Short communication

Effect of the production system and stage of lactation on the microbiological and biochemical characteristics of camel milk

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Abstract

This study was conducted to assess the physicochemical and microbiological status of camel milk from different production systems (intensive, semi-intensive, extensive) and to monitor its quality during the different stages of lactation. Lactic Acid Bacteria (LAB) was also isolated and identified from milk samples from the different production systems and at different stages of lactation. Although the physicochemical characteristics of milk from the different production systems were not statistically different except for fat and proteins, the microbiological analysis revealed significant differences in total counts of mesophilic bacteria, yeast and molds and total coliforms. The differences in physicochemical and microbiological characteristics between lactation stages was significant. The diversity of LAB was also affected by these two parameters.

Keywords: camel milk, production system, lactation stage, lactic acid bacteria

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Introduction

In Tunisia, camel farming is conventionally extensive, a method perfectly suited to the biology of the species, and concentrates in the southern areas (Moslah et al., 1989). In recent years, there has been a growing interest in the development of intensive camel farming operations mainly for milk production. This system is based on the adoption of technologies developed and applied to other species of animals, such as cattle, aimed at optimizing the animal's production capacity (Faye, 2004). The semi-intensive, or integrated, system, created in response to the decrease in pasture and feed, is also being used (Diallo, 1989).

The nature and significance of the microflora contained in camel milk, are determined by the health status of the animal, the milking conditions, and the temperature and handling of milk after milking. The dominant and most beneficial microflora in camel milk are mainly lactic acid bacteria (LAB). This group of bacteria is considered to be a potential source of biological agents for use in dairy technology (Khedid *et al*., 2009). This study aimed to determine the impact of lactation stage and production system on the physicochemical characteristics and microbiological quality, especially LAB concentration, on camel milk.

Material and methods

Source of sampling

This study was carried out during the period from January to November. It involved fifteen healthy she camels at different stages of lactation (early, mid and late) from three different breeds of camel (Medenine, Chenchou, Ben Ghilouf and Tataouine). The camels were kept under three different management systems (intensive, semi-intensive and extensive). Milk samples were collected from individual camels in sterile bottles (50 ml). Each sample was immediately labeled, kept on ice and transported to the laboratory of Livestock and Wildlife in Arid Land Institute (Medenine, Tunisia) for analysis**.**

Physicochemical analysis

The acidity and pH of the milk samples were measured immediately after their arrival at the laboratory. The viscosity (in cP) was determined by a Brookfield type viscometer (model DV-E, MA, USA). Dry matter, ash and total nitrogen contents were determined by dry combustion in a furnace (850 °C) that was purged with O_2 gas according to the method proposed by Dumas (AFNOR, 1993). The fat content was measured by an acid-butyrometric method using a "neusol solution".

Microbiological analysis

A total plate count (TPC) was carried out on Plate Count Agar (PCA), (Scharlau Chemie S.A.), incubated at 37°C for 72 h, and yeast and moulds on Sabouraud Chloramphenicol (Pronadisa) incubated at 25°C for 3 to 5 days. Total coliforms were grown in Violet Red Bile Agar (AppliChem) in a double layer. Lactic Acid Bacteria (LAB) were plated on De Man-Rogosa-Sharpe (MRS) agar (Scharlau Chemie S.A.) and incubated at 30 °C for 48 h.

Isolation and identification of lactic acid bacteria

LAB was isolated on MRS agar (Pronadisa) and incubated at 30 °C for 24 to 48 h in order to apply the conventional identification tests. All isolates were initially examined for Gram staining and catalase reaction. Only Gram-positive and catalase-negative isolates were considered. The biochemical identification was carried out using API systems. API 50 CH was used in conjunction with an API 50 CHL medium for the identification of *Lactobacillus* and related genera strips, in accordance with the manufacturer's instructions (Biomerieux, Marcy l'Etoile, France) (Ghanbari *et al.*, 2009).

Statistical analysis

SAS software (version 9) was used for the statistical analysis of data. Production system and lactation stage were analyzed by ANOVA using the General Linear Model (GLM) for determination of their effect on physicochemical and microbiological characteristics. The means values were compared using the SNK test.

Results and discussion

Effect of the production system

Physicochemical characteristics

The values of pH for the three production systems (i.e. intensive, semi-intensive, extensive) averaged 6.40, which is relatively similar to the average pH value (6.43±0.07) reported by Alwan *et al.* (2014) for Meghrebi Libyan camels kept on different systems (intensive and extensive) and slightly higher than that reported by Ghouri *et al*. (2016) for raw milk (pH=6.0).The values were also similar to those reported by Gnan and Sheriha (1986) (pH=6.2 and 6.8). The lowest pH value was observed in the extensive

system, which might be related to the high content of lactic acid bacteria in milk collected from camels under the same system (Table 1).

Table 1. Effect of breeding system on the physicochemical parameters of camel milk.

Parameter	Production system			
	Intensive	Semi-intensive	Extensive	P value
pH	6.46±0.16[a]	6.46±0.17[a]	6.29 ±0.088[a]	NS
Acidity (°D)	16.75±1.83[a]	16.12±2.41[a]	17.50± 2.08[a]	NS
Viscosity (cP)	3.65±0.54[a]	3.85±0.87[a]	4.62±1.06[a]	NS
Fat (g/l)	21.37±6.9[b]	26±8.78[ab]	34.75±10.68[a]	*
Dry matter (g/l)	117.2±10.28[a]	118.7±7.75[a]	116.3±8.27[a]	NS
Ash (g/l)	9.56±1.8[a]	9.79±1.65[a]	9.44±1.8[a]	NS
Protein (g/l)	31.59±2.48[c]	35.86±4.21[b]	43.65±4.007[a]	**

[a,b,c]means in the same line followed by the same letter are not statistically different P>0.05; NS: Not significant ; *: P<0.05 ; **: P<0.01.

The production system had no effect (P>0.05) on DM and Ash contents of milk. Dry matter values were similar to those reported by Alwan *et al*. (2014) for milk representing intensive and extensive production systems (119.2 vs 117.4 g/l), while the ash content was slightly higher (9.6 vs 8.1). Fat content of milk from the extensive system was higher (P<0.05) than that of the intensive system, but differences between the intensive and semi-intensive systems were not significant (P>0. 05). However, raising camels under intensive or semi-extensive systems produced milk with lower (P<0.01) protein contents (Table 1). Similar results were reported by Alwan et al. (2014) comparing protein contents in milk from intensive with extensive systems (24.5 vs 31.9 g/l).

Microbiological characteristics

Significant differences were observed in the microbial load among the different production systems. The highest bacterial load was marked in the extensive system, which can be due to the environment, processing conditions and a transportation time from milking to analysis (Table 2). This result is similar for cow milk; Srairi *et al*. (2005) reported that milk quality is affected by the livestock's production system. The presence of LAB in camel milk was predictable because milk provides an optimal natural environment for the growth of this group of bacteria irrespective of the milk source (sheep, goat, or cattle).

Table 2. Effect of production system on microbiological parameters

Microorganisms	Production system			
	Intensive	Semi-intensive	Extensive	P value
TPC (log_{10} CFU/ml)	2.96±1.24[b]	4.13±0.98[b]	5.47±0.64[a]	**
Yeast/mould (log_{10} CFU/ml)	1.36±1.25[b]	0.45±0.64[b]	6.36±2.44[a]	**
Total coliform (log_{10} CFU/ml)	2.17±1.68[b]	2.53±1.53[b]	5.38±0.26[a]	**
LAB (log_{10} CFU/ml)	0.13±0.08[a]	1.13±0.20[a]	1.43±0.87[a]	NS

[a, b]means within the same line followed by different letters are statistically different P<0.05; **: P<0.01; NS: Not significant; TPC: total plate count; LAB: lactic acid bacteria.

Effect of the lactation stage

Physicochemical characteristics

It was observed that the pH content of camel milk varied significantly during lactation (Table 3). Colostrum presented a low pH due to its high protein content (Benkerroum, 2008). Chemical composition of camel milk changed with stage of lactation with fat content being higher (P<0.01) in late lactation while protein content was lower (P<0.01). The variation in protein content during lactation was similar to results reported by Riyadh *et al.* (2012) being higher (P<0.01) in early lactation compared with mid- and late lactation.

Table 3. Effect of lactation stage on physicochemical characteristics

Parameters	Lactation stage				
	First week	Early lactation	Mid-lactation	Late lactation	P
pH	6.35±0.18[bc]	6.45± 0.13[ab]	6.52 ± 0.13[b]	6.64±0.11[a]	**
Viscosity (cP)	6.61±2.55[a]	5.82±3.12[ab]	3.37±0.45[c]	4.17±0.53[bc]	**
Fat content (g/l)	11.72±9.78[c]	20.30±5.19 [b]	21.40±4.77[b]	28.44±8.67[a]	**
Dry matter (g/l)	127.70± 12.38[a]	113.35±8.16 [b]	106.09±5.24[c]	105.68±4.01[c]	**
Ash (g/l)	10.74±2.42[a]	7.95±1.50 [b]	7.07±1.23[b]	6.96±0.89[c]	**
Acidity (°D)	24.78±5.63[a]	17.39±4.04[b]	15.40±3.08[b]	16.33±2.64[b]	**
Protein (g/l)	43.07±2.11[a]	33.85±2.26[b]	27.93±0.87[d]	31.34±1.61[c]	**

[a,b,c]means within the same line followed by different letters are statistically different (P<0.05).

Microbiological characteristics

The TPC fluctuated over the different lactation stages, increasing in the early stage of lactation and decreasing in mid-lactation before increasing again at the end (Table 4). Values ranged between 2.64 and 2.30 log_{10} CFU/ml. The yeast and moulds content in Moroccan camel's milk was found to be higher, with an average count of 4.6 log_{10} CFU/ml (Benkerroum *et al.*, 2003).

LAB counts ranged between 1.62 and 2.79 log_{10} CFU/ml, with significant differences found between lactation stages and maximum levels in colostrum. LAB was the predominant microflora in camel milk, which could be beneficial due to their antagonistic capabilities toward other microorganisms (Daeschel, 1989).

Table 4. Effect of lactation stage on microbiological characteristics.

	Lactation stage				
Parameters	First week	Early lactation	Mid-lactation	Late lactation	P
TPC (log_{10} CFU/ml)	2.64±0.80^{a}	2.30± 1.05^{a}	2.48± 0.99^{a}	2.41± 0.76^{a}	NS
Yeast and molds (log CFU/ml)	2.14±0.99^{a}	2.17± 0.30^{a}	1.65± 0.82^{ab}	1.28± 1.24^{b}	NS
LAB (log_{10} CFU/ml)	2.79±0.53^{ab}	2.00± 1.05^{bc}	1.62± 1.23^{c}	2. 48± 0. 66^{a}	**

^{a,b,c}means within the same line followed by different letters are statistically different ($P<0.05$); NS: not significant; ** $P<0.01$; TPC: total plate count; LAB: lactic acid bacteria.

Isolation and identification of lactic acid bacteria

Regarding carbohydrate fermentation, the strains were divided in two groups (Table 5). The first group was dominated by regular rods ($SCC_{1,8}$, $SCC_{1,7}$, $SCC_{1,15}$, $SCC_{1,2}$) and tentatively identified as *Lactobacillus plantarum, Lactobacillus pentosus* and *Lactobacillus brevis*. The second group was coccoid in shape (SLC_{ch14}, SLC_{ch6}, $SCC_{1,13}$, $SCC_{1,33}$, and $SCC_{1,6}$) and tentatively identified as *Lactococcus lactis1* and *Pediococcus pentosaceus*. Earlier studies have reported the presence of the *L. plantarum* and *L. brevis* in fermented Sudanese camel milk (Ashmaig *et al.*, 2009). Sun *et al.*, (2010) isolated the *L. plantarum* and *Lactococcus lactis* from traditional fermented milk in Mongolia.

Conclusion

The physicochemical and microbiological characteristics in camel milk, including lactic acid bacteria counts, are affected by production systems and stages of lactation. The physicochemical characteristics of camel milk samples obtained from different production systems revealed highly significant variations between these systems in the content of fat and protein. These characteristics also varied through the different stages of lactation.

Table 5. Fermentation profiles of lactic acid bacteria isolated from camel milk.

Isolates	Carbohydrate (carbon source)							Identity
	Glycerol	L-sorbose	D-sorbitol	Amygdaline	Esculin	D-Melioidoses	Amidon	
SLC ch6	+	-	-	-	+	-	-	*L. lactisssp lactis1*
SCC 1, 7	-	-	-	+	+	-	W	*L. plantarum*
SLC ch14	W	-	-	+	W	-	+	*L. lactisssp lactis1*
SCC 1, 13	W	-	-	W	W	-	W	*L lactisssp lactis1*
SCC 1, 33	+	W	-	W	+	-	+	*L. lactisssp lactis1*
SCC 1, 15	W	-	+	+	+	-	-	*L. pentosus*
SCC 1, 24	-	-	-	-	W	-	W	*L. lactisssp lactis1*
SCC 1, 6	W	-	+	+	W	+	-	*Pediococcus pentosaceus*
SCC 1, 8	W	-	-	-	+	-	W	*L. plantarum*
SCC 1, 2	+	-	-	-	W	W	W	*L. brevis*

+ = growth; W = weak growth; - = no growth after 48 h of incubation at 37 °C; SLCch = strain camel milk Chenchou; SCC= strain camel colostrum.

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