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Effect of Storage Period on Weight Loss, Chemical Composition, Microbiological and Sensory Characteristics of Sudanese White Cheese (Gibna Bayda)

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Abstract: Effect of storage period on weight loss, chemical composition, microbiological and sensory characteristics of Sudanese white cheese (Gibna Bayda) is studied. The cheese was made from fresh raw cow's milk with 6% salt, preserved by it's own whey in anti-acid containers and stored at room temperature (35-37°C) for 240 days. Weight loss increasedsignificantly ($p<0.05$) increased through out the storage period. Crude proteins, total solids and ash contents significantly ($p<0.05$) increased from day zero to day 120 then decreased onwards. Soluble proteins, tyrosine, tryptophane and Volatile Fatty Acids (VFA) increased. Total Bacterial Count (TBC), coliforms, *E. coli*, *Staphylococcus aureus* and psychrotrophic bacterial counts significantly ($p<0.05$) decreased during storage, while yeasts and moulds increased as storage time progressed. Texture, flavour and colour of the cheese samples significantly ($p<0.05$) improved during storage until day 120 then decreased in scores there after.

Key words: Storage period, weight loss, chemical composition, microbiological and sensory characteristics, Sudanese white cheese, Gibna Bayda

Introduction

In Sudan cheese processing is a major preservation method of surplus milk in rural areas especially during rainy season when plenty of milk is available. The product is an important nutrient for humans especially under conditions where other animal proteins are not available (Kosikowski, 1982). White cheese is the most popular type of cheese produced in Sudan, locally known as (Gibna Bayda). It is a pickled soft cheese that is stored under anaerobic conditions in air tight containers filled with whey (Kur, 1984). White cheese (Gibna Bayda) is a unique among pickled cheese varieties in that high concentrations of sodium chloride are added to the milk before renneting. When manufactured from raw milk no starter culture is added, the natural lactic acid bacteria present in the raw milk will carry the fermentation process required for the cheese maturation. The cheese is usually left over night to drain the whey with or without pressing. Ripening takes place while the cheese was submerged in whey (Osman, 1987). Its appearance, texture and flavour closely resembles the Greek Feta (Ibrahim, 1970).

Cheese maturation includes chemicals, physical and biological changes. Mature cheese processes include glycolysis, proteolysis and lipolysis (Edward and Kosikowski, 1983). The total nitrogen and water-soluble-nitrogen in cheese increased during maturation (Tarakci and Kucukoner, 2006). Nuser (2001) studied the effect of storage period on chemical composition of fresh white cheese, he found the following results 25.13% fat, 23.26% protein, 48.46% total solids, 3.5% ash and 0.65 titratable acidity. Litopoulou-Tzanetaki and Tzanetakis (1992) studied the chemical composition and ripening of white-brine cheese and found that aerobic, lactic acid, proteolytic, lipolytic and psychrotrophic bacterial counts increased after 75 days of ripening. They also found that the pH 4.5 and NaCl (5.8-6.2%) contents in cheese inhibited microbial growth causing bacterial counts to decrease considerably after 3 months.

Chemical qualities of the cheese including titratable acidity, pH, water-soluble-nitrogen, ripening index and tyrosine values had significant ($p<0.05$) effects on flavour (Guler and Uraz, 2004). The coliform bacterial counts in cheeses are expected to decrease during ripening due to low pH and antagonistic action of lactic acid bacteria (Babel, 1977). A significant ($p<0.05$) negative correlation ($r = -0.4014$) was found between yeast-mould counts and coliform counts, probably due to acid forming ability (Ceylan *et al*., 2003). The textural attributes of Turkish white cheese were significantly ($p<0.05$) affected by the ripening period. The highest overall acceptance scores were found at the 60 days of ripening (Topcu and Saldamli, 2006).

The objective of this study was to determine the effect of storage period on the weight loss, chemical composition, microbiological and sensory characteristics of Sudanese white cheese.

Materials and Methods

Cheese manufacture: White cheese (Gibna Bayda) produced from cow's milk was ripened over 240 days at room temperature (35-37°C). Cheese manufacture was

Table 1: Effect of storage period on weight loss and chemical composition of white cheese (Gibna Bayda)

	---------- Weight loss and chemical composition ----------				
Storage period (days)	Weight loss (%)	TA (%)	Total solids (%)	Crude protein (%)	Fat (%)
Zero	0.0^{a}	0.39±0.09^{a}	42.38±1.77^{c}	15.08±0.55^{c}	18.90±0.76^{c}
60	22.64±16.7^{c}	1.09±0.61^{d}	43.38±7.01^{b}	16.06±3.38^{b}	21.71±5.44^{a}
120	26.25±15.1^{ab}	1.24±0.41^{b}	44.17±5.7^{a}	17.85±3.51^{a}	22.16±5.13^{a}
180	27.19±17.72^{a}	1.50±0.63^{a}	41.49±16.83^{d}	12.70±5.40^{d}	19.97±9.50^{b}
240	25.64±19.41^{b}	1.17±0.73^{c}	34.40±20.56^{e}	12.60±7.70^{d}	17.31±11.31^{d}
Level of significance	***	***	***	***	***

	---------- Weight loss and chemical composition ----------				
Storage period (days)	Soluble protein (%)	Ash (%)	VFA 0.1 N mL NaoH/100 gm cheese	Tyrosine mg/100 gms cheese	Tryptophure mg/100 gms cheese
Zero	0.20±0.03^{e}	3.50±0.19^{b}	4.68±0.29^{e}	14.38±3.5^{e}	5.5±1.83^{e}
60	0.38±0.11^{d}	3.62±0.71^{a}	10.91±6.01^{d}	91.13±45.1^{d}	17.85±7.6^{d}
120	0.61±0.09^{b}	3.37±0.65	18.76±14.67^{a}	152.6±66.3^{c}	35.28±18.1^{c}
180	0.63±0.21^{a}	2.93±1.19^{c}	16.52±10.94^{b}	161.11±84.5^{b}	57.59±40.4^{b}
240	0.59±0.33^{c}	2.52±1.59^{d}	11.80±7.57^{c}	180.59±120.06^{a}	117.50±71.09^{a}
Level of significance	***	***	***	***	***

Mean values bearing different letters within colums are significantly different ($p<0.05$); TA = Titratable acidity; VFA = Volatile fatty acid; *** = Means highly significant at ($p<0.001$)

conducted according to Osman (1987). Two experiments were carried out during the period from May 2006 to November 2006. Experiments were performed in the laboratory of the Department of Dairy Production, Faculty of Animal Production, University of Khartoum. Cows' milk was purchased from private dairy farms near by the University campus. The salt was added to the heated (40°C) raw milk before renneting to give 6% salt concentration. Milk was coagulated using commercial rennet (Chr. Hansens Lab., Cobenhagen, Denmark). The milk was stirred for 10 minutes and left undisturbed till completion of coagulation. The curd was cut, the whey drained and pressed over night in wooden moulds. Next day each cheese was removed and cut into small cubes (5×5×5cm). Cheese samples were packed in soldered anti-acid cans, filled with boiled salted whey, stored in triplicates at room temperature (35-37°C) and analyzed when fresh at zero, 60, 120 and 240 days. Weight loss, chemical, microbiological analyses and sensory characteristics were determined.

Chemical analysis: Weight loss, titratable acidity, total solids, crude proteins and ash contents were determined according to AOAC (1990). Fat contents was determined according to Foley *et al.* (1974), soluble proteins according to Ling (1963) and volatile fatty acids according to Kosikowski (1982). Tyrosine and tryptophane content were determined by the method of Vakaleris and Price (1959).

Microbiological analysis: Eleven grams of each cheese samples were weighed aseptically into sterile blender jar (Moulinex 719), then 99 mL of sterile 2% aqueous solution of sodium citrate warmed at 45°C was added and blended for 2 minutes to make 10^{-1} dilutions. Dilutions were made using 0.1% peptone water as diluent (FDA, 1980; Harrigan and McCance, 1976). Plate count agar was used for enumeration of total bacterial and psychrotrophic bacterial counts (FDA, 1980; Frank *et al.*, 1992). Mac Conkey broth and Brilliant green lactose bile broth were used for enumeration of coliforms and *E. coli* most probable numbers according to Marshall (1992). Mannitol salt agar was used for *Staphylococcus aureus* (Rayman *et al.*, 1988). Sabouraud dextrose agar was used for enumeration of yeasts and moulds (Harrigan and McCance, 1976). The cultures with the characteristic properties were calculated as colony forming units (cfu) per ml of cheese (FDA, 1980). Each analysis was carried out in duplicates.

Sensory evaluation: Organoleptic assessment of the cheese was carried by a ten-member untrained panelists team to judge cheese for, texture, flavour, colour and saltiness. The samples were presented to panelists in randomized order after having stood for 4 hrs at room temperature and were graded 1-10. Water was provided for mouth washing between evaluations of samples.

Statistical analysis: All statistical calculations were performed using Statistical Package for Social Studies (SPSS) Software. General Linear Models were used to estimate the effect of storage period on weight loss, chemical composition, microbiological and sensory characteristics of white cheese. Duncan's Multiple Ranges tests were carried out for mean separation between the treatments.

Results

The gross composition of Gibna Bayda were shown in Table 1. The results indicated that weight loss, titratable acidity and soluble proteins significantly ($p < 0.05$)

Table 2: Effect of the storage period on the microbiological quality (Log cfu/ml) of white cheese (Gibna Bayda)

	---------- Microbiological analysis of white cheese ----------					
Storage period (days)	TBC cfu/ml	Coliforms MPN/ml	*E.coli* MPN/ml	*Staph.aur.* Cfu/ml	Psychrotroph cfu/ml	Yeast and mould count cfu/ml
0.0	7.523±1.05^{a}	4.04±1.02^{a}	2.14±0.16^{a}	3.02±0.2	6.35±0.08^{a}	2.65±0.47^{d}
60	7.56±0.26^{a}	1.23±1.16^{b}	0.75±0.98^{b}	ND	3.28±3.37^{b}	3.13±2.15^{c}
120	6.74±1.22^{b}	0.24±0.66^{c}	0.16±0.43^{c}	ND	2.02±2.71^{d}	4.79±2.20^{b}
180	6.46±2.58^{c}	0.14 0.38^{d}	ND	ND	2.52±3.35^{c}	4.23±1.54^{b}
240	5.48±3.28^{d}	ND	ND	ND	1.56±2.80^{e}	6.95±2.35^{a}

Mean values bearing different superscripts within columns are significantly different (p< 0.05); = Not detected ND

Table 3: Effect of the storage period on sensory characteristics of the white cheese (Gibna Bayda)

Storage period (days)	Texture	Flavour	Colour	Saltiness
0.0	5.32±0.20^{c}	4.18±0.42^{d}	5.03±0.42^{d}	3.20±0.24
60	5.95±0.75^{b}	5.41±0.66^{b}	5.45±0.80^{b}	3.50±0.73
120	6.19±0.84^{a}	5.77±1.08^{a}	6.05±0.48^{a}	4.00±1.05
180	5.00±2.13^{d}	4.83±2.02^{c}	5.21±2.03^{c}	6.48±12.70
240	4.55±3.04^{e}	4.05±2.51^{d}	4.73±2.76^{e}	2.76±1.79
level of sign.	***	***	***	NS

Mean values bearing different superscripts within columns are significantly different (p<0.05); NS = Non significant

increased from day zero to day 180, then decreased at day 240. Total solids, crude protein and fat contents significantly (P0.05) increased from days zero to 120 then decreased gradually onwards. Volatile fatty acids, significantly (p<0.001) increased till day 120 then decreased at days 180 and 240. Tyrosine and tryptophane contents of the cheese samples were significantly (p<0.001) increased till the end of the storage period.

The Total Bacterial Count (TBA), coliforms, *E. coli*, *Staphylococcus aureus* and yeast and mould counts were shown (Table 2). Total bacterial counts increased from day zero to day 60 then gradually decreased onwards to day 240.

Coliform counts decreased from log count 4.04±1.02 in fresh cheese (day zero) suddenly to log count 1.23±1.16, then gradually decreased to log counts 0.24±0.66 and 0,14±0.38 at storage days 60 and 120, respectively. The coliforms were completely absent at day 240.

Escherichia coil in the cheese samples decreased in numbers from log count 2.14±0.16 in fresh cheese (day zero) suddenly to log count 0.74±0.98 at day 60 and to 0.16±0.43 at day 120 and it was completely absent at days 180 and 240. *Staphylococcus aureus* was detected only at day zero, at log count of 3.04 cfu/ml.

The results showed that psychrotrophic bacterial count significantly (p<0.05) decreased in log numbers from 6.35±0.08 at day zero to 3.28±3.37, 2.02±2.71, 2.052±3.35 and 1.56±2.80 at days, 60, 120, 180 and 240, respectively.

Yeast and moulds significantly (p<0.05) increased with increase in storage time from log count 2.65+0.±0.47 at day zero to 3.13±2.15, 4.79±2.20, 4.23±1.54 and 6.95±2.35 cfu/ml at days 60, 120, 180 and 240, respectively.

The texture, flavour, colour and saltiness of the cheese samples were presented in Table 3. Storage period significantly (p<0.05) affected the texture, flavour, couler of cheese during storage. The texture, flavour and colour scores improved from the fresh state (day zero) to day 60 and day 120, then decreased in days 180 and 240. Storage period significantly (p<0.05) affected the texture, flavour, colour and saltiness of the cheese.

Discussion

The weight loss results were similar to the findings reported by Bilal (2000) and Nuser (2001). The increase in weight loss was possibly due to the continuous movement of moisture from the cheese to the pickling solution. The moisture content decreased in all cheeses throughout the storage period (Aly and Galal, 2002). It might be attributed to degradation of proteins and fats (Zaki *et al.*,1974).

Similar increase in the acidity of the cheese was reported before by Bilal (2000) and Warsama *et al.* (2006). It is probably due to growth of lactic acid bacteria in the cheese (Walstra *et al.*, 1999; Warsama *et al.*, 2006). The lactic acid does not only contribute to the taste of fresh cheese but also helps cheese structure and protects it against some kind of microbiological spoilage (Ceylan *et al.*, 2003).

The increase in total solids of the cheese up to day 120 was likely attributed to continuous loss of moisture from cheese (Aly and Galal, 2002). The decrease might be due to proteolytic and lipolytic effect of microorganisms on proteins and dissolution of fats into pickling whey (Nuser, 2001; Hayaloglou *et al.*, 2005).

The crude protein findings agreed with Nuser (2001) who reported a protein value of 23.26% for fresh white cheese. The results were different from those reported by Ceylan *et al.* (2003) who reported a range of 16-22.7 for Orgu cheese. The initial increase in crude proteins probably due to decrease in moisture contents of the cheese (Khalid and El Owni, 1991; Abdel Razig, 1996), to the contrary, the nitrogen contents were slightly affected by moisture contents of the cheese (Tarakci and Kucukoner, 2006). The reduction in protein content was possibly due to activity of proteolytic agents on protein degradation (Kim *et al.*, 1992).

Fat content was increased from day zero to day 120, this might be due to the observed increase in TS. It might

also be due to contiuous loss of moisture and degradation products from the curd (Hofi *et al*., 1976). The breakdown of the cheese proteins and their loss in the whey (Babiker, 1987; Bilal, 2000). Advancing the ripening time leads to an increase in protein degradation (Hayaloglou *et al*., 2005). However, the decrease in fat content of the cheese samples from day 120 onwards could be attributed to the increased actions of microorganisms at low salt concentration (Nofal *et al*., 1981). Similarly lipolytic activity of microflora on fat resulting in leakage of some fat from the curd into pickling whey (Khalid and El Owni, 1991; Nuser, 2001). The VFA results were similar to those reported by Abdel Razig, 1996. The increase in VFA probably attributed to lipolytic action of organisms on fat contents during ripening (Esponda *et al*., 1983; Abdel Razig, 1996).

These results of the microorganisms in cheese were in line with those of Ahmed (1985) and Aly and Galal (2002). The increase in TBC possibly attributed to rapid growth of microorganisms during early stages of ripening. The differences found between cheese samples might be explained by the microbial counts of raw milk (Ceylan *et al*., 2003). However the decrease was likely due to the effect of high lactic acid in the cheese samples which suppress the growth of microorganisms (Shakeel-Ur Rehman *et al*., 2000).

The high total bacterial counts at day zero (log count 7.523±1.050) might be due to the observed high coliforms counts (4.04±1.02) in the cheese at day zero. While the decrease in their numbers thereafter probably attributed to increase activity of lactic acid bacteria. The coliform bacteria in cheese are expected to decrease during ripening by low pH and by antagonistic action of lactic acid (Babel, 1977). Aly and Galal (2002) found that coliforms decreased in numbers during storage of Domiati cheese from 3.5×10^5 at day zero to 1.2×10^5 at day 120.

The coliform results were in accordance with those of Maher *et al*. (2001) and Ahmed (1985) who stated that during storage of smear-ripened cheese produced from raw milk *E. coli* counts decreased from log 8.4±0.05 cfu/ ml at day one to less than 10 cfu/ at day 21. The decrease in their numbers could be due to the effect of high storage temperature and high salt level on their growth.

The *Staphylococcus aureus* results agreed with Asperger (1994) who reported log counts values of 6.0×10^3 in Norwegian cheese. This result disagreed with Aly and Galal 2002 who reported growth of *Staphylococcus aureus* for up to 90 days in cheese manufactured in raw milk.

The reduction in psychrotrophic count observed in this study was contrary to the findings of Ahmed (1985) who reported that psychrotrophic bacteria was 2.10×10^2 cfu/gm at day zero and increased to 5.0×10^4 cfu/gm after 8 months of storage.

The yeast and moulds counts were significantly ($p<0.05$) affected by storage period as they revealed a continuous increase with progress in storage time. This in agreement with Ceylan *et al*. (2003) who reported a significant ($p<0.05$) negative correlation ($r = -0.4014$) between yeast-mould contents and coliform counts due to acid forming ability. These results were concurred with those of Roostita and Fleet 1996 who found that yeast population of 10^6-10^8 cfu/ML were present when cheeses stored at either 25°C or 10°C.

The results of sensory characteristics were consistent with those reported by Abdel Razig (1996) and Tarakci and Kucukoner (2006). The improvement in texture from day zero to day 120 indicate clearly the effect of storage time. This agrees with Aly and Galal (2002) and Topcu and Saldamli (2006) who reported that the textural attributes of Turkish white cheese were significantly ($p<0.05$) affected by the ripening period. However, the deterioration in the texture thereafter might be due to further hydrolysis of proteins at later stages of ripening. It was likely due to the effect of proteolytic agents on the protein. It contributes to cheese off-flavour and abnormal texture through the breakdown of the released proteolytic products as amino acids, peptides into amines and acids. Advancing the ripening time leads to an increase in protein degradation of the released proteolytic products as amino acids (Hayaloglou *et al*., 2005).

The improvement in flavour was probably attributed to the effect of lactic acid development which control the growth of undesirable organisms (Kosikowski, 1982). The improvement in flvour might be due to the natural flora initially present in raw milk which participate in flvour production as reported by Law (1999). Chemical qualities of the cheeses including titratable acidity, pH, water- soluble- nitrogen ripening and tyrosine value had sinificent ($p<0.05$) effect on flavour (Guler and Uraz, 2004; Aly and Galal, 2002).

The improvement in colour from day zero to day 120 agreed with the findings of Tarakci and Kucukoner (2006) who reported that appearance and colour scores increased generally during ripening. However, the present study disagreed with Nuser (2001) who reported that storage period did not affect the colour of Sudanese white cheese during storage for 45 days.

It would be concluded that storage period significantly affected the weight loss, chemical composition, microbiological and sensory characteristics of Sudanese white cheese.

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