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Research Article

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Microbiological and Sensory Quality of Mozzarella Cheese as Affected by Type of Milk and Storage

Abdel Moneim E. Sulieman[1*], Rasha A. Mohamed Ali[2] and Kamal A. Abdel Razig[3]

Abstract

Mozzarella cheese that had its origin in Italy has recently been introduced by the Sudanese dairy industry. Mozzarella cheese is a good source of nutrients namely protein, fat, minerals and vitamins. The study was performed to prepare and assess the storage stability of mozzarella cheese prepared from cow milk, goat milk and mixed milk (cow milk: goat milk-1:1, w/w). The cheeses were subjected to microbiological analyses and sensory evaluation at storage temperature of 5°C and at intervals of 1, 15 and 30 days. The goat milk mozzarella cheese (GMMC) had significantly higher microbial load than mixed milk mozzarella cheese (MMMC) and cow milk mozzarella cheese (CMMC). The *Salmonella* sp. was not detected in any of the mozzarella cheeses throughout the storage period, while coliform which was initially present could not be detected at 30th day of storage. There was non-significant difference in the sensory attributes of CMMC and MMMC, which was also the case throughout their storage period. Goat's milk cheese had the least overall acceptability sensory score. Manufacture of Mozzarella cheese from mixed cow and goat milk (1:1) is recommended for industrial exploitation.

Keywords

Sensory evaluation; Total viable count; Yeast and Mold; Coliform

Introduction

Cheese is the fresh or matured product obtained by the drainage (of liquid) after coagulation of milk through use of rennet and starter [1]. Cheese is a popular food due to its diversity in application, nutritional value, convenience and typical taste. The diversity is due to an increasing knowledge of the technology of cheese making and the biochemistry and microbiology of cheese ripening [2].

Cheese making is a major milk preservation method in Sudan [3]. The major traditional cheese types produced in Sudan are Gibna Bayda, Gibna Mudaffra and Mish cheeses [4,5]. Such cheeses varies in composition, texture, colour, taste and flavour, due to the varied composition of milk, production methods, microbial flora, type of microbial activity during ripening and the ripening conditions.

Mozzarella cheese which had its origin in the Battipaglia region of Italy has been introduced recently by the Sudanese industry [6]. In Italy, the cheese makers are still preparing Mozzarella from buffalo milk. However, Mozzarella cheese is being made all over Italy, in other European countries and USA from cow milk with certain modifications [7]. Mozzarella belongs to the "Pasta filata" family of cheeses, which involves skillfully stretching the curd in hot water to get a smooth textured cheese. The cheese is soft, white, unripened, that may be consumed shortly after manufacture. Its melting and stretching characteristics are highly appreciated in the manufacture of pizza, where it serves as a key ingredient.

Mozzarella cheese is available in a large number of forms and sizes ranging from 50 g to 50 kg [8,9]. Mozzarella cheese production in Sudan is a small business. Such cheese has been manufactured and introduced to the market due to the recent popularity of Italian dishes, which was first practiced in Khartoum Dairy Product Company (KDPC) in 1992 and then practiced by few other small processing units. There is a great demand for such cheese by the large hotels and pizza centers. However, no standard procedure is adopted by the different producers for its production.

The research on this type of cheese in Sudan is very limited. The objectives of the present study was to produce mozzarella cheese from cow's milk, goat's milk and mixed milk (1:1 w/w), and to assess their microbiological quality throughout its refrigerated storage.

Materials and Methods

Materials

Fresh cow's and goat's milk were obtained from Khartoum North (Hilat koko). They were collected in sterilized glass bottles immediately after milking, and kept in refrigerator at 5°C. Powdered calf's rennet (Hansen Sticks, France) was obtained from local market and fresh Capo yoghurt used as starter culture was also obtained from local market.

Preparation of mozzarella cheese

Mozzarella cheese was made from three types of milk viz., cow's milk, goat's milk and mixed milk. Cow's milk had 3.6% fat, 3.4% protein, 14.0% TS and a pH of 6.8, while goat's milk had 3.7% fat, 3.6% protein, 14.5% TS and a pH of 6.7. The mixed milk had 3.65 % fat, 3.5% protein, 14.0% TS and a pH of 6.8.

Firstly, 124.5 liters of milk (standardized to 3.15% fat) was filtered and heated to 38°C, then 160 g of starter culture (Capo Yoghurt) and 1 g of calf rennet was added to the mixture. The mixture was blended for 10 minutes, cut and left undisturbed for 30-60 minutes until the curd separated from the whey. The curd was collected, broken and laid on a table to drain and to age for about 20 minutes; the pH of the curd was 5.7. Hot water (4000 liter at 90°C) was added to the curd in a bowl; the curd was mixed and kneaded by hand until homogeneous paste was obtained. The curd was immersed in cold (6°C) brine (12% NaCl) for 6 hours, and then allowed to dry for 4 hours. Finally, the cheese curd was packed in pre-sterilized polyethylene plastic bags and kept for 1 month at (4°C) for further study.

Sensory evaluation

All samples of Mozzarella cheese prepared previously were

***Corresponding author:** Abdel Moneim E. Sulieman, Faculty of Science, Department of Biology, University of Hail, Hail, Kingdom of Saudi Arabia, Tel: +966538085439; Email: am.sulieman@uofh.edu.sa

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subjected to sensory evaluation by ten well trained panelists. They were asked to judge the appearance, texture and flavor of mozzarella cheese using 5-point hedonic scale [10]. All testing took place between 1:00 and 2:00 PM in a room maintained at 28°C.

Statistical analysis

Data obtained were subjected to analysis of variance (ANOVA) as shown by Snedecor and Cochran [11] and means were compared using Duncan's multiple-range test with probability $p \leq 0.05$.

Microbial quality assessment

Mozzarella cheese samples were taken for analyses aseptically after few hours of cooling in pre-sterilized glass containers.

The microbial quality of mozzarella cheese prepared from the 3 types of milk (cow's milk, goat's milk and a mixture of both) was assessed using the following tests:

Total viable count

Total viable count was carried out using the plate method described by Harrigan and MacCance [12]. One ml aliquot from a suitable dilution was transferred aseptically into sterile petri dishes. To each dilution, 10-15 ml of melted and cooled (42°C) plate count agar was added. The inoculums were mixed with media and allowed to solidify. The plates were then incubated in an incubator at 37°C for 48 hours. A colony counter (Quebec colony counter, Germany) was used to count the viable bacteria (colonies) and the results were presented as cfu/g of cheese.

Yeast and mould count

From suitable dilution of samples, 0.1 ml was aseptically transferred onto solidified Potato Dextrose Agar (PDA) medium containing 0.1 g cholramphenicol per liter of medium to inhibit bacterial growth. Samples were spread all over the plates using a sterile bent glass rod. The plates were then incubated at 26-28°C for 48 hours [12]. Colony forming units (cfu) were counted using a colony counter.

Coliform count

One ml of the dilution was inoculated aseptically, in triplicates, to 9 ml of sterilized MacConkey broth using the Durham tubes. The tubes were incubated at 37°C for 48 hours. Positive tubes showed gas in the Durham tubes. The positive tubes (Table 1) were subcultured into EC broth medium and then incubated at 37.5°C or 24 hours to determine the faecal coliform bacteria; the tubes showing any degree of gas production were considered positive.

Salmonella count

Ten grams of each sample were weighed aseptically and mixed well with 90 ml of sterile nutrient broth (NB) and incubated at 37°C for 24 hours. A full loop of incubated NB was transferred aseptically into sterilized Selenite broth and incubated at 37°C for 24 hours. A full loop of 24 hours inoculum was streak-plated on Bismuth Sulphate Agar surface and incubated at 37°C for 24 hours. *Salmonella* colonies appeared as grey colonies with black centers (black metallic sheen). To carry out a confirmatory test, a colony was taken and sub-cultured in sugar iron agar. Production of black color at the bottom of the tube confirmed the presence of *Salmonella* [12].

Results and Discussion

Cheese composition and yield

The particulars of mozzarella cheese prepared from cow's milk, goat's milk and mixed milk with regard to their composition and percent yield is furnished in table 2.

Microbiological characteristics

Figure 1 shows the result of total viable bacteria (TVB) during storage of mozzarella cheese prepared from cow's (CMMC), goat's (GMMC) and mixed milk (MMMC) (1:1 cow's milk:goat's milk). The TVB count of CMMC was significantly lower ($\log_{10}$ 5.62 ± 0.1) than that of MMMC ($\log_{10}$ 5.70 ± 0.01) and GMMC ($\log_{10}$ 6.42 ± 0.01) at 1st day of storage. However, the TVB count of GMMC was significantly lower ($\log_{10}$ 8.699 ± 0.01) than that of CMMC ($\log_{10}$ 8.75 ± 0.01) and MMMC ($\log_{10}$ 9.46 ± 0.01) at 30th day of storage. Santos and Genigcorgis reported that the factors contributing to presence of contaminating bacteria in cheese are ineffective starter culture activity, post-pasteurization contamination of milk and post-process (plasticizing of curd) contamination of cheese.

Yeast and mould count at three different intervals of storage (i.e., 1st, 15th and 30th day) for GMMC ($\log_{10}$ 4.42 ± 0.01, $\log_{10}$ 4.6 ± 0.01, $\log_{10}$ 4.82 ± 0.01) was significantly (was statistical analysis done for microbial count?) higher than that for CMMC ($\log_{10}$ 3.42 ± 0.01, $\log_{10}$ 3.67 ± 0.1, $\log_{10}$ 3.97 ± 0.1) and MMMC ($\log_{10}$ 3.02 ± 0.1, $\log_{10}$ 3.03 ± 0.1), $\log_{10}$ 3.30 ± 0.01) (Figure 2). Fleet and Mian [13] reported that low pH, low moisture content, low temperature and high salt levels of cheese during ripening can lead to rapid growth of yeast.

The coliform count of CMMC was significantly lower ($\log_{10}$ 4.33 ± 0.01 cfu/g) than that of MMMC ($\log_{10}$ 3.00 ± 0.1 cfu/g) and GMMC ($\log_{10}$ 4.43 ± 0.01 cfu/g) when assessed at 1st day of storage (Figure 3). However, the coliform count of MMMC was significantly ($P<0.05$) lower ($\log_{10}$ 4.00 ± 0.1 cfu/g) than that of CMMC ($\log_{10}$ 3.67 ± 0.1 cfu/g) and GMMC ($\log_{10}$ 4.60 ± 0.01 cfu/g) at 15th day of storage. No significant difference was found in coliform count of all mozzarella cheeses at 30th day of storage. Kosikowski [14] reported that coliform

Table 1: MPN table for 10 tubes.

Number of positive tubes MPN per 100 mL	MPN per 100 mL
0	<1.1
1	1.1
2	2.2
3	3.6
4	5.1
5	6.9
6	9.2
7	12
8	16.1
9	23
10	>23

Table 2: Proximate composition of Mozzarella cheeses.

Cheese made from	Moisture	Fat	Protein	pH	Yield (%)
Cow milk	45.2	20.3	25.4	5.3	11.4
Goat milk	44.8	22.3	28.1	5.4	12.1
Mixed milk	44.9	21.0	26.4	5.3	11.8

grow well in cold or warm cheese milk, and in cheese curd their number may reach up to 100-500 million per gram if their initial level in the milk was high and especially if the acid development was slow. Coliform do not survive in pasteurized cheese milk, but may be present in the resulting cheese because of post-pasteurization contamination. *Salmonella* was not detected in any of the mozzarella cheeses, when fresh and also during entire storage period.

Sensory evaluation

The sensory scores of the mozzarella cheese prepared from cow, goat and mixed milk are summarized in tables 3 and 4. The appearance includes color, shape and finish of the mozzarella cheese. The result showed that mozzarella cheese prepared from cow's milk had the highest score for appearance, while the one prepared from goat's milk had the least appearance score; such was the case throughout the storage period.

No significant difference (Table 3) was observed for appearance score of mozzarella cheeses prepared from mixed milk and goat's milk. However, there was a significant difference ($P \leq 0.05$) in the appearance score of mozzarella cheese prepared from cow's milk at different storage interval. There was no significant change in the appearance score of mozzarella cheeses when compared between 1st and 15th day of storage.

The color of milk and thus of resultant cheese made thereof can vary with the season, diet of the milk animals and the use of different type of milk i.e., buffalo, cow, goat [15,16].

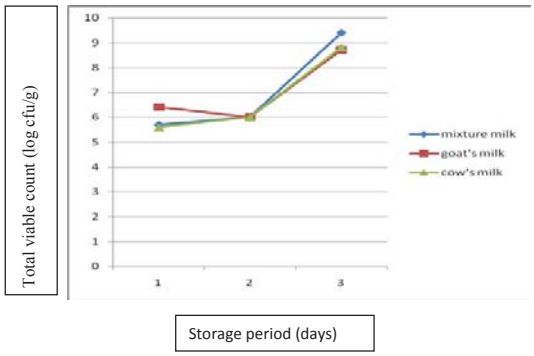

Figure 1: The effect of storage on total viable count of mozzarella cheeses.

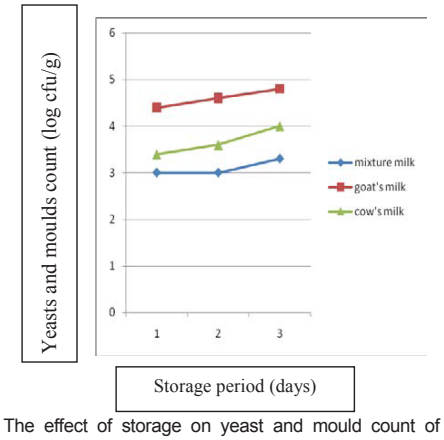

Figure 2: The effect of storage on yeast and mould count of mozzarella cheeses.

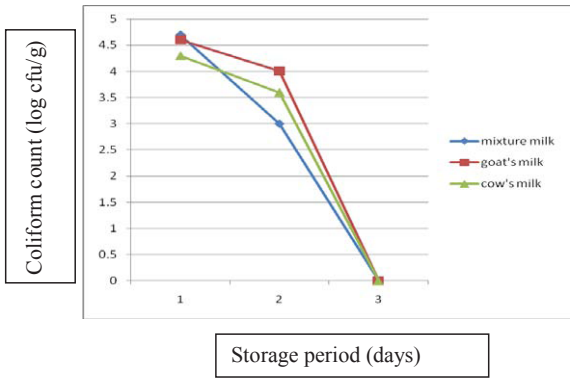

Figure 3: The effect of storage on coliform count of mozzarella cheeses.

Mozzarella cheese prepared from cow's milk had the highest flavor score. But, mozzarella cheese prepared from goat's milk had the least flavor score, which was also the case throughout its storage period (Table 4); this could be attributed to the unpleasant flavor naturally present in goat's milk which are disliked by many people.

Non-significant differences were found in flavor score of mozzarella cheese prepared from mixed and goat's milk. However, there was significant difference ($P \leq 0.05$) on flavor score (decrease in flavor score) of cow's milk mozzarella cheese during the progressive storage. When compared on the same day (i.e., 1st, 15th or 30th day), the flavor score of mozzarella cheeses made from the three type of milk did not differ significantly from each other.

The body and texture of mozzarella cheese is dictated by the chemical composition of the milk (especially protein and calcium content) from which it is made and also to the manufacturing process adopted. Table 3 showed that mozzarella cheese prepared from cow's milk had the highest texture score from amongst the three cheeses prepared. But, mozzarella cheese prepared from goat's milk had the least texture score because of it was too firm and had low elasticity which persisted throughout the storage period.

No significant ($P \leq 0.05$) difference was observed with regard to the texture score of mozzarella cheese prepared from mixed milk and goat's milk. There was a significant difference ($P \leq 0.05$) in the texture score of mozzarella cheese prepared from cow's milk when assessed at various storage intervals, this was beneficial. Significant difference in the texture score of mozzarella cheese prepared from three types of milk was only noted at 30th day as compared to the scores noted at 1st or 15th day; the texture scores were at par with each other when compared between 1st and 15th day.

The results for overall acceptability showed that mozzarella cheese prepared from cow's milk had the highest score (Table 4). Like other sensory attributes, the overall acceptability score of goat's milk mozzarella cheese was least, when compared to other two cheeses.

Goat's milk and mixed milk mozzarella cheese were rated at par with regard to the overall acceptability. Likewise, significant difference was not noted for overall acceptability scores of mozzarella cheese prepared from cow's milk and mixed milk. The overall acceptability score for goat's milk cheese was least; such score was significantly lower compared to the scores of other two cheeses at 1st, 15th and 30th day of storage.

Table 3: The appearance and flavor scores of mozzarella cheeses as affected by storage.

Type of cheese	Storage period of cheese					
	1st day		15th day		30th day	
	Appearance	Flavour	Appearance	Flavour	Appearance	Flavour
Cow milk	4.8 ± 0.42^{Aa}	4.8 ± 0.42^{Aa}	4.1 ± 0.87 ^{Aab}	4.1 ± 0.87^{Aa}	3.8 ± 0.78^{Aa}	3.8 ± 0.78^{Aab}
Goat milk	3.7 ± 0.82^{Ab}	3.7 ± 0.82^{Aa}	3.6 ± 0.69^{Ab}	3.6 ± 0.69^{Aa}	3.3 ± 0.48^{Aa}	3.3 ± 0.48^{Aa}
Mixed milk	4.0 ± 0.9^{Ab}	4.0 ± 0.94^{Aa}	4.6 ± 0.52^{Ba}	4.6 ± 0.52^{Aa}	4.2 ± 0.63^{Ba}	4.2 ± 0.63^{Ba}

Value mean ± standard deviation not sharing capital letter in a row is significantly different (P ≤ 0.05) from each other. Value mean ± standard deviation not sharing small letter in a column is significantly different (P ≤ 0.05) from each other (storage temperature was 5°C).

Table 4: Body and texture and overall acceptability score of mozzarella cheeses as affected by storage.

Type of cheese	Storage period of cheese					
	1st day		15th day		30th day	
	Body and texture	Overall acceptability	Body and texture	Overall acceptability	Body and texture	Overall acceptability
Cow milk	4.0 ± 0.94^{Aab}	4.3 ± 0.67^{Aab}	4.1 ± 0.87^{Aa}	4.3 ± 0.67$^{A\,a}$	3.8 ± 0.79$^{A\,b}$	4.0 ± 0.47$^{A\,a}$
Goat milk	3.7 ± 0.83^{Ab}	3.9 ± 0.67^{Ab}	3.6 ± 0.69^{Ab}	3.7 ± 0.67^{Ab}	3.3 ± 0.48$^{A\,b}$	3.2 ± 0.42^{Ab}
Mixed milk	4.8 ± 0.24^{Aa}	4.6 ± 0.52^{Aa}	4.6 ± 0.52$^{A\,a}$	4.5 ± 0.53^{Aa}	4.2 ± 0.63^{Ba}	4.3 ± 0.82$^{B\,a}$

Value mean ± standard deviation not sharing capital letter in a row is significantly different (P ≤ 0.05) from each other. Value mean ± standard deviation not sharing small letter in a column is significantly different (P ≤ 0.05) from each other (storage temperature was 5°C).

Conclusion

The yield of MMMC was significantly higher than that of GMMC and CMMC at the first day, 15th day and 30th day of storage period. Highest total viable counts as well as yeast and mould count were found in goat's milk mozzarella cheese which persisted throughout its refrigerated storage period. Coliforms which were evident in all fresh cheese samples could not be detected at 30th day of refrigerated storage. Mozzarella cheese prepared from cow's milk and mixed milk (cow+goat, 1:1) compared well with regard all sensory attributes; such scores were superior over goat's milk cheese. Mozzarella cheese preparation from goat milk is not recommended.

It is recommended to encourage dairy industries to utilize goat's milk as blend with cow's milk for the production of mozzarella cheese, with attendant yield improvement over cow milk cheese.

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Author Affiliations

Top

[1]*Faculty of Science, Department of Biology, University of Hail, Hail, Kingdom of Saudi Arabia*
[2]*Faculty of Engineering and Technology, Department of Food Science and Technology, University of Gezira, Wad-Medani, Sudan*
[3]*Faculty of Agriculture, Department of Food Science, University of Elzaeem Elazhari, Khartoum, Sudan*