Journal of Food and Dairy Sciences

Journal homepage: www.jfds.mans.edu.eg
Available online at: www. jfds.journals.ekb.eg

Fotification of Karish Cheese with Calcium by Means of Using Calcium Chloride or Casein Co-Precipitates Containing High Calcium

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ABSTRACT

The objective of the present work was to increase calcium content of Karish cheese by means of adding calcium chloride of high calcium casein co-precipitates (HCCCP) to cheese milk. In treatments 1 and 2 (T1 and T2) calcium was increased to 200 mg/100 ml of cheese milk, while in treatments 3 and 4 (T3 and T4) calcium was increased to be 300 mg/100 ml of milk. Calcium chloride was used in T1 and T3, while HCCCP was used in T2 and T4. The attained results revealed that the control fresh cheese had the lowest significant yield (26.17%), while the highest yield was recorded when calcium chloride was used since the values in T1, T2, T3 and T4 were 33.14, 31.26, 37.67 and 33.15% respectively. Cheese from T3 and T4 had the highest values for total solids, protein, SN/TN, ash and calcium, whereas the values were the minimum in control cheese (C). This was the same with acidity, whereas the trend of pH was the opposite. The differences in most cases were statistically significant (P≤0.05).Fortification of cheese milk with calcium increased hardness, cohesiveness, gumminess and chewiness of cheese and decreased adhesiveness and springiness. T3 and T4 were more effective in this respect as well as had adverse impact on the organopeltic properties of the resultant cheese. Moreover, the use of HCCCP caused more improving in this respect.

Keywords: Karish cheese, fortification with calcium.

INTROIDUCTION

Calcium has many benefits, such as lowering blood pressure particularly among young people, hypertensive disorders of pregnancy, prevention of osteoporosis and colorectal adenomas, lowering blood pressure in the progeny of mothers talking sufficient calcium during pregnancy and lowering cholesterol values (Cormick and Belizan, 2019). It is important nutrient to maximize peak bone mass for osteoporosis prevention (Marcinow *et al.*, 2017) and also effective on non skeletal, especially cardiovascular (Tankeu *et al.*, 2017).

The recommended daily intake of calcium varies between 700 and 1200 mg of elemental calcium (Cano *et al.*, 2018).

Dairy products are the best source for calcium because of their low cost and high biovaibility of this mineral. Cheese is rich in nutrients including calcium, protein, vit. D, potassium and phosphorus (Rizzoli, 2014). Unfortunately, calcium levels are comparatively low in soft cheese, because such cheeses are prepared by acid coagulation. This changes the equilibrium between the colloidal and ionic states of calcium in cheese milk (Tunick, 1987). The calcium converts from the colloidal state to the ionic one, which is lost in the whey. In this respect, it was reported that such impact causing at least two-thirds of calcium to be lost in cheese whey (Wong *et al.*, 1976). However, fortification of Karish cheese milk with calcium is quite important since Karish cheese is acid-coagulated soft cheese.

Addition of calcium chloride to dairy products has the potential to influence the flavour, stability, texture and processing characteristics of the final product (Willium *et al.*, 2005). Calcium percentage greater than 0.02% produces maximum yields for cheese (Ustund and Iticks, 1990).

In the present study calcium content of cheese milk was increased by addition of calcium chloride or high calcium casein co-precipitates (HCCCP). The use of HCCCP is a way to increase also protein content. Milk proteins have many benefits for health like hypertension, obesity, type-2 diabetes mellitus and hypercholesterolemia. They have many beneficial role on the intestinal mucosa through the interaction with opioid receptors located at the epithelium, as well as the protection of the intestinal mucosa and anti-inflammatory activity. They play also an important role in the antimicrobial, immunomodulatory, relaxing and antinocipeptive (Miralles *et al.*, 2018).

The aim of this study was to investigate the effect of calcium chloride and high calcium casein co-precipitates (HCCCP) on the composition, texture attributes and sensory characteristics of Karish cheese.

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DOI: 10.21608/jfds.2019.54698

MATERIALS AND METHODS

Materials:

Fresh cow's and buffalo's milk were obtained from the herds of Sakha and Mehalet Moussa Experimental Stations belonging to Animal Production Research Institute, Ministry of Agriculture, respectively. The milk samples were skimmed using milk separator to obtain skim cow's milk (SCM) and skim buffalo's milk (SBM) in order. The commercial yoghurt starter culture containing *Streptococcus thermophilus* and *Lactobacillus delbrueckii* sub ssp. bulgaricus (YCX-11) in a freeze-dried direct vat set (DVS) was obtained from CHR-Hansen Lab., Denmark. Food-grade calcium chloride was purchased from the local market whereas, high calcium casein co-precipitates (HCCCP) was prepared from SBM according to the method given by Mulvihill (1992). The HCCCP was a mixture of casein and whey proteins while it contained 42.3% total solids, 16.2% protein, 0.1% fat, 4.68% ash and 238 mg calcium /100 g.

Karish cheese making:

Karish cheese was made from SCM according to the method given by Abd El-Khabir *et al.* (2007). In this respect, the prepared SCM was divided into five equal portions. The first one was kept without any additives and served as control (C) whereas calcium chloride was added to milk to increase its calcium content to 200 (T1) and 300 (T3) mg/100 ml respectively. In treatment 2 (T2) and 4 (T4), the prepared HCCCP was added to increase calcium content of the prepared milk to be 200 and 300 mg/100 ml respectively. The additives were thoroughly mixed in milk samples using electric mixer and heat treatment (90^{o}C /5 min) was done before cooling milk to 42^{o}C to be ready for adding yoghurt starter culture. The resultant cheese was salted (1% w/w), packed in polyethylene pouches and kept in refrigerator. The yield of cheese was calculated from the following equation:

$$\text{The yield} = \frac{\text{Weight of cheese}}{\text{Weight of milk used}} \times 100$$

Analysis:

Total solids (TS), total nitrogen (TN), soluble nitrogen (SN), ash and titratable acidity were determined according to AOAC (2005). The pH value was measured using pocket pH meter (Scientific, USA, Model Ia_{125}). Calcium was determined by atomic absorption according to the method mentioned by Sparks (1998).

Cheese samples for texture profile analysis (TPA, Talogo, CNS-Farnell, England) were placed in plastic cups, sealed (to prevent dehydration) and tempered to 12±0.5^{o}C prior to analysis. The conditions of analysis were as follows: Trigger loat was 7 g, test speed and return speed were 1 mm / s, cycles were 2. Pretest speed was 2 mm/s, probe was TA_7 and fixture was Ta – RT – KI.

Hardness, adhesiveness, cohesiveness, springiness, gumminess and chewiness were evaluated in triplicate according to the defination given by IDF (1991).

The organoleptic properties included general appearance (10 points), body & texture (30 points) and flavour (60 points) were evaluated as given by El-Nawasany *et al.* (2015). The organoleptic properties were done by 10 panelists.

Statistical analysis including analysis of variance and Duncan's test as well as average and standard error (SE) were carried out using SPSS computer program (SPSS, 1999).

RESULTS AND DISCUSSION

Table (1) shows yield of fresh Karish cheese made from milk treated with yoghurt starter and calcium chloride or high calcium casein co-precipitates (HCCCP).

There were significant differences in cheese yield between different treatments (Table 1). The control cheese had the lowest significant yield (26.17%) than the yield of all other treatments. The results for treatments 1, 2, 3 and 4 were 33.14%, 31.26%, 37.67% and 33.15%, respectively. The use of calcium chloride or HCCCP at 300 mg Ca/100 ml cheese milk (T3 and T4) gave the highest yield, while decreasing the amount of Ca to 200 mg /100 ml of cheese milk decreased yield of the resultant cheese.

Making cheese by using calcium chloride improved the curd and increased the yield and reduced the costs (Ernstrom *et al.*, 1958). This agrees with our results. However,, the use of HCCCP in the present study significantly decreased the yield of Karish cheese (T2) as compared with the corresponding yield due to the use of calcium chloride (T1). The same trend was recorded at any amount of the both additives. In general, increased the amount of calcium in cheese milk by means of adding calcium chloride or HCCCP significantly increased the yield of Karish cheese comparing with cheese made from skim milk without any fortification.

Table 1. Yield (%) of fresh Karish cheese made from milk fortified with calcium from calcium chloride or high calcium casein co-precipitates (Average±SE of 3 replicates).

Item	Treatments**				
	C	T1	T2	T3	T4
Yield (%)	26.17±0.64^{d}	33.14±0.63^{b}	31.26±0.41^{c}	37.67±0.63^{a}	33.15±0.13^{b}

Averages with unlike superscripts are significantly different (P<0.05).

**** C, control cheese,**

T1 and T3, cheese made from milk supplemented with calcium chloride to increase it's calcium content to 200 and 300 mg/100 ml, respectively,

T2 and T4, cheese made from milk supplemented with HCCCP to increase it's calcium content to 200 and 300 mg/100 ml, respectively,

Table (2) shows the effect of calcium chloride and HCCCP on total solids, protein and SN/TN. The results showed that there were significant differences between all treatments. The values of cheese from all treatments were higher than the control. The use of calcium chloride gave higher values than the use of HCCCP. The addition of calcium is important in modification the salt distribution between aqueous and micellar phases. It was reported that addition of 10 mM calcium chloride to milk, about 80% of calcium was associated with casein micelles (Neville & Watters, 1983 and Gaucheron, 2005).

Table 2. Total solids (TS), protein % and SN/TN of fresh Karish cheese made from milk fortified with calcium from calcium chloride or high calcium casein co-precipitates (Average±SE of 3 replicates).

Item	Treatments				
	C	T1	T2	T3	T4
Total solids,%	30.59±0.05^{d}	35.28±0.08^{c}	34.62±0.44^{d}	37.56±0.43^{a}	36.81±0.20^{b}
Protein, %	10.03±0.86^{e}	14.72±0.71^{c}	14.02±0.86^{d}	16.15±0.84^{a}	15.43±0.18^{b}
SN/TN, %	15.73±0.66^{c}	15.91±0.12^{b}	15.87±0.70^{bc}	16.13±0.09^{a}	16.02±0.15^{ab}

*** See legend to Table (1) for details.**

Table (3) shows ash and calcium of cheese as affected by the applied treatments. The use of calcium chloride increased both of them more than the use of HCCCP while the control cheese had the lowest significant values being 1.34% for ash and 132 mg/100 g for calcium content. Treatment 3 and 4 recorded ash content of 2.49 and 2.45% while calcium contents were 248 and 240 g/100 g respectively. In literature, calcium chloride addition increased ionic calcium (Philippe *et al.,* 2003 and Boumpa *et al.,* 2008). However, richness of the prepared HCCCP with calcium (238 mg/100 g) and ash (4.68%) was responsible for the present results in cheese.

Table 3. Ash and calcium contents of fresh Karish cheese made from milk fortified with calcium from milk fortified with calcium from calcium chloride or high calcium casein co-precipitates (Average±SE of 3 replicates).

Item	Treatments				
	C	T1	T2	T3	T4
Ash, %	1.34±0.06^{c}	1.95±0.05^{b}	1.83±0.13^{b}	2.49±0.18^{a}	2.45±0.06^{a}
Ca, mg/100 g	132±0.90^{c}	188±1.1^{b}	175±0.94^{b}	248±0.83^{a}	240±0.91^{a}

*** See legend to Table (1) for details.**

Table (4) shows pH and acidity of Karish cheese as affected by the applied treatments. The results showed that pH value decreased and the acidity increased with calcium. Calcium chloride also decreased the pH rather than the HCCCP. Calcium chloride addition reduced pH (Philippe *et al.,* 2003 and Boumpa *et al.,* 2008). However, lowering the pH of milk by adding calcium chloride was demonstrated from long time ago by Ernstrom et al. (1958).

Table 4. The pH value and acidity of fresh Karish cheese made from milk fortified with calcium from milk fortified with calcium from calcium chloride or high calcium casein co-precipitates (Average±SE of 3 replicates).

Item	Treatments				
	C	T1	T2	T3	T4
pH value	4.80±0.12^{a}	4.69±0.23^{c}	4.73±0.11^{b}	4.50±0.23^{e}	4.58±0.05^{d}
acidity, %	0.79±0.15^{e}	0.88±0.24^{c}	0.86±0.21^{d}	1.05±0.32^{a}	0.98±0.05^{b}

*** See legend to Table (1) for details.**

Table (5) reveals TPA parameters of the tested cheese samples including the primary parameters (hardness, adhesiveness, cohesiveness and springiness) and texture secondary parameters (gumminess and chewiness). Hardness was the lowest in control cheese which was characterized by the highest moisture content (69.41%, Table 2). This agrees with Mehanna and Posztor-Huszar (2012). The highest significant hardness was observed in T3. On the other hand, the higher was calcium content in cheese, the higher were the values of hardness since hardness of cheese from T3 and T4 were higher than those from T1 and T2. Moreover, T1 and T3 gave corresponding higher hardness than T2 and T4 suggesting impact of HCCCP on decreasing hardness of Karish cheese. This is quite important for soft cheese in general.

Table 5. The rheological properties of fresh Karish cheese made from milk fortified with calcium from milk fortified with calcium from calcium chloride or high calcium casein co-precipitates (Average±SE of 3 replicates).

Item	Treatments				
	C	T1	T2	T3	T4
Hardness, (G)	223±0.03^{e}	283±0.08^{c}	256±0.11^{d}	488±0.08^{a}	398±0.05^{b}
Adhesiveness, G	62±0.08^{a}	40±0.08^{c}	48±0.14^{b}	10±0.13^{e}	24±0.13^{d}
Cohesiveness, ratio	0.43±0.06^{d}	0.47±0.05^{c}	0.47±0.11^{c}	0.63±0.08^{a}	0.53±0.11^{b}
Springiness, mm	10.09±0.03^{a}	9.93±0.08^{b}	9.98±0.12^{b}	9.45±0.03^{c}	9.93±0.09^{b}
Gumminess, G	106±0.24^{e}	213±0.18^{b}	121±0.09^{d}	270±0.13^{a}	135±0.15^{c}
Chewiness, G/mm	9.8±0.20^{d}	12.4±0.11^{c}	11.8±0.05^{c}	36.2±0.13^{a}	21.0±0.15^{b}

*** See legend to Table (1) for details.**

Adhesiveness and springiness were the maximum in control cheese, whereas cheese from T3 had the lowest significant values being 10.0 g and 9.45 mm respectively. Calcium chloride increased the values of gumminess and chewiness ($P \leq 0.05$) and the increase was proportional with the amount of calcium chloride added. Moreover, such addition gave higher significant values than those recorded with adding HCCCP. Cohesiveness had values of 0.43, 0.47, 0.47, 0.63 and 0.53 for control cheese and fresh cheese from T1, T2, T3 and T4 respectively.

The foregoing results suggest that fortification of cheese milk with calcium –in general– increased hardness, cohesiveness, gumminess and chewiness of the resultant fresh Karish cheese and decreased only adhesiveness and springiness. More hardness (T3 and T4) adversely affected the organopeltic properties of Karish cheese. Such correlation was previously given by Adhikari et al. (2003).

Table (6) reveals that control cheese (C) was ranked the lowest significant scores for the general appearance as compared to cheese made from milk containing 200 mg Ca/100 ml. Thus, T1 and T2 had the maximum scores, while adding more calcium to cheese milk caused adverse impact in this respect. T3 had 7.11 points out of 10 suggesting the undesirable impact of using more calcium chloride, while the use of HCCCP was much better for the fortification. This was also true for body and texture and flavour of the resultant cheese. Cheese from T2 had the highest significant scores for both body and texture as well as the flavour attributes.

Table 6. The organoleptic scoring of fresh Karish cheese made from milk fortified with calcium from milk fortified with calcium from calcium chloride or high calcium casein co-precipitates (Average±SE of 10 panelists).

Item	Treatments				
	C	T1	T2	T3	T4
General appearance (10 points)	8.45±0.32^{c}	8.91±0.38^{b}	9.54±0.45^{a}	7.11±0.42^{e}	8.31±0.31^{d}
Body & texture (30 points)	25.31±0.18^{c}	27.15±0.23^{b}	28.38±0.34^{a}	19.13±0.39^{e}	22.41±0.41^{d}
Flavour (60 points)	55.82±0.38^{b}	56.11±0.21^{b}	58.18±0.3^{a}	50.15±0.19^{d}	53.82±0.42^{c}

*** See legend to Table (1) for details**

Such cheese was characterized by pleasant appearance, soft body, smooth texture and accepted flavour, while fortification of cheese milk with more calcium chloride (T3) gave hard cheese with compact body and rough texture. More details about the undesirable impact of calcium chloride on sensory properties of some dairy products were given by Williams et al. (2005), while improving impact of caseinate on texture properties of yoghurt was mentioned by Mistry (2002). More recent, Akalin et al. (2012) recommended using of sodium calcium caseinate to improve sensorial properties of yoghurt.

In conclusion, the present work suggests performance of fortification of Karish cheese milk with calcium via high calcium casein co-precipitates rather than calcium chloride to make calcium fortified Karish cheese.

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تدعيم الجبن القريش بالكالسيوم بإضافة كلوريد الكالسيوم او الكازين ومرافقاته المترسبة عالية المحتوى من الكالسيوم

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الجبن القريش جبن طرى يصنع من اللبن الفرز بالتجبن الحامضى عن طريق اضافة بادىء اليوجورت مما يؤدى الى تحول جزء كبير من الكالسيوم الغروى الى الصورة الايونية (الذائبة) والتى تفقد بدورها فى الشرش . وتهدف هذه الدراسة الى تدعيم الجبن بالكالسيوم عن طريق زيادة محتواه فى لبن الجبن الى ٣٠٠/٢٠٠ مجم/١٠٠ مل عن طريق الاضافة المباشرة لكلوريد الكالسيوم (معاملة ١ ، ٣) او الكازين ومرافقاته المترسبة عالية الكالسيوم والمحتوية على الكازين وبروتينات السيرم والكالسيوم والتى تضمن بدورها اضافة بروتين بجانب الكالسيوم الى الجبن (معاملة ٢ ، ٤).اوضحت النتائج ان تصافى جبن المقارنة (دون اى اضافات) كان اقل (٢٦.١٧%) من تصافى جبن المعاملات حيث كان التصافى ٣٣.١٤ ، ٣١.٢٦ ، ٣٧.٦٧ ، ٣٣.١٥% للمعاملات ١ ، ٢ ، ٣ ، ٤ على الترتيب ، كما احتوى جبن المعاملات ٣ ، ٤ على جوامد كلية ، بروتين ، نتروجين ذائب / نتروجين كلى ، رماد ، كالسيوم بأرقام فاقت ارقام المعاملات ١ ، ٢ والتى بدورها كانت اعلى من ارقام جبن المقارنة ، نفس اتجاه النتائج كان مع الحموضة وعكس ذلك مع الرقم الهيدروجينى وكانت الفروق معنوية احصائيا. تأثرت الخواص الريولوجية معنويا بزيادة المحتوى من الكالسيوم وخاصة من المعاملات ٣ ، ٤ حيث زادت صلابة الجبن وقوة التماسك والصمغية والقابلية للمضغ فى حين انخفضت نتائج الالتصاق والمطاطية (الليونة) .. وقابل ذلك تأثيرا واضحا على الخواص الحسية للجبن الناتج حيث حازت جبن المعاملات ١ ، ٢ على اعلى درجات التحكيم للمظهر العام والقوام والتركيب والنكهة مقارنة بجن المقارنة او جبن باقى المعاملات.