American Journal of Food Technology

ISSN 1557-4571

Academic Journals Inc.

www.academicjournals.com

American Journal of Food Technology 2 (1): 44-47, 2007
ISSN 1557-4571
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The Effect of Short-term Frozen Storage on the Chemical Composition and Coliform Microflora of Wara Cheese 'Wara Cheese under Frozen Storage'

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Abstract: The effect of short-term frozen (-20°C) storage on the chemical composition, pH and coliform bacterial count (cbc) of Wara cheese was investigated. Wara cheese at day old contained 31.00% total solids, 38.26% fat, 33.14% protein, 3.39% ash, 69.00% moisture, 376.50 * 10^5 cfu g^{-1} coliform bacteria and a pH of 5.04. At the end of 3 days of frozen storage, average total solids, fat, protein, moisture and ash were found, respectively, 33.25, 35.45, 36.63, 66.75 and 3.88%, while values for pH and cbc were 4.65 and 63.50 * 10^5 cfu g^{-1}. Daily differences obtained in the parameters observed during frozen storage were similar except for the total solids content that was significantly different ($p<0.01$). A superior and highly significant correlation ($r = 0.664$, $p<0.01$) was found between pH and cbc, while cbc had a positive correlation ($r = 0.366$) with moisture content.

Key words: Wara cheese, frozen storage, chemical composition, coliform bacterial count (cbc), pH

Introduction

Cheese production is a household operation in many developing countries. Livestock farming in general and milk products in particular still play an important socio-economic role in many developing countries (FAO, 1990). Cheese provides a useful service in extending the shelf life of a valuable human foodstuff - milk. Absence of standard processing methods explains the variations observed in cheese physico-chemical characteristics (Turkoglu *et al*., 2003; Belewu, 2001). The general standard of hygiene applied to milk production in developing countries is poor and as a result, the quality of milk and milk products is poor (Aneja, 1989; IFST, 1997). Coliform organisms in cheese indicate poor sanitation and process control although their presence does not threaten public health, (Nielson, 1976).

Freezing of foods, though helps to preserve their shelf life, color, flavor andnutritive value (Kuo and Gunasekaran, 2003), it also bring about certain physical and organoleptic changes, which may or may not be desirable (Hall and Alcock, 1987). Commercially produced cheese are frozen and stored to decrease the rate of ripening and prolong shelf life during marketing (Fontecha *et al*., 1996). However, little information is available on the effect of freezing on cheese microflora (Bricker and Van Hekken, 2004). The Nigerian soft cheese Wara is produced locally and consumed fresh. There is therefore the need to evaluate the changes in the chemical and some microbiological properties of Wara cheese under frozen storage in search of means of extending its shelf life. This study was therefore carried out to evaluate the effect of frozen storage on the chemical composition, pH andthe coliform bacterial count of Wara cheese.

Materials and Methods

Studies on the effect of short-term frozen storage on the chemical composition, pH andthe coliform bacterial count of Wara cheese were carried out during the late dry season between January

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and February in the Dairy Microbiology Laboratory of the Department of Animal Science, University of Ibadan, Nigeria. The soft cheese samples used in this study were purchased from local sellers in Ibadan, Nigeria. Collections were in four batches and each batch served as a replicate of the experiment. Samples were kept without delay under frozen temperature (-20°C) and subsequent chemical and microbiological analysis were carried out at the 15th, 39th, 63rd and 87th h of frozen storage treatment.

Appropriate dilutions of homogenized samples (11 g in 99 mL of sterile distilled water) were incubated in Violet Red Bile Agar (VRBA) (Oxoid) for coliform bacterial counts. pH was measured directly on a Metrohm-Herisau (Metrohm Ltd., Herisau, Switzerland) pH meter. Fat was analyzed by the standard Gerber method, ash and moisture contents by standard methods (AOAC, 1980), while the protein content was determined by the micro Kjedahl method. Results were statistically evaluated by analysis of variance, while differences among means were detected using Duncan's multiple range test.

Results and Discussion

The chemical composition, pH and coliform bacteria count of Wara cheese during frozen storage are presented in Table 1 and Fig. 1. The initial concentrations of total solids, fat, protein and ash were found, respectively, 31.00, 38.26, 33.14 and 3.39% while values for pH and cbc were 5.04 and 376.50* 10^5 cfu g^{-1}. The initial values observed for the moisture, fat and protein content fall within the range reported by Afolabi (1991), Ogundiwin and Oke, (1983) and Okomanyi, (1997). They reported that Wara cheese contained between 60.00-76.60% moisture, 30.00-48.00% fat and 36.63-41.10% protein. However, the 3.69% ash observed in this present study fall below the range (5.00-12.00%). Seasonal variations in the mineral content of pasture consumed by dairy animals could be attributed to the differences observed in the ash content of milk and milk products. The decline (38.26 to 35.45; $p>0.05$) observed in % fat might probably be attributed to sub-optimal lipolytic activities of microorganisms inherent in Wara cheese. Loney and Bassette (1970) had earlier reported an increase in fatty acid concentration during cheese storage. This result suggests that consumers who prefer less fat in diet may freeze Wara cheese for 3 days. On the other hand, the increase observed in the protein content of Wara cheese could be of microbial origin as a result of microbial proliferation most especially the lactic acid bacteria which has been reported to be the most predominant in fermented milk products by low pH (Gomez *et al*., 1989).

Wara cheese under frozen storage was observed to significantly retain more minerals than those kept in whey at room temperature. Average values were 3.69 and 2.83% (Alalade and Adeneye, 2006) for frozen and room temperature storage. Initial pH value of 5.04 was in agreement with previously reported range of 4.0-6.8 (Mormur *et al*., 1994; Kalogridou-Vassiliadou *et al*., 1994; Alalade and Adeneye, 2006). Wara is a slightly acidic milk product. The pH of Wara kept under frozen storage tends to be inversely proportional to the storage period. The increasing acidity in stored Wara would probably increase the shelf life and make it safer for human consumption by preventing the growth of coliform as well as pathogenic microorganisms. High levels of coliform bacteria have been associated with cheese produced via traditional processing methods. An average value of 229.06 * 10^5 cfu g^{-1} was observed during frozen storage. This result corroborates the findings of Mormur *et al*. (1994), Gomez *et al*. (1989) and Tzanetaki *et al*. (1987) who reported that cheese contained 10^2-10^8 cfu g^{-1}. The wide variation in CBC was a probable reflection of the unhygienic and unstandardized method of processing. CBC values declined by 83.13% ($p>0.05$) at the end of the 3-day frozen temperature treatment. The decline observed was in agreement with the findings of Gaya *et al*. (1983). Though CBC value declined ($p>0.05$) in Wara cheese, storage in whey at room temperature significantly reduced coliform count from 472.75 to 21.00 * 10^5 cfu g^{-1} over the same period of storage (Alalade and Adeneye, 2006). However, earlier report by Bricker and Van Hekken (2004) that bacteria demonstrated

Table 1: The effect of short-term frozen storage on chemical composition, pH and coliform bacterial count of Wara

Storage period (day)	Moisture (%)	Totol solids (%)	Fat (%)	Protein (%)	Ash (%)	pH	CBC *10^5 (Cfu g^{-1})
0	69.00±1.08^{a}	31.00±1.08^{ab}	38.26±1.65	33.14±0.53	3.39±0.52	5.04±0.44	376.50±118.49
1	72.75±1.25^{ab}	27.25±1.25^{b}	37.95±0.90	36.98±1.61	3.59±0.49	4.91±0.36	268.75±138.42
2	69.00±2.48^{ab}	31.00±2.48^{ab}	38.04±3.94	39.07±2.36	3.60±0.18	4.55±0.35	207.50±102.50
3	66.75±1.38^{b}	33.25±1.38^{a}	35.45±3.93	36.63±2.53	3.88±0.59	4.65±0.26	63.50±25.09
Av	69.36±0.92	30.63±0.92	37.43±1.47	36.46±0.96	3.69±0.23	4.79±0.18	229.06±52.64

^{abc}: Means on the same column differently superscripted differ significantly ($p<0.05$)

Table 2: Correlation analysis of parameters observed during storage

	PH	cbc	Moisture (%)	Fat (%)	Protein (%)	Ash (%)	Total solids (%)
pH		0.66**	0.25	0.05	-0.19	0.30	0.10
cbc			0.37	0.38	-0.13	-0.25	0.07
Moisture				0.46	-0.22	-0.06	0.15
Fat					-0.25	-0.36	0.01
Protein						-0.42	-0.46
Ash							0.15

**: $p<0.01$

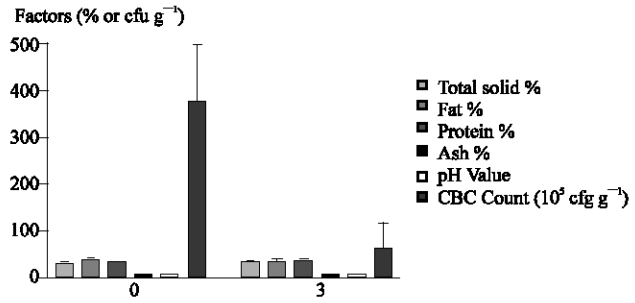

Fig. 1: Effect of frozen storage on the chemical composition and microbiological property of the Nigerian soft cheese on day 0 to 3 (Mean±SEM; n = 3)

similar survival in frozen citratehomogenate, citrate glycerol homogenate and chunk cheese thus suggesting that components in cheese provide cryoprotection for microflora did not hold for Wara cheese during frozen storage. A highly significant correlation ($p<0.01$, $r = 0.664$) was found between pH and cbc (Table 2). Humphrey *et al*. (1993) and Yeow-Lim *et al*. (1996) noted that acid sensitivity of coliform bacteria was a function of the temperature of the medium.

Conclusions

From the results of this study, it could be concluded that frozen storage treatment has no adverse effect on the chemical composition of Wara cheese. Besides, frozen storage, by reducing the coliform bacteria count of Wara cheese could increase its shelf life and makes it safer for human consumption.

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