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Original Research Article

Effect of Calcium Chloride on the Preparation of Low-fat Spreads from Buffalo and Cow Butter

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Abstract

Purpose: *To investigate the effects of $CaCl_2$ on the preparation of low-fat spreads from buffalo and cow butter*
Methods: *Buffalo and cow butter-based low-fat spreads (B-LFS and C-LFS) were treated with $CaCl_2$ (0, 0.02, 0.04, 0.06, and 0.08%) at pH 5.5 and stored at 4°C. They were sampled after 3, 30, 60, and 90 days, and analysed for sensory, morphological, rheological, and melting properties using a 9-point hedonic scale, digital camera, texture analyser TA-XT 2i, Physica MCR 301 rheometer, and differential scanning calorimeter, respectively.*
Results: *Sensory evaluation results showed that control samples were the best of all the treatments; additionally, no phase separation was found in samples treated with 0, 0.02 or 0.04% $CaCl_2$, but separation occurred with 0.06 and 0.08% $CaCl_2$. Generally, hardness and viscosity of samples decreased with increasing $CaCl_2$ concentrations; however, these parameters increased during storage. Increasing $CaCl_2$ concentrations didn't affect the melting profiles of the spreads, but the parameter varied for B-LFS during storage. Furthermore, the temperature range of the high melting zones of the B-LFS samples was greater than that of C-LFS samples.*
Conclusion: *Sensory, morphological, and rheological properties were affected by $CaCl_2$ concentrations but there were negligible effects on the melting behaviour of the spreads.*

Keywords: *Buffalo butter, Cow butter, Calcium chloride Low-fat spread, Sensory, Morphology, Melting*

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INTRODUCTION

Consumers have become increasingly health conscious and respond to the call for a diet that contains less fat, sugar, and salt, but higher fibre. Therefore, this trend has created great challenges for food technologists. For example, low-fat products prepared with a less than 40% fat content have captured increased market interest and extensive attention of food technologists [1].

Previously, there were attempts to produce bread spreads with a high dietary value containing one-half to one-quarter of the fat contained in butter and that also retains its desired appearance, flavor, texture, and sensory characteristics [2]. Obviously, the production of low-fat spreads with an increasingly larger aqueous phase requires the use of proteins and polysaccharides as thickeners or gelling agents. The ability of biopolymers to cross-link and, at high enough concentrations, to form a tangled, interconnected molecular network in water is widely known [3].

During the development of peak bone mass, calcium intake of less than 1 g per day is associated with lower bone mineral density [4]. Nutritionally sufficient levels of calcium in the diet are strongly related to the intake of dairy products, which are the richest sources of highly bioavailable calcium [5].

Therefore, the objective of this work was to study the effects of calcium chloride on sensory, morphological, rheological, and melting properties of buffalo and cow butter-based low-fat spreads (B-LFS and C-LFS).

EXPERIMENTAL

Materials

Buffalo butter (83.48% fat, 2.91% non-fat solids, 13.61% moisture, and 0.145 peroxide value) was obtained from the Department of Dairy Science, Faculty of Agriculture, Suez Canal University (Ismailia, Egypt). Cow butter (82.68% fat, 1.75% non-fat solids, 15.57% moisture, and 0.135 peroxide value), skim milk powder, and sodium chloride (table salt) were purchased from a local market in Wuxi (Jiangsu, China). Halal gelatin (80–280 Bloom) was purchased from Gelatin & Protein Co., Ltd. (Hangzhou, China). Dimodan® HP-C-distilled monoglyceride was obtained from Danisco Co. (Shanghai, China). All other reagents and solvents were of analytical or chromatographic grade to suit analytical requirements.

Preparation of buffalo and cow butter oil

Oil preparation was performed according to Fatouh *et al* [6] with some modifications. Both buffalo and cow butter were melted at 50°C instead of 60°C, and the top oil layer was then decanted and filtered through glass wool. The oil was then refiltered under vacuum to obtain clear buffalo and cow butter oil.

Preparation of B-LFS and C-LFS with different $CaCl_2$ concentrations

$CaCl_2$ treatments (B-LFS and C-LFS treated separately) were performed according to Madsen [7] with some modifications. The treatments consisted of the following (% w/w): 40% buffalo and cow butter oil, 0.5% DIMODAN®HP-C-distilled monoglyceride, 2% halal gelatin, 1% skimmed milk powder, 1% NaCl, $CaCl_2$ (0, i.e., control, 0.02, 0.04, 0.06, and 0.08%), 0.1% k-sorbate, and distilled water (to 100%). The steps for the preparation of $CaCl_2$ treatments were as follows:

1. The ingredients of the water phase (halal gelatin, skim milk powder, NaCl, and k-sorbate) were blended together with distilled water at 70°C for 10 min using a JJ-1B Electric Blender (Changzhou Runhua Electric Appliance Co., Ltd, China).

2. The temperature of the water phase was then reduced to 40°C, and the pH was adjusted with 20% w/w citric acid to 5.5 while mixing.

3. With regard to the fat phase, a portion of the melted buffalo and cow butter oil (~5 × the weight of the emulsifier) was removed and heated to 70°C with blending until the emulsifier dissolved, which was then added back to the melted butter oil at 40°C.

4. The water phase was then slowly added to the fat phase while mixing using a homogeniser (IKA® T18 Basic Ultra-Turrax®, Germany) for 5 min at speed no. 2.

5. The mixture was then pasteurised at 75°C for 10 min in a water bath while blending.

6. The temperature of the mixture was decreased from 75 to 60°C, and then $CaCl_2$ chloride (20% w/w) was blended with the mixture using a homogeniser (IKA® T18 Basic Ultra-Turrax®, Germany) for 3 min at speed no. 2.

7. The mixture was then allowed to pass once through a laboratory homogeniser (model: GYB, Donghua High Pressure Homogenizer Factory, Shanghai, China) at a pressure of 17 MPa and 60°C.

8. Calcium chloride treatments were kept in sterilized plastic cups (30 g) at room temperature for 15 h and then moved to a refrigerator (4°C).

Sensory evaluation

The sensory evaluation tests for $CaCl_2$ treatments (B-LFS and C-LFS) were carried out according to Patange *et al* [8] using a panel of 14 judges selected from Egypt, Sudan, and Yemen. Samples of $CaCl_2$ treatments were approximately 30 g and presented to panelists at refrigeration temperature (4°C). The color and appearance, spreadability, body and texture, flavor, and overall acceptability of the products were rated on a 9-point scale, ranging from 1 (disliked extremely) to 9 (liked extremely). Spreadability was assessed by the panelists using a slice of bread onto which the sample was spread at 4°C.

Morphology evaluation

The morphology evaluation test for $CaCl_2$ treatments (B-LFS and C-LFS) were recorded with a digital camera (Sony Camera T500, Japan).

Rheological measurements

Hardness

Calcium chloride treatments (B-LFS and C-LFS) in plastic cups (diameter × height = 4 × 2.5 cm) were kept in the refrigerator at 4°C before determination of hardness (g). The samples were removed from the refrigerator and quickly placed on the platform of a TA-XT 2i texture analyser (Stable Micro System, Ltd, UK). A puncture test was performed immediately using a probe (P/5: 0.50 cm-diameter cylindrical probe) at a pretest speed of 1 mm/s, test speed of 1 mm/s, post-test speed of 1 mm/s, and data acquisition rate of 200 points/s. The test was stopped when a penetration of 12 mm had been reached.

Apparent viscosity

Both B-LFS and C-LFS with $CaCl_2$ were removed from the refrigerator (4°C) and placed for 1 h at room temperature; then, the apparent viscosity was measured at 25°C with the 5 cm parallel-plate geometry of the Physica MCR 301 Rheometer (Anton Paar, Austria). The shear rates were from 0 to 200/s, whereas the apparent viscosity (Pas) was determined at a shear rate of 100/s.

Melting behavior

Differential scanning calorimetry (DSC Q2000 V24.9 Build 121, TA Instruments, New Castle, DE, USA) was used to determine the melting behaviour for the $CaCl_2$ treatments (B-LFS and C-LFS). The system was purged with nitrogen gas at 20 mL/min during the analysis, and liquid nitrogen was used as a refrigerant to cool the system. Calibration was performed with indium, eicosane, and dodecane standards. An empty aluminum pan was used as a reference. The samples (5 – 8 mg) were hermetically sealed in an aluminum pan, heated to 80°C, and held for 5 min to destroy completely the previous crystal structure. The samples were then cooled to -40°C and maintained for 5 min. Following this step, the melting profiles were obtained by heating the samples to 80°C at a rate of 10°C/min. DSC melting curves were recorded from - 40 to 80°C.

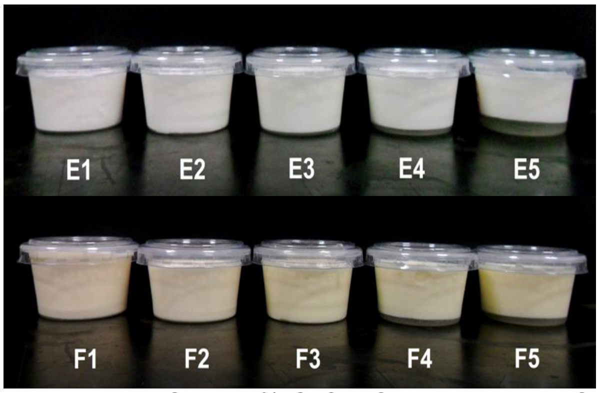
Statistical analysis

Calcium chloride treatments (B-LFS and C-LFS) were analyzed separately, and values of different tests were expressed as mean ± standard deviation (SD). One-way analysis of variance using SPSS 16 for windows (SPSS Inc, Chicago, USA) was performed on all experimental data sets. Duncan analysis was applied to evaluate the significance of differences.

RESULTS

Effect of $CaCl_2$ concentration on sensory and morphological properties of B-LFS and C-LFS

Sensory evaluation scores of B-LFS and C-LFS with different $CaCl_2$ concentrations are summarized in Table 1a and b. Results of sensory evaluation tests (color and appearance, body and texture, spreadability, flavor, and overall acceptability) revealed that the acceptance of these parameters by the panelists decreased gradually with increasing $CaCl_2$ concentrations (0, i.e., control, 0.02, 0.04, 0.06, and 0.08%). In addition, all $CaCl_2$ treatments showed decreased defects ($p < 0.05$) compared with the evaluation at the beginning (3 days) of the storage period. The morphology evaluation (Figure 1) of treatments showed no separate phases in the butters at $CaCl_2$ concentrations 0, 0.02, and 0.04%, whereas both 0.06 and 0.08% $CaCl_2$-treated butters separated, with the phase separation in 0.08% $CaCl_2$ more than in 0.06% $CaCl_2$.



Key: E1) B-LFS with 0% $CaCl_2$ (Control), E2) B-LFS with 0.02% $CaCl_2$, E3) B-LFS with 0.04% $CaCl_2$, E4) B-LFS with 0.06%$CaCl_2$, E5) B-LFS with 0.08% $CaCl_2$ F1) C-LFS with 0% $CaCl_2$ (Control), F2) C-LFS with 0.02% $CaCl_2$, F3) C-LFS with 0.04% $CaCl_2$, F4) C-LFS with 0.06% $CaCl_2$, F5) C-LFS with 0.08% $CaCl_2$

Figure 1: Effect of different $CaCl_2$ concentrations on the sensory evaluation of B-LFS and C-LFS

Table 1(a): Effect of $CaCl_2$ concentration on sensory and morphological properties of B-LFS

	Storage (days)	Sensory evaluation[a]				
		B-LFS				
		$CaCl_2$ 0% (control)	**$CaCl_2$ 0.02%**	**$CaCl_2$ 0.04%**	**$CaCl_2$ 0.06%**	**$CaCl_2$ 0.08%**
Color & Appearance	3	8.67 ± 0.10^{aA}	8.59 ± 0.20^{aA}	8.22 ± 0.14^{aB}	7.39 ± 0.08^{aC}	6.42 ± 0.14^{aD}
	15	8.53 ± 0.07^{abA}	8.37 ± 0.23^{abA}	8.17 ± 0.34^{abA}	7.33 ± 0.09^{abB}	6.30 ± 0.30^{abC}
	30	8.41 ± 0.13^{bcA}	8.35 ± 0.11^{abA}	8.14 ± 0.21^{abA}	7.25 ± 0.25^{abB}	6.22 ± 0.08^{abcC}
	45	8.37 ± 0.10^{bcdA}	8.31 ± 0.13^{abcA}	8.11 ± 0.10^{abB}	7.14 ± 0.08^{abcC}	6.11 ± 0.12^{abcdD}
	60	8.35 ± 0.12^{bcdA}	8.25 ± 0.12^{bcAB}	8.00 ± 0.29^{abB}	7.04 ± 0.07^{bcC}	6.00 ± 0.05^{bcdD}
	75	8.23 ± 0.23^{cdA}	8.11 ± 0.22^{bcA}	7.95 ± 0.12^{abA}	6.92 ± 0.27^{cB}	5.94 ± 0.12^{cdC}
	90	8.15 ± 0.08^{dA}	8.05 ± 0.10^{cAB}	7.82 ± 0.13^{bB}	6.88 ± 0.21^{cC}	5.87 ± 0.29^{dD}
Body & Texture	3	8.57 ± 0.11^{aA}	8.67 ± 0.28^{aA}	8.45 ± 0.09^{aA}	8.11 ± 0.07^{abB}	7.77 ± 0.20^{aC}
	15	8.47 ± 0.11^{abA}	8.61 ± 0.11^{abA}	8.41 ± 0.12^{abA}	8.12 ± 0.14^{aB}	7.63 ± 0.25^{abC}
	30	8.45 ± 0.28^{abA}	8.54 ± 0.09^{abA}	8.32 ± 0.13^{abAB}	8.05 ± 0.12^{abB}	7.64 ± 0.10^{abC}
	45	8.31 ± 0.15^{bcA}	8.46 ± 0.13^{abcA}	8.28 ± 0.20^{abAB}	8.00 ± 0.08^{abB}	7.55 ± 0.11^{abcC}
	60	8.24 ± 0.07^{bcA}	8.33 ± 0.32^{bcdA}	8.16 ± 0.12^{abcA}	7.97 ± 0.34^{abA}	7.41 ± 0.08^{bcB}
	75	8.16 ± 0.10^{cA}	8.21 ± 0.11^{cdA}	8.15 ± 0.21^{bcA}	7.83 ± 0.07^{abB}	7.39 ± 0.14^{bcC}
	90	8.11 ± 0.10^{cA}	8.07 ± 0.10^{dA}	7.98 ± 0.14^{cA}	7.70 ± 0.47^{bAB}	7.33 ± 0.08^{cB}
Spreadability	3	8.74 ± 0.41^{aA}	8.44 ± 0.13^{aAB}	8.68 ± 0.12^{aAB}	8.27 ± 0.10^{aBC}	7.86 ± 0.28^{aC}
	15	8.63 ± 0.08^{abA}	8.39 ± 0.05^{abAB}	8.66 ± 0.07^{abA}	8.22 ± 0.30^{aB}	7.81 ± 0.10^{aC}
	30	8.56 ± 0.11^{abA}	8.32 ± 0.25^{abAB}	8.51 ± 0.12^{bcA}	8.17 ± 0.12^{abB}	7.77 ± 0.11^{abC}
	45	8.55 ± 0.09^{abA}	8.27 ± 0.33^{abcAB}	8.43 ± 0.04^{cdAB}	8.11 ± 0.08^{abBC}	7.75 ± 0.27^{abC}
	60	8.42 ± 0.12^{abcA}	8.17 ± 0.13^{abcBC}	8.34 ± 0.07^{deAB}	8.04 ± 0.07^{abcC}	7.64 ± 0.08^{abD}
	75	8.37 ± 0.10^{bcA}	8.10 ± 0.09^{bcBC}	8.21 ± 0.10^{efAB}	7.94 ± 0.10^{bcC}	7.58 ± 0.13^{abD}
	90	8.19 ± 0.23^{cA}	7.96 ± 0.13^{cAB}	8.11 ± 0.12^{fA}	7.82 ± 0.10^{cB}	7.50 ± 0.12^{bC}
Flavor	3	8.70 ± 0.12^{aA}	8.61 ± 0.17^{aA}	7.61 ± 0.14^{aB}	6.15 ± 0.07^{aC}	4.30 ± 0.10^{aD}
	15	8.64 ± 0.08^{aA}	8.54 ± 0.20^{abA}	7.50 ± 0.12^{abB}	6.11 ± 0.13^{abC}	4.24 ± 0.25^{aD}
	30	8.65 ± 0.10^{aA}	8.43 ± 0.10^{abcA}	7.43 ± 0.24^{abB}	6.00 ± 0.27^{abC}	4.17 ± 0.10^{abD}
	45	8.53 ± 0.17^{abA}	8.38 ± 0.12^{abcdA}	7.32 ± 0.11^{abcB}	6.05 ± 0.07^{abC}	4.00 ± 0.13^{bcD}
	60	8.42 ± 0.10^{bcA}	8.27 ± 0.31^{bcdA}	7.22 ± 0.10^{bcdB}	5.93 ± 0.31^{abC}	3.94 ± 0.07^{bcdD}
	75	8.30 ± 0.09^{cdA}	8.22 ± 0.13^{cdA}	7.12 ± 0.24^{cdB}	5.87 ± 0.05^{abC}	3.86 ± 0.14^{cdD}
	90	8.18 ± 0.09^{dA}	8.11 ± 0.12^{dA}	6.93 ± 0.16^{dB}	5.72 ± 0.39^{bC}	3.72 ± 0.08^{dD}
Overall Acceptability	3	8.79 ± 0.24^{aA}	8.66 ± 0.19^{aA}	7.90 ± 0.15^{aB}	6.77 ± 0.07^{aC}	4.60 ± 0.10^{aD}
	15	8.68 ± 0.06^{abA}	8.51 ± 0.19^{abA}	7.84 ± 0.11^{aB}	6.73 ± 0.31^{abC}	4.51 ± 0.13^{aD}
	30	8.60 ± 0.12^{abA}	8.43 ± 0.10^{abcA}	7.75 ± 0.23^{abB}	6.64 ± 0.26^{abcC}	4.43 ± 0.11^{abD}
	45	8.52 ± 0.13^{bcA}	8.34 ± 0.21^{bcdA}	7.77 ± 0.08^{abB}	6.61 ± 0.08^{abcC}	4.37 ± 0.27^{abD}
	60	8.44 ± 0.13^{bcdA}	8.22 ± 0.12^{cdB}	7.56 ± 0.05^{bcC}	6.50 ± 0.06^{abcD}	$4.21\pm0.05b^{cE}$
	75	8.28 ± 0.11^{cdA}	8.14 ± 0.09^{dA}	7.43 ± 0.10^{cB}	6.41 ± 0.07^{bcC}	4.10 ± 0.12^{cdD}
	90	8.20 ± 0.10^{dA}	8.08 ± 0.15^{dA}	7.38 ± 0.14^{cB}	6.33 ± 0.24^{cC}	3.93 ± 0.10^{dD}

Effects of $CaCl_2$ concentrations on hardness of B-LFS and C-LFS

Effects of $CaCl_2$ concentrations on the texture evaluation of B-LFS and CLFS are presented in Table 2. The texture evaluation showed that the differences in hardness among all $CaCl_2$ treatments (B-LFS and C-LFS) were similar to control samples, but the hardness with 0.06 and 0.08% $CaCl_2$ were clearly lower than the control. Furthermore, within all treatments, the hardness significantly increased ($p < 0.05$) during storage. Additionally, the hardness of $CaCl_2$ treatments (B-LFS) was slightly higher than the C-LFS treatments.

Effects of $CaCl_2$ concentrations on the viscosity of B-LFS and C-LFS

Effects of $CaCl_2$ concentrations on the viscosity of B-LFS and C-LFS samples are reported in Table 3. The viscosity among B-LFS and C-LFS separately with levels of $CaCl_2$ (0.02%, 0.04%, and 0.06%) was not significant, except for 0.06% $CaCl_2$ with B-LFS at 3 days and 0.04% $CaCl_2$ with B-LFS and C-LFS at 60 days. In addition, $CaCl_2$ treatments (0.08%) were clearly lower ($p < 0.05$) compared to the control samples. Moreover, the viscosity of all samples treated with $CaCl_2$ significantly increased during storage at 4°C.

Table 1(b): Effect of $CaCl_2$ concentrations on sensory and morphological properties of C-LFS

	Storage (days)	Sensory evaluation[a] C-LFS				
		$CaCl_2$ 0% (control)	$CaCl_2$ 0.02%	$CaCl_2$ 0.04%	$CaCl_2$ 0.06%	$CaCl_2$ 0.08%
Color & appearance	3	8.59±0.11^{aA}	8.43±0.11^{aAB}	8.31±0.11^{aB}	7.46±0.08^{aC}	6.54±0.09^{aD}
	15	8.31±0.07^{bcA}	8.38±0.12^{abA}	8.32±0.08^{aA}	7.37±0.27^{abB}	6.45±0.18^{abC}
	30	8.37±0.13^{bA}	8.35±0.07^{abA}	8.24±0.07^{aA}	7.26±0.08^{abcB}	6.41±0.10^{abC}
	45	8.24±0.14^{bcA}	8.26±0.14^{abA}	8.13±0.10^{aA}	7.17±0.12^{bcB}	6.33±0.12^{bcC}
	60	8.16±0.12^{cA}	8.22±0.07^{abA}	8.14±0.24^{aA}	7.12±0.10^{cdB}	6.21±0.10^{cdC}
	75	8.14±0.12^{cA}	8.17±0.10^{abA}	8.12±0.09^{aA}	7.13±0.11^{cdB}	6.11±0.06^{deC}
	90	8.10±0.13^{cA}	8.12±0.34^{bA}	7.88±0.13^{bA}	6.90±0.07^{dB}	5.93±0.10eC
Body & texture	3	8.46±0.08^{aA}	8.61±0.11^{aA}	8.46±0.14^{aA}	8.04±0.11^{aB}	7.80±0.11^{aC}
	15	8.45±0.11^{aAB}	8.53±0.09^{abA}	8.33±0.08^{abB}	8.00±0.10^{aC}	7.74±0.12^{abD}
	30	8.36±0.09^{abA}	8.43±0.31^{abcA}	8.28±0.11^{abcA}	7.94±0.11^{abB}	7.66±0.13^{abcB}
	45	8.26±0.22^{abcA}	8.37±0.10^{abcA}	8.26±0.17^{abcA}	7.87±0.07^{abcB}	7.62±0.13^{abcdB}
	60	8.21±0.11^{bcA}	8.21±0.28^{bcA}	8.27±0.09^{abcA}	7.81±0.10^{bcB}	7.51±0.13^{bcdC}
	75	8.11±0.12^{cA}	8.22±0.14^{bcA}	8.17±0.15^{bcA}	7.72±0.10^{cdB}	7.43±0.09^{cdC}
	90	8.12±0.08^{cA}	8.16±0.10^{cA}	8.06±0.23^{cA}	7.61±0.10^{dB}	7.40±0.22^{dB}
Spreadability	3	8.67±0.08^{aA}	8.54±0.12^{aA}	8.58±0.09^{aA}	8.37±0.13^{aA}	7.98±0.31^{aB}
	15	8.53±0.10^{abA}	8.41±0.14^{abAB}	8.46±0.08^{abAB}	8.34±0.10^{aB}	7.97±0.09^{aC}
	30	8.49±0.13^{abA}	8.38±0.07^{abA}	8.36±0.31^{abcA}	8.31±0.10^{aA}	7.81±0.09^{abB}
	45	8.51±0.12^{abA}	8.33±0.12^{bA}	8.31±0.09^{bcA}	8.32±0.31^{aA}	7.70±0.11^{bB}
	60	8.43±0.31^{abA}	8.28±0.10^{bA}	8.30±0.09^{bcA}	8.17±0.12^{abA}	7.63±0.11^{bB}
	75	8.41±0.06^{bA}	8.27±0.07b^{AB}	8.23±0.11^{bcB}	8.16±0.08^{abB}	7.60±0.09^{bC}
	90	8.31±0.06^{bA}	8.07±0.14^{cAB}	8.12±0.14^{cAB}	7.93±0.22^{bB}	7.56±0.10^{bC}
Flavor	3	8.52±0.10^{aA}	8.53±0.12^{aA}	7.56±0.12^{aB}	6.11±0.10^{aC}	4.20±0.07^{aD}
	15	8.45±0.09^{abA}	8.44±0.11^{abA}	7.47±0.09^{aB}	6.04±0.10^{aC}	4.16±0.09^{aD}
	30	8.42±0.11^{abA}	8.39±0.26^{abcA}	7.42±0.10^{abB}	5.96±0.09^{abC}	4.11±0.13^{abD}
	45	8.41±0.25^{abA}	8.32±0.07^{abcA}	7.33±0.09^{abcB}	5.81±0.10^{bcC}	3.94±0.13^{bcD}
	60	8.40±0.11^{abA}	8.24±0.10^{abcA}	7.19±0.25^{bcdB}	5.73±0.12^{cdC}	3.88±0.08^{cdD}
	75	8.38±0.11^{abA}	8.20±0.14^{bcA}	7.12±0.11^{cdB}	5.66±0.11^{cdC}	3.72±0.08^{deD}
	90	8.27±0.10^{bA}	8.10±0.31^{cA}	6.00±0.23^{dB}	5.58±0.08^{dC}	3.60±0.22^{eD}
Overall acceptability	3	8.43±0.10^{aA}	8.35±0.12^{aA}	7.77±0.12^{aB}	6.80±0.10^{aC}	4.51±0.09^{aD}
	15	8.35±0.09^{abA}	8.28±0.11^{aA}	7.71±0.08^{abB}	6.71±0.12^{aC}	4.43±0.08^{abD}
	30	8.31±0.14^{abcA}	8.24±0.05^{aA}	7.63±0.31^{abB}	6.63±0.08^{abC}	4.31±0.12^{bcD}
	45	8.26±0.15^{abcA}	8.23±0.10^{aA}	7.50±0.07^{abcB}	6.52±0.32^{abcC}	4.20±0.12^{cD}
	60	8.18±0.32^{abcA}	8.16±0.10^{abA}	7.44±0.23^{bcB}	6.41±0.11^{bcdC}	4.14±0.11^{cD}
	75	8.13±0.09^{bcA}	8.12±0.10^{abA}	7.32±0.09^{cB}	6.30±0.10^{cdC}	3.94±0.07^{dD}
	90	8.06±0.09^{cA}	7.94±0.34^{bA}	7.27±0.15^{cB}	6.20±0.18^{dC}	3.80±0.10^{dD}

Capital letters: Average values with different letters are statistically significant ($p < 0.05$) within each row. Small letters: Average values with different letters are statistically significant ($p < 0.05$) within each column. ^{a}mean±S.D,; n = 14.

Table 2: Effect of $CaCl_2$ concentrations on hardness of B-LFS and C-LFS

Storage (days)	Hardness (g)[a]				
	B-LFS				
	$CaCl_2$ 0% (control)	*$CaCl_2$ 0.02%*	*$CaCl_2$ 0.04%*	*$CaCl_2$ 0.06%*	*$CaCl_2$ 0.08%*
3	58.31±0.39^{dA}	52.03±0.78^{cC}	56.02±0.50^{cB}	37.64±0.44^{dD}	37.33±0.59^{cD}
30	61.42±0.30^{cA}	53.33±0.60^{cC}	56.36±0.61^{bcB}	38.86±0.46^{cD}	37.95±0.61^{cD}
60	64.39±0.56^{bA}	56.36±0.92^{bB}	57.77±0.83^{bB}	43.81±0.65^{bC}	39.41±0.97^{bD}
90	66.46±0.45^{aA}	61.32±0.54^{aB}	60.12±1.11^{aC}	46.85±0.36^{aD}	44.78±0.47^{aE}
	C-LFS				
3	56.95±0.46^{cA}	49.01±0.52^{dC}	52.31±0.50^{cB}	36.77±0.51^{dD}	34.51±0.59^{dE}
30	61.20±0.62^{bA}	52.42±0.62^{cC}	53.87±0.39^{bB}	38.11±0.41^{cD}	36.63±0.61^{cE}
60	62.16±0.66^{bA}	54.67±0.71^{bB}	54.78±0.49^{bB}	40.15±0.48^{bC}	39.99±0.86^{bC}
90	64.00±0.96^{aA}	59.31±0.65^{aB}	57.52±0.78^{aC}	45.36±0.77^{aD}	45.12±0.47^{aD}

Capital letters: *Average values with different letters are statistically significant ($p < 0.05$) within each row. Small letters: Average values with different letters are statistically significant ($p < 0.05$) within each column. ^{a}mean±S.D,; n = 3.*

Table 3: Effect of $CaCl_2$ concentrations on the viscosity of B-LFS and C-LFS

Storage (days)	Apparent viscosity {η_{app} (Pa s) at 100 γ^{s-1}}^{a}				
	B-LFS				
	$CaCl_2$ 0% (control)	$CaCl_2$ 0.02%	$CaCl_2$ 0.04%	$CaCl_2$ 0.06%	$CaCl_2$ 0.08%
3	0.32 ± 0.07^{cA}	0.30 ± 0.03^{cAB}	0.29 ± 0.03^{cAB}	0.23 ± 0.03^{dB}	0.13 ± 0.04^{cC}
30	0.43 ± 0.09^{bcA}	0.43 ± 0.05^{bA}	0.46 ± 0.08^{bA}	0.41 ± 0.03^{cA}	0.26 ± 0.04^{bB}
60	0.49 ± 0.05^{abB}	0.55 ± 0.04^{aAB}	0.58 ± 0.05^{aA}	0.48 ± 0.04^{bB}	0.34 ± 0.03^{aC}
90	0.60 ± 0.08^{aAB}	0.56 ± 0.04^{aB}	0.66 ± 0.05^{aA}	0.55 ± 0.03^{aB}	0.36 ± 0.05^{aC}
	C-LFS				
3	0.25 ± 0.03^{cA}	0.28 ± 0.04^{cA}	0.28 ± 0.07^{cA}	0.21 ± 0.04^{cA}	0.12 ± 0.03^{cB}
30	0.38 ± 0.07^{bA}	0.41 ± 0.07^{bA}	0.45 ± 0.08^{bA}	0.41 ± 0.03^{bA}	0.26 ± 0.04^{bB}
60	0.45 ± 0.08^{bB}	0.53 ± 0.05^{aAB}	0.55 ± 0.07^{abA}	0.44 ± 0.02^{bB}	0.33 ± 0.03^{aC}
90	0.58 ± 0.05^{aA}	0.56 ± 0.05^{aA}	0.63 ± 0.09^{aA}	0.54 ± 0.05^{aA}	0.35 ± 0.03^{aB}

Capital letters: *average values with different letters are statistically significant ($p < 0.05$) within each row.* ***Small letters:*** *average values with different letters are statistically significant ($p < 0.05$) within each column. ^{a}Mean ± SD; n = 3.*

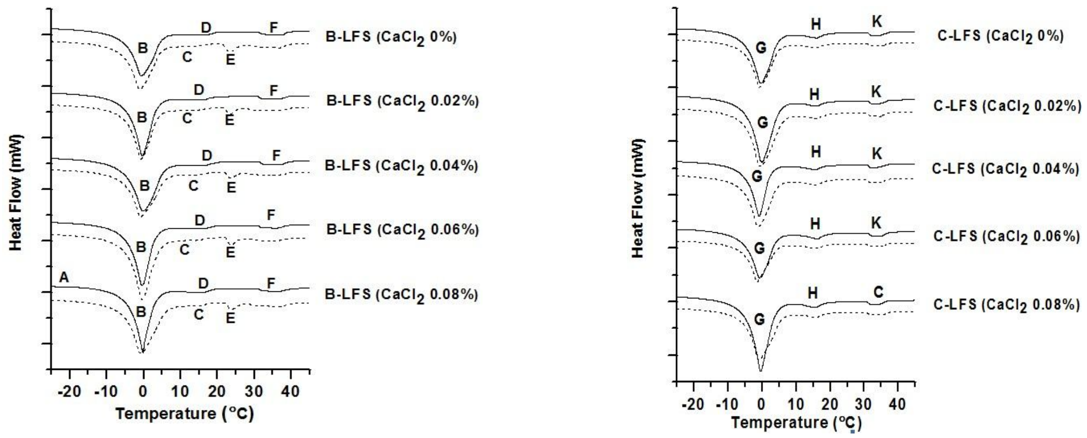


Figure 2: Thermograms of butter containing varying concentrations of calcium chloride

Table 4: Effect of $CaCl_2$ on the melting point behavior of B-LFS

$CaCl_2$ (%)	Storage (days)	B-LFS						C-LFS		
		Melting zone (°C)						Melting zone (°C)		
		A	B	C	D	E	F	G	H	K
0	3	–	-0.72	–	17.14	–	31.99 – 37.90	-0.47	16.14	32.24 to 34.75
	90	–	-1.22	13.49	–	23.56	30.98 – 37.02	-0.97	16.01	31.86 to 35.63
0.02	3	–	-0.48	–	16.78	–	32.15 – 36.81	-0.23	15.90	32.15 to 34.80
	90	–	-0.85	14.37	–	23.68	30.48 – 37.27	-0.72	16.01	31.99 to 34.63
0.04	3	–	-0.23	–	16.91	–	32.40 – 37.57	-0.98	15.77	31.77 to 34.67
	90	–	-0.85	15.00	–	23.81	30.22 – 36.64	-1.35	15.88	31.61 to 35.38
0.06	3	–	-0.48	–	16.78	–	31.90 – 36.81	-0.73	16.28	32.02 to 35.05
	90	–	-0.49	14.63	–	23.81	30.98 – 37.02	-1.60	16.14	31.73 to 35.76
0.08	3	-23.03	-0.23	–	16.65	–	32.15 – 37.32	-0.48	15.52	31.90 to 34.54
	90	–	-0.85	15.00	–	24.06	31.11 – 37.39	-0.97	15.88	31.86 to 35.76

Effect of CaCl2 concentration on thermal behavior of B-LFS and C-LFS – aaa

The thermal profiles of B-LFS and C-LFS with different $CaCl_2$ concentrations are presented in Figure 2 and Table 4. The melting zone (A) was only detected by DSC after 3 days of storage with 0.08% $CaCl_2$ (B-LFS), but disappeared after 90 days of storage. The temperatures of the major peaks (B and G) slightly decreased after 90 days, and the differences between temperatures of melting zones B and G together were slight. Moreover, the temperatures between the endothermic peaks of D and H were similar, but after 90 days, no effects were observed on the melting zone of H. With all $CaCl_2$ treatments (B-LFS), two endothermic peaks (C and E) were detected. Furthermore, the differences in broad shoulders (high melting zones of F and K) among and within $CaCl_2$ treatments (B-LFS and C-LFS separately) were slight. On the other hand, all temperature ranges of the melting zones for F were greater than for K.

DISCUSSION

Phase separation occurred with 0.06 and 0.08% $CaCl_2$, as the attraction potential (Van der Waals' interaction) was greater than the repulsion potential: however, our results were consistent with those of Keowmaneechai and McClements [9], who reported that the droplet aggregation of menhaden emulsions may be due to the combined contributions of both the effects of heating on increased hydrophobic attractions and the influence of $CaCl_2$ on decreased electrostatic repulsions.

Scores for body and texture declined after $CaCl_2$ treatments (B-LFS and C-LFS) during storage due to the proteolytic action of microorganisms in the nonfat portion of the sample [8]. In addition, the changes in spreadability scores of the treatments during storage may be attributed to protein degradation and/or decreased water holding capacity by the nonfat fraction, resulting in increased softening of the spread, particularly towards the end of storage [8]. On the other hand, the decline in flavor scores during storage may be attributed to a loss of freshness [8]. The bitter flavor imparted by $CaCl_2$ treatments (B-LFS and C-LFS) appeared at higher concentrations (0.06 and 0.08%), whereas there was no bitterness detected in either 0.02 or 0.04% $CaCl_2$ when compared with the control samples.

The hardness and viscosity of $CaCl_2$ treatments were significant increasing during the storage periods, due to the slow post-crystallization processes and development of bonds within the fat crystal network that took place during storage [10].

In general, hardness and viscosity decreased with increasing $CaCl_2$ concentrations, presumably because an increase in $CaCl_2$ leads to the weakening of the gelatin, and in addition, because of the attraction and repulsion potentials (see sensory evaluation tests). Panouillé and Larreta-Garde [11] found that the high concentrations of Ca^{+2} in sodium caseinate emulsions could lead to an over-association of alginate chains, resulting in a weakening of the gel.

Hardness was correlated with the viscosity of $CaCl_2$ treatments (B-LFS and C-LFS); however, our results were in agreement with those observed by Glibowski *et al* [12], who reported that hardness was highly correlated with the viscosity of spreads. Furthermore, the solidifying points for both buffalo and cow milk fat were 16.0 – 28.0°C and 15.0 – 23.5°C, respectively [13]. In addition, Patel and Frede [14] found that the crystallization of buffalo milk fat begins at a higher temperature than does cow milk fat. Therefore, it's clear that both solidifying points and crystallization are responsible for the hardness and viscosity test results.

An increase in $CaCl_2$ did not affect the melting profiles of B-LFS and C-LFS separately, but there were differences in the melting profiles of the B-LFS samples during storage. Furthermore, the temperature ranges of the high melting zones of the B-LFS samples were greater than in the C-LFS samples; these results suggest that a slight increase in total high melting species [15] is in total accordance with hardness and viscosity. However, the changes in the shape of the melting profile could be due to changes in polymorphism [16]. Ramamurthy and Narayanan [17] reported that buffalo milk fat has a greater proportion of high melting triglycerides than does cow milk fat (9 – 12% and 5 – 6%, respectively).

CONCLUSION

Treatments with $CaCl_2$ (0, 0.02 and 0.04%) were deemed acceptable by the panelists; whereas higher concentrations (0.06 and 0.08%) were unaccepted. Generally, hardness and viscosity of B-LFS and C-LFS separately treated with $CaCl_2$ decreased with an increase in $CaCl_2$, but increased during storage. With regard to thermal behaviour, we did not notice changes in the melting profiles of B-LFS and C-LFS separately with increasing $CaCl_2$, but we noticed differences in the melting profile of B-LFS samples during storage. In addition, the temperature ranges of the high melting zones of B-LFS were greater than in C-LFS.

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