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Effect of Total and Differential Somatic Cell Counts, Lactation Stage and Lactation Number on Lipolysis and Physicochemical Composition in Camel (*Camelus dromedaries*) and Cow Milk

Research Article

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ABSTRACT

The present study was carried out to investigate the effects of somatic cell counts (SCC), differential SCC (macrophage (MAC), lymphocyte (LYM) and polymorphonuclear leukocytes (PMN)), number and stage lactation on milk composition in camel and cow milk. Camel milk appeared to contain significantly ($P<0.05$) a higher content of minerals. Lipolysis level is similar in camel milk compared to cow milk. Lipolysis level increased as MAC level increased in camel's milk but not in cow's milk. Our results suggest that MAC play a role in the degradation of dromedary milk fat. Mineral compositions were significantly affected by the SCC in camel milk. The milk composition was not affected by lactation number in both species. Total solid, Ca and Na content in camel's milk were gradually decreased through lactation.

KEY WORDS camel milk, lipolysis, physicalchemical composition, somatic cell count.

INTRODUCTION

Milk normally contains some level of somatic cells: neutrophils (PMN cells), lymphocytes (LYM) and macrophages. Macrophages (MAC) comprise the major cell type in milk from healthy udders (Dosogne *et al*. 2003; Lindmark-Månsson *et al*. 2006). When there is bacterial infection, tissue damage, or other inflammation processes affecting the mammary tissue, the SCC in milk dramatically increases (Sharma *et al*. 2011; Katherine *et al*. 2013). This increase in SCC results from the transfer of white blood cells from the blood to the mammary gland (Kelly *et al*. 2000; Sládek *et al*. 2006). In addition, the relative proportions of cell types present in milk change significantly, with an increased in PMN level (up to 90%) to protect the udder from bacterial challenge (Alhussien *et al*. 2016; Kehrli and Shuster, 1994; Zecconi and Smith, 2000). The increase in SCC milk causes the change in the components of cow's milk. The variation of the many components of cow milk with SCC was observed by many authors (Aysan *et al*. 2011; Brandt *et al*. 2010; Kelly *et al*. 2000; Somers *et al*. 2003; Bansal *et al*. 2005; Lindmark-Månsson *et al*. 2006) have reported changes in the composition of milk obtained from cows with infection, but little is known about such changes in camel milk. The relationship between SCC and lipolysis was investigated. It has been suggested that milk cells contribute to the lipolysis of milk fat to provide flavor defects (Azzara and Dimick, 1985; Ma *et al*. 2000; Santos *et al*. 2003; Gargouri *et al*. 2008). On the contrary, other studies indicated no relationship between milk SCC and lipolysis level (Lee *et al*. 1980; Cartier and Chilliard, 1990). Milk composition varies according to factors such as breed, age, mammary gland health, lactation stage, nutritional management and season (Dobranié *et al*. 2008).

Similar, the variation in the constituents of camel milk may be attributed to factors such as breed, age, the number of calving, nutrition, management, the stage of lactation, and the sampling technique used (Alshaikh and Salah, 1994). The purposes of this study were, firstly, to investigate the relationship among SCC, lipolysis and chemical composition and, secondly, to evaluate the influence of lactation stage on these variables in lactating camels and cows.

MATERIALS AND METHODS

Sampling

The study was carried out using individual milk samples from 36 dromedary animals (*Camelus dromedarius*) of Maghrabi breed from the south and the center of Tunisia. A total of 52 lactating dairy cows housed either in a free stall barn were used. Samples were obtained from each cow at days < 100 (n=15), between 100 and 240 days (n=25) and > 240 (n=12) after parturition. Of the 36 dromedaries, 11 were at early lactation (100 days in lactation), 18 at mid lactation (between 100 and 240 days lactation) and 7 at late lactation (between 100 and 240 days lactation). The camels were fed exclusively on natural browse. For cows, their nutrition is based on forage and concentrates. The milk was collected during the routine morning milking. Bovine samples were obtained by automated milking systems, but dromedary samples were obtained by manual milking. All the animals were free from clinical mastitis during the sampling period. Milk samples were taken to the laboratory immediately after collection and 250 mL were kept at 4 ˚C until the SCC. The rest was stored at -18 ˚C up to the rest of analysis.

Somatic cell counts

Somatic cells were counted using a Fossomatic 5000 (FossElectric, Hillerod, Denmark) according to International Dairy Federation Standard (IDF, 1995).

Milk analyses

Milk was analyzed for pH, titratable acidity (AOAC, 1995), total solids by drying at 102 ˚C (IDF, 1987), milk fat by gerber method (IDF, 1981). The extent of lipolysis in milk was measured using the bureau of dairy industries (BDI) method (IDF, 1991) and was expressed as acid degree value in meq FFA/100 g of fat. The mineral content was estimated using an Automate Synchron CX9 (Beckman coulter®). All analyses were performed in duplicate

Stastics

Statistical evaluations were performed using SPSS software (SPSS, 2011). The effect of lactation stage and lactation number on the different data was analyzed by one-way analysis of variance (ANOVA) and group means were compared by the Tukey's least significant difference test. Secondly, pearson's correlation coefficients (r) were also established to determine the relationships between the various parameters studied. The results were considered significant if the associated P-value was < 0.05.

RESULTS AND DISCUSSION

The overall results of physicalchemical parameters of dromedary and cow milk are resumed in Table 1. The Pearson correlation coefficients between total and differential SCC and physicalchemical parameters of dromedary and cow milks are presented in Tables 2 and 3. The pearson correlation coefficients between stage and number of lactation and physicalchemical parameters of dromedary and cow milk are presented in Table 4.

Milk characteristics

The data obtained showed a wide range of variation in some parameters studied between different individual camel and cow milk samples. There were no significant difference between pH values, titratable acidity and ash content of fresh camel and cow milk (Table 1).

Table 1 Composition of the camel's and cow's milk (Mean±SE)

Component	Dromedary milk	Cow milk	P-value
pH	6.38±0.22	6.7±0.19	0.44
Acidity (%)	16.8±0.36	17.09±0.22	0.08
Ash (%)	0.63±0.04	0.67±0.04	0.53
Total solid (%)	11.32±0.38	10.21±0.22	0.01
Fat (%)	3.71±0.23	3.43±0.13	0.30
Lipolysis (meq/100 mg fat)	2.96±0.39	2.29±0.24	0.13
Mg (mmol/L)	3.45±0.35	4.32±0.40	0.09
Cl (mmol/L)	61.4±3.02	37.58±1.08	0.000
K (mmol/L)	58.72±2.29	41.53±1.09	0.000
Na (mmol/L)	31.86±0.68	21.73±0.94	0.000
Ca (mmol/L)	10.32±0.44	6.37±0.30	0.000
Na^+/K^+	0.573±0.66	0.520±0.019	0.118

SE: standard error.

The obtained result of titrable acidity of camel milk was lower in comparison to the acidity of camel milk reported by Khaskheli *et al.* (2005). These results were similar to those reported by Aljumaah *et al.* (2012) and Hammadi *et al.* (2010). The value of titrable acidity found in camel milk is similar to that of cow. The ash content (0.63%) of camel milk found in this study was similar to that reported by Mehaia *et al.* (1995). The average of total solids content in camel milk was significantly higher (P<0.01) in comparison to cow's milk. The total solids content of camel milk was higher to that reported by others authors (Farag and Kebary, 1992; Ahmed, 1990; FAO, 1982).

According to Mehaia *et al.* (1995), the total solids content ranged from 10.0 to 14.4% in camel milk. The average value of fat in camel milk was 3.7%, which is similar to the content of fat in cow's milk (Table 1). The findings of this study agrees with Konuspayeva *et al.* (2009), who as a result of a meta-analysis of the literature data reported 3.82% an average fat content of camel milk. From the results shown in Table 1, it appears that lipolysis level in camel milk was not significantly different from lipolysis level found in cow's milk. The results of lipolysis of cows' milk were comparable to of the results reported by Andrews (1983). It was estimated from previous research (Bodyfelt *et al.* 1988) that the sensory threshold for detection of off-flavor would be about 1.0 meq/100 g of fat.

Table 2 Correlation coefficients (r) between total and differential SCC and physicochemical parameters of camel's milk. (values noted in bold are significant at P<0.05)

Parameter	SCC[1]	MAC[1]	LYM[1]	PMN[1]
pH	0.256	0.156	0.320	0.165
Ash	0.058	0.247	0.148	0.022
D°	-0.118	-0.217	-0.217	-0.077
TS	0.126	0.283	0.294	0.066
Fat	0.091	0.083	0.142	0.010
Lipolysis	0.213	**0.350**	-0.303	0.168
Mg	**0.404**	0.315	0.293	**0.385**
Cl	0.163	-0.078	0.022	0.193
K	0.015	-0.320	-0.319	0.071
Na	**0.635**	0.093	0.216	0.312
Ca	0.301	0.017	**0.366**	0.304

SCC: somatic cells count; MAC: macrophage; LYM: lymphocyte; PMN: polymorphonuclear leukocytes; D°: acidity (°Doronic) and TS: total solids.

Results from this study are in general higher than this threshold and the average lipolysis neared 2 meq/100 g of fat. The higher lipolysis level has been described as the most important factor that contributed to the lower sensory quality and shorter shelf life of milk (Azzara and Dimick, 1985; Ma *et al.* 2000). The levels of K, Cl, Na and Ca were significantly higher (P<0.05) in dromedary (Table 1), which is in agreement with others studies (Mehaia *et al.* 1995; Sawaya *et al.* 1984). An average Ca concentration in camel milk was 10.32 mmol/L, a little lower than that reported by Faye *et al.* (2008). The K, Na and Cl contents of camel milk were higher than the value reported by Kamoun (1990). Magnesium content of camel milk mean value was higher than the value reported by Ahmed (1990). High variability was observed in some studies regarding the mineral content of camel milk (Dukwal *et al.* 2007; Haddadin *et al.* 2008; Ayadi *et al.* 2009) and it could be attributed to the breed difference, intervals between milking, feeding, analytic procedures and water intake (Haddadin *et al.* 2008; Mehaia *et al.* 1995). Mehaia *et al.* (1995) considered that genetic factors could significantly affect the milk composition, especially under non controlled environmental conditions, as is mostly the case locally. The calculated Na:K ratio in camel milk was higher than that reported by Aljumaah *et al.* (2012). The variation in concentration of minerals and the increments in Na:K ratio were studied in dairy goats (Boutinaud *et al.* 2003) and dairy cows (Stelwagen *et al.* 1999; Delamaire and Guinard-Flament, 2006). Alterations in the Na:K ration could interfere with a number of intracellular processes. Increased Na:K ratio reduce mammary protein system in dairy goats (Stelwagen *et al.* 1999). In dairy camels, the regulatory mechanism seems not to operate (Ayadi *et al.* 2009). Instead, this difference might be related to the adaptation of the camels to the desert conditions.

Effect of SCC

The results show that the SCC and differential cell count did not have any significant correlation with pH, titratable acidity, ash and total solid values in both species. Table 3 shows that there was no significant relationship between lipolysis and total and differential SCC count in cow milk, which in agreement with Chazal and chillard (1986); Lee *et al.* (1980) and Cartier and Chilliard (1990). However, a positive correlation (P<0.05) between lipolysis and MAC was found in dromedary milk (Table 2). This suggests that the macrophages secreted lipolytic enzymes into the gradient while fractions containing polymorphonuclear leukocytes and lymphocytes did not possess lipolytic activity. These results confirm those of Russell *et al.* (1977) and Azzara and Dimick (1985), who found that lipolytic enzymes produced by monocytes and macrophages are believed to play a role in the degradation of cow milk fat ingested by those cells. A positive correlation (P<0.05) between fat, SCC and PMN count was found in cow milk but not in camel milk, which is in accordance with others studies (Aysan *et al.* 2011; Paura *et al.* 2002; Sawa and Piwczynski. 2002).

Table 3 Correlation coefficients (r) between total and differential SCC and physicochemical parameters of cow's milk. (values noted in bold are significant at P<0.05)

Parameter	SCC	MAC	LYM	PMN
pH	0.189	0.105	0.300	0.165
Ash	0.058	0.247	0.148	0.022
D°	0.193	0.144	0.136	0.299
TS	0.129	0.121	-0.014	0.134
Fat	**0.428**	0.195	0.195	**0.408**
Lipolysis	0.097	0.025	-0.191	0.165
Mg	**-0**.076	0.113	0.150	-0.002
Cl	-0.019	0.099	-0.195	-0.026
K	-0.111	-0.027	**-0.373**	-0.031
Na	0.019	0.066	-0.170	0.063
Ca	0.015	0.152	-0.240	0.023

SCC: somatic cells count; MAC: macrophage; LYM: lymphocyte; PMN: polymorphonuclear leukocytes; D°: acidity (°Doronic) and TS: total solids.

This positive correlation reported in this study and others (Barbano *et al.* 1989; Pereira *et al.* 1999; Ma *et al.* 2000)

may be ascribed to the strong reduction in milk production consequently to mammary epithelium damages (Akers and Thompson, 1987). Mineral composition was significantly affected by the SCC in camel milk. Table 2 illustrates the positive correlation between SCC, PMN and Mg content. A high positive correlation was also observed between the SCC and the Na content, which is in agreement with Bruchmaier *et al*. (2004).

However, mineral compositions was not significantly correlated with SCC in cow's milk, except in the case of K content which is in a negative correlation with LYM. Potassium declines because of paracellular passage out of the alveolar lumen between damaged epithelial cells (Harmon, 1994). The ion concentrations in milk may be due to increased blood capillary permeability, the destmetion of tight junctions, and the destruction of the active ion-pumping systems.

Effect of lactation

The pH in camel milk was significantly (P<0.05) affected by the stage of lactation (Table 4), in agreement with Aljumaah *et al*. (2012).

Table 4 Correlation coefficients (r) between stage of lactation, number of lactation and physicochemical parameters of camel's and cow's milk (values noted in bold are significant at P<0.05)

Parameter	Camel milk		Cow milk	
	SL	NL	SL	NL
pH	**0.396**	0.006	0.256	0.004
Ash	0.044	0.145	0.133	0.014
D°	**0.338**	0.007	0.075	0.018
TS	**-0.357**	0.003	0.103	-0.040
Fat	0.102	0.009	0.017	0.080
Lipolysis	0.185	0.094	0.087	**-0.308**
Mg	-0.278	-0.257	0.097	-0.079
Cl	-0.005	0.239	0.251	-0.136
K	0.248	0.214	0.031	0.022
Na	**-0.363**	-0.160	0.102	0.288
Ca	**-0.491**	0.201	0.038	0.067

SL: stage of lactation and NL: number of lactation.

Fat content was not affected by the stage of lactation (SL) in both species, which also observed Abeni *et al*. (2005). The ash content in camel milk was higher in the late stage compared to the initial stage of lactation. These results confirmed those of El-Hatmi *et al*. (2004) and Raziq *et al*. (2011), who reported that the ash content increased during lactation. The higher ash contents during late lactation stage suggest that camel milk can provide a satisfactory level of minerals (Mal *et al*. 2007). There was a negative relationship between total solid and lactation stage in dromedary. This decrease may be due to the increase in the milk water content during the last stage of lactation. These results confirmed those of Zeleke (2007), who demonstrate that total solid of camel milk decreased from 11.7% in the first stage of lactation to 10.1% by the end of lactation. In this study, Ca and Na content in camel milk showed a significant (P<0.05) decrease throughout the lactation, as observed by Aljumaah *et al*. (2012).

The variations in the major mineral contents of camel milk could be due to breed, feeding, stage of lactation, drought conditions, or analytical procedures (Haddadin *et al*. 2008; Farah, 1993; Mehaia *et al*. 1995). There was a negative, but not significant, relationship between lipolysis and lactation number (NL) in cow milk (r=-0.308; P>0.05) and camel milk (r=-0.09; P>0.05). This suggests that lipolysis seems to be higher in primaparous cows than in multiparous. These results showed no effect of lactation number on camel's milk composition.

CONCLUSION

In view of the observed results on the camel milk, it could be concluded that physicochemical properties was comparable to that of cow's milk. However, in present study, cow milk was found to contained lower mineral content compared to camel milk. The higher level of lipolysis was observed in camel's milk that contained a high percentage of MAC. This may also indicate that MAC in milk could play an important part in determining the lipolysis level in camel's milk. Negative relationship between lactation number and lipolysis level was found in cow's milk. For this species, the lactation stage not affected the physicalchemical compostion. The present study emphasizes that the variations in the camel's milk composition could be attributed to SCC and lactation stage.

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