Effects of Type of Starter Culture, Increase of Dry Matter and Microbial Transglutaminase on the Texture and Consumer Acceptability of Fermented Camel Milk

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ABSTRACT

Camel milk has been found to be difficult to process into different dairy products due to slower rate of acidification and other factors related to its composition. Hence, this study was aimed to evaluate effects of two different mesophilic starter cultures, addition of camel milk powder (CMP) and the use of microbial transglutaminase (MTGase) on the texture, viscosity, sensory and physicochemical properties of fermented camel milk. The two mesophilic starter cultures used were R-707 (Lactococcus lactis) and CHN-22 (contain multiple strains of Lactococcus lactis ssp. cremoris, Lactococcus lactis ssp. lactis, Lactococcus lactis ssp. lactis biovar diacetylactis, Leuconostoc mesenteroides and Leuconostoc pseudomesenteroides). All milk samples were heat treated (90 ^oC, 10 min), cooled to inoculation and incubation temperature (27 ^oC) of starter cultures and incubated until pH reaches 4.6. Use of a single strain starter culture (R-707) resulted in fermented milk which was significantly ($P<0.05$) higher in cohesiveness and adhesiveness than a multi-strain (CHN-22) starter culture. But addition of CMP significantly ($P<0.05$) decreased the firmness, cohesiveness and adhesiveness of the fermented camel milk. Use of MTGase has improved the textural attributes of fermented camel milk samples and the effect depended on the starter culture used. The cohesiveness of the fermented camel milk was significantly ($P<0.05$) higher when made using R-707 starter culture with MTGase compared to using CHN-22 starter culture. Fermented camel milk produced using CHN-22 starter culture with MTGase was significantly ($P<0.05$) lower in titratable acidity as compared to that produced using CHN-22 starter culture without MTGase. Therefore, R-707 starter culture was found to be more preferable to improve the textural attributes of fermented camel milk samples, and MTGase can be used by dairy industries and smallholder farmers to improve the textural attributes of fermented camel milk.

Keywords: Camel milk powder, Fermented camel milk, Mesophilic starter cultures, Microbial transglutaminase, Texture.

INTRODUCTION

Camel milk is technically more difficult to process into different products than milk from other domestic animals (Mehaia, 1994; Ibrahim, 2009; Konuspayeva *et al.*, 2014). For instance, Jumah *et al.* (2001) reported that the viscosity of camel milk yoghurt does not change during gelation, and Mohammed *et al.* (1990) observed that camel milk failed to form gel like structure after 18 hours incubation with lactic acid culture. This was attributed to the presence of antibacterial factors such as lysozymes, lactoferrin and immunoglobulin in camel milk (El Agamay *et al.*, 1992). However, a recent report by Tesfemariam Berhe *et al.* (2018) found that the slower speed of acidification in camel milk than bovine milk was due to difference in proteolysis rather than the presence of inhibitory substance in camel milk. The authors concluded that the proteolytic systems of the starter cultures used are unable to support a growth rate in camel milk as fast as in bovine milk. Farah *et al.* (1990) reported that the *Suusa* (traditional fermented camel milk) can be improved by using selective mesophilic lactic acid cultures.

It has been reported that the compositional properties of camel milk attributed to the product quality during processing (Tesfemariam Berhe *et al.,* 2017). The content of heat-stable serum proteins of camel milk which make up 20-25% of the total protein (Desouky *et al.,* 2013) and the weak interaction between denatured serum proteins and casein due to lack of β-lactoglobulin (Shabo *et al.,* 2005), the lower amount of κ-casein (Farah, 1993), the high whey protein to casein ratio (Shamsia, 2009) in camel milk can be attributed to the weak texture and thin consistency of camel milk yoghurt (Tesfemariam Berhe *et al.*, 2017). Compared to bovine milk, camel milk casein has larger micelle size (Bornaz *et al.,* 2009). It has been reported that smaller casein micelles have improved the gelation properties of bovine milk (Glantz *et al.,* 2010). The lower amount of κ-casein, the high ratio of whey protein to casein, and the larger micelle size in camel milk also result in formation of a less firm coagulum and lower yield during cheese processing (Tesfemariam Berhe *et al.*, 2017).

Enzymatic cross-linking of milk proteins is a method that has received increasing attention during the last two decades (Faergemand *et al.*, 1998; Motoki and Seguro, 1998). One of the cross-linking enzymes available for catalysing covalent bond formation between protein molecules on a commercial scale is microbial transglutaminase (MTGase) (Dickinson, 1997). MTGase is a transferase which catalyzes the acyl-transfer reaction between γ-carboxamide groups of peptide or protein bound glutamyl residues and primary amines (Dickinson and Yamamoto, 1996; Bonisch *et al.,* 2007). Cross-linking of milk proteins by MTGase modifies functionality such as hydration ability and rheological as well as emulsifying properties (Motoki and Seguro, 1998; Lorenzen, 2000). MTGase is effective in reducing syneresis in acid milk gels and has been reported as a method of improving the texture and shelf-life of yoghurt (Motoki and Seguro, 1998).

Some attempts have been made to improve the texture and sensory properties of fermented camel milk by increasing the total solids through addition of milk powder (Mortada and Omer, 2013). The commonly used dry dairy ingredients to increase the solids content of yoghurt mix are skim milk powder, whey protein concentrate and sodium caseinate. Milk supplements with milk proteins can affect the texture and the physical properties of the yoghurt (Ibrahim, 2015). For instance, the addition of skim milk powder assisted in increasing the viscosity and gel strength of yoghurt as compared to the unfortified yoghurt (Peng *et al.*, 2009).

The use of selected commercial mesophilic starter cultures for the fermentation of camel milk combined with addition of camel milk powder (CMP) and use of MTGase to improve the texture of fermented camel milk has not previously been investigated. Therefore, the objectives of the present study were to evaluate the effects of mesophilic starter cultures, MTGase and addition of CMP on the texture, viscosity, sensory and physicochemical properties of fermented camel milk.

MATERIALS AND METHODS

Materials

Pooled fresh camel milk used in this study was collected from Errer Valley, Babille district, Eastern Ethiopia. The milk samples were collected from about 10 lactating camels in the early morning. After collection, the milk samples were brought to the Dairy Technology Laboratory of Haramaya

University within two hours of milking. A total of about 20 litres of camel milk was collected on three different occasions.

The mesophilic starter cultures (CHN-22 and R-707) were obtained as freeze-dried multiple and pure cultures from Christian Hansen A/S (Hørsholm, Denmark A/S). CHN-22 contains multiple strains of *Lactococcus lactis* subsp. *cremoris, Lactococcus lactis* subsp. *lactis, Lactococcus lactis* subsp. *lactis* biovar *diacetylactis, Leuconostoc mesenteroides* and *Leuconostoc pseudomesenteroides*, whereas R-707 contains a single strain of *Lactococcus lactis* without biovar *diacetylactis*. The MTGase (ACTIVA® MP) was obtained from Ajinomoto Foods Europe S.A.S (Paris, France). The ingredients of the enzyme include Lactose, Maltodextrin and Transglutaminase. Camel milk powder (CMP) was obtained from *Camelicious* Company in Dubai. The compositions (100 g) of the CMP were 25 g fat, 40 g carbohydrates, 38 g lactose, 25 g protein, 1.6 g salt; different vitamins [vitamin A (87.2 µg), vitamin B1 (0.4 mg), vitamin B2 (0.3 mg), vitamin C (22.6 mg), vitamin D (0.7 µg) and vitamin E (100 µg)]; and 1100 mg calcium.

Inoculum Preparation

Inoculums were prepared according to Tesfemariam Berhe *et al.* (2018). A 50-unit sachet of culture was added in 500 ml autoclaved bovine milk. The cultures were distributed into 100 ml bottles, capped tightly and frozen at -20 °C. During fermented camel milk preparation, 1 ml of the thawed inoculums was added to 400 ml milk.

Treatments and Experimental Design

The experiment was designed as factorial experiment (2*2*2=8) with two starter cultures (R-707 and CHN-22), two levels of camel milk powder [with (5%) and without] and two levels of MTGase [with (0.2 g L^{-1}) and without]. Eight different fermented camel milk samples were prepared as shown in Table 1. The experiment was done in three replications.

Table 1. Fermented camel milk samples

Sample (S)	Starter cultures		CMP	MTGase
	R-707	CHN-22		
S1	+	-	-	-
S2	+	-	+	-
S3	+	-	-	+
S4	+	-	+	+
S5	-	+	-	-
S6	-	+	+	-
S7	-	+	-	+
S8	-	+	+	+

Note: + = added (with); - = not added (without); CHN-22 =starter culture containing mixed strains of *Lactococcus lactis* subsp. *cremoris, Lactococcus lactis* subsp. *lactis, Lactococcus lactis* subsp. *lactis* biovar *diacetylactis, Leuconostoc mesenteroides* and *Leuconostoc pseudomesenteroides*; R-707 = starter culture containing a single strain of *Lactococcus lactis* without biovar *diacetylactis*; CMP = camel milk powder; MTGase = microbial transglutaminase.

Fermented Camel Milk Production

The flow diagram of the fermented camel milk production is shown in Figure 1. The pooled fresh camel milk brought to Haramaya University Dairy Technology Laboratory was sieved using a muslin cloth and immediately divided into eight portions (each portion was used as individual fermented camel milk sample (S)). The experiment was done in triplicates with the milk collected on three different occasions according to the procedures outlined in Figure 1.

For texture profile analysis, 80 ml of inoculated milk samples was added to three 100 ml beakers, from each treatment. Then, all the milk samples were incubated in a thermostatically controlled water-bath (model WNB 45, D-91126, Memmert GmbH, Büchenbach, Germany) at 27 ^{o}C until the pH reaches 4.6. The acidification progress of the samples was checked by measuring the pH using a pH-meter during the incubation period. After 24 h storage time, the fermented milk samples were analysed for different parameters.

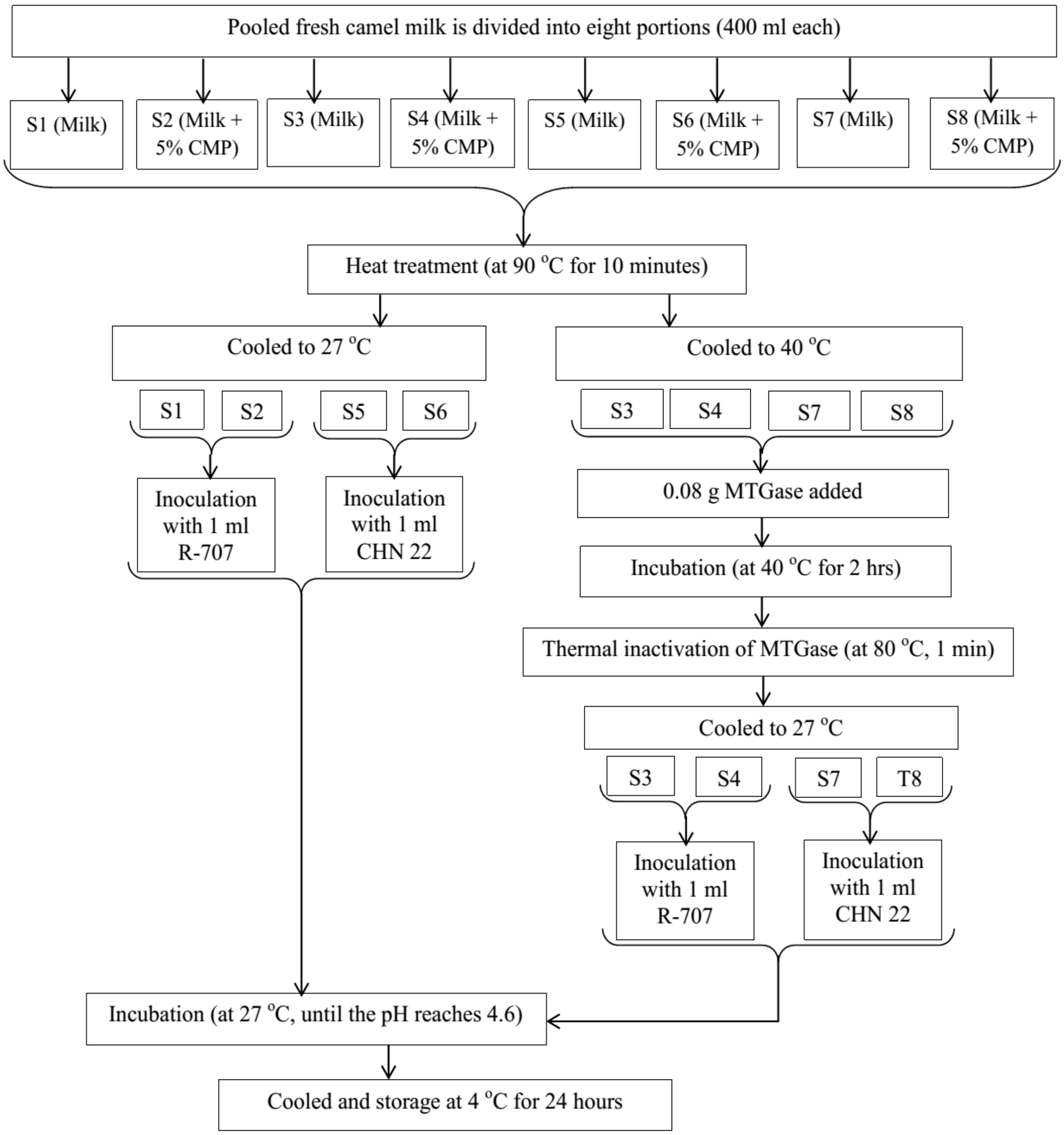

Figure 1. Flow diagram for fermented camel milk production

Physicochemical Analysis of Raw and Fermented Camel Milk

The pH of raw and different fermented camel milk samples was measured using a calibrated digital pH-meter. For determination of titratable acidity, 9 ml of raw or fermented camel milk sample was measured into a beaker and three drops of 0.1% phenolphthalein indicator was added into a sample and then titrated with 0.1N sodium hydroxide (NaOH) solution until faint pink colour persisted. The titratable acidity was expressed as percent lactic acid (Richardson, 1985). Thus, percent lactic acid was calculated as:

$$\% \text{ Lactic acid} = \left(\frac{(ml \text{ of } 0.1\text{N NaOH} \times 0.009 \times 100)}{ml \text{ of milk sample used}}\right)$$

Fat, protein, lactose, total solids (TS), and solids not fat (SNF) contents of raw and different fermented camel milk samples were determined using a MilkoScan FT1 (FOSS Analytical A/S, Hilleroed, Denmark). Eighty millilitres of the raw and fermented milk samples were used for the analysis of fat, protein, lactose, TS, and SNF. Just before analysis, the fermented milk samples in the beakers were thoroughly homogenized using an Ultra-Turrax T18 homogenizer (IKA-Labortechnik, Staufen, Germany). The ash content was determined according to AOAC (1995). The ash content of the raw and different fermented camel milk samples was determined gravimetrically by igniting in a muffle furnace (Fisher Scientific, Model 650-58, Canada). Five grams of the samples was measured into crucibles using sensitive balance and oven dried at 102 °C for 18 hrs. Then, the samples were transferred to the muffle furnace and ignited at a temperature of 550 °C for 3hrs. The samples were taken out of the muffle furnace and put in desiccators for 30 min and then measured on a sensitive balance. Finally, the percentage ash content was calculated as:

$$\text{Percentage Ash} = \left(\frac{\text{Residue weight}}{\text{Sample weight}}\right) * 100$$

The experiment was replicated three times and the measurements were done two times per replication for all physicochemical parameters.

Texture Profile Analysis

The texture profile of the different fermented camel milk samples were measured using a Texture Analyzer (TA.XT plus Stable Micro Systems, Godalming, Surrey, UK) fitted with 30 kg load cell. The 80 ml fermented camel milk samples prepared in 100 ml beakers of 45 mm diameter were individually fitted under the probe and the tests were carried out. After analysis, the following parameters were extracted from the force verses time curves: peak positive force, peak negative force, positive area and negative area as shown in Figure 2 were taken as measurement of firmness (g), elasticity (g), cohesiveness (g.sec) and adhesiveness (g.sec), respectively. The tests were done using 40 mm diameter back extrusion rig with the following settings: pre-test speed = 1 mm/sec; test speed = 2 mm/sec; post-test speed = 10 mm/sec; distance = 20 mm. The experiment was carried out three times and the analysis was done three times per replication. A typical graph for a measurement of fermented camel milk is shown in Figure 2.

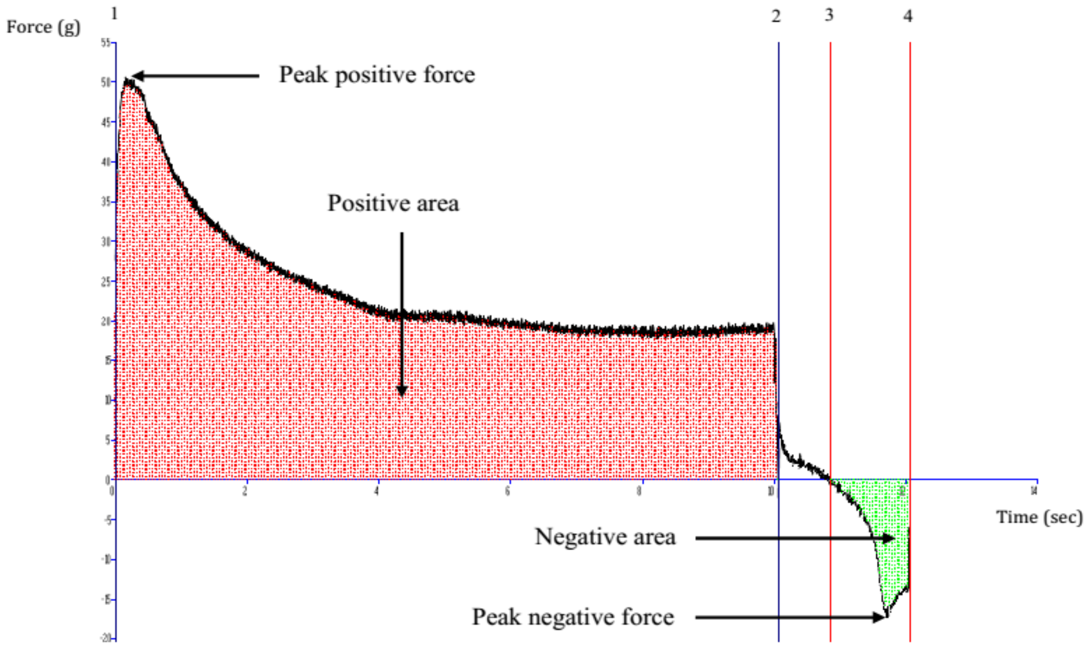

Figure 2. Graphic representation of measurements of textural attributes of fermented camel milk using texture analyser

Consumer Acceptability Test

For sensory analysis, 10 panellists were chosen to evaluate consumer acceptability of the different fermented camel milk samples. The panel members were selected based on the previous experience of evaluating sensory properties of different fermented dairy products and other processed foods. They evaluated all the fermented camel milk samples for sensory parameters such as color, appearance, aroma, taste, flavour, texture and overall acceptability using 7-point hedonic rating scale (7 = like very much; 6 = like moderately; 5 = like slightly; 4 = neither like nor dislike; 3 = dislike slightly; 2 = dislike moderately; 1 = dislike very much). About 40 ml of fermented camel milk samples were served in plastic cups. Pure bottled water was provided for panellists for cleansing palate between samples (Chen *et al.,* 1996).

Viscosity Analysis

Viscosity of fermented camel milk samples was measured using a post-humus funnel. The description of the post-humus funnel used is shown in Figure 3. Before the analysis, the fermented milk samples prepared in the bottles were thoroughly homogenized using an Ultra-Turrax T18 homogenizer (IKA-Labortechnik, Staufen, Germany). During viscosity measurement, the bottom outlet of the post-humus funnel was blocked by a finger and the homogenized fermented milk samples were poured into the post-humus funnel until it reaches the upper mark of the funnel. The bottom outlet was then opened and stop-watch started at the same time of opening the outlet. The time, in seconds, until the metal pin on the lower mark of the post-humus funnel visible was taken as viscosity values.

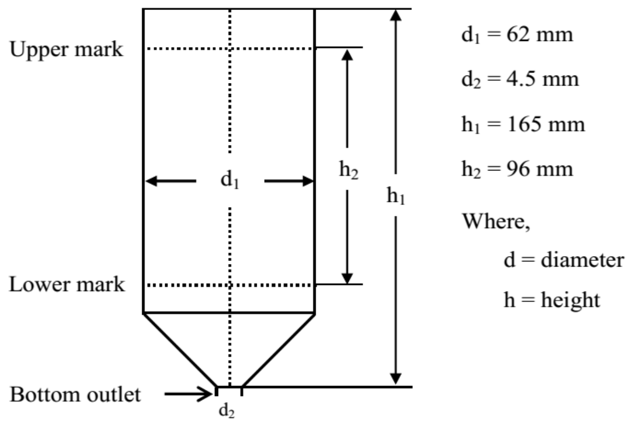

Figure 3. The diagram of a post-humus funnel used for measurements of viscosity of fermented camel milk

Statistical Analysis

The data were analysed with factorial analysis of variance (ANOVA) and the differences between means were assessed with Least Significant Difference (LSD) method. Statistical analysis was performed using SAS (2002) version 9.0. The level of significance for all analysis was done at *P<0.05.*

The statistical model was:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + \gamma_k + (\alpha\beta)_{ij} + (\alpha\gamma)_{ik} + (\beta\gamma)_{jk} + (\alpha\beta\gamma)_{ijk} + \varepsilon_{ijk}$$

Where,

Y_{ijk} = the response variable

μ = overall mean; α_i = effect of starter culture; β_j = effect of camel milk powder

γ_k = effect of MTGase; $(\alpha\beta)_{ij}$ = interaction effects of starter culture and camel milk powder; $(\alpha\gamma)_{ik}$ = interaction effects of starter culture and MTGase; $(\beta\gamma)_{jk}$ = interaction effects of camel milk powder and MTGase; $(\alpha\beta\gamma)_{ijk}$ = interaction effects of starter culture, camel milk powder and MTGase; and ε_{ijk} = random error

RESULTS

Physicochemical Properties of Raw and Fermented Camel Milk

There was no significant difference (*P>0.05*) in the chemical composition (fat, protein, lactose, total solids and ash) of fermented camel milk due to the different starter cultures used or by the addition of MTGase (Table 2). However, fermented camel milk produced by the addition of CMP was resulted in significantly (*P*<0.05) higher content of protein, lactose, TS, SNF, ash and titratable acidity (% lactic acid).

Table 2. Effects of starter cultures, CMP and MTGase on the physicochemical properties (%) of fermented camel milk (Mean ± SE)

		Starter culture		CMP		MTGase	
Parameters	Raw camel milk	R-707	CHN-22	With	Without	With	Without
Fat	3.46	3.79 ± 0.31	3.70 ± 0.33	4.19 ± 0.29	3.30 ± 0.29	3.75 ± 0.32	3.74 ± 0.32
Protein	2.71	3.11 ± 0.22	3.15 ± 0.22	3.58 ± 0.18[a]	2.68 ± 0.17[b]	3.14 ± 0.22	3.13 ± 0.22
Lactose	4.51	3.75 ± 0.18	3.55 ± 0.12	4.01 ± 0.12[a]	3.29 ± 0.11[b]	3.70 ± 0.16	3.60 ± 0.16
TS	11.54	12.60 ± 0.66	12.44 ± 0.69	14.16 ± 0.46[a]	10.88 ± 0.46[b]	12.55 ± 0.67	12.49 ± 0.68
SNF	7.87	8.78 ± 0.40	8.67 ± 0.40	9.92 ± 0.17[a]	7.53 ± 0.17[b]	8.74 ± 0.40	8.71 ± 0.39
Ash	0.83	0.97 ± 0.05	0.98 ± 0.05	1.13 ± 0.02[a]	0.81 ± 0.03[b]	0.97 ± 0.06	0.98 ± 0.05
TA	0.14	0.90 ± 0.04	0.89 ± 0.05	1.00 ± 0.027[a]	0.79 ± 0.03[b]	0.86 ± 0.04	0.93 ± 0.04
pH	6.55						

Means with different superscripts in the same row are significantly different at P < 0.05; CMP = camel milk powder; MTGase = microbial transglutaminase; TS = total solid; SNF = solid not fat; TA = titratable acidity.

The titratable acidity of the fermented camel milk was significantly (*P<0.05*) higher when MTGase was used with R-707 starter culture than when used with CHN-22 starter culture (Table 3). The use MTGase with CHN-22 starter culture significantly (*P<0.05*) lower the titratable acidity of the fermented camel milk than that of without MTGase for the same starter culture. Therefore, the use of MTGase with CHN-22 starter culture can be an alternative method for reducing excess acid production.

Table 3. The interaction effects of starter cultures and MTGase on the titratable acidity (%) of fermented camel milk (Mean ± SE)

Starter culture	MTGase	
	With	Without
R-707	0.91 ± 0.07^{a}	0.90 ± 0.05^{b}
CHN-22	0.82 ± 0.05^{bB}	0.97 ± 0.06^{aA}
$LSD_{(0.05)}$	0.0782	

MTGase = microbial transglutaminase; LSD = least significant difference; Means in the same row having different capital letter superscripts are significantly different at *P<0.05*; Means in the same column having different small letter superscripts are significantly different at *P< 0.05*.

Textural Properties of Fermented Camel Milk

Firmness

Addition of MTGase significantly increased gel hardness (Table 4). But addition of CMP, irrespective of the addition of MTGase, resulted in a significantly (*P<0.05*) lower firmness in the fermented camel milk (Table 4 and 5). In addition, when MTGase was applied, the resulting decrease upon addition of CMP was much more pronounced, illustrating that CMP, and hence extra camel milk protein, interfered with the positive effect of MTGase.

Table 4. The interaction effects of CMP and MTGase on the firmness (g) of fermented camel milk (Mean ± SE)

CMP	MTGase	
	With	Without
With	32.98 ± 4.46^{b}	21.54 ± 2.45^{b}
Without	86.77 ± 10.21^{aA}	36.29 ± 5.10^{aB}
$LSD_{(0.05)}$	14.202	

CMP = camel milk powder; MTGase = microbial transglutaminase; LSD = least significant difference; Means in the same row having different capital letter superscripts are significantly different at *P<0.05*; Means in the same column having different small letter superscripts are significantly different at *P<0.05*.

Table 5. Effects of starter cultures, CMP and MTGase on the textural attributes of fermented camel milk (Mean ± SE)

Parameters	Starter culture		CMP		MTGase	
	R-707	CHN-22	With	Without	With	Without
Firmness (g)	46.54 ± 7.81	42.25 ± 9.37	27.26 ± 2.98[b]	61.53 ± 9.35[a]	59.88 ± 9.69[a]	28.91 ± 3.50[b]
Elasticity (g)	-15.49 ± 0.52	-16.00 ± 0.75	-15.27 ± 0.65	-16.23 ± 0.61	-17.32 ± 0.33[a]	-14.17 ± 0.53[b]
Cohesiveness (g.sec)	207.63 ± 9.07[a]	136.21 ± 7.97[b]	154.54 ± 11.56[b]	189.31 ± 13.75[a]	178.46 ± 15.72	165.38 ± 11.08
Adhesiveness (g.sec)	-7.54 ± 0.60[a]	-3.68 ± 0.48[b]	-4.72 ± 0.69[b]	-6.51 ± 0.80[a]	-6.59 ± 0.83[a]	-4.64 ± 0.64[b]

Means with different superscripts in the same row are significantly different at *P<0.05*; CMP = camel milk powder; MTGase = microbial transglutaminase.

Elasticity

There was no significant difference (*P>0.05*) in elasticity between fermented camel milk prepared using R-707 and CHN-22 starter cultures (Table 5). The addition of CMP also did not significantly (*P>0.05*) affect the elasticity of the fermented camel milk. However, significantly (*P<0.05*) higher elasticity was observed for the fermented camel milk produced by the addition of MTGase (Table 5).

Cohesiveness

The cohesiveness of the fermented camel milk produced by R-707 starter culture was significantly higher than that produced by CHN-22 starter culture while the addition of CMP significantly reduced the cohesiveness of the fermented camel milk as compared to that prepared without CMP (Table 5). On the other hand, the interaction of starter culture and MTGase had a significant effect (*P<0.05*) on the cohesiveness of fermented camel milk (Table 6). The use of MTGase with R-707 starter culture significantly (*P<0.05*) improved the cohesiveness of the fermented camel milk as compared to the R-707 starter culture without MTGase (Table 6). Generally, the use of MTGase with R-707 starter culture has improved the cohesiveness of the fermented camel milk. However, MTGase did not improve the cohesiveness when it is used with CHN-22 starter culture.

Table 6. The interaction effects of starter cultures and MTGase on the cohesiveness (g.sec) of fermented camel milk (Mean ± SE)

	MTGase	
Starter culture	With	Without
R-707	224.08 ± 13.18^{aA}	191.18 ± 8.96^{aB}
CHN-22	132.84 ± 9.00^{b}	139.58 ± 13.92^{b}
$LSD_{(0.05)}$	19.547	

MTGase = microbial transglutaminase; LSD = least significant difference; Means in the same row having different capital letter superscripts are significantly different at *P<0.05*; Means in the same column having different small letter superscripts are significantly different at *P<0.05*.

Adhesiveness

Fermented camel milk prepared with R-707 starter culture showed significantly (*P<0.05*) higher adhesiveness than that prepared with CHN-22 starter culture (Table 5). The addition of CMP significantly decreased the adhesiveness of fermented camel milk. Significantly (*P<0.05*) higher adhesiveness was observed for the fermented camel milk samples treated with MTGase as compared to the samples without MTGase. In general, in the present study, the addition of MTGase improved the textural attributes of fermented camel milk.

Consumer Acceptability of Fermented Camel Milk

Fermented camel milk samples produced with the addition of CMP and R-707 starter culture had significantly (*P<0.05*) lowered likeability of aroma as compared to that produced from R-707 starter culture without CMP (Table 7). Moreover, fermented camel milk produced using R-707 starter culture without CMP had significantly (*P<0.05*) better aroma score than that of CHN-22 starter culture without CMP.

Table 7. The interaction effects of starter cultures and CMP on the aroma of fermented camel milk (Mean ± SE) (n = 10)

Starter culture	CMP	
	With	Without
R-707	5.40 ± 0.31^{B}	6.25 ± 0.31^{aA}
CHN-22	5.80 ± 0.30	5.25 ± 0.34^{b}
$LSD_{(0.05)}$	0.6374	

LSD = least significant difference; CMP = camel milk powder; Means in the same row having different capital letter superscripts are significantly different at *P<0.05*; Means in the same column having different small letter superscripts are significantly different at *P<0.05*.

Table 8. The interaction effects of starter cultures, CMP and MTGase on the sensory properties of fermented camel milk (Mean ± SE) (n = 10)

Parameters	Starter culture	CMP	MTGase		$LSD_{(0.05)}$	P-Value
			With	Without		
Color	R-707	With	6.5 ± 0.22	6.3 ± 0.21	0.4196	0.2389
		Without	6.5 ± 0.31	6.1 ± 0.28		
	CHN-22	With	6.4 ± 0.22	6.5 ± 0.17		
		Without	5.6 ± 0.56	6.5 ± 0.22		
Appearance	R-707	With	5.9 ± 0.35	6.1 ± 0.23	0.4116	0.1506
		Without	6.4 ± 0.22	6.1 ± 0.23		
	CHN-22	With	6.3 ± 0.15	6.3 ± 0.21		
		Without	5.5 ± 0.52	6.2 ± 0.25		
Aroma	R-707	With	5.1 ± 0.50	5.7 ± 0.37	0.6374	0.8762
		Without	5.9 ± 0.59	6.6 ± 0.16		
	CHN-22	With	5.8 ± 0.39	5.8 ± 0.49		
		Without	5.3 ± 0.47	5.2 ± 0.51		
Taste	R-707	With	5.1 ± 0.61	5.9 ± 0.43	0.6481	0.2853
		Without	5.8 ± 0.47	5.6 ± 0.22		
	CHN-22	With	5.5 ± 0.54	5.1 ± 0.50		
		Without	5.5 ± 0.45	5.5 ± 0.34		
Flavor	R-707	With	5.1 ± 0.62	5.4 ± 0.64	0.7166	0.3004
		Without	6.0 ± 0.26	5.7 ± 0.56		
	CHN-22	With	5.7 ± 0.47	5.0 ± 0.54		
		Without	5.3 ± 0.47	5.5 ± 0.40		
Texture	R-707	With	5.8 ± 0.25	6.0 ± 0.26	0.4869	0.4760
		Without	6.2 ± 0.25	5.7 ± 0.42		
	CHN-22	With	5.7 ± 0.50	5.8 ± 0.39		
		Without	6.0 ± 0.37	6.1 ± 0.23		
Overall acceptability	R-707	With	5.7 ± 0.50	5.9 ± 0.48	0.5562	0.3276
		Without	6.2 ± 0.33	6.3 ± 0.15		
	CHN-22	With	6.1 ± 0.41	5.5 ± 0.45		
		Without	5.4 ± 0.43	5.8 ± 0.29		

CMP = camel milk powder; MTGase = microbial transglutaminase; LSD = least significant difference.

The interaction of the three factors (starter cultures, CMP and MTGase) did not significantly (*P>0.05*) affect the sensory attributes such as color, appearance, aroma, taste, flavour, texture and overall acceptability of the fermented camel milk samples (Table 8). However, from the comments given by the panellists, none of the fermented camel milk samples was considered as unacceptable.

Viscosity of Fermented Camel Milk

Significant difference (*P<0.05*) was observed in viscosity between the fermented camel milk produced with starter cultures and CMP (Table 9). The significantly higher viscosity was recorded for fermented camel milk prepared using CHN-22 starter culture with CMP than R-707 starter culture with CMP. The viscosity was significantly (*P<0.05*) higher when CHN-22 starter culture interacts with CMP than when R-707 starter culture interacts with CMP. Moreover, the use of CMP with CHN-22 starter culture has significantly (*P<0.05*) improved the viscosity of the fermented camel milk than CHN-22 starter culture without CMP (Table 9).

Table 9. The interaction effects of starter cultures and CMP on the viscosity (seconds) of fermented camel milk (Mean ± SE)

	CMP	
Starter culture	With	Without
R-707	18.26 ± 0.14^{b}	18.02 ± 0.13
CHN-22	18.74 ± 0.17^{aA}	17.97 ± 0.06^{B}
$LSD_{(0.05)}$	0.245	

CMP = camel milk powder; LSD = least significant difference; Means in the same row having different capital letter superscripts are significantly different at *P<0.05*; Means in the same column having different small letter superscripts are significantly different at *P<0.05*.

The use of MTGase with CHN-22 starter culture had significantly (*P<0.05*) improved the viscosity of the fermented camel milk than R-707 starter culture with MTGase (Table 10). Fermented camel milk produced using CHN-22 starter culture with MTGase was significantly (*P<0.05*) higher in viscosity as compared to that of CHN-22 starter culture without MTGase (Table 10).

Table 10. The interaction effects of starter culture and MTGase on the viscosity (seconds) of fermented camel milk (Mean ± SE)

	MTGase	
Starter culture	With	Without
R-707	18.03 ± 0.13^{b}	18.24 ± 0.15
CHN-22	18.54 ± 0.24^{aA}	18.17 ± 0.14^{B}
$LSD_{(0.05)}$	0.245	

MTGase = microbial transglutaminase; LSD = least significant difference; Means in the same row having different capital letter superscripts are significantly different at *P<0.05*; Means in the same column having different small letter superscripts are significantly different at *P<0.05*.

There was significant difference (*P<0.05*) in viscosity between fermented camel milk samples produced by the addition of CMP and without CMP (Table 11). The viscosity of the fermented camel milk produced by the addition of CMP was significantly (*P<0.05*) higher than that of without CMP (Table 11).

Table 11. The main effects of starter cultures, CMP and MTGase on the viscosity (seconds) of fermented camel milk (Mean ± SE)

Starter culture	R-707	18.14 ± 0.10
	CHN-22	18.35 ± 0.15
CMP	With	18.50 ± 0.13[a]
	Without	17.99 ± 0.07[b]
MTGase	With	18.29 ± 0.15
	Without	18.20 ± 0.10

Means with different superscripts in the same column are significantly different at *P<0.05*; CMP = camel milk powder; MTGase = microbial transglutaminase.

DISCUSSIONS

Physicochemical Properties of Raw and Fermented Camel Milk

The average fat content of raw camel milk used in the present study is higher than 2.95% that was reported by Haddadin *et al.* (2008) and in agreement with that of El Zubeir *et al.* (2012) (3.5%). The average protein content of raw camel milk in the present study is higher than 2.54% that was reported by Khaskheli *et al.* (2005), and lower than that of Shamsia (2009) (3.46%) and El Zubeir *et al.* (2012) (3.7%). These variations could be attributed to various factors such as analytical measurement procedures, camel breed, stage of lactation, age, health status, parity, herd management practices, environmental conditions, geographical origin and seasonal variations (Al Haj and Al Kanhal, 2010; Khaskheli *et al.*, 2005; Konuspayeva *et al.*, 2009).

It was expected and also reported by Farnsworth *et al.* (2006), there was no significant difference in the chemical composition of fermented camel milk as a result of the different starter cultures used or by addition of MTGase. The higher titratable acidity of the fermented camel milk produced with CMP is perhaps due to the increased buffering capacity of the additional proteins, phosphates, citrates, lactates and other milk constituents (Walstra and Jenness, 1984). The titratable acidity of the fermented camel milk was significantly ($P < 0.05$) higher when MTGase was used with R-707 starter culture which might be attributed to the acid producing strain of homo-fermentative *Lactococcus lactis* (Walstra *et al.*, 2006) of R-707. Tesfemariam Berhe *et al.* (2018) found that R-707 starter culture acidified camel milk faster than CHN-22. Use of MTGase with CHN-22 starter culture significantly lowered titratable acidity of the fermented camel milk which might be due to the inter- or intra-molecular cross-linking of milk proteins by transglutaminase between a γ-carboxyamide group of glutamine residues and an ε-amino group of lysine residues which leads to the formation of an ε-(γ-glutamyl) lysine iso-peptide bond with generation of one molecule of ammonia per crosslink (Folk and Finlayson, 1977). On the contrary, Jooyandeh *et al.* (2015) reported that there was no significant difference in acidity between MTGase-treated yoghurts and control sample.

Therefore, the use of MTGase with CHN-22 starter culture can be an alternative method for reducing excess acid production. The cross-linking of low molecular weight peptides and amino acids required for the growth of starter bacteria was a possible reason of slow growth of starter bacteria and this causes slower acidity development in yoghurt products (Ozer *et al.*, 2007).

Textural Properties of Fermented Camel Milk

Firmness

Addition of MTGase has significantly increased gel hardness and similar results were found for camel milk yoghurt reported by Abou-Soliman *et al.* (2017). But addition of CMP resulted in a significantly lower firmness which is contra-intuitive as addition of skim milk powder to bovine milk is well known to increase hardness and enhance texture (Lucey, 2002); and addition of bovine skim milk powder has indeed been shown to have this effect in camel milk yoghurt (Abou-Soliman *et al.*, 2017). However, Attia *et al.* (2000) studied the glucono delta-lactone induced acidification of dromedary milk and they found that the casein micelles show a very marked initial drop in hydration (to approx. 50% of the initial value, compared to a drop of only 10-20% for bovine milk). These authors also noted that the solvation of minerals proceeded somewhat differently compared to bovine milk and found initial higher amounts of soluble calcium at neutral pH (~15%) compared to bovine (~7%). The demineralization of micelles in dromedary milk started at around pH 5.8 whereas in bovine milk demineralization this initiated at the onset of acidification, and it exhibits a more pronounced, sharper drop. Formation of hydrogen as well as electrostatic bonds, which are important in providing structure to acid coagula from bovine milk (Lucey, 2002) could be restricted in dromedary milk, resulting in the more fragile curd observed which appears to be formed from disassociated casein micelles (Attia *et al.*, 2000). Addition of CMP could possibly further aggravate this phenomenon by supplying additional soluble calcium and increased casein concentration, resulting in casein aggregates less prone to interact with each other.

In addition, when MTGase was applied, the resulting decrease upon addition of CMP was much more pronounced, illustrating that CMP, and hence extra camel milk protein, interfered with the positive effect of MTGase. This is also in stark contrast to the results of Abou-Soliman *et al.* (2017) who found that addition of bovine skim milk powder together with MTGase treatment markedly improved the texture of camel milk yoghurt. Our observed result could possibly be due to the MTGase enzyme preferentially acting within the micelle, binding casein molecules together and consequently changing the internal structure of the micelle, instead of binding micelles together and form aggregates (Mounsey *et al.*, 2005).

Elasticity

Elasticity (springiness) is a measure of ability of food to return to its original form after being compressed (Prakasan *et al.*, 2015). In the present study higher elasticity was observed for the fermented camel milk produced by the addition of MTGase, and Dinkcei (2012) reported that the addition of 1.85 U MTGase g^{-1} of protein significantly increased the cohesiveness of strained yogurt as compared to control sample. On the contrary, Prakasan *et al.* (2015) reported that there was no significant change observed in elasticity characteristic of MTGase treated *paneer*.

Cohesiveness

Cohesiveness indicates the strength of internal bonds making up the body of food and the degree to which a food can be deformed before it breaks (Radocaj, 2011). In the present study the cohesiveness of the fermented camel milk produced using R-707 starter culture with MTGase was significantly higher while the addition of CMP significantly reduced its cohesiveness. This is in line with the report by Iličić *et al.* (2013) who elucidated the cohesiveness of fermented milk samples were improved by the addition of MTGase as compared to control sample.

Adhesiveness

Fermented camel milk prepared with R-707 starter culture and MTGase showed significantly higher adhesiveness and the increased values in adhesiveness for camel milk yogurt treated with MTGase. In general, in the present study, the addition of MTGase improved the textural attributes of fermented camel milk. This could be attributed to the strengthening of the network structure by the enzyme as a result of forming inter- and intra-molecular isopeptide bonds in and between all types of milk proteins (Romeih *et al.,* 2014). It was unexpected that the addition of CMP did not improve the textural attributes of the fermented camel milk samples. Jooyandeh *et al.* (2015) concluded that the cross-linking of milk proteins by means of MTGase seems to be an acceptable alternative instead of addition of extra protein or stabilizer in yogurt production.

Consumer Acceptability of Fermented Camel Milk

The interaction of the three factors (starter cultures, CMP and MTGase) did not significantly affect the sensory attributes. However, from the comments given by the panellists, none of the fermented camel milk samples was considered as unacceptable. Farah *et al.* (1990) reported that fermented camel milk samples made with mesophilic lactic cultures was clearly preferred by panellists. The treatment with MTGase did not have a negative effect on aroma and flavour (Şanli, 2015). Similarly, Prakasan *et al.* (2015) suggested that MTGase treatment did not lead to any objectionable change in odor and appearance of the products, which could lead to rejection of products by the consumer.

Viscosity of Fermented Camel Milk

The use of MTGase with CHN-22 starter culture has significantly improved the viscosity of the fermented camel milk which might be attributed to MTGase cross-linking reaction that improved the viscosity of skimmed milk yoghurt (Aprodu *et al.,* 2012). Significantly higher viscosity values were also obtained for MTGase-treated yoghurt (Ozer *et al.,* 2007). The production of polysaccharides by lactic acid bacteria can greatly enhance the viscosity of fermented dairy products (Hati *et al.,* 2013).

CONCLUSION

We found that R-707 starter culture was better than CHN-22 starter culture in improving the texture of fermented camel milk particularly the cohesiveness and adhesiveness. Addition of R-707 starter culture with MTGase has improved the cohesiveness of fermented camel milk. The addition of CMP had a negative effect on the firmness, cohesiveness, adhesiveness and/or it decreased the textural attributes. The use of MTGase has improved the textural attributes of the fermented camel milk. Therefore, MTGase can be used by dairy industries and smallholder farmers to improve the textural attributes of fermented camel milk.

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