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Quality and Shelf Life of Ready to Cook Mozzarella Cheese Stick Snack with Different Packaging Materials and Methods under Refrigerated Storage

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ABSTRACT

Background: The effect of packaging material (polyethylene and laminated pouches) and method of packaging (atmospheric and vacuum packaging) on the shelf-life of mozzarella cheese stick snacks stored under refrigerated conditions was studied.

Methods: The work was carried out in 2019-20 when the mozzarella cheese preparation and the process to make a battered and breaded mozzarella cheese sticks snacks were standardized. The snacks were then studied for its quality and shelf life in different packaging materials (polythene and laminated pouches) under atmospheric and vacuum conditions under refrigerated conditions.

Result: There was a significant increase in acidity, free fatty acids (% oleic acid) and non-protein nitrogen of the samples during refrigerated storage. Though the chemical changes were not so great as to render the product unacceptable, however the refrigeration temperature resulted in a rapid increase in the microbial count. As a result, the samples with atmospheric packaging showed visible microbial growth on the 12th day while those with vacuum packaging spoiled on the 13th day.

Key words: Mozzarella cheese sticks snacks, Packaging, Pre-frying, Quality, Refrigerated storage, Shelf-life.

INTRODUCTION

Mozzarella cheese is a soft unripened cheese variety of the pasta-filata family and has a unique property called stretchability- ability to form fibres or strings when hot (Jana and Tagalpallewar, 2017). Though mozzarella cheese is used in a variety of preparations in foreign countries such as *lasagne*, veal cutlet, *allaparmagiana*, *bruschetta*, *etc.,* its use in India is limited as toppings on pizza. However, apart from the traditional pizza topping mozzarella cheese can be used for the preparation of fried snack foods, a product similar to the traditional *paneer pakora*, but yet different from it due to the inherent stretching and melting characteristics of mozzarella cheese (Mijan *et al*., 2010). The snack market is predominant with potato-based products in the vegetarian section which are invariably high in carbohydrates and lacking in protein. Cheese has high protein content and ready-to-cook snacks made with it will fill this gap in the market. Moreover, convenient ready-to-cook snacks are getting popular owing to the ease attached to them. The present investigation aimed at developing a convenient mozzarella cheese sticks snack and studying their shelf-life stability and quality under refrigerated storage.

MATERIALS AND METHODS

Raw materials

The work was carried out in the Department of Food Science and Technology, Punjab Agricultural University, Ludhiana in the year 2019-20. Fresh, whole buffalo milk, commercial citric acid, gram flour, wheat flour, eggs, breadcrumbs, salt, black pepper, sugar, Bakery shortening (*Gagan* Brand, manufactured by *Gagan Vanaspati* Ltd.), Baker's yeast (*Saccharomyces cerevisiae*) and frying oil (Rice bran oil of brand "Ricella") were obtained from local market. Commercial powdered microbial rennet was procured from Valiren Valley Inc., U.S.A. and kept at refrigerated temperature.

Preparation of mozzarella cheese

All equipment were washed and sanitized with 50 ppm of chlorine. Pasteurized water was used during all stages of cheese making. For the preparation of desirable quality cheese, milk with fat percentage of 1, 3 and 5% were taken initially. Snacks containing cheese with 1 per cent fat milk showed significantly lower mean sensory scores for flavour, texture and chewability and hence these were rejected. There was no significant difference in the sensory scores for cheese snacks prepared from milk adjusted to 3 and 5 per cent fat levels. But in preparation of cheese with

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5 per cent fat milk, there were significantly higher losses of fat and total solids in whey, cooking water and stretching water. Hence 3 per cent fat milk was selected for mozzarella cheese preparation in this study. Fig 1 depicts the flow diagram for the preparation of mozzarella cheese.

Preparation of mozzarella cheese stick snacks

For preparation of mozzarella cheese stick snacks, mozzarella cheese sticks were cut (5×1×1 cm size), dipped in batter of gram flour + water (1:1), coated with breadcrumbs, patted and rested. The battered and breaded sticks were pre-fried, cooled and packed. For packaging and storage of snacks two parameters were considered.

1 Packaging material: Polyethylene (200 gauge) and Laminated pouches (200 gauge).

2 Packaging method: Atmospheric and Vacuum.

The samples were kept at refrigeration (4±2°C) conditions.

Analysis of samples

The raw material and mozzarella cheese snacks were analyzed for their proximate composition according to AOAC (2000). Carbohydrate content was calculated by difference. The mozzarella cheese snacks were further analyzed for total solids, acidity, free fatty acid content and non-protein nitrogen according to standard procedures prescribed by AOAC (2000).

Sensory evaluation

Organoleptic evaluation of the cheese snacks was performed by a panel of 10 semi-trained judges. The stored mozzarella sticks snacks were taken out of refrigerated storage, deep fried in vegetable refined oil at 180°C till golden brown color of the crust was achieved, drained and served hot to the panelists. The sensory evaluation was carried out for appearance, flavour, texture and overall acceptability using the 9-point hedonic rating scale (Amerine *et al.,* 1965).

Microbiological analysis

Microbiological analysis of the stored refrigerated samples was carried out at regular intervals for total plate count, Yeast and mould count. The total plate count was determined by using the nutrient agar media while the yeast and mould count was determined using glucose yeast agar media.

Statistical analysis

The data collected from the studies was statistically analyzed and subjected to analysis of variance using CPCS1 software.

RESULTS AND DISCUSSION

The proximate composition of raw material and mozzarella cheese snacks is given under Table 1. It is observed that the developed product has a comparable amount of fat and carbohydrates along with an appreciable amount of protein

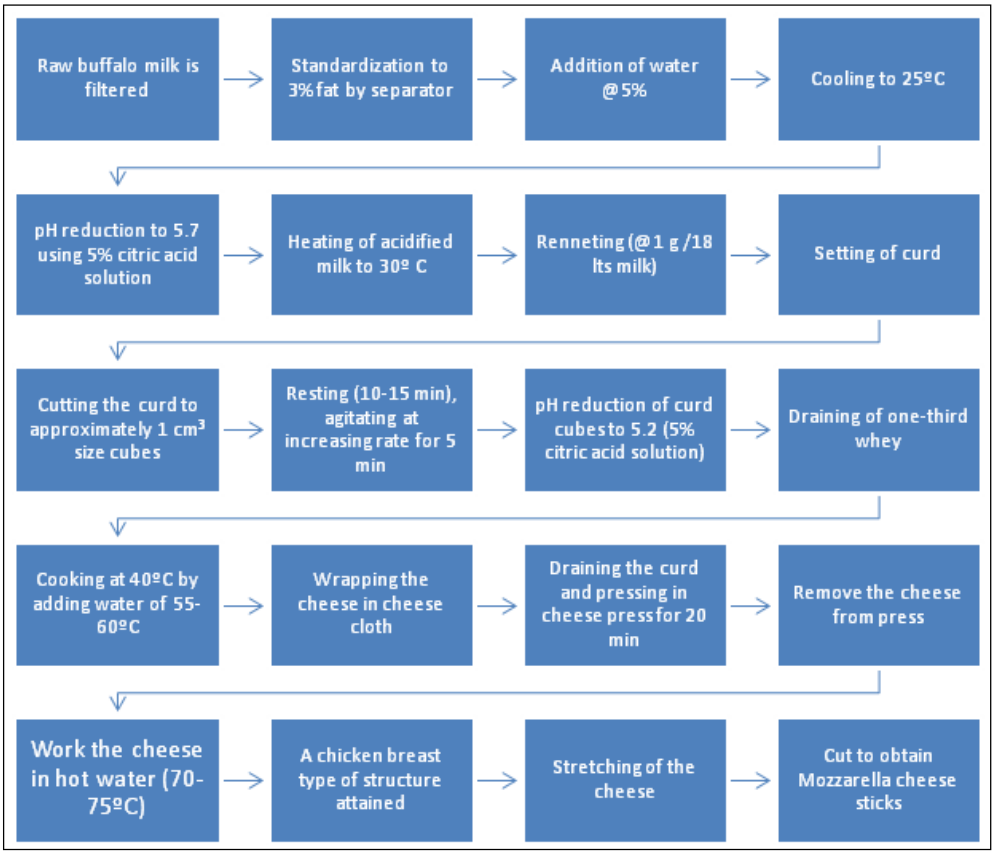

Fig 1: Flow diagram depicting preparation of mozzarella cheese.

as well. The snacks available in the market are deficient in protein content.

Effect of packaging material and method of packaging on the chemical characteristics of pre-fried snacks stored under refrigerated conditions

The effect of packaging material and method of packaging on the various parameters of pre-fried mozzarella cheese stick snacks stored under refrigerated conditions is given in Table 2. The initial moisture content of the snacks was 49.30 per cent and after 11 days of refrigerated storage it was found to be 49.0 per cent, hence, no significant reduction in moisture content was observed. The lowest moisture content (49%) was for snacks stored in atmospheric sealed polyethylene pouches and highest (49.12%) for laminated pouches sealed under vacuum conditions after 11 days.

There was a significant increase in the titrable acidity of the samples from 0.52 per cent to 0.561, 0.558, 0.555 and 0.550 per cent for samples stored in atmospheric packaged polyethylene, laminated, vacuum packaged polyethylene and laminated pouches, respectively. However, there was no significant difference between acidities of all the samples after 11 days.

There was a significant increase in FFA of snacks from 0.14 per cent to 0.30, 0.27, 0.25 and 0.20 per cent after 11 days of refrigerated storage, for samples in atmospheric packaged polyethylene, laminated, vacuum packaged polyethylene and laminated pouches, respectively. The FFA denotes the rancidity parameter indicating the oxidation of fat during storage of food products which cause off-flavor. The FFA was significantly greater for samples in polyethylene than laminated pouches for both packaging methods. Similarly, in case of both the packaging materials, significantly higher FFA content was observed for samples packed under atmospheric than vacuum conditions. Maity *et al.* (2012) reported a steady increase in FFA per cent in the ready-to-fry frozen vegetable snack during its storage studies. NPN content increased from 0.04 per cent to a maximum of 0.32 per cent on the 11th day, for samples packed in atmospheric packaged polyethylene pouches.

Table 1: Chemical composition of raw material and standardized mozzarella cheese sticks snack.

Parameters (%)	Milk	Gram flour	Wheat flour	Breadcrumbs	Snack
Total solids	13.32	82.18	85.76	88.09	50.66
Protein	4.53	19.73	10.08	11.89	6.89
Fat	3.00	0.89	0.86	0.95	22.11
Ash	0.83	0.68	0.59	2.26	0.39
Carbohydrates	4.96	60.88	74.23	72.99	21.27

Table 2: Effect of packaging material and method of packaging on the parameters of pre-fried mozzarella cheese stick snacks stored under refrigerated conditions.

Parameter	Storage days	Atmospheric		Vacuum		CD (5%)
		P	L	P	L	
Moisture (%)	0	49.30	49.30	49.30	49.30	Storage time NS; Packaging method NS; Storage time × Packaging method NS; Packaging material NS; Storage time × Packaging material NS; Packaging method × Packaging material NS; Storage time × Packaging method × Packaging material NS
	4	49.23	49.26	49.26	49.28	
	7	49.12	49.17	49.19	49.20	
	11	49.00	49.04	49.09	49.12	
Acidity (%)	0	0.52	0.52	0.52	0.52	Storage time 0.008; Packaging method NS; Storage time × Packaging method NS; Packaging material NS; Storage time × Packaging material NS; Packaging method × Packaging material NS; Storage time × Packaging method × Packaging material NS
	4	0.537	0.535	0.534	0.531	
	7	0.548	0.546	0.543	0.539	
	11	0.561	0.558	.555	0.550	
Free fatty acids (% oleic acid)	0	0.14	0.14	0.14	0.14	Storage time 0.02; Packaging method 0.01; Storage time × Packaging method 0.02; Packaging material 0.01; Storage time × Packaging material NS; Packaging method × Packaging material NS; Storage time × Packaging method × Packaging material NS
	4	0.20	0.19	0.18	0.16	
	7	0.25	0.24	0.22	0.19	
	11	0.30	0.27	0.25	0.20	
Non-protein nitrogen (%)	0	0.04	0.04	0.04	0.04	Storage time 0.02; Packaging method 0.01; Storage time × Packaging method 0.02; Packaging material 0.01; Storage time × Packaging material NS; Packaging method × Packaging material NS; Storage time × Packaging method × Packaging material NS
	4	0.11	0.1	0.1	0.09	
	7	0.23	0.21	0.19	0.18	
	11	0.32	0.29	0.27	0.23	

P: Polythene; L: Laminated.

The minimum NPN content (0.23%) was in case of vacuum packaged laminated pouches. NPN was significantly greater for samples packed in polyethylene than laminated pouches, and under atmospheric than vacuum packaging for similar packaging methods and packaging material, respectively. The extent of proteolysis was greater in all the samples as compared to similar samples stored under frozen conditions. Similar results were observed by Ghosh and Singh (1991) who reported that proteolysis in refrigerated samples followed the same trend as deep-frozen but were considerably faster. Faster proteolysis observed in refrigerated samples may be due to fast enzymatic action as reported by Alinovi *et al* (2020).

Effect of packaging material and method of packaging on the sensory characteristics of pre-fried snacks stored under refrigerated conditions

The data regarding the effect of packaging material and method of packaging on the sensory scores of snacks stored under refrigerated conditions is given in Table 3. There was a significant reduction from initial appearance scores (8.85) to samples stored for 11 days of refrigerated storage, the highest scores (8.40) were for samples stored in polyethylene pouches under atmospheric conditions and lowest (7.05) in vacuum packaged laminated pouches. After 11 days of refrigerated storage the flavour scores for snacks packed in atmospheric packaged polyethylene, laminated, vacuum packaged polyethylene and laminated pouches were 7.25, 7.45, 7.70 and 7.80, respectively. The flavour scores were related to acidity, FFA and NPN development in the samples. The samples packed under vacuum packaging had significantly higher flavour scores than atmospheric packaging, irrespective of the packaging material. However, no significant effect of packaging material was observed on the flavour scores of the samples.

After 11 days of refrigerated storage, minimum texture scores (6.85) were recorded for sample stored in vacuum packaged laminated pouches and these were significantly less than atmospheric packaged polyethylene (7.45), laminated (7.30) and vacuum packaged polyethylene pouches (7.20). Alam *et al.* (2016) while studying the effect of MAP on textural properties of mozzarella cheese also reported that the cheese packed in 100% CO_2 showed minimum changes in the textural quality during storage.

The scores for overall acceptability decreased significantly upon refrigerated storage and after 11 days the highest scores (7.70) were obtained for samples stored in polyethylene pouches packaged under atmospheric conditions. The minimum chemical degradation was observed for samples in vacuum packaged laminated pouches, but it had lower overall acceptability scores due to significantly low scores for appearance as well as texture.

Although the sensory scores of all the samples differed significantly after 11 days of refrigerated storage, all scores were found to be acceptable. However, there was fungal growth on samples stored in atmospheric packaged polyethylene and laminated pouches on the 13th day and on vacuum packaged polyethylene samples on the 14th day. The sample stored in vacuum packaged laminated pouches did not have any visible fungal growth; but gave a typical off smell on 14th day. So, further analyses of these samples were discontinued.

Table 3: Effect of packaging material and method of packaging on the sensory scores of pre-fried mozzarella cheese stick snacks stored under refrigerated conditions.

Parameter	Storage period (days)	Atmospheric		Vacuum	
		Polyethylene	Laminated	Polyethylene	Laminated
Appearance	0	8.85	8.85	8.85	8.85
	4	8.75	8.55	8.30	8.10
	7	8.45	8.25	7.90	7.55
	11	8.40	8.10	7.75	7.05
Flavor	0	8.65	8.65	8.65	8.65
	4	8.40	8.40	8.45	8.45
	7	7.40	7.45	7.90	7.85
	11	7.25	7.45	7.70	7.80
Texture	0	8.60	8.60	8.60	8.60
	4	8.20	8.25	8.10	7.65
	7	7.95	7.60	7.60	7.25
	11	7.45	7.30	7.20	6.85
Overall acceptability	0	8.70	8.70	8.70	8.70
	4	8.35	8.37	8.29	8.07
	7	8.03	7.87	7.80	7.55
	11	7.70	7.62	7.55	7.23

CD (5%): Storage time 0.16; Packaging method 0.11; Storage time × Packaging method 0.22; Packaging material 0.11; Storage time × Packaging material NS; Packaging method × Packaging material NS; Storage time × Packaging method × Packaging material NS.

Table 4: Effect of packaging material and method of packaging on the total plate count (TPC) (dilution 10^{-1}) and yeast and mould (Y&M) count (cfu/g) of pre-fried mozzarella cheese stick snacks stored under refrigeration conditions.

Storage period (days)	Packaging							
	Atmospheric				Vacuum			
	Polyethylene		Laminated		Polyet		Laminated	
	TPC	Y and M	TPC	Y and M	TPC	Y and M	TPC	Y and M
0	31	-	31	-	31	-	31	-
4	57	-	55	-	55	-	53	-
7	78	48	75	47	71	45	69	41
11	120	76	120	75	100	63	95	58
13	Visible growth	Fungal	Visible growth	Fungal	-		-	
14	-		-		Visible growth	Fungal	Off-odor	

Effect of packaging material and method of packaging on the microbiological condition of pre-fried snacks stored under refrigerated conditions

Experimental data showing the effect of packaging material and method of packaging on the total plate count of pre-fried snacks stored under refrigerated conditions is given in Table 4. There was a significant increase in the total plate count during refrigerated storage and the final TPC after 11 days was maximum (1.2×10^3 cfu/g) for sample stored in atmospheric packaged polyethylene pouches and minimum (9.5×10^2 cfu/g) for vacuum packaged laminated pouches. Alam and Goyal (2011) also reported an increase in the standard plate count of mozzarella cheese from 6.1 log cfu/g to 6.8 log cfu/g which was packed in cryovac and LDPE pouches. Mozzarella cheese maintained its microbiological characteristics until 7 days from production when stored at 18-20°C as studied by Losito *et al.* (2014).

Experimental data for the effect of packaging material and method of packaging on the Yeast and Mould Count of snacks stored under refrigerated storage is shown in Table 4. After 11 days, the snacks stored in vacuum packaged laminated pouches recorded minimum (58 cfu/g) and atmospheric packaged polyethylene samples maximum (76 cfu/g) count of yeast and mould. A similar trend of increase in the mean value of yeast and mould counts were reported by Alam and Goyal (2011) during the packaging studies of mozzarella cheese stored in different packages. A gradual increase in the yeast and mold counts during the period of 4 weeks was observed by Rehman *et al*. (2017) in mozzarella cheese made by *L. kefiranofaciens*. The growth of yeast is due to the low pH, low moisture content, low temperature and high salt levels as deduced by Sulieman *et al*. (2013).

CONCLUSION

To meet the demand of consumers of good nutritive quality convenient snacks, mozzarella cheese snacks are ideal. It was found that pre-fried breaded mozzarella cheese sticks can be stored under refrigeration up to 11 days. Hence, refrigerated storage can be used for mozzarella cheese sticks when there is a continuous demand for the product. This value-added product will help to utilize the surplus milk, add income to the farmer and provide a vegetarian snack with good protein content as well as taste to the consumers.

Conflict of interest

All authors declare that they have no conflict of interest.

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