MICROBIOLOGICAL QUALITY OF SUDANESE WHITE CHEESE DURING STORAGE USING DIFFERENT PACKAGING MATERIALS

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Abstract
Sudanese white cheese was purchsed from El Dueim town, then it was packed into five different packaging materials: tin containers, metal tin lined with polyethylene bags, plastic buckets, plastic buckets lined with polyethylene bags and metal gallon as a control. Each pack weighs 2 kg. The cheese samples were stored at room temperature (30± 5°C) for 6 months (180 days) so as to study the effect of packaging materials on the microbiological quality (total bacterial count, moulds and yeast, coliform, Clostridium spp., Staphylococcus spp. and Salmonella spp.) during the storage period.
Generally the cheese samples kept in plastic lined with polyethylene bags containers were significantly ($P \leq 0.05$) lower in TBC, coliforms and yeasts and moulds counts compared to those packed into metal tins, plastic non lined containers and metal gallons during the storage. Also the maximum level of moulds, yeasts and bacteria were reported in cheese stored in plastic and metallic gallon, this increased the risk of an earlier deterioration of these samples compared to the other backaging materials. Staphylococcus aureus was detected at day zero as 2.8×10^5 then it disappeared after 60 days storage. Salmonella spp. was detected at zero time and then it disappared after 60 days of the storage, while Clostridum botulinum showed negative results in all types of packaging.
It is concluded that metal tin containers and polyethylen lined containers made either of plastic or metal would improve rhe quality of Sudanese white soft cheese.

Keywords: Sudanese white cheese, storage, different packaging materials, microbiological quality

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1. INTRODUCTION

Cheese provides a useful service in extending the shelf-life of milk (Alalade and Adeneye, 2006). Cheese making in Sudan is the major preservation method for surplus milk in rural areas especially during rainy season when plenty of milk is available (El Owni and Hamid, 2007; El Owni and Hamid, 2008). Sudanese white cheese is the most popular type of cheese produced in Sudan, locally known as *Gibna bayda* (El Owni and Hamid, 2008). 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 (Idris and Alhassan, 2010). Abdalla and Mohamed (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. In a laboratory trial, Nour El Diam and El Zubeir (2007) reported that glass packaging was more acceptable compared to plastic packaging (70% and 30%, respectively). Ceylan et al. (2003) studied the microbiological quality of sikma cheese (white pickled Turkish cheese) and found that the average coliform count was 5.99 log cfu/g, the high coliform content was attributed to the post-contamination during

storage. El Owni and Hamid (2008) found that total bacterial count, coliform, *E. coli* and psychrotrophic bacteria were decreased significantly (P≤ 0.05) during the storage period of Sudanese white cheese, while yeast and molds increased significantly (P≤ 0.05). Significant difference (P<0.05) in total bacterial viable count, coliforms, *E. coli*, molds and yeasts counts of cheese in different packaging materials was also observed, with the highest values been in plastic packaging (Idris and Alhassan, 2010).

The average microbiological properties were: 9.71 log cfu/ml for total bacterial count; 3.28 log MPN/ml coliforms; 0.70 log MPN/ml *E. coli;* 1.73 log cfu/ml *Staphylococcus aureus;* 5.06 log cfu /ml lactic acid bacteria; 3.72 log cfu /ml streptococci and 4.46 log cfu/ml for yeast and mould counts (Hamid and El Owni, 2007). Moreover positive isolates for *Salmonella typhi*, *Salmonella paratyphi Staphylococcus aureus and E. coli* were detected in Sudane*se* white cheese (Warsama et al., 2006). The US Food and Drugs Administration (FDA, 2005) stated that, soft raw-milk cheese can cause serious infectious diseases including listeriosis, brucelleosis, salmonellosis and tuberculosis

Recently, the consumer desire for a healthier microbiologicaly safer foods has been increased, therefore the importance of production of cheese being properly packed in convenience, smaller, family size packages, and longer product shelf life is a must. Hence the present study is conducted to evaluate the traditional and improved packaging materials used for packing Sudanese white cheese.

2. MATERIALS AND METHODS

Source of Sudanese white cheese samples

The white cheese used in this study was purchased from a traditional factory at Galaja 70 kilometers south El Dueim and 350 kilomers southwest of Khartoum in Sudan. The cheese was packaged into 5 different types of packing; metal tins (lined with polyethylene and non lined), plastic containers (lined with polyethylene and non lined) and petroleum gallons), Each was 2 kg in size. The tin containers were made from tinplate; the inside were coated by golden lacquer and the out side was coated by white paint. The tin was square, with push-on-closures. Plastic containers were white, square, the cover lined with adhesive tape. The petroleum gallon were sealed by soldering.

Microbiological examination

The selection and preparation of the media and reagents, sterilization, serial dilution and culturing methods, icubation and counting were done as described by Harrigan and McCance (1976).

Plate count agar (Himedia 400086) and nutrient agar (Scharlau, 9006) were used for determination of TBC by poured plate method, (Richardson, 1985). For total moulds and yeast count malt extract agar medium (Oxoid) was used (Harrigan and McCance, 1976). MacConkey broth (Scharlan, 8743) and Brilliant green lactose bile broth (Merck, 736) were used for enumeration of coliforms most probable number (MPN) per gram of the sample tested using the special tables and formula described by Marshall (1992). Staphylococcus 110 medium (Oxoid) and manitol salt agar (Oxoid) were used for the enumeration and detection of *Staphylococcus aureus* (Harrigan and McCance, 1976). Cooked meat broth and blood agar were used for *Clostridium botulinum* enumeration (Silliker and Greenberg, 1969). Deoxycholate citrate agar (DCA) (Oxoid, 435) was used for detection of salmonellae in the cheese samples according to Thatcher and Clark (1968).

Statistical analysis

Data generated was subjected to Statistical Package for Social Sciences (SPSS, 1998). Means were tested using two-factors Analysis of Variance (ANOVA), and then separated using Duncan´s Multiple Range Test (DMRT) according to Mead and Gurnow (1983).

3. RESULTS AND DISCUSSION

Total Bacterial count

After 15 days storage the bacterial count of the cheese samples kept in plastic lined with polyethylene bags containers were significantly ($P \leq 0.05$) lower (2×10^{10}) than those packed into metal tins, plastic and metal gallons (Table 1). The significantly ($P \leq 0.05$) lower level of TBC correlated well with their lower pH. El Owni and Hamid (2009) attributd such decrease in TBC of cheese to the effect of high acidity of the cheese samples. On the other hand, Abdalla and Ahmed (2010) reported that the high acidity of raw milk cheese could be due to the fact that storage temperature activated the natural microflora of raw milk and resulted in the development of acidity as the result of lactose fermentation. The TBC in cheese samples kept in metal gallons and plastic non lined containers increased significantly ($P \leq 0.05$) at day 30 of storage and it was found to correlate well with their higher ripening indices (not shown data). These resuls were in the same line with Idris and Alhassan (2010) who observed a continuous increase in viable bacterial count during the storage period.

Moulds and yeasts

Table 2 showed that moulds and yeasts counts of cheese samples kept in metal tins were significantly ($P \leq 0.05$) higher (2.25×10^{13}) at day 15 of storage then decreased gradually to reach 3.7×10^{8} at the end of the storage period (180 days). After 30 days storage, moulds and yeasts counts of cheese samples kept in metal gallon and plastic non lined containers were 5.5×10^{8} and 4.5×10^{8} cfu, respectively. Then they both increased to 1.0×10^{12} at day 150, where some of the cheese samples were spoiled. These results were higher that found by El Owni and Hamid (2009) who attributed the high yeast count of white cheese to its high acidity. The constant increase of moulds and yeasts during storage might be due to the fact that yeasts and moulds counts could metabolize lactic acid and lower pH value (Turkoglu et. al., 2003). However Nour El Diam and El Zubeir (2006) found that the heat treatment and processing improve the cheese quality via reducing the counts of yeasts and moulds.

Coliform Bacteria

Table 3 shows the effect of packing type on the coliform bacterial count of the Sudanese cheese samples during the storage. After 45 days of storage, the coliform count of cheese samples kept in metal gallons (4.25×10^{13}), lined and non lined plastic containers (3.5×10^{13} and 3.5×10^{13}, respectively) were significantly ($P \leq 0.05$) higher compared to lined and non lined metak tins cans (4.00×10^{12} and 1.25×10^{12}, respectively). El Zubeir et al. (2006) was able to indentify different species of coliform bacteria in the cheese samples from restaurants in Khartoum State. El Owni and Hamid (2009) found that cheese samples kept in plastic containers had higher coliform and *E. coli* counts than those stored in anti-acid cans. Idris and Alhassa (2010) observed similar results and stated that the coliform bacteria in plastic packages were significantly ($P \leq 0.05$) higher than in metal packages.

The high bacterial load found in the cheese samples investigated during the present study (Table 1, 2 and 3) supported Elkhider et al. (2011) who reported that cheese samples collected from different producers in rural atras of eastern Sudan indicate that the level of hygiene and production methods, source of raw milk and its handling could be the main factors attributed to this high loads which might affect the quality of cheese.

Clostridium botulinum, Staphylococcus aureus and *Salmonella*

The presence of *Clostridum botulinum*, *Staphylococcus aureus* and *Salmonella spp.* in Sudanese white cheese stored in different types of packaging materials are shown in Table 4. *Staphylococcus aureus* was detected at day zero as 2.8×10^{5} then it disappeared after 60 days storage. This result supported El Owni and Hamid (2009) who stated that *Staphylococcus aureus* count in Sudanes

Table 1: Changes in total bacterial count (cfu) of Sudanese white soft cheese during storage period as affected by type of packaging*

	Storage period (days)										
	0.0	7	15	30	45	60	75	90	120	150	180
MT	1.90×10^{6ijkl}	5.00×10^{10cdefgh}	3.70×10^{10efghijk}	3.00×10^{12ghijk}	5.60×10^{11cdefg}	2.50×10^{11hijkl}	7.50×10^{12bc}	100.0×10^{11a}	1.75×10^{8ijkl}	6.00×10^{7bcdef}	3.20×10^{7fghijk}
MTL	1.90×10^{6ijkl}	4.00×10^{10defghij}	4.00×10^{10defghij}	3.00×10^{12ghijk}	5.60×10^{11cdefg}	8.50×10^{11b}	2.00×10^{12ijkl}	2.75×10^{13hijkl}	2.25×10^{8hijkl}	6.40×10^{7bcde}	3.50×10^{7fghijk}
P	1.75×10^{6ijkl}	4.50×10^{10defghi}	2.50×10^{10hijkl}	3.50×10^{12fghijk}	5.60×10^{11cdefg}	1.50×10^{11jkl}	3.40×10^{12fghijk}	6.50×10^{13bcd}	1.00×10^{3kl}	ND	ND
PL	1.90×10^{6ijkl}	8.50×10^{10b}	2.00×10^{10ijkl}	4.50×10^{12defghi}	5.60×10^{11cdefg}	1.50×10^{11jkl}	1.00×10^{12kl}	3.35×10^{12fghijk}	ND	ND	ND
MG	1.90×10^{6ijkl}	7.50×10^{10bc}	3.00×10^{10ghijk}	4.50×10^{12defghi}	5.60×10^{11cdefg}	6.50×10^{11bcd}	1.00×10^{12kl}	$3.75\times10^{13defghijk}$	ND	ND	ND

In this and the following tables:
* Mean values having different superscript letters in columns and rows differ significantly ($P \leq 0.05$).
Where:
MT = Metal tin
MTL = Metal tin lined with polyethylene bags
P = Plastic
PL = Plastic lined with polyethylene bags
MG = Metal gallon
ND= Not detected (samples discarded)

Table 2: Changes in mould and yeast count (cfu) of Sudanese white cheese during storage as affected by type of packaging*

Packaging type	Storage period (days)							
	0.0	7	15	30	60	75	150	180
MT	1.50×10^{3hii}	2.25×10^{10g}	2.25×10^{13g}	1.00×10^{8i}	1.00×10^{12i}	1.00×10^{12i}	2.00×10^{9gh}	3.70×10^{8d}
MTL	1.00×10^{3i}	3.50×10^{10de}	1.50×10^{13hi}	1.00×10^{8i}	4.00×10^{12cd}	1.00×10^{12i}	3.00×10^{9ef}	2.40×10^{8fg}
P	1.50×10^{3hi}	4.00×10^{10cd}	1.00×10^{13i}	1.00×10^{8i}	$<100\times10^{11a}$	1.00×10^{12i}	ND	ND
PL	1.50×10^{3hi}	1.00×10^{11i}	1.50×10^{13hi}	4.50×10^{8c}	$<100\times10^{11a}$	1.00×10^{12i}	ND	ND
MG	1.000×10^{3i}	2.000×10^{11gh}	2.000×10^{13gh}	5.500×10^{8b}	$<100\times10^{11a}$	1.000×10^{12o}	ND	ND

Table 3: Changes in coliforms count (cfu) of Sudanese white soft cheese during storage period as affected by types of packaging*

Packaging material	Storage period (days)									
	0.0	7	15	30	45	60	90	120	150	180
MT	5.50×10^{3a}	2.50×10^{13cde}	2.50×10^{13cde}	1.00×10^{8ef}	1.25×10^{12ef}	1.00×10^{9ef}	1.00×10^{5ef}	4.00×10^{3abcd}	$2.50^{cde}\times10^{2}$	4.10×10^{2abcd}
MTL	5.50×10^{3a}	3.50×10^{13abcd}	3.00×10^{12bcde}	1.00×10^{8ef}	4.00×10^{12abce}	1.00×10^{9ef}	1.00×10^{5ef}	4.00×10^{3abcd}	$0.00^{f}\times10^{2}$	3.90×10^{2abcd}
MP	5.50×10^{3a}	5.25×10^{13a}	2.00×10^{13def}	2.50×10^{8cde}	3.50×10^{13abcd}	1.00×10^{9ef}	1.00×10^{5ef}	5.00×10^{4ab}	ND	ND
PL	5.50×10^{3a}	4.25×10^{13abc}	2.00×10^{13def}	4.50×10^{8abc}	3.50×10^{13abcd}	1.00×10^{8ef}	1.00×10^{5ef}	ND	NDE	ND
MG	5.50×10^{3a}	4.50×10^{13abc}	4.00×10^{13abcd}	3.50×10^{8abcd}	4.25×10^{13abc}	1.00×10^{8ef}	1.00×10^{5ef}	ND	ND	ND

Table 4: Detection of selected pathogenic bacteria (*Staphylococcus spp.*, *Salmonella spp.* and *colistridum spp.*) in Sudanese white soft cheese

Type of bacterial	*Staphylococcus*	*Salmonella*	*Colistridum spp.*
0 day	2.8×10^{5}	+ve	-
60 days	ND	-ve	-
150 days	-	-	-ve

white cheese was detected at zero time before storage and completely disappeared after 60 days of storage. Ali and Galal (2000) attributed the disappearance of *Staphylococcus aureus* to the increase acidity of the cheese and the high salt level.

The presence of *Salmonella spp.* was examined at zero time and then after 60 days of the storage, the results were found to be positive and negative, respectively. Warsama et al. (2006) found some of *Salmonella spp.* in the some samples of Sudanese white cheese. Similar finding was reported by Amran and Abbas (2011) who reported that pathogenic flora such as *Salmonella* and *Listeria* were detected in some samples and disappeared at the end of storage period.

Clostridum botulinum in cheese samples was investigated after 120 days storage and showed negative results in all types of packaging. The absent of *Clostridum botulinum* from cheese samples might be attributed to the fact that the raw milk was free from it, beside the high acidity of the cheese samples. Banwart (1981) reported that there is no growth of *Clostridum botulinum* at pH 4.7 or less.

4. CONCLUSION

The present result found that the microbiological features remain the limiting factor in shelf life of cheese. However it is concluded that metal tin containers whether lined or non-lined would secure safety measures for Sudaese white cheese during storage for maturity. Also the use of polyethylen lined containers made either of plastic or metal was highly recommended for packaing of Sudanese white soft cheese. Hence this study supported the previous recommendation drawn by Elkhider et al. (2011) that intervention and training of cheese producers will be needed to improve the quality of white Sudanese cheese in rural areas of Sudan. On the other hand, Hussein et al. (2011) encourages manufacturing of processed cheese in Sudan in order to utilize huge amount of raw milk produced and also to minimize the cost of cheese importation.

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