.03	8.2±0.00	8.27±0.03

Mean±S.E

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