International Journal of

Dairy Science

ISSN 1811-9743

Academic
Journals Inc.

www.academicjournals.com

International Journal of Dairy Science 5 (3): 128-134, 2010
ISSN 1811-9743

Effect of Packaging Material on Microbiological Properties of Sudanese White Cheese

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Abstract: Sudanese soft white cheese was made from cows' milk using the traditional method and packaged in each of 16 plastic and 16 metal press lid packages. Half of the packages were stored at room temperature and the other half at refrigerator temperature. Samples were analysed at zero time, then after 21, 42, 63 and 84 days for total bacterial viable count, coliforms, *E. coli*, molds and yeasts counts. Significant difference ($p<0.05$) in total bacterial viable count, coliforms, *E. coli*, molds and yeasts counts of cheese in different packaging materials was observed. The highest values were in plastic packages. The total bacterial viable count was significantly ($p<0.05$) affected by storage temperature, the highest value was at room temperature, yeast, molds, coliforms and *E. coli* counts were not significantly ($p<0.05$) affected. The total bacterial viable count, yeasts and molds increased during storage period, while coliforms and *E. coli* counts decreased. Packaging of cheese in metal containers was found to be better as low coliforms, *E. coli* and yeast counts were obtained.

Key words: Sudanese white cheese, packaging material, storage, microbiological properties

INTRODUCTION

Cheese making is the main preservation method for milk in rural areas of Sudan particularly during rainy seasons when plenty of milk is available (El-Owni and Hamid, 2008). Soft white cheese locally known as Jibna-Beyda is the major type of cheese in Sudan, beside Mudaffara, Mozzarella and Gouda which are introduced recently (Ibrahim, 2008). However, production of processed cheese has also been tried (Nour- El-Daim and El-Zubeir-Ibtisam, 2007). Jibna-Beyda is a pickled cheese to which salt (6-20%) has been added (Abdalla *et al.*, 1993) for preservation and contribution to flavor. Salt controls microbial growth, enzyme activity, biochemical changes during ripening and development of flavor and aroma of cheese (Guinee, 2004). Jibna-Beyda has mean total solids, fat, crude protein and ash of 52.77, 22.8, 22.50 and 4.87 (2009%, respectively (El-Owni and Hamed, 2007). However, Abdel Razig and Babiker (2009) reported values of 49.48 -53.32, 20.10-22.91, 20.08-23.83 and 2.03-3.53% for

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total solids, protein, fat and ash, respectively, for Sudanese white soft cheese made by direct acidification. Jibna-Beyda is usually packaged in tins filled up with salted whey and the majority of which is stored at room temperature which may exceed 30°C during Summer. According to Bishop and Smukowski (2006) to maintain safety, storage and display temperatures of cheese must not exceed 30°C.

Cheese production in Sudan was estimated as 72470 tons in 1994 (FAO, 1995) and increased to 152000 tons in 2003 (FAO, 2005). The increase in cheese production in Sudan witnessed a retreat in packaging, as the metal containers were reused several times and sealed by soldering. However, soldering of cheese metal packages was prohibited and accordingly the packaging of cheese was changed to metal and plastic press lid containers. The effect of these packaging materials and designs on microbiological quality of the Sudanese white pickled cheese is not yet known. There are scarce published research findings for packaging of Sudanese pickled cheese particularly plastic packaging. Warsama *et al.* (2006) identified some potentially food borne pathogens in Sudanese white cheese in Khartoum North market, which might be partially attributed to packaging practices. Osman *et al*. (2009) investigated the effect of vacuum packaging on chemical composition and sensory properties of white soft cheese and found that sensory properties are gradually improved. However, vacuum packaging is currently not feasible in rural areas of Sudan where the majority of cheese is produced.

The objective of this study is to evaluate the effect of packaging in press lid metal and plastic containers on microbiological quality of Sudanese white soft cheese.

MATERIALS AND METHODS

Materials

Fresh whole cow's milk was obtained from Kuku Dairy Plant, Khartoum North. Commercial rennet tablets [Chris Hansen's Laboratory, Copenhagen Denmark] were brought from Khartoum Dairy Products Company. Clean fine sodium chloride was purchased from the local market. A two-piece round shape cans made from tinplate with side seam welder (Soudronic-Swiss) were brought from Aza Metal Factory, Khartoum, Sudan. Plastic containers polypropylene (PP) round shape with closure were brought from Mabco Plastic Factory, Khartoum, Sudan.

Cheese Manufacture

This study was conducted at the Department of Food Science and Technology, College of Agricultural Studies, Sudan University of Science and Technology, during the period from March to December 2005. Cheese was manufactured by the traditional method according to Ibrahim (1970) with some modifications. Milk was salted at the rate of 6% (w/w). The salted milk was strained through a piece of cheese cloth to remove any foreign matter and heated to 40°C Rennet tablets (1 tablet/45 kg milk) were dissolved in about 50 mL distilled water in a beaker and the solution was added to the warm salted milk in the cheese vat and stirred thoroughly. The cheese vat was covered and left for about 1 h for milk to coagulate. After coagulation, the curd was cut into approximately 2 cm cubes with stainless steel knives. The curds were transferred to wooden moulds (50×50×20 cm) lined with cheese cloth the edges of the cloth were overlapped firmly. The curd was pressed for about 24 h using weights of about 15 kg. The molded curd was then cut into small rectangular blocks (400-500 g) and 500 g of these blocks were packaged in each of the 16 tin plate containers and 16 plastic containers (PP) and about 200 mL of the whey collected was added to the curd in each container. Five hundred grams of cheese were transferred to the lab for analysis to represent zero time. After packaging, one half of the containers (8 tinplates and 8 plastic) were stored at room temperature, the other half was stored at refrigerator temperature. The analyses were carried out in triplicate at 0, 21, 42, 63 and 84 day intervals.

Microbiological Analysis

The culture media were prepared according to the manufacturer's instructions (Oxoid) and sterilized in an autoclave at 121°C for 15 min at 15 pound pressure. Cheese samples were taken aseptically and weighed in sterile stomacher bags. Eleven grams of cheese were added to 99 mL of peptone water and blended by the stomacher for 2 min. One milliliter of the bag contents was pipetted into separate tubes containing 9 mL of peptone water; the liquids were then mixed carefully by aspirating 10 times with a sterile pipette. With the same pipette 1 mL was transferred to another dilution tube containing 9 mL of peptone water and mixed with fresh pipette. Serial dilutions of 10^{-1}-10^{-6} were obtained by repeating the steps of mixing and transferring. Plate count agar was used for enumeration of total bacterial viable count according to Richardson (1985). McConkey broth and Brilliant green bile broth were used for enumeration of coliforms and *E. coli* Most Probable Number (MPN) per gram of sample (Harrigan and McCance, 1976). Potato Dextrose Agar (PDA) was used for enumeration of yeasts and molds counts (Harrigan and McCance, 1976).

Statistical Analyses

Statistical analyses were performed using the Genstat 5 Release 3.2. Factorial Experiment in a randomized block design was used to determine the effect of storage period (0, 21, 42, 63 and 84 days), type of containers and storage temperature. Means were separated using Least Significant Differences of means with a $p<0.05$.

RESULTS AND DISCUSSION

Significant variations ($p<0.05$) were observed in bacterial viable count of cheese during storage in the two types of package. The mean of total bacterial viable count of cheese in plastic packages was significantly higher ($p<0.05$) at 21, 42 and 84 days of storage at room temperature compared to those in metal packages (Table 1). This suggests that plastic press lid packages may be of high microbial contamination compared with metal packages. Barbo *et al*. (1966) observed that during manufacture and handling electrostatic charges can occur on the plastic. These charges attract air-borne materials such as dust and microorganisms. Atasever *et al.* (2003) reported higher total viable counts in cheese vacuum packaged in plastic compared to that packaged in tin containers.

Table 1: Effect of storage period, temperature and type of packaging on total bacterial viable count of Sudanese white soft cheese

	Total viable count (cfu g^{-1})			
	Room temperature		Refrigerator temperature	
Storage period (days)	Plastic	Metal	Plastic	Metal
0.0	2.60×10^{5def}	2.6×10^{5def}	2.6×10^{5def}	2.60×10^{5def}
21	5.50×10^{5bef}	3.0×10^{5def}	2.9×10^{5def}	3.00×10^{4f}
42	6.20×10^{5bef}	4.3×10^{5def}	3.6×10^{5def}	1.47×10^{5ef}
63	1.58×10^{6cd}	7.8×10^{5defc}	1.5×10^{6cbe}	2.40×10^{5def}
84	4.80×10^{6a}	3.1×10^{6b}	2.1×10^{6bc}	5.60×10^{5def}

Means with different superscript letters are significantly different ($p<0.05$)

Significant differences ($p<0.05$) were also observed in bacterial viable count of cheese during storage at room temperature and refrigerator temperature. The viable bacterial count of cheese at room temperature significantly increased ($p<0.05$) during storage than at cold storage. El-Owni and Hamed (2009) reported increase in total bacterial count of Jibna Beyda stored at room temperature. A significant increase ($p<0.05$) was observed in bacterial viable count during storage period. The mean bacterial viable count in cheese at zero time was 2.6×10^5. The bacterial viable count progressively increased after 21 days of storage. The value continued to rise till 84 days of storage. These results were lower than those of Ahmed (1985) who obtained higher total bacterial count values of 5.2×10^8-5.3×10^9 in fresh cheese made in the laboratory. This was attributed to starter culture used in cheese manufacture (Ahmed, 1985). The result was in disagreement with Beshir (1999), who found that the bacterial viable count of cheese made from whole milk with 20% soymilk was 3.0×10^4 at zero hour and this was attributed to the presence of soymilk which develops acidity in cheese and increased the lactic acid bacteria which inhibited growth of the other organisms. However, the high bacterial load is in agreement with Hadi-Sulieman (2007) who found that bacterial load of Jibna-Beyda made from raw milk is high compared to that made from pasteurized and boiled milk.

Table 2 and 3 show changes in number of coliforms bacteria and *E. coli* in Sudanese white cheese. Significant variations ($p<0.05$) were observed in coliforms bacteria in cheese as affected by packages. The coliforms bacteria in plastic packages were significantly higher ($p<0.05$) than in metal packages, in agreement with results reported by Atasever *et al.* (2003).

Significant differences ($p<0.05$) were also observed in *E. coli* in cheese packaged in the two types of packages. *E. coli* in cheese in plastic packages was significantly higher ($p<0.05$) than that in metal packages. This is because the pH tended to be more acidic in metal packages than in plastic packages. There were significant differences ($p<0.05$) in coliforms bacteria between cheese stored at refrigerator and room temperature. Significant differences

Table 2: Effect of storage period, temperature and type of packaging on coliform bacterial count of Sudanese white soft cheese

	Coliform bacteria (General) MPN g^{-1}			
	Room temperature		Refrigerator temperature	
Storage period (days)	Plastic	Metal	Plastic	Metal
0	1100^{a}	1100^{a}	1100^{a}	1100^{a}
21	200^{b}	200^{b}	230^{b}	140^{c}
42	98^{cd}	84^{cde}	77^{def}	57^{defg}
63	84^{cde}	43^{efg}	52^{defg}	38^{efg}
84	25^{fg}	18^{g}	24^{fg}	15^{g}

Means with different superscript letters are significantly different ($p<0.05$)

Table 3: Effect of storage period, temperature and type of packaging on *E. coli* Count of Sudanese white soft cheese

	E. coli MPN g^{-1}			
	Room temperature		Refrigerator temperature	
Storage period (days)	Plastic	Metal	Plastic	Metal
0	75^{a}	75^{a}	75^{a}	75^{a}
21	48.5^{b}	40.5^{bc}	41.5^{bc}	36.5^{cd}
42	34.5^{cd}	21.5^{efg}	28.5^{de}	19.5^{efgh}
63	24.5^{ef}	12^{ghij}	16.1^{fghi}	11.1^{hij}
84	<10	<10	<10	<10

Means with different superscript letters are significantly different ($p<0.05$)

($p<0.05$) were also observed in *E. coli* count between cheese stored at refrigerator and room temperature. Kosikowski (1982) reported that coliforms grow well in cold or warm cheese milk.

The coliforms bacteria in cheese decreased significantly ($p<0.05$) with storage time after 42 days of storage. The coliforms bacteria at zero time were 1.1×10^3. It rapidly dropped after 21 days of storage and the decline continued gradually till day 84 of storage. This result was attributed to the decrease in pH of cheese throughout the storage period. *E. coli* in Sudanese white cheese also decreased significantly ($p<0.05$) as the storage time progressed. *E. coli* at zero time was 75. It rapidly dropped after 21 days of storage and the decline continued gradually till day 84 of storage. This could be due to decline in pH of cheese throughout the storage. Ahmed (1985) reported that no coliforms bacteria were detected in market cheese and they attributed this to low pH in cheese which is not suitable for the presence of coliforms bacteria. However, Hamid and El-Owni (2007) detected coliforms and *E. coli* in Sudanese white cheese collected in West Darfur. It is likely that the low pH of cheese was the inhibitory factor to the presence of coliforms bacteria. In cheese curd coliforms number may reach up to 100-500 million per gram if their initial level in milk was high and curd acid development was slow (Kosikowski, 1982). Coliforms do not survive in pasteurized cheese milk, but may be present in the resulting cheese because of post pasteurization contamination (Kosikowski, 1982; Yang and Jones, 1969).

Table 4 shows changes in yeasts count of Sudanese white cheese. A significant difference ($p<0.05$) was observed in yeasts count in cheese as a result of storage in the two types of package. The yeast count in plastic packages was significantly higher ($p<0.05$) than in metal packages. No significant differences ($p>0.05$) were observed between yeasts count of cheese in metal package stored at refrigerator and room temperature. Significant differences ($p<0.05$) were observed in yeasts count during storage period. As revealed by the results, yeasts were detected at zero time, yeasts were found after 21 days of storage, increased after 42 days and thereafter it declined at 63 days of storage. These results disagree with those of Ahmed (1985) who obtained lower yeasts count values of 2.5×10^3-zero in fresh Sudanese white cheese and that stored at 7 and 37°C. This is due to the packaging cheese in anti acid cans closed by double seaming. In this study cheese was packaged in plastic and metal containers and closed by cover.

Yeasts count found by Beshir (1999) was 6.1×10^4 in cheese made with traditional method obtained from El Dueim area and 7.5×10^3 in cheese made in the laboratory. Ibrahim (1971) attributed the high yeast count of white cheese to the high acidity of the product.

Table 5 shows changes in molds count of Sudanese white cheese. The results showed significant difference ($p<0.05$) between cheese stored in plastic and metal packages. There is significant difference ($p<0.05$) in molds count between cheese stored at refrigerator and room temperatures. Generally molds were found in cheese at 21 days storage period storage,

Table 4: Effect of storage period, temperature and type of packaging on yeasts count of Sudanese white soft cheese

	Yeasts (cfu g^{-1})			
	Room temperature		Refrigerator temperature	
Storage period (days)	Plastic	Metal	Plastic	Metal
0.0	<10	<10	<10	<10
21	5.5×10^{5abcd}	2.8×10^{5bcd}	8.8×10^{4bcd}	1.6×10^{5bcd}
42	7.3×10^{5ab}	4.9×10^{4cd}	1.1×10^{6a}	3.6×10^{4cd}
63	2.2×10^{4d}	2.4×10^{5bcd}	1.6×10^{5dcd}	7.0×10^{4bcd}
84	6.6×10^{5abcd}	4.9×10^{5abcd}	7.0×10^{5abc}	5.1×10^{5abcd}

Means with different superscript letters are significantly different ($p<0.05$)

Table 5: Effect of storage period, temperature and type of packaging on molds count of Sudanese white soft cheese

	Molds (cfu g^{-1})			
	Room temperature		Refrigerator temperature	
Storage period (days)	Plastic	Metal	Plastic	Metal
0.0	<10	<10	<10	<10
21	<10	4.1×10^{5ab}	3.1×10^{5ab}	1.1×10^{5b}
42	5.6×10^{5ab}	1.9×10^{5b}	4.5×10^{5ab}	5.6×10^{4b}
63	4.1×10^{4b}	4.3×10^{5ab}	1.6×10^{5b}	3.6×10^{5ab}
84	5.0×10^{5ab}	<10	7.4×10^{5a}	3.9×10^{5ab}

Means with different superscript letters are significantly different ($p<0.05$)

increased after 42 days, fell after 63 days and increased again after 84 days. Cheese in plastic package has the highest molds count after 84 days of storage at both storage temperatures. Ahmed (1985) obtained mold count of 2.0×10^2 in fresh cheese which decreased throughout storage till 1.5×10^1 at 4 months of storage at 7°C and obtained no mold at 37°C.

CONCLUSIONS

It could be concluded that packaging of Sudanese white cheese in metal containers is better than that in plastic containers as low total bacterial, coliforms, *E. coli* and yeast counts were obtained in cheese packaged in metal containers. Cold storage of Sudanese white cheese is better than storage at room temperature as it resulted in better microbiological characteristics compared to room temperature storage. The use of seamed containers is recommended as press lids appear to allow contamination of cheese.

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