J. King Saud Univ., Vol. **8**, *Agric. Sci.* (1), pp. 107-117, Riyadh (A.H. 1416 / 1996).

Heat Coagulation of Camel Milk

Abdulrahman A. Al-Saleh
Food Science Dept., College of Agriculture, King Saud University, Riyadh, Saudi Arabia.

(Received 12/9/1414; accepted for publication 18/1/1415)

Abstract. Heat denaturation of whey proteins and the heat coagulation time (HCT) of camel and cow milks were investigated. The milk was heated to 63, 80, 90 and 120°C for 30 min. and nitrogen distribution was determined. The non casein nitrogen (NCN) in raw milks of camel and cow was 28 and 26 mg/100g, respectively. Whey protein nitrogen (WPN) was decreased significantly and after 30 min. of heating at 90°C the whey protein denaturation of camel milk was nearly half that of cow milk. Compared to cow milk, camel milk whey proteins had higher heat stability. Camel milk exhibit poor HCT and modification of the protein level and salt composition of the milk similar to that of cow milk did not improve the HCT. Suspension of camel milk micelles in cow ultracentrifugal whey (UCW) gave a milk unstable to heat. The system consisting of serum protein free casein (SPFC) of both camel and cow milks in their own milk diffusate, SPFC of camel milk in cow milk diffusate and SPFC of cow milk in camel milk diffusate, all became very unstable to heat at all pH values.

Introduction

The ability of milk to withstand relatively high processing temperature is very important from a technological viewpoint. The heat coagulation of cow milk has been the subject of considerable research since the pioneering work of Sommer and Hart [1]. The literature has been reviewed by Fox and Morrissey [2] and Fox [3, P, 189]. Camel milk is an important component of the human diet in many parts of the world, it contains all essential nutrients [4]. Camel milk has been shown [5] to have different composition from cow milk. In Saudi Arabia the population of camels is more than 0.6 million [6] and their milk is consumed fresh or in the form of fermented milk. Pasteurized camel milk has introduced to the local market on a very limited scale.

The heat coagulation of camel milk has received very little attention and heat treatments such as sterilization as means of preserving camel milk is unknown.

Changing in heat stability induced by addition of permitted stabilizer have been investigated for camel milk (Al-Saleh, unpublished data). It was found that addition of stabilizers was ineffective in improving the heat stability of camel milk. For this reason a study has been carried out for manipulating the heat stability of camel milk by modifying composition or interchanging constituents of camel and cow milk.

Materials and Methods

Milk samples

Camel and cow milk from animals in the farm of College of Agriculture, King Saud University were used. The milks were defatted by centrifugation at 2500g for 30 min at 4°C (Hermlez 320K, Germany). The skim milk so obtained was used fresh or after storage at 4°C.

Nitrogen distribution

The nitrogen distribution in the milk samples was determined by micro-kjeldahl method [7]. The following N-fractions were determined: total protein nitrogen (TN), non casein nitrogen (NCN) and non protein nitrogen (NPN soluble in 12% trichloracetic acid). Denaturation of whey protein nitrogen (WPN) was calculated by the difference of the whey protein nitrogen (NCN minus NPN) before and after heating of the milk. One portion was kept raw (control) and the rest was heated at 63, 80, 90 and 120°C for 30 min.

Camel milk with modified protein and salt composition

Cow milk (Protein 3.53%) was diluted with water so that the diluted milk had the same protein level (2.79%) as camel milk used in the study. The camel milk was dialyzed against 30 vol. of cow milk for 3 days at 4°C to adjust its salt composition similar to cow milk.

Integrated systems of casein micelles and ultracentrifugal wheys

Camel and cow milks were separately centrifuged at 105,000g for 30 min at 4°C (Beckman L 8-80 Ultracentrifuge, USA). The camel milk casein micelles were resuspended in cow milk ultracentrifugal whey and vice versa by stirring for 20h at 4°C.

Milk diffusate

Distilled water was dialyzed against 30 vol. of cow and camel milk for 3 days at 4°C (three replacement of the milk were carried out using No.1 Spectra/por membrance, MWCO, 6000 - 8000, (Fisher Scientific Co., USA) as given by Fox and Hoynes [8].

Preparation of serum protein-free systems

Serum protein-free systems were prepared by the method of Fox and Hoynes [8] with some modifications. Cow and camel milk casein micelles were prepared by centrifugation of milk samples at 105,000g for 30 min at 4°C. The supernatants were poured off and the pellets resuspended in milk diffusate by stirring for 20h at 4°C.

Determination of heat stability

The pH of samples was adjusted to various values in the range 6.2 - 7.2 at 4°C using 2N-HCl or 2N-NaOH and held for 1h before assay. Heat coagulation time (HCT) was determined at stated temperatures in a thermostatically controlled oil-bath according to the method of Davis and White [9].

Results and Discussion

Effect of heat treatment on nitrogen distribution in camel and cow milk

The denaturation of whey proteins is important in understanding the changes in the properties of milk that occur with heat treatment.

The effect of heat treatment on the distribution of nitrogen fractions in samples heated at 63°, 80°, 90°C and 120°C for 30 min. is shown in Table 1.

The value of NCN (expressed as percentage of total milk nitrogen) consists of whey proteins and NPN. The NCN in raw milk samples of camel and cow milk was 28% and 26%, respectively and showing no significant differences. As the temperature increased up to 90°C the NCN was decreased in both milks with the amount of 19% for camel milk which was lower than that of cow milk, 10%. The NPN was not affected by heat treatment for both milks. WPN was decreased significantly with heating for both types of milks and the content of WPC cow milk was 19 mg/100g which was significantly lower than that of camel milk, 51 mg/100g.

Denaturation of whey proteins was expressed as percentage relative to the control raw milk. It was apparent that cow milk showed higher whey protein denaturation. At low temperature (63°C) which is normally used for pasteurization, there was little whey protein denaturation. At higher temperature (90°C) the denaturation was 82 mg and 44 mg/100g for cow and camel milk, respectively. Moderate heat treatment (60-70°C range) generally resulted in structural unfolding of the proteins [10]. At higher temperatures, protein aggregation occurred [11]. Pasteurization processes cause very small amounts of denaturation, while sterilization causes almost complete denaturation [12, p. 69]. The present observation suggests that camel milk whey pro-

Table 1. Effect of heat treatment on the distribution of N-fraction in camel and cow milk

Temp. (°C) for 30 min	TN		NCN				NPN				WPN		Percentage denatured WP	
	mg/100g		mg/100g		% of TN		mg/100g		% of TN		mg/100g		mg/100g	
	Cow	Camel	Cow	Camel	Cow	Camel	Cow	Camel	Cow	Camel	Cow	Camel	Cow	Camel
raw	534	431	139	121	26	28	35	31	6.5	7.0	104	90	-	-
63	"	"	133	112	25	26	35	30	6.5	6.9	98	82	6	9
80	"	"	58	90	11	21	35	31	6.5	7.0	23	59	78	34
90	"	"	54	82	10	19	35	31	6.5	7.0	19	51	82	44
120	"	"	52	N.D.	10	N.D.	36	N.D.	6.7	N.D.	16	N.D.	85	N.D.

teins had higher heat stability and whey protein denaturation of camel milk was nearly half that of cow milk whey proteins.

Heat coagulation time (HCT) of camel and cow milk

The heat stability of milk can be defined either in terms of the time required to induce coagulation at a given temperature or the temperature required to induce coagulation in a given time. For convenience, a fixed temperature (120°C) was chosen.

Figure 1 shows the heat coagulation time (HCT)/pH curve of camel and cow milk. It was quite clear that camel milk was less heat stable than cow milk. HCT of camel milk became and remained low with increasing pH up to pH 7.0 and the HCT increased gradually with no maximum and minimum. Unlike camel milk HCT of cow milk increased progressively as the pH was increased from 6.2 to 7.2. The shape of the curve for camel milk at high temperature (120°C) was the same as those at low temperature (100°C). Cow milk showed pronounced maximum in the HCT at pH 6.6 and minimum at pH 6.8 and this was in agreement to previous finding [13; 14]. Many investigators had confirmed the important relation between the pH and heat stability. Fox and Hearn [15] concluded that the surface charge is important in determining the stability of milk and pH change could result in low stability. Singh and Fox [16] reported that stability increased at higher pH values ($\geq$ 7.1) owing to increased negative charge on the K-casein micelles, thereby reducing the maximum and minimum in the heat coagulation time.

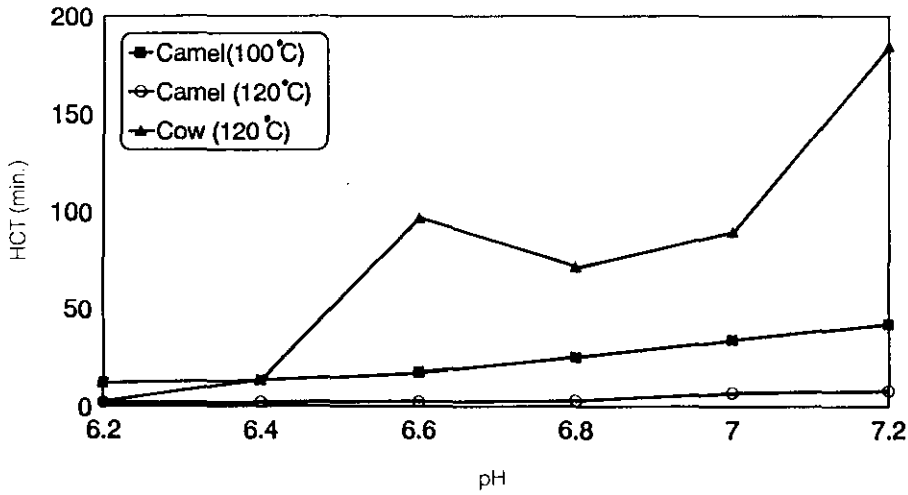

Fig. 1. Heat coagulation time (HCT)/pH curve of camel and cow milk .

Heat induced interaction between β-lactoglobulin (B-lg) and K-casein [2]. Rose [17] considered that β-lg/K-casein interaction was responsible for the maximum in the HCT/pH curve and failure of this interaction to occur was responsible for the minimum in HCT. According to Farah and Riesen [18] Larsson-Raznikiewicz and Mohamad [19] camel milk contained so little protein fraction corresponding to K-casein. This might be responsible for the low heat stability of camel milk.

HCT of camel milk with modified protein and salt composition

Dialysis of camel milk against diluted skim cow milk for 3 days to adjust its salt composition similar to cow milk had no effect on HCT of camel milk (Fig. 2). However, the heat stability of the dialyzed cow milk was decreased, but the shape of HCT/pH curve was not altered. The ash content of camel milk was 0.88% before dialysis and 0.80 after dialysis. It was found that the ash content of cow milk, before and after 3 days of dialysis, was 0.78% and 0.87% respectively. Additionally, the protein content for both camel and cow milks was not altered. The results showed that partial removal of ions from camel to cow milk caused a decrease in HCT of cow milk and no effects on the overall shape of HCT/pH curve. Sweetsur and Muir [20] found that the heat stability could be altered markedly by including small changes in the levels of soluble calcium or phosphate ions. Kudo [21] reported that variation in the amounts of calcium phosphate deposited on casein micelles could be one of the factors which might affect heat stability.

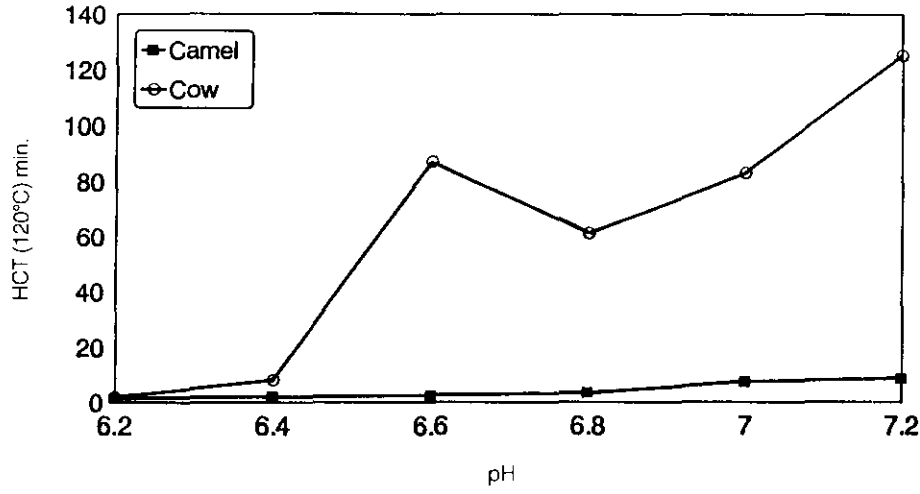

Fig. 2. Heat coagulation time (HCT)/pH curve of camel milk with modified protein and salt composition.

HCT of interchanged system between casein micelles and ultracentrifugal whey of camel and cow milk

The influence of interchanging casein micelles and ultracentrifugal wheys (UCW) from camel and cow milk was studied to know which constituents mainly affected the heat stability of camel milk. Table 2 shows that suspending camel casein in cow UCW gave a milk with low heat stability at all pH values. On the other hand, the system consisting of cow casein micelles in camel UCW was heat stable at pH $\geq$ 6.6. The HCT/pH profile was not altered, but the HCT was less than that of cow skim milk, which could be ascribed to the serum compositional differences (Table 1 and Fig.1)

Table 2. Effect of interchanging of casein micelles in ultracentrifugal wheys from camel and cow milk on heat coagulation time (HCT)

pH	Camel casein*		Cow casein**	
	2.0%	2.6%	2.0%	2.6%
	min		min	
6.2	2.00	1.50	2.05	2.12
6.4	2.10	1.35	2.00	2.20
6.6	2.15	2.05	83.20	84.50
6.8	22.45	2.30	63.20	66.10
7.0	5.10	4.19	75.10	79.15
7.2	5.30	4.45	180	182

HCT at 120°C

*Ultracentrifugal camel casein was resuspended in cow ultracentrifugal whey.

**Ultracentrifugal cow casein was resuspended in camel ultracentrifugal whey.

This suggests that the principal component which caused the low heat stability of camel milk might be its casein micelle. Heat induced changes in milk casein preceding coagulation when subjected to severe heat treatment and some type of interaction occurred between B-lactoglobulin and casein on heating which markedly affects heat stability [22]. According to Farah [23] K-casein and β-lactoglobulin play an important role in the stability of bovine milk and the absence and deficiency of these two proteins in camel milk might be a cause of its poor stability at high temperature. Mehriz and Ganguli [24] found that interchanging buffalo and cow casein micelle subunits (CSU) between their UCW resulted in insignificant increase in the heat stability of buffalo CSU suspended in cow UCW, while that of cow CSU in buffalo UCW predominately increased.

Effect of adding ultracentrifugal cow casein on HCT of camel milk

The casein content of camel milk (2.0%) was adjusted above the original level by adding bovine micellar casein (0, 0.2, 0.4 and 0.6%). The HCT of camel milk was not increased and HCT obtained with the casein concentrated samples was similar to those obtained without addition of casein. The poor heat stability of camel milk was very much dependent on the compositional differences between camel and cow milk which was mentioned in previous reports [18; 19; 25].

HCT of casein systems with interchanged casein micelles and milk diffusates

The data in Table 3 shows the HCT of camel and cow serum protein free casein micelles (SPFCM) in interchanged milk diffusate at different pH values. The SPFCM of both camel and cow was unstable to heat at all pH values over the range 6.2 - 7.2. As shown in Table 3 when sedimented camel or cow milk was suspended in their own milk diffusate the HCT was decreased markedly. The decrease in HCT of all milk samples might be attributed to the absence of serum proteins (β-lactoglobulin) and the state of salt balance. The failure of the β-lg/K-casein interaction to occur and salt balance shifts could be responsible to the minimum HCT. The whey proteins concentrations and the relative concentration of ions (salt-balance) are likely to be responsible for this phenomenon, since by dialysis only soluble constituents were interchanged.

Table 3. Heat coagulation time (HCT) of camel and cow serum protein free casein micelles (SPFC) in interchanged diffusate

pH	A	B	C	D
6.2	1.12	1.08	1.04	1.08
6.4	1.34	1.38	1.12	1.45
6.6	1.33	2.15	1.16	1.46
6.8	1.25	2.09	1.17	1.51
7.0	1.14	2.17	1.13	1.31
7.2	1.25	2.13	1.21	1.27

where:
HCT : in minutes (120°C)
A : Camel casein in camel milk diffusate
B : Camel casein in cow milk diffusate
C : Bovine casein in cow milk diffusate
D : Bovine casein in camlel milk diffusate

According to Fox [3, p. 195] factors in the serum rather than in the colloidal phase were primarily responsible for variations in heat stability. Ganguli [26] found that bovine casein micelles were less heat stable in their milk diffusate. On the other hand, Rose [17] observed increase in heat stability of micelles resuspended in their milk diffusate with increasing pH values. Pouliot and Boulet [14] reported that the use of higher temperature had undoubtedly accelerated some physiochemical changes such as salt balance shifts.

In general, camel milk exhibited poor heat stability and differs from that of cow milk. Interchanging casein micelles, ultracentrifugal whey and milk diffusate of camel and cow milk did not improve the heat stability of camel milk. Therefore, more research is needed to study casein submicelles and whey proteins of camel milk with regard to their presence and effect on heat stability.

References

[1] Sommr, H. and Hart, E. "The Heat Coagulation of Milk". *J. Biol. Chem.*, 40, (1919), 137-151.

[2] Fox, P. and Morrissey, P. "Reviews of the Progress of Dairy Science: The Heat Stability of Milk." *J. Dairy Res.* 44, (1977), 627-646.

[3] Fox, P. "Heat-Induced Coagulation of Milk." In: *Development in Dairy Chemistry-1-Proteins.* P. Fox, (Ed.) London: Applied Science Publishers, 1982.

[4] Yagil, R. *Camels and Camel Milk.* Rome: FAO, Animal Production and Health. Paper No.26, 1982.

[5] Abu-Lehia, I.H. "Composition of Camel Milk". *Milchwissenschaft.*, 42, (1987), 368-371.

[6] Chapman, M.J. "Bactrians and Dromedaries." *Asian Affairs,* 18, (1987), 276.

[7] AOAC. *Official Methods of Analysis.* 15th ed. Washington, D.C.: AOAC, 1990.

[8] Fox, P. and Hoynes, M. "Heat Stability Characteristics of Ovine, Caprine and Equine Milks." *J. Dairy Res.*, 43, (1976), 433-442.

[9] Davis, D. and White, J. "The Stability of Milk Protein to Heat. 1. Subjective Measurement of Heat Stability of Milk." *J. Dairy Res.*, 33, (1966), 67-81.

[10] Schmidt, R.H.; Packard, V.S., and Morris, H.A. "Effect of Processing on Whey Protein Functionality." *J. Dairy Sci.*, 67, (1984), 2723-2733.

[11] Wit, J.N. "Structure and Functional Behavior of Whey Proteins." *Neth. Milk Dairy J.* 35, (1981), 47.

[12] Burton, H. *Ultra-High Temperature Processing of Milk and Milk Products.* London: Elsevier Applied Science Publishers., 1988.

[13] Rose, D. "Factors Affecting the pH-Sensitivity of the Heat Stability of Milk from Individual Cows." *J. Dairy Sci.*, 44, (1961), 1405.

[14] Pouliot, Y. and Boulet, M. "Seasonal Variations in the Heat Stability of Concentrated Milk: Effect of Added Phosphate and pH Adjustment." *J. Dairy Sci.*, 74, (1991), 1157-1162.

[15] Fox, P.F. and Hearn, C.M. "Heat Stability of Milk: Influence of K-casein Hydrolysis." *J. Dairy Res.*, 45, (1978), 173.

[16] Singh, H. and Fox, P.F. "Heat Stability of Milk: pH-Dependent Dissociation of Micellar K-casein on Heating Milk at Ultra-High Temperatures." *J. Dairy Res.*, 52, (1985), 529.

[17] Rose, D. "Heat Stability of Bovine Milk." *A Review. Dairy Sci. Abst.*, 25, (1963), 45-52.

[18] Farah, Z. and Farah-Riesen, M. "Separation and Characterization of Major Components of Camel Milk Casein." *Milchwissenschaft,* 40, (1985), 669-671.

[19] Larsson-Raznikiewicz, M. and Mohamed, M.A. "Analysis of the Casein Content in Camel (*Camelus dromedarius*) Milk." *Swedish J. of Agricultural Research,* 16, (1986), 45-52.

[20] Sweetsur, A.W. and Muir, D.D. "Natural Variation in the Heat Stability of Concentrated Milk Before and after Homogenization." *J. Soc. of Dairy Technol.*, 35, (1982), 120.

[21] Kudo, S. "The Heat Stability of Milk: Formation of Soluble Proteins and Protein-Depleted Micelles at Elevated Temperatures." *Newzeland J. of Dairy Sci. and Tech.*, 15, (1980), 255.

[22] Fox, P.F. "Heat Induced Changes in Milk Preceding Coagulation." *J. Dairy Sci.*, 64, (1981) 2127-2137.

[23] Farah, Z. "Heat Coagulation of Camel Milk." *Dairy Res.*, 59, (1992), 229-331.

[24] Mehriz, A.M. and Ganguli, N.C. "Heat Stability of Casein Micelles from Buffalo Milk." *Milchwissenschaft.*, 39, (1980), 547-578.

[25] Farah, Z. "Composition and Characteristics of Camel Milk." *J. Dairy Res.*, 60, (1993), 603-626.

[26] Ganguli, N.C. "Stability of Buffalo Casein Micelles." *J. Dairy Res.*, 46, (1979), 401-405.

الثبات الحراري لحليب الإبل

عبدالرحمن عبدالله الصالح

قسم علوم الأغذية، كلية الزراعة، جامعة الملك سعود، الرياض، المملكة العربية السعودية.

(قُدم للنشر في ١٤١٤/٩/١٢هـ؛ وقبل للنشر في ١٤١٥/١/١٨هـ)

ملخص البحث. أجريت دراسة للتغيرات في طبيعة بروتينات الشرش (الدنترة) وكذلك الثبات الحراري لحليب الإبل مقارنة بحليب الأبقار. بعد رفع درجة الحرارة إلى ٦٣، ٨٠، ٩٠، ١٢٠°م لمدة ٣٠ دقيقة تم حساب توزيع النيتروجين في الحليب. وقد أوضحت الدراسة أن النيتروجين غير الكازيني في الحليب الخام للإبل والأبقار حوالي ٢٨، ٢٦ مجم/١٠٠ جم على التوالي. ولقد لوحظ أن النيتروجين في بروتينات الشرش يقل بارتفاع درجة الحرارة، كما أن التغيرات في طبيعة بروتينات الشرش (الدنترة) في حليب الإبل كانت حوالي نصف تلك المقدرة في حليب الأبقار مما يدل على أن الثبات الحراري لبروتينات الشرش في حليب الإبل أعلى من تلك في حليب الأبقار.

وبمقارنة النتائج وجد أن الثبات الحراري لحليب الإبل أقل بكثير من ذلك لحليب الأبقار كما أن تغيير مستوى البروتينات والأملاح في حليب الإبل إلى مستوى مشابه لذلك الموجود في حليب الأبقار لم يحسن من درجة الثبات الحراري لحليب الإبل. وجد أيضًا أن إضافة جسيمات كازين حليب الإبل إلى شرش الحليب البقري أدى إلى انخفاض الثبات الحراري وأوضحت النتائج أيضًا أن إضافة جسيمات كازين الحليب إلى ناضح مكونات الحليب البقري أو الإبل أدى إلى انخفاض كبير في الثبات الحراري.