

Development and storage stability of RTE Bengal gram (*Cicer arietinum*) based spiced snacks – Chana Nibble

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Article history

Received: 8 December 2014

Received in revised form:

5 May 2015

Accepted: 12 June 2015

Abstract

A Ready-to-Eat (RTE) spiced Bengal gram snack (Chana Nibble) with 50.33±0.15% moisture, 17.92±1.12% protein, 9.36±0.56% crude fat, 20.39±1.12% carbohydrate, 2.27±0.05% ash, 16.54±1.25% total dietary fibers was developed. The effect of different heat treatments & preservatives on the RTE Bengal gram snack was evaluated with a view to stabilize the product at ambient storage conditions for more than three months. The process of Chana Nibble was based on soaking, cooking, and seasoning followed by in-pack preservation techniques. The product scored superior acceptability (8.3±0.1) on 9 point hedonic scale. Different heat treatments applied to the product packed in PP pouches (75 µ thickness) were able to increase shelf life from 2 days to 115 days with respect to microbial spoilage at ambient temperature. Thermal treatment with steam under pressure at 121±0.1°C for 15 minutes was able to not only prevent microbial spoilage but also to impart shelf stability to the product for more than 12-14 weeks whereas thermal treatment with boiling water was able to stabilize product for 7 days and heat treatment by heating the product in cabinet dryer at 90°C for 120 minutes remained stable for 12 weeks with respect to microbial spoilage.

Keywords

Bengal gram

Shelf stable

Antioxidant

Water absorption

In pack-pasteurization

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Introduction

Chickpea (*Cicer arietinum*) is a legume of the family Fabaceae, subfamily Faboideae. Other common names for the species include Garbanzo bean, Ceci bean, Chana and Bengal gram, Hummus (Arabic) (Wikipedia, 2014). India is among largest producer of this legume with a record production as high as 8.88 million tons in 2012-13. Bengal gram is among most popular and well accepted legumes in Indian dietary (annual consumption 6-6.5 million tons) for many reasons inclusive of its sweet/mellow taste and its nutritional value (Yadav and Patki, 2014). Preliminary research has shown that chickpea consumption may lower blood cholesterol owing mainly to its fibre content (Pittaway *et al.*, 2008).

Mature chickpeas (cooked) are used to prepare salads, stews, various fried/steamed/fermented snacks (Dhokala). Chickpea grains ground into flour (gram flour/chickpea flour/Besan) and used commonly in Indian cuisines such as Missie Roti, Mirchi Bhajji, Pakodas, Bonda, Boondi, Kadhi, Falafel, Farinata/Panella (Sharma *et al.*, 2006). Chickpeas are used to make curries and are one of the most popular vegetarian foods in the Indian Subcontinent and in diaspora communities of many other countries (Bhat and Bhattacharya, 2001; Ramasamy *et al.*, 2004;

Semwal *et al.*, 2005).

Armed forces are the largest among institutional consumer of legumes and pulses. They have to operate in adverse conditions where cooking of these products become difficult. Any reduction in cooking time of such grains is of immense help to the consumers (Onwuke, 1983; Patki, 1984; Alexeevich *et al.*, 2006) generally, high moisture product gets spoiled due to various microorganisms such as yeasts, molds, and bacteria. Several attempts were carried out to stabilize layered snacks, RTE cereals, egg Halwa, Rava Idli and healthy snack food using thermal treatments and cost effective processes for their preparation (Madhura *et al.*, 1998; Kaur and Ahmed, 2000; McCormick *et al.*, 2003; Juneja, 2003; Lakshmi Devi *et al.*, 2005; Bin and Holdridge, 2008; Bows *et al.*, 2008). Submersion heating of RTE Deli meats at 90.6°C to 96.1°C for 2 min could readily provide 2-log reductions in microbial count and increased stability at ambient temperature (Mauriana *et al.*, 2002). The effectiveness of in-pack stabilization in inactivating food spoiling organisms depends upon package size and the texture of the product matrix (Muriana *et al.*, 2002; Murphy *et al.*, 2003a; Murphy *et al.*, 2003b). Civilian population in general and Armed forces in specific require RTE convenience products with minimum preparatory

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steps along with balanced nutrition. Bengal gram based RTE snack was developed using cost effective user-friendly preservation technique with a shelf life of three months under ambient field conditions. The purpose of the present investigation was to develop a RTE semi dehydrated snack by blending cooked Bengal gram grains with seasoning, spice mixture, salt, oil hydro in an optimized ratio while stabilizing the product with user friendly and cost effective techniques (combination of heat treatment and low levels of permitted class II preservative).

Materials and Methods

Raw Materials and pretreatments

Good quality whole grains of Bengal gram (*Cicer arietinum*) were procured from Agricultural Produce Marketing Corporation (APMC) of Mysore. The brown colored grains were cleaned to remove objectionable materials such as fodder, straw, dust, soil, or stone pieces, sorted to screen infested, hollow, and fragmented grains. The spice mix was prepared by grinding and mixing good quality whole spices in a defined ratio (Table 1). Refined ground nut oil was used in the preparation of Chana Nibble, and is the source of major lipid content of the product.

Chemicals and reagents

Chemicals used in the study were obtained from MERCK, SD FINE and SIGMA ALDRICH and were of analytical grade.

Processing method

The Savoir-faire of the product is described in the flow chart (Figure 1).

Spice composition

Ingredient formulation as used for the development of RTE Chana Nibble was enumerated in descending order (Table 1) Good quality whole spices were procured from local market and dried using forced air cabinet dryer to a moisture content of 5- 6% at 50°C, powdered fine using electro-mechanical flour mill. The powdered spices were sieved (100 μ) and packed in an airtight PFP Pouches and stored in a cool and dry place for further use.

Seasoning of Bengal gram with spices

Soaked and drained Bengal gram grains were sautéed in pre heated (170°C) fresh refined oil. The temperature of the oil immediately lowered to 95°C after adding Bengal gram, further, spice and condiment mix was homogenized properly to coat over the grains uniformly.

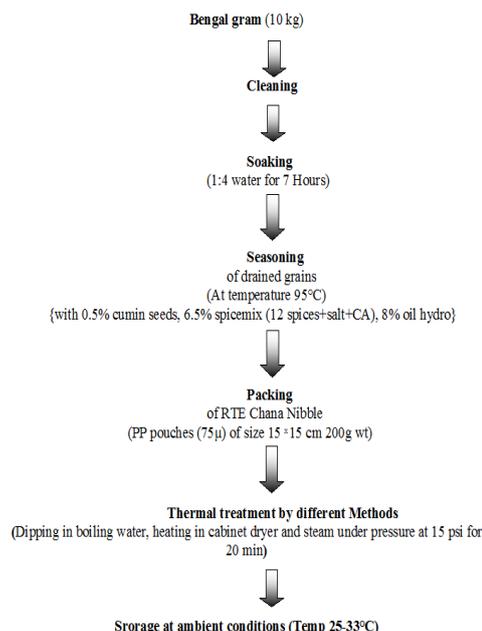


Figure 1. Process flow Chart for the preparation of RTE Chana Nibble

Proximate composition

Percent moisture and total ash content in the samples were estimated following the official methods (AOAC, 1984; AOAC, 1990). Total protein was estimated by Kjeldahl method (Hawk, 1965) using Gerhardt nitrogen digestion (Turbotherm) and distillation (Vapodest) auto apparatus, Bio-incorporation, India. Lipid content was estimated by using programme controlled automatic fat extraction apparatus; Soxtherm Fat Extraction System (SE-416, Germany). Water activity (a_w) of the sample was determined using a pre-equilibrium conditioned water activity meter (Labmaster- a_w , Novasina, Switzerland) at 25°C. pH of homogenized sample was determined by using digital pH meter (Cyber scan 510, Singapore).

Fibre fractions and mineral estimation

Crude fibre in all the samples was estimated using Fibretherm Instrument (Gerhardt FT-12, Germany). Dietary fibres were estimated in the sample using well-established method (Asp *et al.*, 1983). Iron was determined calorimetrically by using methods as described by Hill (1930) and Piper (1950) at 540 nm and Calcium was estimated using methods of Siong *et al.* (1989). Other minerals were estimated using Ultima-2 ICP-Optical Emission Spectrometer loaded with ICP 5.4.2 Win-IMAGE software (Horiba Scientific Jobin Yvon France).

In vitro antioxidant activity (DPPH radical scavenging method)

Antioxidant activity of Chana Nibble was

Table 1. Composition of spices mix/100 g cooked Bengal gram grains

Spices and other ingredients	Quantity in (g)
Oil Hydro (Refined Groundnut Oil)	7.58
Common Iodized salt	2.00
Coriander Powder	1.50
Dry Mango Powder	1.00
Red Chili Powder	0.80
Whole cumin	0.50
Cumin Powder	0.22
Turmeric Powder	0.20
Citric acid	0.15
Black Pepper Powder	0.10
Mace Powder	0.10
Nutmeg Powder	0.10
Cinnamon Powder	0.10
Bay Leaves Powder	0.10
Clove powder	0.05
Black Cardamom powder	0.05

determined as per method described by Braca *et al.* (2001), Dasgupta and De (2007) using stable, 1 diphenyl-2-picryl hydrazyl (DPPH) radical. BHA, BHT were used as standard. The antioxidant activity of the extract was measured at 515 nm. The antioxidant activity of the extract was calculated using formula

$$\frac{\text{Absorbance sample} - \text{Absorbance control} \times 100}{\text{Absorbance control}}$$

Lipid Oxidation

Peroxide value of the samples were analysed during processing and storage at room temperature. PV of the sample was analysed according to the standard methods of AOCS (1973). Changes in free fatty acid content of the processed samples were analysed during storage. The free fatty acid content of the processed samples was also measured according to the standard procedures of AOCS (1973). Oxidative stability of processed samples was determined by estimating the changes in Thiobarbituric acid reactive substances content (Tarladgis *et al.*, 1960).

Microbiological Analysis

Standard methods of APHA (1992) were used to establish the microbial quality of the product. The analysis was carried out to map total viable counts (TVC), coliforms, *E. coli*, yeast and molds (Y&M).

Organoleptic and statistical evaluation

Sensory analysis was carried out as per procedure described by Murray *et al.* (2001). Samples were evaluated by 30 semi trained panelists to establish its sensory acceptability with respect to their colour, aroma, taste, texture and overall acceptability (OAA)

during product development as well as during storage on a 9 point hedonic scale. The 9-point Hedonic scale grading was as follows: 9=Excellent, 8=Very good, 7=Good, 6= Good above fair, 5= Fair, 4= Fair above poor, 3= Poor, 2= Very poor, 1= extremely poor. (9-like extremely, 1- dislike extremely). Samples were randomly drawn, coded, and served to the panelists. The statistical analysis for significance was carried out using GraphPad InStat 3 (Trial version).

Results and Discussion

The preparation of shelf stable RTE Chana Nibble involves soaking, pressure cooking, seasoning followed by in-pack thermal stabilization of Bengal gram using different heat treatments viz. boiling water, forced air cabinet dryer and batch type microwave oven.

Water absorption array and cooking

Moisture uptake during soaking is one of the regulating parameter for optimum cooking in general and edible acceptability in specific for product development (Bhattacharya, 1985; Ravi *et al.*, 2011). Each grain behaves differently for moisture uptake due to its explicit physico-chemical characteristic. Optimum soaking is important for the reason of preventing lump formation due to higher moisture uptake, whereas inadequate soaking results into lower water absorption leading to undercooking or incomplete gelatinization and hard gritty texture (Singh and Rao, 1995) however, it was observed that beyond optimum time of soaking, the rate of moisture gain usually becomes very slow (Engels *et al.*, 1986). Initially Bengal gram was analyzed to have 7.10±0.32% moisture whereas minimum 45-55% moisture is required for complete gelatinization of starch and to keep discrete shape and integrity of individual grains (Patki, 1996). Soaking of Bengal gram in tap water (at 27°C and grains to water ratio 1:4 w/w) for 16 h resulted in increase in moisture content from 7.10±0.32% to 55.95±0.25%, however, optimum soaking time was observed to be 7 h as grains attained (54.21±0.21%) near to maximum moisture content (55.95±0.25%). There was minor increase in the moisture content during further soaking from 7 to 16 h (3%) indicating saturation in moisture absorption limits at conditions prescribed for the experiment. Rate of moisture uptake by the grains is graphically represented in Figure 2. During cooking, moisture content of the soaked grains remained constant (55.95±1.01%).

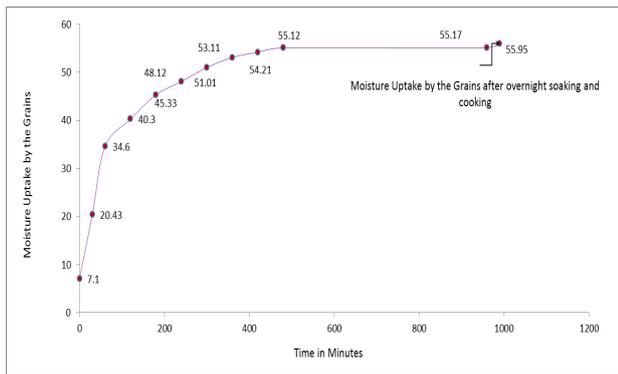


Figure 2. Moisture uptake pattern in Bengal gram grains

Stabilization of RTE Chana Nibble

Various attempts performed and the methodology adopted in the investigation with subsequent stability achieved is tabulated in Table 2. The results obtained clearly indicate that neither boiling water treatment at $90\pm 5^\circ\text{C}$ for 30 min nor analogous treatment with Sorbic acid was efficient to destroy the spoiling biological factors (enzymes and microbes); therefore the maximum shelf stability at room temperature attained was 2 and 5 days respectively. However Ingham *et al.* (2005) revealed efficiency of small-scale hot water post packaging pasteurization (PPP) as a post lethality (post cooking) treatment for *Listeria monocytogenes* on ready-to-eat beef snack sticks and natural-casing wieners. Result showed an average reduction of 2 log units for *L. monocytogenes* with heating times of 1.0 min. Conversely, the keeping quality of Chana Nibble processed in cabinet dryer and steam under pressure was comparable. In both the cases the product stabilized with Sorbic acid accompanied by thermal treatment was found to have improved microbial stability with fresh palatability. Finally, Chana Nibble stabilized in open pack treated with steam under pressure was taken for future studies because of its sustained microbial activity during initial trials. The packs were processed with a small cleavage as a vent to out flow the steam generated during processing as packs develop bulging, irregular shape, and openings near to sealed area if sealed completely.

Proximate and nutritional components analysis

RTE Chana Nibble contains $50.33\pm 0.15\%$ moisture and could be categorized as semi-dehydrated product with 0.982 ± 0.006 water activity (aw) at 25°C . At this moisture level it has well acceptable palatability (8.40 ± 0.10) owing to its soft & pliable texture in consort with ease in mastication. Chana Nibble was analyzed to have a fat content of $9.36\pm 0.56\%$ which was majorly contributed by added oil and seconded by lipid of Bengal gram and spices. It was analyzed to have $17.92\pm 1.12\%$ protein (db),

Table 2. Stabilization methods and their respective stability with respect to visible growth of spoiling organisms

Stabilization Method	Preservative Status	Shelf Life
In a Boiling Water bath ($90\pm 5^\circ\text{C}$) for 30 min	No Preservative	2±1 days
	With Sorbate (1000 ppm)	5±2 days
In a Cabinet Dryer ($90\pm 1^\circ\text{C}$) for 120 min	No Preservative	21±2 days
	With Sorbate (1000 ppm)	84±3 days
In Pressure Cooker (121°C , 15 lb P for 15 Min) (open pack processing)	No Preservative	
	With Sorbate (1000 ppm)	110±5 days

Values are mean±SD; n=3

$20.39\pm 1.12\%$ carbohydrate, $2.27\pm 0.05\%$ total ash, and 237.48 Kcal calories. The finished product was acidic due to added (1.12%) citric acid having pH 4.8 which was essential in stabilizing process. The product contains $15.89\pm 0.26\%$ crude fibers which comprises largely insoluble dietary fibre contributed majority by seed coat and the spice mix. Insoluble dietary fibers are essential to maintain good health of digestive tract as it helps in peristaltic movement of the bowl and large intestine (Elia and Cummings, 2007; Buttriss and Stokes, 2008). RTE Chana Nibble contains total dietary fibre (%) as 16.54 ± 1.25 , insoluble dietary fibre (%) as 15.53 ± 1.15 and soluble dietary fibre (%) as 1.01 ± 0.012 respectively. The product is rich in essential minerals (db) and contained Calcium-Ca 396.14 ± 15.27 ; Iron-Fe 11.18 ± 2.23 ; Copper-Cu 1.72 ± 0.02 ; Potassium-K 1327.06 ± 19.15 ; Magnesium-Mg 268.94 ± 3.06 ; Manganese-Mn 4.1 ± 0.05 ; Sodium-Na 2066.26 ± 15.12 and Zinc-Zn 29.16 ± 2.03 . It was found that approximately 25% RDA requirement of these minerals may be complimented with a serving (100 g) of this product.

Antioxidant activity (AOA)

Results showed that there was non-significant difference in the AOA of Bengal gram before (18.42 ± 1.1) and after cooking (18.22 ± 2.1) indicating no loss in the antioxidant contributing fractions of the Bengal gram while processing. However, existing antioxidant activity of raw and cooked chickpea may be contributed to present carotenoids, 28.12 ± 1.10 µg/g (Stahl and Sies, 2003), total polyphenol, and tannins (Scalbert *et al.*, 2005). Antioxidant activity of these fractions is well documented by many researches (Barampama and Simard, 1995; Kanatt *et al.*, 2011). Antioxidant activity was estimated by measuring free radical scavenging activity by different fractions of the Chana Nibble using 2, 2-diphenyl-1-picryl hydrazyl (DPPH) as free radical producing agent. It was observed that major AOA is contributed by the spice mix ($91.95\pm 2.6\%$) which was comparable with Butylated Hydroxy Anisole-BHA ($92.63\pm 1.2\%$) and Butylated Hydroxy Toluene-BHT ($92.81\pm 1.8\%$). Chana Nibble was also found to have comparable antioxidant activity ($38.38\pm 3.1\%$).

Herbs and spices have better antioxidant activity owing to their antioxidant content in the form of polyphenols, carotenoids, tocopherols, tocotrienols, glutathione, ascorbic acid, and certain enzymes with antioxidant activity (Shobana *et al.*, 2000), which helps to protect them from hazardous oxidative damage (*invitro/invivo*) (Kahkonen *et al.*, 1999). Clove, cardamom, cinnamon, and asafoetida are among spices having stronger antioxidant activity attributable to the presence of aromatic essential oils and phenolic compounds (Duh *et al.*, 1999; Aaby *et al.*, 2004).

Storage stability

Autoxidation of lipids is another major cause of spoilage during storage of dehydrated/semi hydrated RTE convenience foods (Premavalli *et al.*, 1987; Johnston *et al.*, 2005). Lipid oxidation is a dynamic three-stage process and peroxide value of the product usually changes as a factor of time (Kerler and Grosch, 1996).

Peroxide value and storage stability

Slow increase in peroxide values from 17.83 ± 1.54 meq O_2/Kg to 25.89 ± 2.45 meq O_2/Kg , 28.24 ± 3.21 meq O_2/Kg and 30.42 ± 1.14 meq O_2/Kg after 1, 2 and 3 months respectively, indicates high resistance to autoxidation of spiced bengal gram due to the addition of spices during preparation. Antioxidant activity of spices has been reported by many researches, which probably among one of the potential reasons for shelf stability of intermediate/semi dehydrated products (Shobana *et al.*, 2000). Detection of peroxide gives the initial evidence of rancidity in products prepared with unsaturated fats and oils. Peroxide value is one of the chemical markers to judge autoxidation and to assess the extent to which spoilage has advanced (Robards *et al.*, 1988). It measures the extent to which sample has undergone primary oxidation. The unsaturation found in non-visible fat of the grain and visible fat used for the preparation of such product, play a role in autoxidation and lipids with high degree of substitution are most susceptible to autoxidation (Coupland and McClements, 1996). Auto oxidation is a free radical reaction involving oxygen that leads to deterioration of fats and oils, which form off flavors and off odors (Kubow, 1990). Peroxides are intermediate in the auto oxidation reaction. Lipid oxidation may also be controlled for a time by various methods including oxygen barrier packaging, absence of light, modified atmosphere packaging and temperature-controlled storage. It was established by Semwal *et al.* (2005) that polypropylene has better barrier properties for oxygen compared to

Polyethylene but lesser compared to Paper Foil Polyethylene (PE/PF) laminates.

Acid value and storage stability

As Chana Nibble is a semidehydrated product, and undergone thermal processing, hence maximum lipase activity assumed to be stopped during cooking and stabilization, therefore increase in free fatty acid (FFA) content from 2.12 ± 0.21 to 2.54 ± 0.32 , 3.12 ± 0.23 and $4.42 \pm 0.54\%$ oleic acid after storage period of 1, 2 and 3 months may have resulted from hydrolysis of triglycerides and secondary degradation products of lipid oxidation (Gan *et al.*, 2005). FFA is a shelf life indicator test for hydrolytic rancidity and acidity of oily product is dependent on the amount of FFAs present (Vyletelova *et al.*, 2000a; Vyletelova *et al.*, 2000b). This in turn is dependent on the degree of hydrolysis of the oil or the nature of the processing which the product/oil may have undergone (Teoh, 2006). The increase in FFA content in stored chana Nibble results from lipolysis of lipids owing to presence of higher quantity of fat along with moisture in the finished product. Refined oils, often referred to as neutralized with low acidity but raw and crude oils are naturally hydrolyzed and consequently have a comparatively higher acid value (Tandy *et al.*, 1984). As Bengal gram contains relatively higher amount of unsaturated fat, hence initial values are higher, however, spices and refined groundnut oil incorporated to finished product has reduced formation of much FFAs.

Thiobarbituric acid reactive substances (TBARS) and storage stability

TBARS are formed as a byproduct of lipid peroxidation that is as degradation products of fats which can be detected by the TBARS assay using thiobarbituric acid as a marker reagent. Increase in TBARS value from initial 0.231 ± 0.002 to 0.242 ± 0.002 , 0.251 ± 0.001 and finally 0.271 ± 0.003 in Chana Nibble after a storage period of 1, 2 and 3 months respectively suggests that there is minor formation of thio-barbituric acid reactive substances during processing and storage. As reactive oxygen species (ROS) have extremely short half-lives, they are difficult to measure directly. Instead, measurable species are several products of the damage produced by oxidative stress, such as malondialdehyde (MDA) present in the sample, as well as malondialdehyde generated from lipid hydroperoxides by the hydrolytic conditions of the reaction (Zipser and Watts, 1962). MDA is one of several low-molecular-weight end products formed via the decomposition of certain primary and secondary lipid peroxidation products

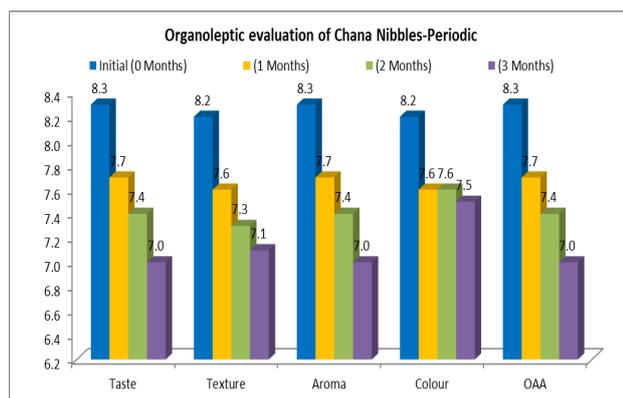


Figure 3. Periodic sensory evaluation of Chana Nibble (Draper *et al.*, 1993).

Microbiological stability

The shelf life and the storage stability of the product were established on the basis its microbial load at same time it was correlated with the sensory score obtained by semi trained panelists. It was observed that treatments such as in pack pasteurization using boiling water bath (with and without sorbic acid) develops visible growth of molds and possess objectionable flavour after 2 and 5 days respectively hence were discarded and not considered for further study. A meager delay in the spoilage period may be attributed to the antimycotic properties of sorbic acid in the formerly described batch. However, batches thermally treated with cabinet dryer and autoclave (with sorbic acid) was stable with respect to microbial counts and organoleptic profile (Figure 3). The batch preserved using cabinet dryer resulted in significant loss in moisture content during subsequent storage, hence scored significantly low value (for texture <7.0 and OAA <7.0) on hedonic scale during its shelf life period. The results indicate that total plate counts for the autoclave processed batch were $10 \pm 1 \times 10^1$, $16 \pm 2 \times 10^1$, $20 \pm 1 \times 10^1$, $31 \pm 3 \times 10^1$, $43 \pm 2 \times 10^1$ cfu/g besides Yeast & Mold count of 5 ± 2 , 11 ± 3 , 18 ± 5 , 24 ± 1 and 32 ± 3 cfu/g at the storage period of 0, 1, 2, 3 and 4 months respectively. However, Coliforms counts were absent indicating hygienic handling during processing of Chana Nibble. It is clear from the above result that these batches processed with steam under pressure have microbial count well within the acceptable range. As per World Health Organization (WHO) norms permissible microbial profile of processed foods is as follows; TPC (cfu/g) should be below 10000).

Sensory analysis

Chana Nibble preserved using steam under pressure was considered as final batch and periodically analyzed for its organoleptic profile by

20 semi-trained panelists during storage of 1, 2, 3, and 4 months. It was found that there were changes, in the taste, mouth feel, consistency, and flavour during storage, which were within acceptable limits of hedonic scale however; color of the product has not varied significantly (Figure 3). Chana Nibble was acceptable well within the sensory limits (7.0 ± 0.1) after a storage period of 3 months. Product's acceptability reduced drastically after a period of 15 weeks due to unacceptable taste which may be attributed to enhanced rancidity, staling of starch, moisture loss; however, its microbial quality was within safe limits.

Conclusion

RTE foods are gaining importance owing to their suitability/adaptability to address the need of rapidly intervening urbanization and changing dietary habits of all spheres of society. But at the same time it is obligatory to maintain its nutritive aspect. Then only food preservation of processed foods can be considered as protracted modus operandi. Alternative to retort technology where food is exposed to pressurized thermal energy and lead to destruction of most of the nutrients, the present indigenous and user-friendly technique of preservation could be adapted as cottage scale preservation scheme to support various self-help groups, small scale entrepreneurs and pilot plants where similar products could be made shelf stable while preserving most of intact nutrient of the product.

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