

Storage studies on *Low calorie burfi* incorporated with *Sucralose* and *Costus speciosus* extract

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Abstract: In the present study, changes in compositional, physico-chemical, sensory and microbial properties (SPC, coliform count and yeast and mould count) of *low calorie burfi* packed in low density polyethylene (LDPE) film was monitored. The changes were studied at $37 \pm 1^\circ\text{C}$ and $4 \pm 1^\circ\text{C}$ on every 7th day of storage till the products became unacceptable. The sucralose content in *low calorie burfi* was estimated and found to be 150 ppm. The pH was found to significantly ($p < 0.05$) decreased during the storage period whereas acidity, FFA, TBA value and soluble nitrogen contents increased significantly ($p < 0.05$). The standard plate counts and yeast and mould counts were found to increased progressively during storage whereas coliform count was absent in the product throughout the storage period. Shelf life study showed that developed *low calorie burfi* had a shelf life of 20 days under refrigeration and 7 days at ambient temperature.

Keywords: *Costus speciosus*, Sucralose, Low calorie burfi

Introduction

In the present scenario, consumers are becoming more health conscious and the demand for health foods has been increasing rapidly. There has been a considerable interest in extending the

use of herbal extracts in dairy foods, fruits juice based products and pharmaceuticals (Mann et al. 2018; Idowu et al. 2021). Further, now it has become important to look for an economical as well as therapeutically effective treatment especially for developed and developing countries. During the search for alternate antidiabetic foods, it was found that some of the herbs have potential antidiabetic activity. *Costus speciosus* plant known as insulin plant (diosgenin compound), belongs to *Costaceae* family, and is a medicinal plant. In recent times, these plant leaves are commonly being incorporated in various food products because of its exceptional health benefits. *Burfi* is one of the most popular *khoa* based sweet in India. Once confined to household production, *burfi* is gaining an international market in recent years owing to its delicious taste, flavour and texture (Aneja et al. 2002). *Khoa* has a unique adaptability in terms of flavour, body and texture to blend with a wide range of ingredients resulting in the development of a wide range of varieties of *burfi*. Several varieties of *burfi* are available in the market such as plain or *mawa/khoa burfi*, fruit and nut, cashew *burfi*, chocolate, saffron and *rava burfi* (Sarkar et al. 2002; Prasad et al. 2017; Prasad et al. 2018). It is prepared from a mixture of *pindi khoa* and sugar, heating to near homogenous consistency. Beating and whipping operations prior to cooling are sometimes practiced to obtain a product with smooth texture and closely knit body. It is white to light cream in colour with firm body and smooth texture with very fine grains (Patil and Pal, 2005). However, high levels of sugar are used in the preparation of *burfi* contributes to multiple health-related issues. Therefore, various low-calorie sweeteners such as saccharin, acesulfame K, aspartame and sucralose have been permitted in the dairy products like *khoa*, *burfi*, and *rasogolla* (FSSAI, 2012). Shelf-life of *burfi* is most important from manufacturing and consumer point of view. To make *burfi* as commercially viable product, it should have sufficient shelf-life. The growth of the microorganisms brings about various changes in the product and spoils the taste of the product during storage. Moreover, dairy products containing plant based extracts and low calorie sweeteners in different packaging materials could be a major factor that affects its storage stability under different storage temperatures.

Khan et al. (2008) studied the changes in quality of groundnut *burfi* packed in polypropylene (PP, 75 μ) and metallized polyester

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(12 μ) low density/linear low density (MP, 75 μ) during storage in order to assess the shelf life. The samples without sorbic acid spoiled within 30 days of storage due to mold growth and fermented odour. Sachdeva and Rajorhia (1982) studied the chemical and microbiological changes in plain *burfi* during storage at room and refrigerated temperatures using two packaging materials viz., parchment paper and tin containers. The shelf life of *burfi* when stored in parchment paper was found 10 days at 30°C and 50 days at 5.0 \pm 1°C whereas *burfi* packed in tin containers had a shelf life of more than 105 days at 5.0 \pm 1°C. *Burfi* with high moisture develops a hard structure and crystallization of sugar during long storage (Anon, 1979; 2012). Vijayalakshmi et al. (2005) reported that a free O₂ absorber coupled with high – barrier materials like metalized films/foil laminates gave more than 45 days' shelf life to *burfi* at 27 °C. Prasad et al. (2017) investigated the effect of different packaging materials and essential oils on storage stability of *burfi* and reported that *burfi* containing mixed essential oils (Turmeric/ginger/cardamom) and packed in HDPE boxes had shown shelf life of 25 days at 4 \pm 1°C. However, limited reports are available on the storage stability of sucralose added *burfi* in presence of *costous speciosus* extract. Hence the present study was aimed to evaluate the storage stability of *low calorie burfi* at two different storage temperatures i.e. refrigeration and room temperature.

Materials and Methods

Preparation of *low calorie burfi*

The *low calorie burfi* was prepared with *costus speciosus* extract (63.12 ppm) and sucralose (151.85) as per the optimized procedure described in Anupama et al. (2020). Samples were moulded into flat round shaped pieces and wrapped in parchment paper which was further packaged in LLDPE pouch. After packaging, pouches were stored at room temperature 37°C and refrigerated temperature 4°C.

In the preparation of traditional *burfi*, sucralose was replaced with sucrose (35 per cent w/v of milk) and no extract was added.

Estimation of sucralose in *low calorie burfi*

Sucralose content in *low calorie burfi* was quantified as per the procedure described by Arora (2010). Briefly, 1.75g of *low calorie burfi* was weighed and ultra-sonificated at 40°C for 20 min. After cooling to room temperature, two millilitres of Carrez solution No. 1, Carrez solution No. 2 and one millilitre of HPLC grade methanol was added. Then solutions were allowed to stand at room temperature for 10 min, and subsequently filtered using Whatman No.1 filter paper. The filtrate containing sucralose was analysed by HPTLC. Ten microlitres of the standard sucralose solution in methanol (1 μ g/10 μ l) and 10 μ l of the sample filtrate was applied at a distance of 1-1.5 cm from the sides of silica gel 60 F₂₅₄ (Kiesel gel 60 F₂₅₄) HPTLC aluminium sheets (which have been previously activated for 30 min in an oven at 100°C). The

spots applied were dried simultaneously using a hot air blower. The plates were then developed in a vertical chamber consisting of dichloromethane: methanol (4:1), as a developing solvent system, till a distance of 1 cm remained from the top edge of the plate. The separation was accomplished within 15 min. The developed plates were then removed from the chamber and dried using a hot air blower, and sprayed with 15 per cent (v/v) methanolic sulphuric acid and dried again. Subsequently the plates were heated for 10 min at 100°C. At this temperature, sucralose appears as charred spots, having an R_f value between 0.40- 0.60. The charred spots obtained were scanned and quantified using the known amount of standard sucralose solution spot by Bio-Rad quantity one software.

Preparation of standard curve

Standard sucralose solution was prepared by dissolving 10 mg of sucralose in 100 ml of HPLC grade methanol to give a concentration of 1.0 μ g/10 μ l. The standard solutions at concentration of 2.5 μ l, 5.0 μ l, 7.5 μ l, 10 μ l and 12.5 μ l was applied on the HPTLC silica gel plates at a distance of one centimetre (previously activated at 100°C for 30 min in an oven). The plates were then dried, developed, sprayed, scanned and quantified to get the standard curve for sucralose. The standard curve was plotted using the Bio-rad software.

Physico-chemical analysis of *burfi*

The acidity of *burfi* was determined by method described in BIS (IS: 1166-1968) for condensed milk. The pH of *burfi* was measured using Systronic digital pH meter, Model 335. The method prescribed by Deeth et al. (1975) was used to estimate the FFA content of *burfi*. The extent of oxidation of fat in *burfi* was measured in terms of TBA value. TBA value was expressed as absorbance (OD) at 532 nm. The soluble nitrogen contents of *burfi* sample were determined by the procedure outlined by Kosikowaski (1982).

Microbiological Analysis of *low calorie burfi*

Standard plate count, Coliforms and Yeast and mould count of *burfi* samples were estimated by pour plate technique, as described in IS: SP: 1224 (part I and II), 1981.

Sensory Evaluation of *low calorie burfi*

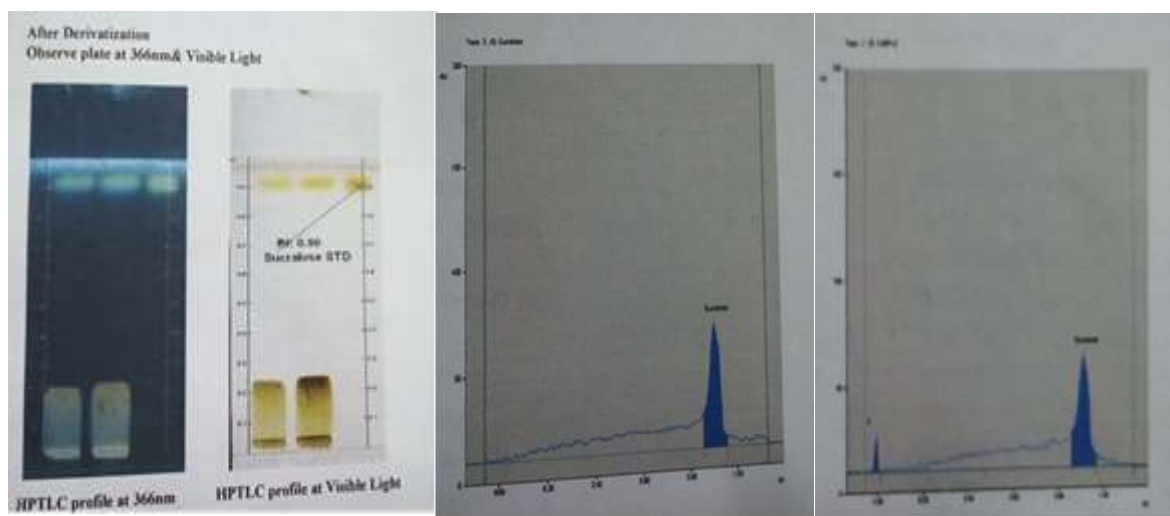
For the organoleptic evaluation of *burfi*, judges who were familiar with desirable attributes of *burfi* were selected. The selection criterion was kept as the judges were a regular consumer of the dairy sweets as well as their similar behavior between sensory evaluation sessions. The *burfi* samples were tempered at room temperature for 1-2 hour before judging. Sensory evaluation of the samples was conducted in isolated booths illuminated with incandescent light and maintained at 28 \pm 2 °C.

Statistical analysis

Two different burfi samples viz. traditional burfi and low calorie burfi were analysed and compared for changes under both room and refrigerated temperatures during storage. Data pertaining to physico-chemical and microbiological qualities was analysed statistically using repeated measures of ANOVA and the sensory qualities was analysed using Friedman’s test and Mann-Whitney u-test.

Results and Discussion

Fig 1 (a). HPTLC plates of sucralose standard and sucralose content in *Low calorie burfi*
(b): Peak response of sucralose standard
Fig 1(c): Peak response of sucralose in *Low calorie burfi*



Quantification of sucralose in low calorie burfi

HPTLC plates of sucralose standard and sucralose content in *low calorie burfi* are presented in Fig 1 (a). The peak responses curves obtained for the standard sucralose and sucralose in low calorie burfi are delineated in Fig 1(b) and (c). The quantitative analysis of standard sucralose was performed over silica gel HPTLC plates HPTLC. The regression equation was drawn from the obtained data and it is presented in equation 1. The coefficient of correlation for the regression equation was 0.996.

Table 1. Effect of storage on physico-chemical properties of traditional and *Low calorie burfi* at 37±1°C

Sample	Days of storage	
	0 th day	7 th day (spoiled)
	Changes in moisture	
Traditional burfi	13.11±0.16 ^{ax}	12.25±0.18 ^{bx}
<i>Low calorie burfi</i>	18.033±0.16 ^{ay}	17.26±0.18 ^{by}
	Changes in pH	
Traditional burfi	6.42±0.08 ^{ax}	5.31±0.08 ^{bx}
<i>Low calorie burfi</i>	6.40±0.08 ^{ay}	5.18±0.05 ^{by}
	Changes in acidity (% Lactic acid)	
Traditional burfi	0.36±0.01 ^{ax}	0.47±0.01 ^{bx}
<i>Low calorie burfi</i>	0.35±0.01 ^{ay}	0.42±0.1 ^{by}
	Changes in FFA (µeq/g)	
Traditional burfi	11.05±0.11 ^{ax}	13.27±0.019 ^{bx}
<i>Low calorie burfi</i>	12.46±0.09 ^{ay}	14.25±0.19 ^{by}
	Changes in TBA	
Traditional burfi	0.18±0.01 ^{ax}	0.27±0.05 ^{bx}
<i>Low calorie burfi</i>	0.21±0.06 ^{ay}	0.27±0.05 ^{by}
	Changes in tyrosin value (mg/100g)	
Traditional burfi	7.70±0.20 ^{ax}	7.77±0.21 ^{bx}
<i>Low calorie burfi</i>	8.77±0.21 ^{ay}	9.82±0.18 ^{by}

Figures are mean ± standard error of three replications, ^{a-d} Means with different superscript vary significantly within a row, ^{x-y} Mean with different superscripts vary significantly within a column

$$Y = 763.7 + 1680.08 * X \text{-----Equation I}$$

Concentration of sucralose in the product was calculated from the regression equation and it was found out to be 150 ppm which is under maximum limit of sucralose (750 ppm) in traditional milk products as per regulations of FSSAI, 2012. The lower detection level of sucralose than the added amount (i.e. 151.85) could be because of accuracy of the detection for the adopted method. The recovery percentage of sucralose in the low calorie burfi was observed to be 98.78% which is in range for the recovery % (96-99.2%) suggested by George et al. (2010) for the followed method.

Effect of storage on physico-chemical properties

The storage changes taking place in the composition of burfi samples during storage at room and refrigerated temperature are presented in Table 1 and Table 2 respectively. It can be seen from Table 1 that, moisture content of low calorie burfi significantly (P<0.01) decreased from an initial moisture content of 18.03% to 17.26% during the storage period up to 7 days and thereafter the product was unacceptable due to visible mould growth. It can be seen that there was a progressive significantly decreasing trend (p<0.05) in the moisture content of the product when stored at refrigerated temperature from 18.03% to 16.58% (Table 3). After 21st day of storage at refrigeration temperature the product was unacceptable due to quality changes in the product. During storage period, the moisture content of low calorie burfi

decreased rapidly at room temperature as compared to refrigerated temperature. Similar pattern in decline of moisture level for khoa and khoa products has been reported by several researchers (Londhe et al. 2012; Jha et al. 2013 & 2014; Singh et al. 2021). The decrease in moisture content during refrigerated storage might be due to drying at low temperature (7±2°C) and surface evaporation (Sharma et al. 2003). Sachdeva and Rajorhia (1982) also reported a decrease of moisture content during storage of burfi at 30±2 °C and 7±2 °C. A decline in the pH was observed in low calorie burfi with values ranging from 6.4 to 5.49 with significant decrease (p<0.05) at refrigeration temperature (Table 2). At room temperature storage, pH varied from 6.40 to 5.18 in low calorie burfi samples with significant decrease (p<0.01) from 0th day to 7th day. This can be correlated with the findings of Londhe et al. (2012) wherein pH of lal peda samples packed in paper board boxes and stored at 30 °C decreased from 6.3 to 5.2 in 20 days.

It can be seen that the acidity of low calorie burfi was significantly influenced by storage period when stored at room temperature (Table 1). During storage of low calorie burfi at room temperature a significant (P<0.01) increase in acidity content from 0.35% LA at 0thday to 0.42 % LA at 7th day was observed. Similar results were also observed when the product was stored at refrigerated temperatures (Table 2). During storage of low calorie burfi at refrigerated temperature a significant (P<0.01) increase in acidity content from 0.35% LA at 0thday to 0.49 % LA at 21st day was observed. It can be seen that the titratable acidity of low calorie burfi increased at faster rate during storage period

Table 2 Effect of storage on physico-chemical properties of traditional and low calorie burfi at 4±1°C

Sample	Days of storage			
	0 th day	7 th day	14 th day	21 st day (spoiled)
	Changes in moisture			
Traditional burfi	13.11±0.16 ^{ax}	12.45±12.25 ^{bx}	12.25±0.18 ^{cx}	11.35±0.14 ^{dx}
Low calorie burfi	18.03±0.16 ^{ay}	17.88±0.13 ^{by}	17.26±0.18 ^{cy}	16.58±0.14 ^{dy}
	Changes in pH			
Traditional burfi	6.42±0.08 ^{ax}	6.18±0.05 ^{bx}	5.61±0.05 ^{cx}	5.24±0.01 ^{dx}
Low calorie burfi	6.40±0.08 ^{ay}	6.18±0.08 ^{by}	5.68±0.01 ^{cy}	5.49±0.01 ^{dy}
	Changes in acidity (% lactic acid)			
Traditional burfi	0.36±0.01 ^{ax}	0.39±0.01 ^{bx}	0.47±0.11 ^{cx}	0.53±0.01 ^{dx}
Low calorie burfi	0.35±0.01 ^{ay}	0.37±0.01 ^{by}	0.42±0.01 ^{cy}	0.49±0.01 ^{dy}
	Change in FFA (µeq/g)			
Traditional burfi	11.05±0.09 ^{ax}	13.39±0.13 ^{bx}	12.74±0.08 ^{cx}	13.55±0.09 ^{dx}
Low calorie burfi	12.46±0.09 ^{ay}	13.00±0.13 ^{by}	13.52±0.08 ^{cy}	14.53±0.09 ^{dy}
	Changes in TBA			
Traditional burfi	0.18±0.04 ^{ax}	0.25±0.01 ^{cx}	0.27±0.08 ^{cx}	0.29±0.01 ^{dx}
Low calorie burfi	0.21±0.01 ^{ay}	0.24±0.10 ^{cy}	0.27±0.01 ^{cy}	0.29±0.01 ^{dy}
	Changes in tyrosin value (mg/100g)			
Traditional burfi	7.70±0.21 ^{ax}	8.87±0.11 ^{bx}	9.43±0.12 ^{cx}	10.56±0.16 ^{dx}
Low calorie burfi	8.77±0.21 ^{ay}	9.42±0.11 ^{by}	10.33±0.12 ^{cy}	11.50±0.16 ^{dy}

Figures are mean ± standard error of three replications, ^{a-d} Means with different superscript vary significantly within a row, ^{x-y} Mean with different superscripts vary significantly within a co

at room temperature as compared to refrigerated temperature. The acidity development could be attributed to production of acids like formic acid, acetic acid, lactic acids and other organic acids. Similar trend was reported by Londhe et al. (2012) who recorded an increased acidity of *Lal* peda samples stored at 30 °C from 0.75 per cent to 1.52 per cent in 20 days. Maillard reaction also produces many organic acids which are also responsible for increment in acidity (Goyal and Shrinivasan, 1988). Similar findings were reported by Sachdeva and Rajorhia (1982) in *burfi* during storage. Increase in titratable acidity was also observed during storage of *khoa* (Choudhary et al. 2019).

Effect of storage on FFA content

Lipolysis, regardless of cause seriously degrades the quality of the stored product by imparting off flavours and is also responsible for the development of rancidity. In stored dairy products, lipolysis by microbial lipase is of the greatest significance (Downey and Murphy, 1970). The influence of storage period on FFA content of *low calorie stored burfi* at room temperature as shown in Table 1 reveals that the FFA values of *low calorie burfi* stored increased from 12.46 % to 14.25 % during 7 days of storage and this increase was observed in *low calorie burfi* in 21 days of storage at refrigeration temperature. The FFA values of both traditional *burfi* and *low calorie burfi* samples increased significantly ($p < 0.01$) at both storage temperatures. During storage of *low calorie burfi* at $37 \pm 1^\circ\text{C}$, a significant increase in FFA content up to 7th day was observed and thereafter both the *burfi* samples found to be unacceptable due to visible mould growth. This increase in FFA content may be due to the higher SPC and Yeast and mould counts observed at higher temperatures as evident in Table 5 and Table 6. This increase in FFA content could be attributed to hydrolysis of fat which is primarily affected by the growth of yeasts and molds. In the present investigation also the increase in FFA could be due to increase in yeast and mold count. A similar trend of increase in FFA content during storage was noticed in *burfi* by Tiwari (2013). Vijaykher and Patel (1983) also reported an increase in free fatty acids in *Peda* during storage at ambient temperature (25-29°C), using polyethylene bags of various densities.

Effect of storage on soluble nitrogen content

Soluble nitrogen is the measure of water soluble nitrogenous portion of protein. This may result from the degradation of proteins because of proteolysis and hence it serves as an important constituent for monitoring the proteolysis in fermented milk products like chesses. In heat desiccated products such as *burfi*, it may serve as an indicator of storage related deterioration of milk proteins and some minor solubilization of micellar proteins due to vigorous heat and agitation employed in the process of manufacture of *burfi*. Tabulated values revealed that the soluble nitrogen of *low calorie burfi* samples (Table 1) stored at room temperature was significantly ($P < 0.01$) affected by storage period.

As the storage period advanced, soluble nitrogen increased in *low calorie burfi*. It can be seen that soluble nitrogen content of fresh *low calorie burfi* was significantly ($P < 0.01$) increased from 8.77 % at 0th day to 9.82 at the 7th day of storage at ambient temperature. At refrigerated temperature of storage, a similarly the soluble nitrogen content of *low calorie burfi* increased significantly ($p < 0.01$) from 8.77% to 11.50% in 21 days. The higher soluble nitrogen content observed could be attributed to the heat treatment employed. The phenomenon of the heat treatment on degradative changes in protein is well established (Jenness and Patton, 1969). On the other hand, survival of heat resistant groups of bacteria and heat stable enzymes capable of protein breakdown could be also considered for proportionately higher soluble nitrogen content during storage. The increase in soluble nitrogen content on storage might be the direct consequence of degradation of protein content of *low calorie burfi*. Kumar (2010) reported the tyrosine value of *khoa* samples to be 12.02 mg/100 g of *khoa*. A lesser tyrosine value in standardized product implies to a lower level of protein breakdown as compared to other treatments which can be correlated to lower heat treatment and lesser microbial count.

Effect of storage on TBA value

TBA determination is one of several analytical methods for the evaluation of the degree of oxidation of oils and fats. 2-thiobarbituric acid forms red-coloured products with malonaldehyde, some polyunsaturated aldehydes, dioxolanes and furan derivatives. The intensity of colouration is correlated with the rancidity degree of fats and oils. It can be revealed from the Table 2 that, TBA content of *low calorie burfi* was significantly ($P < 0.01$) increased from an initial value of 0.21 to 0.29 over a period of 21 days at refrigeration temperature, whereas the increase in TBA values noted from 0.21 to 0.27 over a period of 7 days for *Low calorie burfi* at room temperature. At both temperatures the TBA content of *low calorie burfi* significantly increased during the storage period. However, it can be concluded that the TBA values of *low calorie burfi* during storage period increased more rapidly at room temperature than at refrigeration temperature. The increase in TBA values might be due to oxidation of milk fat of *low calorie burfi* during storage. Increase in TBA values during storage were also noticed by Sachdeva and Rajorhia (1982) in *burfi*. In yet another study conducted by Jha et al. (2014), the initial TBA value of 0.179 and 0.184 (absorbance at 532 nm) for *lal/brown peda* samples increased to 0.274 (at 4 °C) and to 0.281 (at 37 °C) in 31 and 9 days respectively.

Effect of storage on sensory properties

The sensory attributes have profound effect on the consumer's preference. Different food products undergo deterioration in sensory profile as a consequence of various chemical and biochemical changes that progress during storage. The effect of

storage period on sensory attributes of *low calorie burfi* stored at ambient temperature is presented in Table 3. The mean value presented revealed that flavour score of *low calorie burfi* was significantly ($P<0.01$) reduced during the storage period. During storage of *low calorie burfi*, flavour score up to 7th day was observed and thereafter the product became unacceptable due to visible mould growth. From the Table 3 and Table 4, it can be seen that the flavour score decreased rapidly at room temperature as compared to refrigerated temperature during storage. The decrease in flavour score could be attributed to slight loss of freshness, which is inherent with any food product.

In fresh product, the compounds formed during browning reactions are responsible for the typical flavour of the product, but as storage period progresses, the chemical reactions disturbed the delicate balance of the compounds. The findings of the present study are in accordance with the result reported by Londhe et al. (2012) for brown *peda* and Sharma et al. (2003) in *Malai Peda* samples during storage study. Similar observations were recorded on stored *kalakand* by Rao and Goyal (2007) and stored *doda burfi* by Chawla et al. (2015).

It can be seen from Table 3 that body and texture score of *low calorie burfi* was significantly ($P<0.01$) influenced by the storage period at room temperature. During storage of *low calorie burfi*, the body and texture score decreased significantly from 7.0 at 0th day to 6.53 at the 7th day of storage. Similarly, at the refrigerated temperature, the body and texture score of *low calorie burfi* was found significantly ($P<0.01$) decreased during storage period (Table 4). The initial mean body and texture score of 7.33% at 0th day decreased to 6.0% at 21st day of storage (Table 4). The decrease in body and texture score of samples was observed in both the samples kept at different temperatures and it was much faster in the product kept at room temperature compared to the samples kept at refrigeration temperatures. A similar decrease in body and texture scores were observed in *multigrain halwa* samples kept at ambient and refrigerated conditions of storage (Itagi et al. 2011).

At room temperature the integrity of the grains remained intact, but the grains became harder and chewier becoming conspicuous in the product as the moisture content reduces. At refrigerated temperature the product became dry, hard, sandy and brittle which might be ascribed to the loss of moisture due to addition of sucralose. This is because of dynamic structural and conformational changes, which may or may not be dependent on changes in moisture content (Navajeevan and Rao, 2005) and can be attributed to decline in hydrophilic groups. Therefore, body and texture was considered as important criteria for determining the acceptability of *low calorie burfi* during storage study particularly at refrigerated temperature. The mean values presented reveals that colour and appearance score of *low calorie burfi* was significantly ($P<0.01$) decreased during the storage period. During storage of *low calorie burfi* at room temperature, decreased in colour and appearance score from 7.83±0.16 at 0th day to 6.98±0.10 at the 7th day of storage was observed. From the Table 3 and Table 4 it can be observed that the changes in colour and appearance scores decreased rapidly at room temperature than at refrigeration temperature. The decline in scores during storage of *low calorie burfi* can be attributed to microbial, chemical and textural changes in the product. During storage the samples became drier in appearance and lacked the greasy appearance desired in good quality *burfi* which resulted in a steady decrease in colour and appearance scores. The colour and appearance of the product became dull and darker with dry appearance. Moreover, in the present study, evaporation of moisture during storage might have aggravated the appearance of the *low calorie burfi* as presence of moisture enlivens the appearance of the product by reflecting incident light. Londhe et al. (2012) reported decrease in colour and appearance score during storage study of brown *Peda* at 30±2 °C using different packaging materials. These results are in accordance with those observed by Chawla et al. (2015) who also noted a decrease in colour and appearance scores of *doda burfi* on storage.

The overall acceptability of stored samples depends upon several factors like degree of proteolysis, extent of lipolysis, flavour

Table 3. Effect of storage on the sensory quality of traditional and *Low calorie burfi* at 37±1°C

Attribute	Sample	Days of storage		Chi square value
		0 th day	7 th day	
Flavour	Traditional burfi	7.33±0.21 ^a	7.50±0.22 ^{ab}	6.00 ^{**}
	<i>Low calorie burfi</i>	8.0±0.001 ^a	7.83±0.21 ^{ab}	6.00 ^{**}
Body and Texture	Traditional burfi	7.33±0.21 ^a	7.0±0.22 ^{ab}	6.00 ^{**}
	<i>Low calorie burfi</i>	7.83±0.16 ^a	6.53±0.16 ^{ab}	6.00 ^{**}
Colour and appearance	Traditional burfi	7.67±0.21 ^a	7.83±0.16 ^{ab}	6.00 ^{**}
	<i>Low calorie burfi</i>	6.50±0.22 ^a	6.98±0.10 ^{ab}	6.00 ^{**}
Sweetness	Traditional burfi	7.33±0.21 ^a	7.50±0.22 ^{ab}	6.00 ^{**}
	<i>Low calorie burfi</i>	8.0±0.01 ^a	8.50±0.01 ^{ab}	6.00 ^{**}
Overall acceptability	Traditional burfi	7.33±0.21 ^a	7.53±0.22 ^{ab}	6.00 ^{**}
	<i>Low calorie burfi</i>	7.67±0.21 ^a	7.98±0.21 ^{ab}	6.00 ^{**}

Figures are mean ± standard error of three replications, ^{**}-Significant at one per cent level ($p<0.01$), ^{a-d} Means with different superscript vary significantly within a row

changes and microbial activity. Statistical analysis indicated a significant ($P < 0.01$) difference among the treatment, viz. type of *burfi* and storage period for both the temperatures studied viz. $37 \pm 1^\circ\text{C}$ and $4 \pm 1^\circ\text{C}$. As observed from the Table 3 and Table 4, the overall acceptability score decreased rapidly at room temperature compared to refrigeration temperature. The decline in overall acceptability scores of *low calorie burfi* was due to changes in flavour and body and texture characteristics. The influence of storage period and temperature of storage was significant for changes in flavour and body and texture and thus, overall acceptability scores. All deteriorative changes, i.e. oxidative, proteolytic, lipolytic, browning, acid development, microbial and textural changes were collectively reflected in sensory quality and thus led to unacceptability of the stored product after a definite period. Low temperature always promotes a longer shelf life of many products and the same was confirmed in this study. This could be attributed to the lower rate of lipid oxidation and non-enzymatic browning reactions as well as reduced rate of unwanted microbial growth which decreases the shelf life of products stored at elevated temperatures (Rossini et al. 2011).

Effect of storage on microbial quality

Most of the milk products are perishable commodities. The perishability of milk products is mostly ruled by microbiological quality of that product. According to BIS (IS: 5550:2005) standards laid down for Burfi, the standard plate count should not be more than 30,000/g and the yeast and mould count not more than 10/g burfi. The microbial count influences the acceptability and hence, shelf life of any product affecting its colour and appearance, flavour and body and texture of the product. The shelf life of product like *low calorie burfi* depends on the growth of microorganisms in the product during storage. Most of the physico-chemical changes as like FFA content, soluble nitrogen, acidity development, change in pH etc., are affected by the presence and growth of various microorganisms. Increase in FFA and soluble nitrogen content signifies lipolytic and proteolytic activity caused by microorganisms. Therefore, the stored samples of *low calorie burfi* were subjected to microbiological analysis for standard plate count (SPC), yeast and mold count (YMC) and coliform count. The influence of period of storage temperature $37 \pm 1^\circ\text{C}$ on the SPC of *low calorie burfi* is presented in Table 5. The mean value

Table 4. Effect of storage on the sensory quality of traditional and *Low calorie burfi* at $4 \pm 1^\circ\text{C}$

Attribute	Sample	Days of storage			Chi square value
		0 th day	7 th day	14 th day	
Flavour	Traditional burfi	7.33±0.21 ^a	7.50±0.22 ^{ac}	5.83±0.30 ^{bc}	8.45 ^{**}
	<i>Low calorie burfi</i>	8.0±0.001 ^a	7.83±0.21 ^{ac}	5.83±0.30 ^{bc}	10.57 ^{**}
Body and Texture	Traditional burfi	7.33±0.21 ^a	7.50±0.22 ^{ac}	6.17±0.30 ^{bc}	7.17 ^{**}
	<i>Low calorie burfi</i>	7.83±0.16 ^a	7.83±0.16 ^{ac}	7.67±0.21 ^{bc}	0.50 ^{ns}
Colour and appearance	Traditional burfi	7.67±0.21 ^a	7.83±0.16 ^{ac}	7.33±0.21 ^{bc}	3.50 ^{ns}
	<i>Low calorie burfi</i>	6.50±0.22 ^a	6.50±0.22 ^{ac}	6.33±0.21 ^{bc}	4.0 ^{ns}
Sweetness	Traditional burfi	7.33±0.21 ^a	7.50±0.22 ^{ac}	7.50±0.22 ^{bc}	5.50 ^{ns}
	<i>Low calorie burfi</i>	8.0±0.01 ^a	8.00±0.01 ^{ac}	7.67±0.21 ^{bc}	4.0 ^{ns}
Overall acceptability	Traditional burfi	7.33±0.21 ^a	7.50±0.22 ^{ac}	7.33±0.21 ^{bc}	0.66 ^{ns}
	<i>Low calorie burfi</i>	7.67±0.21 ^a	7.67±0.21 ^{ac}	7.67±0.21 ^{bc}	0.23 ^{ns}

Figures are mean ± standard error of three replications, ^{**}-Significant at one per cent level ($p < 0.01$), ^{a-d} Means with different superscript vary significantly within a row

Table 5 Effect of storage on microbial quality of traditional and *Low calorie burfi* at $37 \pm 1^\circ\text{C}$

Sample	Days of storage	
	0 th day	7 th day
	Standard plate count (\log_{10} cfu/g)	
Traditional burfi	2.47±0.06 ^{ax}	4.46±0.14 ^{bx}
<i>Low calorie burfi</i>	2.49±0.06 ^{ay}	5.08±0.14 ^{cy}
	Coliform count (\log_{10} cfu/g)	
Traditional burfi	Absent	Absent
<i>Low calorie burfi</i>	Absent	Absent
	Yeast and mold count (\log_{10} cfu/g)	
Traditional burfi	Absent	1.93±0.05 ^{by}
<i>Low calorie burfi</i>	Absent	1.32±0.05 ^{cy}

Figures are mean ± standard error of three replications, ^{a-d} Means with different superscript vary significantly within a row, ^{x-y} Mean with different superscripts vary significantly within a column.

Table 6. Effect of storage on microbial quality of traditional and *Low calorie burfi* at 4±1°C

Sample	Days of storage			
	0 th day	7 th day	14 th day	21 st day (spoiled)
		Standard plate count (log ₁₀ cfu/g)		
Traditional burfi	2.47±0.06 ^{ax}	3.58±0.06 ^{bx}	4.46±0.11 ^{cx}	4.76±0.10 ^{dx}
<i>Low calorie burfi</i>	2.49±0.06 ^{ay}	3.73±0.06 ^{by}	4.58±0.11 ^{cy}	5.08±0.10 ^{dy}
		Coliform count (log ₁₀ cfu/g)		
Traditional burfi	Absent	Absent	Absent	Absent
<i>Low calorie burfi</i>	Absent	Absent	Absent	Absent
		Yeast and mold count (log ₁₀ cfu/g)		
Traditional burfi	Absent	0.91±0.03 ^{bx}	1.45±0.06 ^{cx}	1.93±0.39 ^{dx}
<i>Low calorie burfi</i>	Absent	0.92±0.31 ^{by}	1.11±0.06 ^{cy}	1.42±0.03 ^{dy}

presented reveals that SPC of *low calorie burfi* was significantly ($P < 0.01$) influenced by storage period. The initial mean SPC of 2.49±0.06 log cfu/g at 0th day increased to 5.08±0.14 log cfu/g at the 7th day of storage in *low calorie burfi* at 37±1°C. During storage of *low calorie burfi* at refrigerated temperature also, a significant ($P < 0.01$) increase in SPC up to 21st day was observed and thereafter the product was found unacceptable due to visible mold growth. From the Table 5 & 6, it can be seen that the SPC increases rapidly at room temperature compared to refrigeration temperature during storage period. Jha et al. (2015) reported that SPC of *lal peda* samples increased from initial value of 4.60 to 6.38 log₁₀ cfu/g in 30 days when stored at 10 °C. Londhe et al. (2012) reported an increase of 2.6 to 4.3 log₁₀ cfu/g for SPC in *lal peda* in 20 days when stored at 30 °C. The coliform count was absent in *low calorie burfi* both temperatures stored at 4 ± 1°C and 37±1°C during the storage period of 21 and 7 days respectively. Gautam et al. (2012) reported that coliform count was absent in retort processed *chhana kheer* during 90 days storage period. For most of the intermediate moisture Indian dairy foods such as *Peda*, *Burfi*, *Kalakand*, etc. mould growth tends to be a major problem and often most important single factor limiting their shelf life. The influence of period of storage at room temperature (37±1°C) on the yeast and mold count of *low calorie burfi* is presented in Table 5. The mean values presented reveals that Yeast and mould count of *low calorie burfi* was found nil on 0th day of storage. During further storage of *low calorie burfi*, increase in yeast and mold count on 7th day was observed and thereafter the product was found unacceptable due to visible mold growth. The influence of period of storage at refrigeration temperature (4±2°C) on the yeast and mold count of *low calorie burfi* is presented in Table 6. The mean values presented reveals that yeast and mould count of *low calorie burfi* was found nil in 0th day of storage. During further storage of *low calorie burfi*, increase in yeast and mold count up to 21st day was observed and the product was found unacceptable due to visible mold growth. From the Table 5 and Table 6, it can be seen that the yeast and mold count increased rapidly with storage period at

room temperature compared to refrigerated temperature. The colonies obtained in the present study at room temperature storage were white and green colonies. The numbers of the fungal colonies obtained during present investigation are similar to various workers who had analysed the milk products like *Peda*, *burfi* and *Kalakand*. The results are in harmony with findings of Ghayal et al. 2014 in dietetic *rabri*. Jha et al. (2013) reported an increase in yeast and mold count of *peda* samples from 2.70 to 3.16 log₁₀ cfu/g in 30 days of storage at 10°C. The product was found to be free from coliforms during storage at both temperatures.

Conclusion

Low calorie burfi packed in low density polyethylene (LDPE) has shown shelf life of 7 and 20 days at storage at room temperature (37±1°C) and refrigerated temperature (4±1°C) respectively. The sucralose content estimated by HPTLC had shown recovery of ~98%. The combination of *costus speciosus extract* and sucralose had not shown marked influence on the sensory parameters. However further investigations are required for establishing scientific knowledge on antidiabetic properties of *costus speciosus extract* added low calorie burfi.

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