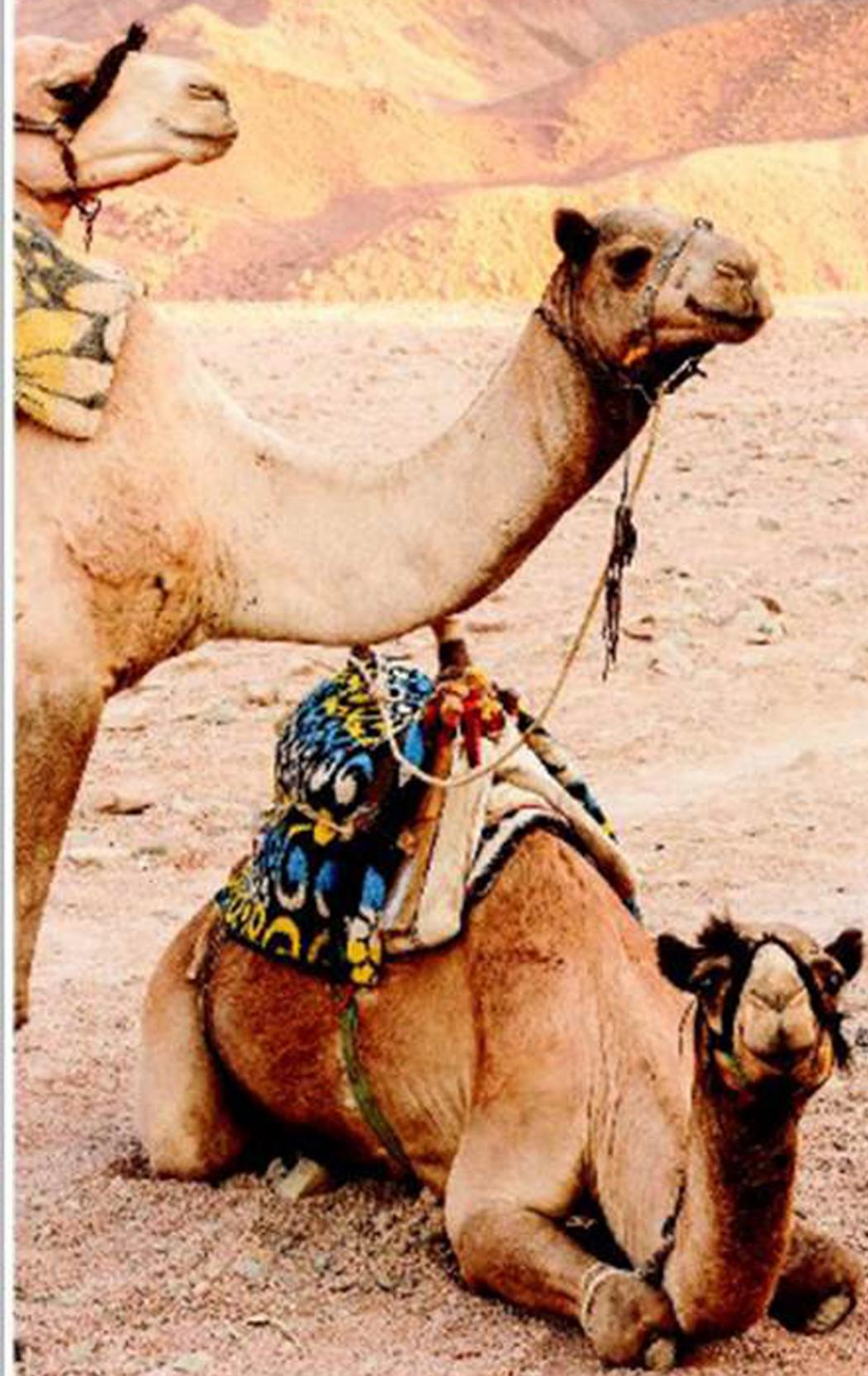


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Special issue on “Camel Milk and Related Research”

Guest Editor

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CONTENTS

EDITORIAL

Page No.

Special issue on “Camel Milk and Related Research”

Bernard Faye

ARTICLES

Fast protein liquid chromatography of camel α -lactalbumin fraction with radical scavenging activity

El Hatmi Halima, Jrad Zeineb, Khorchani Touhami, Dary Annie and Girardet Jean-Michel..... 309

Nutritional value and sanitary evaluation of raw Camel's milk

Ahmed Abdel-Hameid Ahmed, Rasha Galal Sayed and Mohammed Sayed..... 317

Microflora identification of fresh and fermented camel milk from Kazakhstan

Shynar Akhmetadykova, Almagul Baubekova, Gaukhar Konuspayeva, Nurlan Akhmetadykov and Gérard Loiseau..... 327

Impact of husbandry, stages of lactation and parity number on milk yield and chemical composition of dromedary camel milk

Wafa I. A. Babiker and Ibtisam E. M. El-Zubeir..... 333

Effect of Selenium injection in pregnant camels on selenium status of their new-born and milk

Bernard Faye, Ossamah Althamma, Abdelgadir Musaad, Gaukhar Konuspayeva and Mohammed Bengoumi... 342

Some lipid components of the camel milk and blood in intensive farm in Saudi Arabia

Gaukhar Konuspayeva, Bernard Faye and Abdelgadir Musaad..... 349

Some parameters to process camel milk into cheese

Gaukhar Konuspayeva, Benedicte Camier, Frederic Gaucheron and Bernard Faye..... 354

Camel milk value chain in Northern Saudi Arabia

Bernard Faye, Hassani Madani and Sageed A. H. El-Rouili..... 359

Comparison of dairy performances between dromedaries, bactrian and crossbred camels in the conditions of South Kazakhstan

Moldir Nurseitova, Gaukhar Konuspayeva and Stefan Jurjanz..... 366

Morphological diversities and eco-geographical structuring of Ethiopian camel (*Camelus dromedarius*) populations

T. Yosef, K. Kefelegn, Y. K. Mohammed, U. Mengistu, A. Solomon, D. Tadelle and J. Han..... 371

EDITORIAL

Special issue on “Camel Milk and Related Research”

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The interest for camel milk is growing both in arid countries where camel is originated and in Western countries where it is regarded as an original model for research. Indeed, while the gross composition of camel milk is similar to that of cows' milk, its fine composition is quite original: regarding proteins for example, camel caseins have only 60% homology with cow caseins, the micelles are big (>300 nm), β -lactoglobulin is lacking (explaining the hypo-allergic properties of camel milk), acidic whey proteins are present, and there is a high concentration of non-protein nitrogen. Camel milk is rich in iron, and sometimes in chloride when the animals graze halophytes. The fat globules are smaller than those of cow milk and the proportion of unsaturated fatty acids is higher. Its richness in vitamin C is regularly reported. Much believes and health assertions are running about the true or expected “medicinal” virtues of camel milk. However some solid scientific arguments exist to support this idea. Regarding the camel milk processing, the suitability for cheese manufacture being low, the main form of consumption is fresh, pasteurized or fermented milk. However, with specific chymozyme available nowadays on the market, camel cheese making is in development. So, the Emirates Journal of Food and Agriculture published in a country where camel plays a pivotal role in the livestock economy, pays naturally a high attention to this “white gold of the desert” as said Pr Wernery. In the present special issue devoted to camel milk within its different dimensions (composition, medical properties, processing, market etc.,), ten papers are proposed. They tackled a wide range of questions from the physico-chemical characteristics up to the camel milk sector organization.

The paper of El-Hatmi et al. from Morocco, proposes a tool for the study of the biological property of one component of the milk protein, the β -lactalbumin while the nutritional value of camel milk is assessed by the team of Ahmad et al. Those papers revealed the specific richness of camel milk

allowing protein accessibility for the population in desert areas. The health allegations of the camel milk are the main reason of its success among the milk drinkers population. It is an important challenge for the research to describe the mechanisms of this renowned aspect of camel milk. The antidiabetic properties, regularly supported in many recent papers, are approached again here based on an experiment on mice where the placebo effect cannot be evoked (Sayed et al.). In the paper of Akhmetadykova et al., 138 strains were isolated in shubat, the fermented camel milk produced in Central Asia and 37 were identified, leading to a potential use in industry for conducting specific fermentation process. Selenium is an important trace element for the maintenance of the mineral equilibrium in all type of livestock. The selenium supplementation by injection in camel is a way, widely used in Saudi Arabia by the camel owners for enriching the camel milk in this trace element and avoid the symptoms of the selenium deficiency (white muscle disease), especially on the young animals (Faye et al.). If the proteins are an important part of the camel milk composition, the fat part is also quite predominant. The fatty acid composition in milk, cholesterol and liposoluble vitamins content could change in proportion due to the intensification of the camel production system. It is the suggestion of Konuspayeva et al. The ability of camel milk to be processed into cheese was in the past an important technological challenge because the difficult clotting. Nowadays, this problem is solved thanks to special camel rennet available on market, but the making of camel cheese still needs specific parameters (as calcium, pH, time of maturation) to be determined as suggested in the second paper of Konuspayeva et al. Milk and milk products are targeted for marketing, but the main part of this production is still used for self-consumption, even in rich countries like Saudi Arabia. In many case, camel milk sector is not well structured, but before to say that, the study of the camel milk value chain is

essential (Faye et al.). The impact of camel farming system (nomadic, semi-intensive or intensive) is not negligible on the milk composition of the camel and on the milk yield. Based on the monitoring of those three types of farms, Babiker and El-Zubeir in Sudan, confirm some results regarding the variability of the camel milk composition. Dromedary (one-humped camel) and Bactrian (double-humped camel) are the both large Camelid species sharing the arid countries of the world. However, in spite of their genetic proximity underlined by the existence of fertile hybrids, the camel milk composition and milk productivity potential of these two species differ significantly (Nurseitova et al.). However, the variability exists also within a single population of camels. Based on a clear phenotype description, Kelefelegn et al. (Ethiopia) was able to distinguish clearly different breeds and describe them.

We expect that the present new issue of the Emirates Journal of Food and Agriculture on camel milk will contribute to a better wide knowledge regarding this animal product by different approaches and will arouse further studies as these published results raise more questions than answers.

REGULAR ARTICLE

Fast protein liquid chromatography of camel α -lactalbumin fraction with radical scavenging activity

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Abstract

The aim of this study was to investigate the radical-scavenging properties towards a stable radical cation, ABTS, of *Camelus dromedarius* whey proteins (CWP) separated onto a cation-exchanger by fast protein liquid chromatography. The highest activities were found for CWP and fraction F1 mainly composed of α -lactalbumin. Fractions F2, F3 and F4 contained a mixture of lactoferrin, immunoglobulins G and probably camel whey basic protein (CWBP). These three fractions displayed low radical-scavenging activities. Lactoferrin was eluted almost pure in the last fraction (F5) but did not possess detectable radical-scavenging activity. The present results suggested that the cation-exchange chromatography is of great interest to yield, in a single step, whey protein fractions with various biological activities, i.e. a highly-enriched α -lactalbumin fraction displaying efficient antioxidant activity, a fraction (pool of F2-F4) mainly composed of heavy-chain immunoglobulins potentially interesting for human therapy and a fraction of pure lactoferrin having numerous biological activities such as antimicrobial and immunomodulating properties.

Key words: ABTS, α -Lactalbumin, Antioxidant activity, Camel milk, Radical scavenger

Introduction

It is generally well established that the food constituents can be used to reduce the risk of developing or aggravating human disease conditions. In this regard, functional foods and nutraceuticals have emerged as adjuvant or alternative to chemotherapy especially in the prevention and management of human diseases and for maintaining optimum health state (Kris-Etherton et al., 2002). Interest in the camel milk for human nutrition is increasing due to its distinct composition and unique biofunctional properties (e.g. antidiabetic properties; Sboui et al., 2012).

Camel milk possesses vital role in human nutrition in hot regions and countries. It contains the essential nutrients found in bovine milk, though some of them are found in higher concentrations

such as vitamin C, iron, and unsaturated fatty acids (Al Haj and Al Kanhal, 2010). Besides caseins, camel whey proteins (CWP) constitute 20–25% of the total camel milk proteins (Khaskheli et al., 2005), the majority of them having various biological activities not found or in a lesser extent in the bovine milk protein fraction. In contrast to bovine milk whey proteins, CWP contain large amounts of heavy-chain antibodies IgG2 and IgG3 which are devoided of light chains, and thus have the potential to inhibit efficiently enzymes and micro-organisms (Harmsen and De Haard, 2007; Daley-Bauer et al., 2010). Lactoferrin (Lf) is present in much larger amount in camel milk than in bovine milk (ca. 0.3 g L⁻¹ and 0.1 g L⁻¹, respectively; El Hatmi et al., 2006; Konuspayeva et al., 2007). A number of preventive properties is attributed to Lf such as antibacterial, antiviral, fungistatic, antiparasitic, antithrombotic and immunomodulatory effects (Darewicz et al., 2011). β -Lactoglobulin known for its allergenic potential is lacking in camel whey (Elagamy et al., 2009), whereas α -lactalbumin (α -LA; SwissProt accession number P00710) constitutes the main component

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(2.2 g L⁻¹ of milk; El Hatmi et al., 2006). An advantage of α -LA may be its beneficial role in the antioxidant system of the neonate (Lien, 2003). The bovine α -LA (Sadat et al., 2011) and the camel α -LA (Salami et al., 2009; 2010) are a source of free radical-scavenging peptides. Therefore, attention is being focused on producing α -LA-enriched formulae because α -LA might have an ability to attenuate oxidative stress occurring in inflammatory bowel disease after oral administration (IBD; Rezaie et al., 2007). The protein or its peptides generated by gastrointestinal digestion might act directly on the inflammatory site in the gut without passing through the intestinal barrier.

This study was undertaken to prepare an α -lactalbumin-enriched fraction possessing a free radical-scavenging activity much better than that of CWP. This activity was investigated spectrophotometrically with the 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) or ABTS method. In this work, cation-exchange chromatography performed by fast protein liquid chromatography (FPLC) revealed a single step efficient method to readily produce α -LA-enriched fraction. In addition, two other protein fractions possessing biological activities of great interest such as fractions containing IgGs with potent therapeutic applications and Lf having antimicrobial properties were also obtained.

Materials and Methods

Sample collection

Milk samples from 5 healthy camels (*Camelus dromedarius*) were collected and mixed together. The animals, all belonging to experimental herd of the Livestock and Wildlife Laboratory (Institute of Arid Land, Médenine, Tunisia) were in the third month of lactation (Atigui et al., 2013). Samples were collected manually in sterile bottles once per day usually in the morning. Three aliquots of each sample were immediately stored at -20°C until used.

Preparation of whey proteins and chromatography

The milk was firstly skimmed by centrifugation (4500 g at 30°C for 20 min). Then, the casein fraction was precipitated at pH 4.2 with 1 M HCl and discarded by centrifugation performed in the same conditions. The supernatant (milk whey) was neutralized with 1 M NaOH, dialyzed against distilled water at 4°C for 72 h and CWP were lyophilized.

Fractionation of CWP was performed by cation-exchange fast protein liquid chromatography

(FPLC) with the ÄKTA-FPLC technology (GE Healthcare, Uppsala, Sweden) by passing sequentially through three Hitrap CM (carboxymethyl) 5/5 columns (1.5 x 2.5 cm) equilibrated in 20 mM tris(hydroxymethyl) aminomethane hydrochloride (Tris/HCl) buffer, pH 8.0 containing 0.02% sodium azide. Volumes of 10 mL of whey proteins (10 g L⁻¹ of Tris/HCl buffer) were loaded onto the three columns and a 0–1 M linear gradient of NaCl in the same buffer was applied at 1 mL min⁻¹. Eluted proteins were detected at 280 nm.

Electrophoresis

Whey proteins of the different FPLC fractions were separated by sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) in the presence of 1.1% SDS and 5% 2-mercaptoethanol according to the method of Laemmli and Favre (1973) with a 4.9% stacking gel and a 15.4% resolving gel running in 0.125 M Tris/HCl buffer, pH 6.8 and 0.38 M Tris/HCl buffer, pH 8.8, respectively. Volumes of 20 μ L of samples at 2 g L⁻¹ proteins were loaded in the gel. Proteins were stained for 30 min by 0.1% Coomassie blue R250 in a mixture of 50% ethanol and 10% acetic acid followed by overnight destaining in a solution of 30% ethanol, 7.5% acetic acid and 5% trichloroacetic acid. Molecular mass standards (Precision Plus Protein All Blue Standards) were from Bio-Rad (Hercules, CA, USA).

Protein concentration determination

The protein concentration was determined by the Bradford method. The bovine serum albumin was used as standard. The results of the assay depend on the number of basic amino acid residues of each protein (Ku et al., 2013) and the data are expressed as mg L⁻¹ equivalent (eq.) to BSA. Each measurement was carried out in triplicates.

ABTS⁺ radical-scavenging assay

The radical-scavenging assay was carried out according to Sadat et al. (2011), a method adapted from that of described by Re et al. (1999). The stable radical cation ABTS⁺ was produced by dissolving 7 mM ABTS⁺ in 2.45 mM potassium persulfate and by keeping the mixture in the dark for 15 h at room temperature. The ABTS⁺ radical reagent was then diluted with 5 mM sodium phosphate buffer, pH 7.4 to reach an absorbance of 0.70 ± 0.02 at 740 nm. The radical cation was stable in phosphate buffer for at least 1 h at 22°C. The decrease in absorbance in the presence of protein fractions was measured at 740 nm with an

MRX® microplate reader (ThermoLabsystems, Chantilly, VA, USA). Volumes of 150 μ L of protein fractions (0-100 mg L⁻¹ eq. BSA) or of Trolox or gallic acid (0-30 μ M) dissolved in phosphate buffer were added to 150 μ L of the ABTS⁺ reagent and the mixture was incubated for 10 min at 30°C before absorbance measurement. All the assays were carried out five times. The radical-scavenging activity was calculated as follows:

$$\text{Activity (\%)} = [1 - (A_r - A_b) / (A_i - A_b)] \times 100 \quad [1]$$

Where: A_i = the absorbance of the initial ABTS⁺ radical, A_r = the absorbance of the remaining radical and A_b = the absorbance of the blank (phosphate buffer, $A_b=0.09$).

The IC₅₀ value is defined as the concentration of sample able to transform 50% of ABTS⁺ to ABTS⁺ *i.e.* when the absorbance of the remaining radical was equal to the scavenged radical. Thus, log (IC₅₀) corresponds to the x-intercept of the curve of log [(A_r - A_b)/ (A_i - A_r)] *vs.* log (concentration of sample).

The Trolox-equivalent antioxidant capacity (TEAC) measures the free radical scavenging capacity of a given substance, as compared to the standard, Trolox. The TEAC (in μ mol Trolox equivalent or TE per μ mol of a given substance) is the ratio of the gradient of the plot of activity *vs.* concentration of the given substance over the gradient of the plot of Trolox (Re et al., 1999).

Results and Discussion

In the present study, we proposed a simple method of separation of CWP by cation-exchange chromatography with the ÄKTA-FPLC technology in order to prepare in single step different fractions containing biologically active proteins *i.e.* α -LA-, IgGs- and Lf-enriched fractions and to investigate their potential free radical-scavenging activity. Although the anion-exchange chromatography (Ochirkhuyag et al., 1998) or size-exclusion chromatography (Si Ahmed et al., 2013) allow to obtain pure α -LA, these methods have not been revealed enough suitable to recover the IgGs and Lf (Si Ahmed et al., 2013). Elagamy et al. (1996) have achieved the purification of camel milk IgGs by protein affinity chromatography. The preparation of heavy-chain antibodies (IgG2 and IgG3) from camel milk is of great interest. Indeed, after immunization of *Camelidae* species, milk instead of blood serum might be a dietary source of single-domain antibody fragments (V_HHs) able to bind therapeutic targets. For example, llama's V_HHs can specifically target the cell receptor domains of toxins of *Clostridium difficile* (Hussack

et al., 2010). In addition, the V_HHs of small size (*ca.* 15 kDa) are especially suited for oral immunotherapy because of their stability against very acidic pH, proteolysis and high concentrations of denaturing agents (Harmsen and De Haard, 2007).

Fractionation of CWP by ÄKTA-FPLC chromatography

After separation of the CWP, the chromatographic fractions were analyzed by SDS-PAGE electrophoresis (Figure 1). Fraction F1 mainly contained the major soluble protein of camel whey *i.e.* α -LA and camel serum albumin (CSA). As expected, these proteins were not adsorbed onto the cation-exchanger due to their acidic isoelectric points (pHi). The theoretical pHs are 5.01 and 5.60 for the camel α -LA and bovine serum albumin (SwissProt accession number P02769), respectively (the CSA sequence is not available in the databank). The fractions F2 and F3 mainly contained heavy chains H45 and H42 of IgG2 and IgG3, respectively, these IgGs being devoided of light chains (Lauwereys et al., 1998; Daley-Bauer et al., 2010) whereas IgG1, which consisted of both heavy chains H55 and light chains L30 was recovered in fraction F4. The IgGs have generally near-neutral or basic pHs (pHis 6.5-9.5; Igawa et al., 2010) and could be adsorbed onto the cation-exchanger and then desorbed all along the ionic strength gradient. The fraction F4 might also contain the camel whey basic protein (CWBP) isolated for the first time by Ochirkhuyag et al. (1998). According to these authors, CWBP displays apparent molecular mass and pHi of 20 kDa and 9.30, respectively. The fractions F2-F4 also contained Lf at estimated molecular mass of 78 kDa by Elagamy et al. (1996). IgGs and Lf were eluted in several fractions because of their microheterogeneity of their glycan moiety (Zinger-Yosovich et al., 2011). However, fraction F5 contained almost pure Lf highly retained onto the cation-exchange column as shown by SDS-PAGE, which is in accordance to our previous work (El Hatmi et al., 2007). Like CWBP, the camel Lf is a basic protein and has a theoretical pHi of 8.63 (unglycosylated form; UniProt/SwissProt accession number Q9TUM0). As expected, these two proteins were strongly retained onto the cation-exchanger, whereas the acidic α -LA and CSA were directly eluted in the void volume of the column.

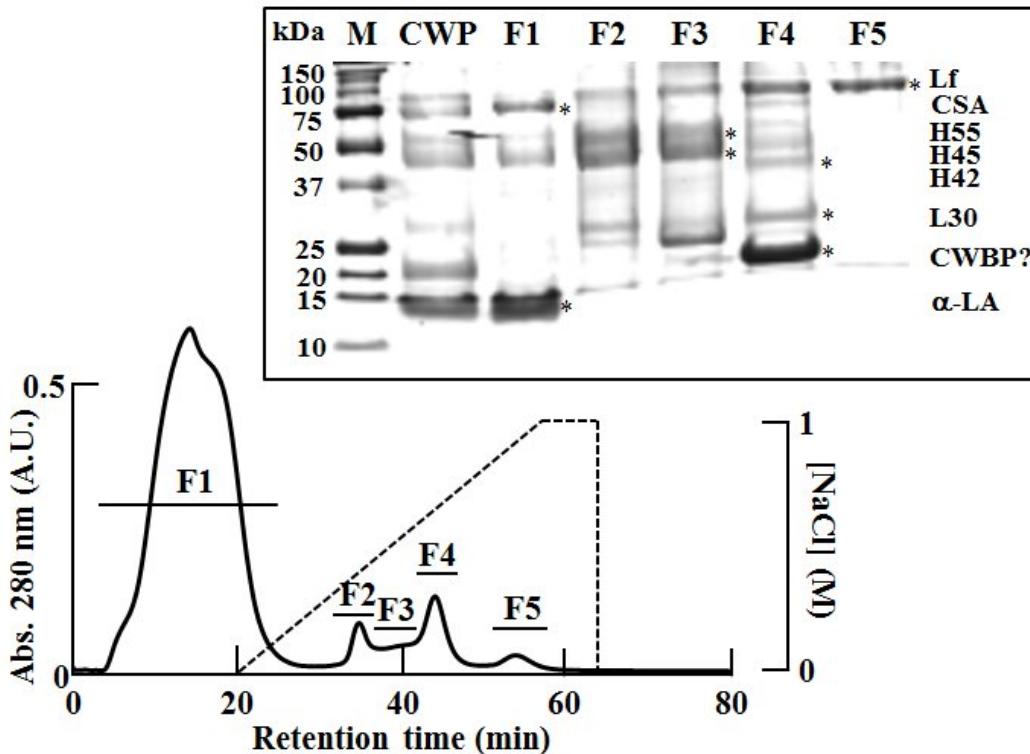


Figure 1. Cation-exchange fast protein liquid chromatography of camel whey proteins (CWP) onto three successive Hitrap CM columns connected to an ÄKTA-FPLC system and SDS-PAGE analysis of the collected fractions F1–F5. The ionic strength gradient is in dashed line and the chromatogram in solid line. Electrophoretically identified bands are indicated by an asterisk. A.U.: absorbance unit; M: molecular mass standards; Lf: lactoferrin; CSA: camel serum albumin; H55, H45, and H42: heavy-chains of immunoglobulins G of 55, 45, and 42 kDa, respectively; L30: light chains of immunoglobulins G of 30 kDa; CWBP: camel whey basic protein; α -LA: α -lactalbumin.

Investigation of radical-scavenging activity

Gallic acid and Trolox (soluble analog of vitamin E) are strong radical scavengers that were used in this study as positive controls. A linear relationship was found from the concentration response curve in the range of 0–5 μ M gallic acid and 0–10 μ M Trolox (Figure 2A). In the present study, the TEAC value of gallic acid was 3 μ mol TE μ mol⁻¹ showing that gallic acid was a greater free radical scavenger than Trolox as evident from its three-fold higher antioxidant power. Its IC₅₀ was 2.0 μ M, close to the IC₅₀ of 2.5 μ M determined by Sadat et al. (2011).

Chen et al. (2003) found that the ABTS method was most suitable and sensitive to determine the antioxidant capacity of bovine milk proteins. This method was thus used in this study to assess the

free radical scavenging activity of CWP. The activities of the different fractions were estimated by determination of the IC₅₀ values (Table 1). The best activities were found for CWP and F1 (Figure 2B), respectively, whereas the other fractions did not display any interesting activity. The α -LA of CWP was fully recovered in F1 and was probably responsible of the respective free radical scavenging activities of CWP and F1. It was noteworthy that the basic proteins, IgGs, CWBP and Lf did not possess interesting scavenging power. Particularly, Lf did not show any detectable radical scavenging activity (Table 1). The basic amino acid residues Lys and Arg are not reported to be efficient free-radical scavengers (Hernandez-Ledesma et al., 2005) and might not confer such activity to the basic proteins containing them.

Table 1. IC_{50} and TEAC values of gallic acid, camel whey proteins (CWP) and the different chromatographic fractions.
n.d.: not determined.

| Sample | IC_{50} (g L ⁻¹ eq. BSA) | TEAC (μmol TE μmol ⁻¹) |
|-------------|---------------------------------------|------------------------------------|
| Gallic acid | 2 μmol L ⁻¹ | 3 |
| CWP | 0.15 | n.d. |
| F1 | 0.20 | 1 |
| F2 | 0.45 | n.d. |
| F3 | 0.35 | n.d. |
| F4 | 0.31 | n.d. |
| F5 | 0.3 10 ⁶ | 0.01 |

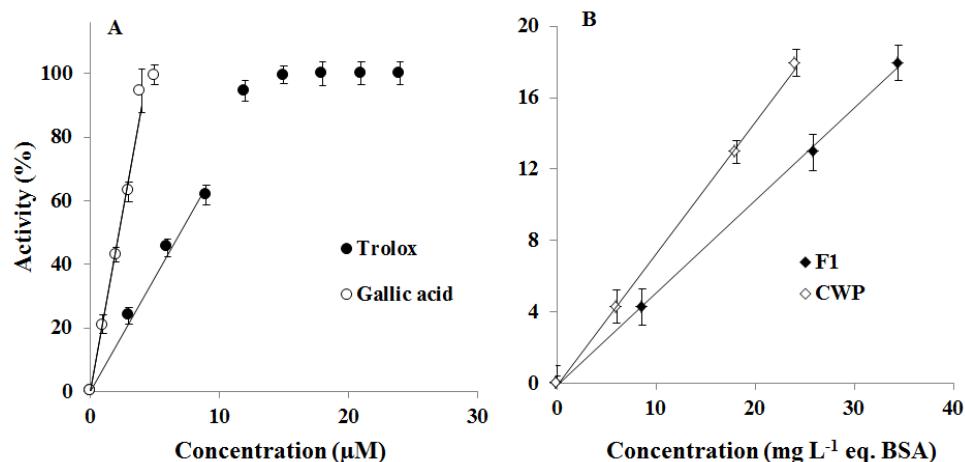


Figure 2. ABTS•+ radical scavenging activity determined at 740 nm of (A) Trolox and gallic acid, (B) the camel whey proteins (CWP) and the fraction F1 recovered from the cation-exchange chromatography separation of CWP. The equations of the curves are: (A) $y = 22.24 x$ ($R = 0.99$) for gallic acid and $y = 7.146 x$ for Trolox ($R = 0.99$) and (B) $y = 0.737 x - 0.118$ for CWP ($R = 0.99$) and $y = 0.516 x - 0.118$ for F1 ($R = 0.9$).

The TEAC of a protein mixture could not be compared to another one (the TEAC depends on the molecular mass of the compound tested rather than its weight expressed in g). However, the TEAC of F1 and F5 was calculated on the basis of the molecular masses of α -LA and Lf, the two proteins being considered to be the principal compounds of F1 and F5, respectively.

The TEAC value of F1 was 1 μmol TE μmol⁻¹, showing that the antiradical power of F1 was identical to that of Trolox. This value was, however, lower than that found by Salami et al. (2009) for the camel α -LA (3 μmol L⁻¹). This difference could be explained by the fact that F1 was a mixture of several proteins that underestimated the TEAC value of α -LA contained in F1. Recently, Sadat et al. (2011) have reported that bovine α -LA is a source of five highly antioxidant peptides and amongst them, Leu-Asp-Gln-Trp and Ile-Asn-Tyr-Trp exhibit remarkable free radical-scavenging activities towards ABTS^{•+}. These two peptides possess a Trp residue at their

carboxy-terminal extremity. According to Tsopmo et al. (2011), in the presence of free radicals, Trp can lose the labile hydrogen linked to the nitrogen of its indole ring leading to produce a radical stabilized by electron delocalization. For these authors, Trp plays a crucial role in the ability of proteins or peptides to scavenge free radicals. The addition of an extra Trp residue at the amino-terminal extremity of peptide Ile-Ser-Glu-Leu-Gly-Trp significantly increases its antioxidant power (Tsopmo et al., 2011). The camel α -LA possesses five Trp residues on its sequence, whereas the bovine counterpart contains only four. The presence of an additional Trp residue in the case of the camel sequence might contribute to its better radical scavenging power than that of the bovine protein reported by Salami et al. (2009).

On the other hand, Salami et al. (2010) have reported that CWP are a source of hydrolysate with significantly higher free radical-scavenging properties than bovine whey protein hydrolysate. Hernandez-Ledesma et al. (2005) have reported

that the lowering of the number of peptide bonds has an increasing effect on the antioxidant activity of the constituent amino acids of small peptides (typically with molecular masses lower than 1000 Da). In the case of the bovine species, α -LA hydrolysate obtained by thermolysin action displays a high and similar free radical-scavenging power than the source protein, since no improvement of the activity has been observed after enzyme treatment (Sadat et al., 2011). By taking into consideration the results reported by Salami et al. (2010), it would be thus interesting to determine in a further work if enzyme hydrolysis of the camel proteins contained in F1, mainly α -LA, would be required to enhance the antioxidant activity of this protein fraction.

The TEAC value (0.01 $\mu\text{mol TE } \mu\text{mol}^{-1}$) of pure Lf eluted in F5 was very low indicating that this protein did not possess any antiradical properties. It is however reported that Lf possesses antioxidant properties. In fact, these properties are rather related to its capacity to bind iron and therefore to inhibit the Fenton reaction than to any free radical-scavenging activity (Belizy et al., 2001). The main property of Lf is that it is a source of antimicrobial peptide named lactoferricin (Lfcin; Gifford et al., 2005). The Lfcin is produced by the gastric protease pepsin and it would be thus interesting to investigate the possibility of camel Lf to be a source of Lfcin-like peptide.

Conclusion

The cation-exchange chromatography enabled us to produce an α -LA-enriched fraction that was not retained on the column. Thus, this method may be adapted for high volumes of camel whey with *e.g.* fractionation onto CM Sephadex medium to readily prepare large quantities of α -LA-enriched fraction. The latter displayed a greater antioxidant power and might therefore have capability to attenuate oxidative stress occurring in IBD after oral administration. The other fractions did not display any interesting free radical-scavenging activity and this might seem to be related to their basic property. However, the strongest adsorbed protein, Lf, was recovered almost pure and may be used for its various biological activities *i.e.* antimicrobial, antithrombotic and immunomodulatory effects, whereas the intermediate fractions containing the heavy-chain IgG2 and IgG3 may also be valorized in immunotherapy.

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REGULAR ARTICLE

Nutritional value and sanitary evaluation of raw Camel's milk

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Abstract

The present study was carried out to investigate the nutritional value and hygienic status of fresh camel's milk collected for a period of 12 weeks (on weekly basis). The milk samples were divided into two portions under sterile conditions. The 1st portion was examined for the gross composition (total solids, solids non fat, moisture, fat, protein, lactose and chloride). The 2nd portion was examined for the sanitary condition through monitoring sensory evaluation, acid value and determination of fecal contamination. Wide variation was observed in the chemical analysis of the different milk constituent. The global mean values of total solids, solids non fat, fat, protein, lactose, chloride, and moisture were 10.8 ± 0.3 , 7.9 ± 0.2 , 2.84 ± 0.2 , 4.02 ± 0.1 , 3.8 ± 0.1 , 0.15 ± 0.003 , and $89.5 \pm 0.4\%$ respectively. The results of sensory evaluation indicated that the color was the most accepted attribute has the best score 7.9 and graded very good, then odor scored 6.8 and graded as slight good. The taste, over all acceptability (OAA) and flavor had fair grades and scored 5.4, 5.4 and 5.3 respectively. The average content of titratable acidity was $0.21 \pm 0.01\%$. The bacteriological analysis revealed that coliforms, fecal coliforms and *E. coli* were detected among the study period with incidence varied from 28.6 to 100% for coliforms and 28.6 to 71.4% for both fecal coliform and *E. coli*. Also, this study revealed presence of a relation between frequency distribution of coliforms and sensory scores.

Key words: Camel's milk, Nutritional value, Sanitary, Sensory evaluation, Growth composition

Introduction

In Egypt the majority of people consume cow's milk regularly than camel milk, due to the fact that cows and buffalos give much more milk and require less maintenance and labor. Unfortunately, people are unaware about the nutritional facts and healthy benefits of camel's milk. Camel's milk composition is different from that of ruminants (Al-Haj and Al-Kanhal, 2010) as is their physiology (Shabo et al., 2005). The value of camel's milk is due to its high concentration of volatile acid especially linoleic acid and poly unsaturated fatty acid which are essential for human nutrition, rather it is rich in mono-unsaturated fatty acid (Gast et al., 1969; Karry et al., 2005; Konuspayeva et al., 2008). Camel's milk is regarded to be abundant source of protein for people living in arid lands of the world. This protein is rich in protective component include lysozyme, lactoferrin, Lactoperoxidase

(LP) and peptidoglycan recognition protein (PGRP) which only detected in camel's milk (Singh et al., 2006), IGA and IgG immunoglobulins that are compatible with human ones and provide effective defense against several viral and bacterial pathogens (Khitam, 2003). The fact that camel's milk is low in different β -caseins (Beg et al., 1986) and without β -lactoglobulin (Merin et al., 2001) the 2 powerful allergens in cow's milk makes it attractive for those suffering from milk allergies (Mankinen and Palosuo, 1992; Shabo et al., 2005). Camel's milk is a rich source of chloride (Khaskheli et al., 2005) and its lactose is easily metabolized by persons suffering from lactose intolerance (Hanna, 2001). The vitamin C levels are more than three times that of cow milk and one-and-a-half that of human milk (Konuspayeva et al., 2011). Camel's milk is also having low sugar, low protein and high minerals (sodium, potassium, iron, copper, zinc, selenium and magnesium) (Konuspayeva et al., 2008). Camel's milk consumption may also be helpful in reducing the nutritional deficiencies and morbidities in adult community (Agrawal et al., 2005; Singh et al., 2009).

In Egypt camel's milk is produced in traditional way by hand milking, handled and transported under low hygienic measures. In view of its health benefits, there is a fast growing demand for raw

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camel's milk around the world (Faye and Bonnet, 2012) and further it is introduced recently as a new functional food in the European market. Therefore, there is a high necessity to find out about the present hygienic situation and nutritional value of raw camel's milk in Egypt.

The objective of this work was to study the nutritional value and sanitary condition of raw camel's milk.

Materials and Methods

Animals

It is extremely difficult to study a large number of camels on a regular basis taking into account the distance between the study area and the laboratory, lack of sufficient number of camels at one place and continues movement of herds. Therefore, this study was conducted on seven lactating dromedary she-camels (*Camelus dromedaries*) from a private camel herds belonging to Ebel El-Kher farms in Marsa Matroh, Egypt reared under satisfactory conditions and grazing on natural grass that grow in the desert

Samples collection

Seven fresh raw camel's milk samples (250 ml each) were collected individually weekly for 12 weeks. The samples were kept in ice box during transportation to the laboratory where they examined as soon as possible with a minimum of delay. Every individual sample thoroughly stirred before the analysis to obtain representative result for chemical and microbiological parameters.

Chemical analysis of camel's milk

Total solids (T.S. %)

A total solid was carried according to AOAC (1990).

Ten ml of camel's milk sample were placed in a previously weighed flat bottom porcelain dish (w), and then placed on a steam bath for 15 min, followed by heating in hot air oven at 100°C for 3 h. Heated samples were placed in a dissector for cooling then weighing (w'). Reading was taken at constant weight. T.S. % was calculated according to the following equation:

$$T.S. \% = \frac{(Weigh\ of\ dish + milk) - weigh\ of\ dish}{Weigh\ of\ sample} \times 10$$

Determination of moisture %

It was calculated by subtracting T.S. % from 100.

Determination of fat % (APHA, 1985)

Gerber method was used to determine fat %. Briefly, 10 ml of concentrated sulphuric acid were

placed into a clean and dry milk butyrometer, 11 ml of camel's milk sample were added followed by adding 1 ml of amyl alcohol into the butyrometer. The rubber stopper was firmly inserted and the butyrometer was shaken longitudinally very carefully and inverted several times until the curd is digested. The butyrometer was then placed in centrifuge and spun at 1500 rpm for 4 min. After which the fat content was read on the butyrometer scale at the lower part of meniscus.

Determination of solids non fat % (S.N.F. %)

S.N.F.% was calculated by subtracting the fat% from T.S.% and calculated according to the following equation:

$$S.N.F. \% = T.S. \% - \text{fat}\%$$

Determination of protein %

Total Protein % was determined by formal titration method modified by Mumm (1970). Twenty-five ml of milk sample was added into a beaker. Then, 1 ml potassium oxalate solution (28%) and 0.25 ml phenolphthalein (2%) was added into the milk. After mixing, the solution was titrated against NaOH (N/7) until faint pink color appeared, and then 5 ml of neutralized formalin solution (40%) was added to the beaker in which the faint pink color disappeared. A second titration against NaOH (N/7) was preformed until the faint pink color appears again and the second reading was recorded as protein%.

Lactose % (Harvey and Hill, 1967)

Lactose % was estimated by quantitative Benedict method. In a cylinder (100 ml capacity), 10 ml milk sample, 40 ml distilled water, 10 ml of sulphuric acid 2/3N, 5 ml of sodium tungestate 10% were added. The mixture was brought up to 100 ml by addition of 35 ml distilled water. The cylinder was left to stand for 10-15 min to allow the formation of precipitate. The solution was then filtered through filter paper and a clear filtrate was then transferred to burette. 25 ml of standard Benedict solution, 5 g anhydrous sodium carbonate and 50 ml distilled water were added in a porcelain dish. The mixture was boiled and titration against the filtrate was carried out during boiling until disappearance of blue color and appearance of white precipitate. The reading was recorded and multiplied by factor 0.067 (Each 0.067 g lactose reduces 25 ml Benedict).

$$\text{Lactose \%} = 67/R$$

Chloride % (Ling, 1963)

10 ml milk sample, 5 ml nitric acid 25% (freshly prepared), 5 ml silver nitrate N/10, 1 ml saturated iron alum solution (indicator). The

solution was mixed thoroughly by glass rod and titrated against ammonium thiocyanate N/10 until brownish color (end point) was obtained and persisted for 1-2 min.

$$\text{Chloride\%} = (5 - R) \times 0.003546 \times 10$$

1 ml silver nitrate N/10 = 1 ml ammonium thiocyanate

$$1 \text{ ml silver nitrate N/10} = 0.003546 \text{ g chloride}$$

R = amount of thiocyanate N/10

5 = amount of silver nitrate N/10

5 - R = amount of silver nitrate N/10 combined with chloride

Sanitary evaluation of camel's milk

2.2.1. Sensory evaluation: All camel's milk samples were sensory evaluated by untrained panelists using a 9-points hedonic scoring scales (9 = excellent, 8 = very good, 7 = good, 6 = slightly good, 5 = fair, 4 = slightly bad, 3 = bad, 2 = very bad, 1 = extremely bad) (Abdel Rahman et al., 2009). The samples were evaluated for color, smell, taste, flavor and overall acceptability (OAA). Also the panelists were asked to list any defects in the samples. All samples were subjected to clot on boiling test before testing its flavor and taste.

Determination of acidity value (Pearson, 1972)

Ten ml of well mixed camel's milk sample were placed into a clean dry beaker then 1 ml phenolphthalein 0.5% was added and titrated against NaOH N/10 until faint pink color appeared and persisted for at least 5 sec (end point) and the reading was recorded.

$$\text{Lactic acid \%} = R/10.$$

Examination of camel's milk for fecal contamination: according to AOAC (1975)

Preparation of milk samples: Camel's milk samples were stirred thoroughly several times and then 10 ml was added to 90 ml of sterile peptone water (1/10 dilution), in which decimal serial dilutions were prepared according to APHA (1992).

Coliform count, fecal coliform count and E.coli count were determined using three tubes most probable number (MPN) method.

Coliforms count (MPN/ml)

Presumptive test: 1 ml of the previous prepared 1:10, 1:100 and 1:1000 dilutions was inoculated into 3 replicate tubes of lauryl sulphate tryptose (LST) broth supplied with inverted Durham's tubes. The inoculated tubes were incubated at 35°C and scored for gas formation at 24 and 48 hr.

Confirmatory test: All positive LST tubes were subculture into brilliant green lactose bile (BGLB) broth with inverted Durham's tube by means of 3

mm loop and were incubated at 35°C for 48±2 hr. the most probable number for total coliform bacteria per ml was computed by scoring the number of gas positive BGLB tubes at each dilution and calculated from MPN table.

Fecal coliforms count (MPN/ml): Using a 3 mm loop, samples from gassing BGLB tubes were transferred to EC broth tubes with inverted Durham's tubes and incubated at 45.5 °C in covered water bath for 48±2 hr.

E. coli count (MPN/ml)

Gas positive EC broth tubes were streaked to Levine's eosin methylene blue (LEMB) agar plates and incubated at 35°C for 24±2 hr. typical nucleated dark center colonies with metallic sheen were considered to be *E. coli* positive and were selected for confirmation.

Data analysis: were expressed as mean ± standard error using SAS program (SAS, 1997).

Results and Discussion

Chemical analysis of camel's milk

Compositional analysis of fresh camel's milk was carried out for a period of twelve weeks (on weekly basis). Mean values for total solid contents of camel's milk varied from 9.7±0.3 to 12.5±0.7% with grand mean of 10.8±0.3% (Table 1). These results were comparable to Faye et al. (2008) and Farah (1993) while, they were lower than those reported by Moustafa et al. (2000); El Shaer and El Ganzoury (2008); and higher than what reported by Omer and El-Tinay (2009); Shuiet et al. (2008).

The moisture content mean values which varied from 87.5±0.8 to 91.6±0.6% with grand mean of 89.5±0.4% is in agreement with results of Omer and El-Tinay (2009) and Meiloud et al. (2011).

The grand mean of fat (in %) in camel's milk was 2.8±0.2 and ranged from 2±0.1 to 3.4±0.3% (Table 1). Fat content obtained in this study agreed with the value reported by Shuiet et al. (2008); Haddadin et al. (2008); Meiloud et al. (2011) while, Attia et al. (2001); Omer and El-Tinay (2009) reported lower values. Our results were lower than those reported by Al-Haj and Al-Kanhal (2010) and Konuspayeva et al. (2009). The grand mean value of S.N.F. was 7.9±0.2% and ranged from 7.1±0.6 and 9.5±0.8%. This result was similar to those recorded by Guliye et al. (2000) and Mal et al. (2006, 2007) and lower than those recorded by Iqbal et al. (2001) and El Zubeir and Ibrahim (2009). Camel's milk is considered to be abundant source of protein for people living in arid lands of the world. Our results showed that the grand mean value of protein was found to be 4.02±0.1% and

ranging from 3 ± 0.3 to $4.5\pm0.2\%$ (table 1). Protein content recorded in this study was agreed with the value reported by Faye et al. (2008) and Konuspayeva et al. (2010) while it was higher than that reported by Guliyev et al. (2000); Moustafa et al. (2000); Iqbal et al. (2001); El Shaer and El Ganzoury (2008) and El-Zubier and Ibrahim (2009). Lactose is the major carbohydrate in the milk. The average lactose content was $3.8\pm0.1\%$ and varied between 3.3 ± 0.2 to $4.7\pm0.3\%$. These results were comparable to Haddadin et al (2008); Bakheit et al., (2008) and were lower than that recorded by Guliyev et al (2000). The chloride content of camel's milk as shown in Table 1 varied from 0.14 ± 0.008 and $0.16\pm0.003\%$ with grand mean of $0.15\pm0.003\%$. These results were in the same line with Moustafa et al. (2000), while Khaskheli et al. (2005) recorded a higher result.

In general, the present study showed wide variations in the gross composition of camel's milk. These variations could be due to several factors including analytical measurement procedures, water availability, stage of lactation, age, breeds and number of calving, camel's diet and climate. Our study was done in the period from June to September, i.e. at summer time. Yet, camel having a seasonal reproductive cycle, the summer time is corresponding with the lactation peak when fat and protein in milk are at their lower values (Musaad et al., 2013).

Sanitary evaluation of camel's milk

Sensory evaluation

Good quality milk should have a pleasant sweet and clean flavor without distinct aftertaste

Camel's milk is generally opaque white (Yagil and Etzion, 1980; Desai et al., 1982), with normal odor and has faint sweet taste, a sweet but sharp (Ohri and Joshi, 1961), sometimes it is salty in taste (Rao et al., 1970; Desai et al., 1982). Due to practical reasons it was extremely difficult to recruit more people available to share in the sensory test on a regular basis for consecutive 12 weeks. Therefore, the sensory analysis of the examined camel's milk samples was performed by four untrained panelist comprising staff member and master student in the food hygiene department, faculty of veterinary medicine, Assuit University, Egypt. They were informed and trained to understand the used words such as flavor, OAA and sensory scores. Among all sensory attributes color had the best score during the twelve weeks with grand mean score 7.9 and were graded very good (Table 2). This may be attributed to the low content of carotene (Wernery, 2006); also camel's milk fat completely

homogenized giving the milk a smooth white appearance (Abu-lehia, 1998). Odor had grand mean score 6.8 and were graded slight good, both taste and over all acceptability (OAA) had the same grand mean score 5.4 and were graded fair. The flavor had the lowest grand mean score 5.3 and was graded fair.

Acid value

Measuring the acidity is an important test used to determined milk quality (AOAC, 1990). The grand mean value of acidity was $0.21\pm0.01\%$ and varied from 0.16 ± 0.01 to $0.27\pm0.03\%$ for a period of twelve weeks (Table 1). This result was in agreement with those recorded by El-Shaer and El-Ganzoury (2008); El-Zubier and Ibrahim (2009). Titratable acidity in the present study was higher than those recorded in other studies. This might be due to the relatively high temperature of milk after collection (Yagil and Etzion, 1980).

Fecal contamination of camel's milk

It is worth to mention that there are no microbiological standards specified to camel's milk. Therefore, the microbiological limit value for cow's milk is used to assess the quality of camel's milk (El-Ziney and AL-Turki, 2007). In this study, the microbiological results of camel's milk samples were compared with parameters laid down by European Union (EU) standards commission (Anonymous, 1992).

Most of examined samples were positive for total coliforms. The highest prevalence were found between the 3rd to 8th weeks (100%) for total coliforms, 2nd and 11th weeks (71.4%) for fecal coliforms and in the 11th week (71.4%) for *E.coli*. Table 4 shows the microbial distribution in the camel's milk among the twelve weeks. The highest frequency distribution (71.4%) for total coliforms was $<10^2$ in the 7th week, $<10^3$ in the 3rd week and $<10^4$ in the 5th and 10th weeks. While the highest frequency for fecal coliforms and *E. coli* (71.4%) was <10 in the 11th week. The existence of coliforms bacteria may not necessary to indicate direct fecal contamination of milk but precisely as an indicator for poor sanitary practices during milking and further handling processes. More over the presence of fecal coliforms i.e. *E. coli* implies the risk of fecal contamination and possibility of enteric pathogens existence.

Table 1. Chemical composition of camel's milk (n=7) representing 12 consecutive weeks.

| W | T.S.±S.E. | S.N.F.±SE | Moisture±SE | Chloride±SE | Fat±SE | Protein±SE | Lactose±SE | T.A. |
|----|-----------|-----------|-------------|-------------|----------|------------|------------|-----------|
| 1 | 9.7±0.3 | 7.3±0.3 | 90.3±0.3 | 0.14±0.008 | 2.4±0.08 | 4.5±0.2 | 3.7±0.3 | 0.27±0.03 |
| 2 | 12.5±0.7 | 9.5±0.8 | 87.5±0.8 | 0.15±0.003 | 3±0.3 | 4.1±0.2 | 4.7±0.3 | 0.18±0.01 |
| 3 | 11.2±0.2 | 7.9±0.2 | 91.6±0.6 | 0.16±0.002 | 3.3±0.2 | 4.5±0.2 | 4.5±0.2 | 0.17±0.01 |
| 4 | 10.7±0.6 | 7.8±0.6 | 89.3±0.6 | 0.15±0.003 | 3±0.2 | 3±0.3 | 3.9±0.1 | 0.16±0.01 |
| 5 | 11.01±0.7 | 8.2±0.6 | 89±0.7 | 0.16±0.002 | 2.8±0.2 | 3.8±0.1 | 3.8±0.02 | 0.2±0.01 |
| 6 | 10.1±0.4 | 7.5±0.5 | 89.9±0.4 | 0.15±0.003 | 2.6±0.2 | 4.5±0.2 | 3.7±0.07 | 0.2±0.03 |
| 7 | 9.7±0.6 | 7.1±0.6 | 90.3±0.6 | 0.15±0.003 | 2.5±0.1 | 3.4±0.3 | 3.5±0.03 | 0.2±0.03 |
| 8 | 11.2±0.4 | 8.7±0.6 | 88.8±0.4 | 0.16±0.001 | 2.5±0.3 | 4.5±0.2 | 3.6±0.07 | 0.21±0.01 |
| 9 | 9.7±0.5 | 7.7±0.6 | 90.3±0.5 | 0.16±0.002 | 2±0.1 | 3.7±0.2 | 3.8±0.2 | 0.19±0.02 |
| 10 | 11.4±0.4 | 8.08±0.2 | 88.6±0.4 | 0.16±0.002 | 3.4±0.3 | 4.1±0.3 | 3.3±0.2 | 0.27±0.01 |
| 11 | 10.7±0.3 | 7.5±0.2 | 89.3±0.3 | 0.16±0.002 | 3.2±0.2 | 4.1±0.2 | 3.4±0.2 | 0.22±0.02 |
| 12 | 11.1±0.4 | 7.7±0.3 | 88.8±0.4 | 0.16±0.002 | 3.4±0.2 | 4.1±0.1 | 3.5±0.07 | 0.19±0.01 |
| GM | 10.8±0.3 | 7.9±0.2 | 89.5±0.4 | 0.15±0.003 | 2.8±0.2 | 4.02±0.1 | 3.8±0.1 | 0.21±0.01 |

T.S = total solids, S.N.F = solids non fat, T.A. = Titratable acidity

Table 2. Sensory evaluation scores* of camel's milk (n=7) representing 12 consecutive weeks.

| W | Color | Taste | Flavor | Odor | OAA |
|----|----------|-----------|---------|-------------|-------------|
| | Score | Grade | Score | Grade | Score |
| 1 | 6.9±0.5 | Good | 4.7±0.5 | Slight bad | 5.4±0.4 |
| 2 | 8 | Very good | 5.3±0.2 | Fair | 5±0.3 |
| 3 | 8 | Very good | 4.9±0.3 | Slight bad | 4.6±0.2 |
| 4 | 8 | Very good | 6.1±0.3 | Slight good | 5.7±0.2 |
| 5 | 8 | Very good | 5.3±0.2 | Fair | 4.7±0.3 |
| 6 | 8 | Very good | 5.4±0.2 | Fair | 5.4±0.2 |
| 7 | 8 | Very good | 5.6±0.2 | Fair | 5.7±0.1 |
| 8 | 8 | Very good | 5.1±0.2 | Fair | 5±0.1 |
| 9 | 8 | Very good | 6±0.1 | Slight good | 5.6±0.2 |
| 10 | 8 | Very good | 5.1±0.2 | Fair | 4.9±0.2 |
| 11 | 8 | Very good | 5.6±0.3 | Fair | 5.1±0.3 |
| 12 | 8 | Very good | 6±0.1 | Slight good | 6±0.1 |
| GM | 7.9±0.08 | very good | 5.4±0.1 | Fair | 5.3±0.1 |
| | | | | | 6.8±0.1 |
| | | | | | Slight good |
| | | | | | 5.4±0.08 |
| | | | | | Fair |

*scores using 9 point hedonic scales (9= excellent, 8= very good, 7=good, 6= slight good, 5=fair, 4= slight bad, 3= bad, 2= very bad, 1=extremely bad) *
OAA= Over all acceptability.

Table 3. Weekly incidence of coliforms, fecal coliforms and *E.coli* in camel's milk samples.

| Weeks | Total coliforms Positive samples | | Fecal coliforms Positive samples | | <i>E.coli</i> Positive samples | |
|-------|----------------------------------|------|----------------------------------|------|--------------------------------|------|
| | No. | % | No. | % | No. | % |
| 1st W | 5 | 71.4 | 3 | 42.8 | 2 | 28.6 |
| 2ndW | 6 | 85.7 | 5 | 71.4 | 3 | 42.8 |
| 3rd W | 7 | 100 | 3 | 42.8 | 3 | 42.8 |
| 4th W | 7 | 100 | - | - | - | - |
| 5th W | 7 | 100 | 2 | 28.6 | - | - |
| 6th W | 7 | 100 | 3 | 42.8 | 2 | 28.6 |
| 7th W | 7 | 100 | - | - | - | - |
| 8th W | 7 | 100 | - | - | - | - |
| 9th W | 6 | 85.7 | - | - | - | - |
| 10tW | 6 | 85.6 | 1 | 14.3 | 1 | 14.3 |
| 11tW | 6 | 85.6 | 5 | 71.4 | 5 | 71.4 |
| 12tW | 2 | 28.6 | 2 | 28.6 | 2 | 28.6 |

Table 4. Frequency distribution of positive camel's milk samples based on their coliforms, fecalcoliforms and *E.coli* cfu/ml.

| Weeks | Total coliforms | | | | | | | | Fecal coliforms | | | | <i>E.coli</i> | | | |
|-------|-----------------|------|------------------|------|------------------|------|------------------|------|-----------------|------|------------------|------|---------------|------|------------------|------|
| | <10 | | <10 ² | | <10 ³ | | <10 ⁴ | | <10 | | <10 ² | | <10 | | <10 ² | |
| | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % |
| 1st W | 2 | 28.6 | 3 | 42.8 | - | - | - | - | 2 | 28.6 | 1 | 14.3 | 1 | 14.3 | 1 | 14.3 |
| 2ndW | - | - | 1 | 14.3 | 3 | 42.8 | 2 | 28.6 | 2 | 28.6 | 3 | 42.8 | 3 | 42.8 | - | - |
| 3rd W | - | - | 1 | 14.3 | 5 | 71.4 | 1 | 14.3 | - | - | 3 | 42.8 | - | - | 3 | 42.8 |
| 4th W | 2 | 28.6 | 3 | 42.8 | 1 | 14.3 | 1 | 14.3 | - | - | - | - | - | - | - | - |
| 5th W | - | - | - | - | 2 | 28.6 | 5 | 71.4 | 1 | 14.3 | 1 | 14.3 | - | - | - | - |
| 6th W | - | - | 2 | 28.6 | 2 | 28.6 | 3 | 42.8 | 3 | 42.8 | - | - | 2 | 14.3 | - | - |
| 7th W | - | - | 5 | 71.4 | 2 | 28.6 | - | - | - | - | - | - | - | - | - | - |
| 8th W | - | - | 3 | 42.8 | 1 | 14.3 | 3 | 42.8 | - | - | - | - | - | - | - | - |
| 9th W | - | - | 4 | 57.1 | 2 | 28.6 | - | - | - | - | - | - | - | - | - | - |
| 10tW | - | - | - | - | 1 | 14.3 | 5 | 71.4 | - | - | 1 | 14.3 | - | - | 1 | 14.3 |
| 11tW | 3 | 42.8 | 2 | 28.6 | 1 | 14.3 | - | - | 5 | 71.4 | - | - | 5 | 71.4 | - | - |
| 12tW | - | - | 2 | 28.6 | - | - | - | - | 1 | 14.3 | 1 | 14.3 | 2 | 28.6 | - | - |

Sensory analysis is a powerful tool in its own right for quality assurance (Q A). However coupling sensory analysis with chemical and microbiological analysis data can provide even more insights than using either technique alone. Total coliforms recorded the lowest count (28.6%) in the 12th week and it had the best scores for taste, odor, flavor and over all acceptability. Similar sensory score were recorded for milk samples of the 4th, 7th, 8th and 9th. These samples were negative fecal coliforms and *E. coli* and this give an indication that sensory evaluation could be guide for the microbiological level of milk. Color of milk in this study couldn't be used for the judgment as it record high score for all samples. This may be attributed to the low content of carotene (Wernery, 2006); also camel's milk fat completely homogenized giving the milk a smooth white appearance (Abu-lehia, 1998). In the current study there is no relation between chemical and sensory parameters and they are completely independent.

Conclusion

In the present study on limited number of animals, fresh camel's milk had good nutritional values and unique flavor, sensory attributes as color, taste, flavor, odor and OAA. Extensive studies are needed to establish Egyptian standard of chemical parameters for camel's milk. So, it is strongly recommended to apply milking protocol, hygiene measures and sanitization programs to control the contamination of camel's milk during collection, storage, transportation as required for any other milk destined to human consumption.

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REGULAR ARTICLE

Microflora identification of fresh and fermented camel milk from Kazakhstan

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Abstract

In Kazakhstan where Bactrian camel, dromedary camel and their hybrids are cohabiting within same farms, the consumption of camel milk is very popular because its medicinal and dietary properties. This milk is consumed under fermented form, called *shubat*. *Shubat* is still very often made on a small scale in the steppe with a fermentation step driven by wild bacteria. Camel milk and *shubat* were sampled from 4 regions with high number of camel population. As the whole, 26 samples were obtained from 13 selected farms representing the variability of the farming system. Isolated LAB strains were identified by method of a polymorphism determination of 16S ribosome DNA. PCR with using two different pairs of amores (338f/518r; W001/23S1) was done. Majority of microflora were cocci in a both milk products. The following microorganisms were identified: *Enterococcus durans*; *Enterococcus faecalis*; *Enterococcus faecium*; *Lactobacillus casei*; *Lactobacillus casei* subsp. *casei*; *Lactobacillus curvatus*; *Lactobacillus kefiri*; *Lactobacillus paracasei*; *Lactobacillus sakei*; *Lactococcus lactis* subsp. *lactis*; *Leuconostoc mesenteroides*. Diversity of microorganisms in a both products was similar, but percentage of each microorganism changed during fermentation process. Yeast biodiversity in *shubat* was studied by using denaturing gradient gel electrophoresis (DGGE). Target DNA bands were identified according to the reference species scoring. Comigrating bands present in the DGGE profiles were resolved by species-specific PCR. The dominant yeasts in both products included *Kazakhstania unispora*, *Saccharomyces cerevisiae* and *Kluyveromyces marxianus*. Frequently isolated yeast species were *Dekkera bruxellensis* and more rarely *Galactomyces geotrichum*. The results of microflora identification in these products provide a theoretical foundation for developing starter cultures.

Key words: Camel, Fermented camel milk (*shubat*), LAB, Yeast, PCR, DDGE, Kazakhstan

Introduction

Shubat, which is made from unpasteurized fresh camel milk, is the most popular fermented dairy beverage in Kazakhstan. This traditional fermented product is widely consumed also in Mongolia, Uzbekistan, Turkmenistan and some regions of Russia (Konuspayeva and Faye, 2011). For centuries, *shubat* has been regarded not only as an essential food, but also as a nutrient and a medicinal remedy (Urazakov et Bainazarov, 1974; Mal et al., 2000; Mohamad et al., 2009;

Konuspayeva et al., 2003; Yagil et Creveld, 2000; Djangabilov et al., 2000; Chuvakova et al., 2000). Lactic acid bacteria (LAB) and yeasts were proven to be the main components in fermentation process. They play detrimental role to the safety of dairy products. Moreover, the benefits of *shubat* are mainly attributable to these microorganisms which not only were reported to play a major fermentative role on the aroma, texture, and acidity of this product, but also play a major therapeutic role on improvement of digestion properties, against diarrhea and responsible for antimicrobials properties (Puzyrevskaya et al., 2000; Saubenova et al., 2002). The specific microflora of *shubat* directly depends from fresh milk, utilized starters and fermentation conditions (Serikbayeva et al., 2005). In particular, differences in microflora composition of conventional starters originating

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from the respective family environment will result in *shubat* quality instability. Nowadays studying microflora of traditional fermented dairy products as *shubat* and creation of starters is very important. To obtain the *shubat* of better quality and to produce this traditionally fermented product on the industrial level with high quality control starter cultures should be developed. The first step of such ambitious project is the identification of the main microflora strains available in *shubat* of different origin which is the objective of the present paper.

Materials and Methods

Dairy products sampling

Four regions (Almaty, South Kazakhstan, Kyzylorda and Atyrau) of the Kazakhstan were selected according to their importance of camel livestock. As the whole, 13 farms were selected representing the variability of the farming system in the retained regions and overall producing *shubat* with different known organoleptic quality. Each sample (n=26, i.e. two samples per farm) was aseptically transferred to a 500 ml sterile bottle,

transported in ice-box until the laboratory and stored at 4°C.

Microorganisms and growth conditions

LAB strains were isolated on the nutritive media M17 and MRS (Biokar Diagnostics, France) and yeasts on the Saburo media (Himedia, India). The transfers were repeated until to get pure colonies. The pure colony was inoculated in the respective media and conserved at 4°C after incubation at 37°C for LAB and 25°C for yeasts, 48 hours. For long term maintenance of isolates, stock cultures were stored at - 20°C in 30% (v/v) glycerol, with 70% (v/v) M17, MRS and Saburo broth, respectively.

Preliminary identification of microorganisms

The pure strains were characterized by coloration Gram (reagent kit "Color Gram2-E" BioMérieux, France), catalase tests (ID color catalase ID-ASE Biomérieux France) and oxydase tests (Oxydase reagent Biomérieux, France).



Figure 1. Map of Kazakhstan, showing the locations of Almaty, Atyrau, Kyzylorda and South Kazakhstan sampled regions.

DNA extraction

Bacterial DNA extraction was done according to the manual method described by Leesing (2005). Extraction of the yeasts DNA was achieved by using commercial Wizard kit (Promega, France). The DNA extracted was then stored at -20°C. Existence and purity of DNA was verified by electrophoresis in 0.8% (w/v) agarose gel (Promega, France) in TAE 1X buffer.

Amplification of DNA by PCR

The method of a polymorphism determination of 16S ribosome DNA was used. The PCR samples were prepared by performing 2 successive PCR using a DNA Peltier thermal cycler PTC-100 (MJ Research Inc., USA). Firstly, a 237-bp fragment of the 16S rDNA including the V3 region (in *Escherichia coli*, which corresponds to position (338-534) was amplified with primers 338f (5'-ACTCCTACGGGAGGCAGCAG-3') and 518r (5'-ATT ACC GCG GCT GCT GG-3') (Sigma-Genosys, France). Secondly, amores which amplifies the intergenic region (ITS: Internal Transcribed Spacer) between the regions coding RNA16S and RNA 23S (Turpin et al., 2011). A 1500-bp fragment was amplified with the primers W001 (5'- AGA GTT TGA TCM TGG CTC-3') and 23S1 (5'- CNC GTC CTT CAT CGC CT-3'). The PCR reaction mixtures and the 2 above amplification programs were the same as described previously (Ampe et al., 1999; Leesing, 2005) and (Turpin et al., 2011), respectively.

Yeast biodiversity in shubat was studied using polymerase chain reaction denaturing gradient gel electrophoresis (PCR-DGGE) fingerprinting. Target DNA bands were identified according to the reference species scoring, constructed in this study. Comigrating bands present in the DGGE profiles were resolved by species-specific PCR. For DNA amplification, two primers were used: NL1 (GCCATATCAATAAGCGGAGGAAAAG) and LS2 (ATTCCCAAACAACCGACTC) (Sigma-Genosys, France), respectively.

The sizes and quantities of PCR products were determined by 1% (w/v) agarose gel QA TM (Q-Biogene, USA) electrophoresis in comparison with a standard containing DNA fragments of defined length.

Purification and Sequencing of PCR bands

The corresponded bands were excised from the denaturing gels with sterile scalpel. The amplicons of PCR were purified with Wizard PCR Preps DNA Purification system kit (Promega, France) and

stored at -20°C. Sequencing was done by EUROFINS GENOMICS enterprise. Sequence annotation and database searches for similar sequences were performed by using BLAST at the National Center for Biotechnology Information (<http://www.ncbi.nlm.nih.gov/>) to determine the closest known relative species (Altschul et al., 1990).

Results

From the 26 *shubat* samples, 138 strains of microflora were isolated and among them only 37 LAB strains (Table 1) and 12 yeasts strains were identified. The majority of microflora among the 138 isolated strains was cocci (109), 17 bacilli and 12 yeasts. The percentage of similarity for the 37 LAB strains with their affiliations was above 80 % in all the cases except *Enterococcus faecium* (NC_017960.1) which was 81% only (Table 1).

The preponderance of cocci in lactic microflora of camel milk has been already reported by other authors (Grillet, 2006; Kacem et al., 2002). Khedid et al. (2009) listed the dominant species of camel milk as *Lactococcus lactis* subsp. *lactis* (17.5%), *Lactobacillus helveticus* (10%), *Streptococcus salivarius* sub sp. *thermophilus* (9.2%), *Lactobacillus casei* subsp. *casei* (5.8%), *Lactobacillus plantarum* (5%) and *Leuconostoc mesenteroides* subsp. *mesenteroides* (4.2%).

The predominance of enterococci in microflora of *shubat* in our results is in accordance with results of Zadi-Karam and Karam (2005) who, after analyzing eight samples of raw camel milk from eight different animals in five farms of Timimoune and Bechar (South-western Algeria) regions, found 35% of enterococci, *Lc. lactis* ssp *diacetylactis* (28.4%), *Lc. lactis* ssp *cremoris* (4.9%), *Lc. lactis* ssp *lactis* (1.2%), *Leuconostoc lactis* (7.4%), *Leuconostoc dextranicum* (4.9%) and *Lactobacillus plantarum* (18.5%). The presence of enterococci can also be caused by poor hygiene during milking (Khedid et al., 2009, Martin and Mundt, 1972 cited by Stiles and Holzapfel, 1997). For many authors, the presence of enterococci is evidence of possible fecal contamination and therefore a risk to consumers because although these strains are known for their low virulence, they pose serious health problems due to the emergence of many antibiotic-resistant strains, for example strains of *E. faecalis* (Giraffa et al., 2000 cited by Khedid et al., 2009). However, the positive role of these cocci in the development of quality of fermented dairy products should not be forgotten. For example, the proteolytic properties of these strains lead to the

release of casein amino acid precursors of molecules involved in the flavor of cheese (Urbach, 1995 cited by Khedid et al., 2009). Enterococci produce enterocins which have a specific inhibitory activity against some pathogenic bacteria (Sabia et al., 2002). It was also reported that *E. faecalis* produce anti-listeria bacteriocins in milk and cheese. Enterococci contribute significantly to the development of organoleptic properties of cheese mature (Litopoulou-Tzanetaki, 1990) and have a beneficial effect on the growth of other lactic acid bacteria in their proteolytic activity that promotes intense gas production by strains of *Leuconostoc*

and lactic acid production by lactococci, enterococci that's why it is used very often in cheese production in the Mediterranean countries (Macedo et al., 1995; Jovanovic and Sandine-Levata, 1996 cited Zadi-Karam et al., 2011).

Also, five yeasts species were identified in *shubat*. Among them, *Kazakhstania unispora*, *Saccharomyces cerevisiae* and *Kluyveromyces marxianus* (*Candida kefyr*) were predominant. More rarely isolated yeasts species were *Dekkera bruxellensis* (*Brettanomyces*) and *Galactomyces geotrichum*.

Table 1. Phylogenetic affiliations of LAB isolates recovered in *shubat* from four regions in Kazakhstan.

| No. | Closest 16S rRNA sequence in Gene bank | Accession no. | Similarity, % | Affiliation |
|-----|--|---------------|---------------|-------------------|
| 1 | <i>Leuconostoc mesenteroides</i> subsp. <i>mesenteroides</i> | NC_016805.1 | 92 | <i>Firmicutes</i> |
| 2 | <i>Leuconostoc mesenteroides</i> subsp. <i>mesenteroides</i> | NC_016805.1 | 100 | <i>Firmicutes</i> |
| 3 | <i>Leuconostoc mesenteroides</i> subsp. <i>mesenteroides</i> | NC_016805.1 | 97 | <i>Firmicutes</i> |
| 4 | <i>Enterococcus durans</i> | S000004741 | 98 | <i>Firmicutes</i> |
| 5 | <i>Enterococcus durans</i> | S000004741 | 98 | <i>Firmicutes</i> |
| 6 | <i>Enterococcus durans</i> | S000004741 | 99 | <i>Firmicutes</i> |
| 7 | <i>Enterococcus durans</i> | S000004741 | 100 | <i>Firmicutes</i> |
| 8 | <i>Enterococcus faecalis</i> | NC_004668.1 | 90 | <i>Firmicutes</i> |
| 9 | <i>Enterococcus faecalis</i> | NC_018221.1 | 99 | <i>Firmicutes</i> |
| 10 | <i>Enterococcus faecalis</i> | NC_018221.1 | 96 | <i>Firmicutes</i> |
| 11 | <i>Enterococcus faecalis</i> | NC_018221.1 | 99 | <i>Firmicutes</i> |
| 12 | <i>Enterococcus faecium</i> | NC_017960.1 | 81 | <i>Firmicutes</i> |
| 13 | <i>Enterococcus faecium</i> | NC_017960.1 | 95 | <i>Firmicutes</i> |
| 14 | <i>Enterococcus faecium</i> | NC_017960.1 | 99 | <i>Firmicutes</i> |
| 15 | <i>Enterococcus faecium</i> | S000002717 | 99 | <i>Firmicutes</i> |
| 16 | <i>Enterococcus faecium</i> | NC_017960.1 | 99 | <i>Firmicutes</i> |
| 17 | <i>Enterococcus faecium</i> | S000002717 | 99 | <i>Firmicutes</i> |
| 18 | <i>Enterococcus faecium</i> | NC_017960.1 | 98 | <i>Firmicutes</i> |
| 19 | <i>Enterococcus faecium</i> | S000002717 | 100 | <i>Firmicutes</i> |
| 20 | <i>Enterococcus faecium</i> | NC_017960.1 | 98 | <i>Firmicutes</i> |
| 21 | <i>Enterococcus hirae</i> | NC_018081.1 | 99 | <i>Firmicutes</i> |
| 22 | <i>Enterococcus hirae</i> | NC_018081.1 | 99 | <i>Firmicutes</i> |
| 23 | <i>Lactobacillus buchneri</i> | NC_018610.1 | 99 | <i>Firmicutes</i> |
| 24 | <i>Lactobacillus buchneri</i> | NC_018610.1 | 93 | <i>Firmicutes</i> |
| 25 | <i>Lactobacillus casei</i> | S000004550 | 98 | <i>Firmicutes</i> |
| 26 | <i>Lactobacillus casei</i> | S000008152 | 100 | <i>Firmicutes</i> |
| 27 | <i>Lactobacillus casei</i> | HE970764.1 | 98 | <i>Firmicutes</i> |
| 28 | <i>Lactobacillus casei</i> | S000008152 | 96 | <i>Firmicutes</i> |
| 29 | <i>Lactococcus lactis</i> subsp. <i>cremoris</i> | NC_017949.1 | 99 | <i>Firmicutes</i> |
| 30 | <i>Lactococcus lactis</i> subsp. <i>lactis</i> | NC_017486.1 | 98 | <i>Firmicutes</i> |
| 31 | <i>Lactococcus lactis</i> subsp. <i>lactis</i> | NC_017486.1 | 98 | <i>Firmicutes</i> |
| 32 | <i>Lactococcus lactis</i> subsp. <i>lactis</i> | NC_017486.1 | 100 | <i>Firmicutes</i> |
| 33 | <i>Lactobacillus sakei</i> subsp. <i>sakei</i> | NC_007576.1 | 99 | <i>Firmicutes</i> |
| 34 | <i>Lactobacillus sakei</i> | S000261305 | 100 | <i>Firmicutes</i> |
| 35 | <i>Lactobacillus sakei</i> subsp. <i>sakei</i> | NC_007576.1 | 95 | <i>Firmicutes</i> |
| 36 | <i>Lactobacillus sakei</i> subsp. <i>sakei</i> | NC_007576.1 | 95 | <i>Firmicutes</i> |
| 37 | <i>Lactobacillus sakei</i> subsp. <i>sakei</i> | NC_007576.1 | 100 | <i>Firmicutes</i> |

Saccharomyces cerevisiae has also been isolated by Njage et al. (2011) in African fermented camel milk (*suusac*). Gadaga et al. (2007) also founded *Saccharomyces cerevisiae* and *Candida kefyr* in *amasi* - naturally fermented cow milk from Zimbabwe.

The yeast *Dekkera bruxellensis* (*Brettanomyces*) is usually regarded as a contamination organism in wine production and distilleries. But in production of beer and sourdough it is a desirable member of microflora which plays a key role in the spontaneous fermentation and food flavor (Stender et al., 2001; Blomqvist et al., 2010). The yeast *Geotrichum candidum* which was identified in our study is appearing in the early stages of ripening on soft and semi-hard French cheeses. Its lipases and proteases promote flavor development, and its aminopeptidases reduce bitterness imparted by low-molecular-weight peptides in cheese (Marcellino et al., 2001).

Njage et al. (2011) also identified species belonging to the genera *Rhodotorula*, *Cryptococcus*, *Candida*, *Trichosporon*, *Geotrichum* and *Issatchenkia* which weren't founded in our study. Perhaps it's depending of relatively few *shubat* samples taken for this study. Geographic factors, specific natural fermentation processes and hygienic practices could play an important role on the yeast biodiversity in dairy products (Njage et al., 2011).

Conclusion

This study revealed the high biodiversity of microflora available in fermented camel milk. In the perspectives, the identification of the remaining isolated LAB strains should be done to give a definitive idea of microflora diversity in the fermented camel milk in Kazakhstan. This step is essential for selecting in a second step, specific strains according to their role in fermentation process of camel milk. It is expected in that sense, after proper testing, to conduct fermentation with specific starter allowing special flavor and taste of the final product. It is the objective of our further investigations.

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REGULAR ARTICLE

Impact of husbandry, stages of lactation and parity number on milk yield and chemical composition of dromedary camel milk

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Abstract

The present study was designed to assess the impact of husbandry, stage of lactation and parity number on milk yield and chemical composition of camel milk within three different camel farms at Khartoum State, Sudan. Camel milk samples (n=220) were collected from 43 healthy she-camels at different lactation stages (early, mid, late and latest stages of lactation) and parity number (1-7 parities). The overall means of daily milk yield and composition of fat, protein, lactose, solids not fat (SNF), acidity and density were 2.73 ± 1.16 L/day, $3.69 \pm 1.31\%$, $3.32 \pm 0.33\%$, 4.59 ± 0.45 , $8.49 \pm 0.86\%$, $0.19 \pm 0.03\%$ and 1.030 ± 0.017 g/cm³, respectively. Camel milk yield and composition were significantly ($P < 0.05$) affected by husbandry, stage of lactation and parity number. The highest milk yield (3.49 ± 0.89 L/day) was recorded for she-camels kept in the intensive farming system during early stage of lactation (2.96 ± 1.28 L/day). The result showed that the she-camels in the second parity gave the highest milk yield (4.06 ± 1.85 L/day), while the lower milk yield was found at the subsequent parities. The highest means of fat ($4.05 \pm 1.5\%$), SNF ($8.78 \pm 0.74\%$), protein ($3.41 \pm 0.3\%$) and lactose ($4.67 \pm 0.42\%$) were recorded for the milk of she camels in the semi-intensive farming. The highest means of fat, protein, lactose and SNF ($4.46 \pm 1.62\%$, $3.5 \pm 0.27\%$, $4.75 \pm 0.42\%$ and $8.88 \pm 0.89\%$, respectively) were found in camel milk during the early stage of lactation. Moreover the highest means of protein, lactose and SNF ($3.42 \pm 0.33\%$, $4.71 \pm 0.52\%$ and $8.83 \pm 0.86\%$, respectively) were recorded in milk for the she camels at parity number five. This study concluded that husbandry systems, stage of lactation and parity number have impact on milk yield and chemical composition of camel milk. Therefore, factors that cause variations in milk yield and composition should be considered for the nutritional and technological uses of camel milk.

Key words: Camel farming systems, Milk yield, Chemical composition, Husbandry, Stage of lactation, Parity number, Sudan

Introduction

Sudan is rated as the second highest world size of camel population in the world. According to recent estimation of camels in Sudan there are about 4.623 million heads (Ministry of Animal Resources and Fisheries, 2011). In Sudan, four camel management systems were identified. These systems are: Traditional nomadic system (Shuiop et al., 2008; Ishag and Ahmed, 2011); Transhumance or semi-nomadic system (Musa et al., 2006a; Eisa and Mustafa, 2011); Sedentary or semi-sedentary system (Ishag and Ahmed, 2011; Shuiop and El

Zubeir, 2012) and the Intensive system (El Zubier and Nour, 2006; Eisa and Mustafa, 2011). El Zubier and Nour (2006) described camel husbandry and practices in the periurban area of Khartoum State.

Kamoun and Jemmali (2012) reported that the milk yield of camel varies greatly depending on the region. These variation in milk yield due to breed or types (Wernery et al., 2004), stage of lactation (Musa et al., 2006b; Raziq et al., 2008; Al-Saiady et al., 2012); parity numbers (Al-Saiady et al., 2012) and the production systems (Musa et al., 2006b; Bakheit et al., 2008).

Musaad et al. (2013) concluded that camel milk composition showed a wide variability in its constituents depending on the physiological, genetic and environmental factors. Variations observed in camel milk composition could be attributed to several factors such as feeding conditions (Khaskheli et al., 2005) and production systems (Nabag et al., 2006; Sheep et al., 2008;

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Bakheit et al., 2008), seasons (Sheep et al., 2008; Haddadin et al., 2008; Konuspayeva et al., 2008), breeds and stage of lactation (El-Amin et al., 2006; Konuspayeva et al., 2010) and calving number (El-Amin et al., 2006; Zeleke, 2007; Konuspayeva et al., 2010). In Sudan, selling of milk is neither practiced nor accepted by camel herders in the traditional systems (Musa et al., 2006a; Shuijp and El Zubeir, 2012) and there are no well-established camel dairy farms (Shuijp and El Zubeir, 2008). However, currently a new trend towards commercialization of camel milk associated with the new semi intensive camel system has starting in Khartoum State as well as other big towns (Shuijp and El Zubeir, 2012). The objective of this study is to assess the impact of management system, stage of lactation and parity numbers on milk yield and chemical composition of camel milk.

Materials and Methods

Collection of data

This study was carried out during the period from March 2012 to May 2012. A questionnaire was prepared for data collection. The questionnaire included questions regarding general information about the farmers and farms (camel types, herd size

and structure), building and design, farm management (record keeping, culling practices and general hygiene), system of feeding, health care, calf rearing and milk production and reproduction.

Husbandry practices and rearing of the selected camels

The camel husbandry practices of she camel selected for this study include intensive, semi-intensive and grazing + supplement farming systems (Table 1). In intensive farming systems, camels are kept in barns all times. The farm contains also separate fences for cows, goats and chickens. The daily ration consists of a mixture of Alfalfa, *Sorghum bicolor* (Abu70) and groundnut cake. Water supply was taken from the wells. In the semi-intensive farming system, the camels are kept in an open barn and graze around the farm. The lactating female camels are supplemented with concentrates beside good quality ration containing groundnut cake, *Sorghum bicolor* in addition to continuous water supply. In grazing +supplement farming system, the animals graze at open areas surrounding the farm at the morning times until mid-day then they were kept inside the farm for milking and supplement feeding (Table 1).

Table 1. General information of camel Husbandry practices in the selected farms at Khartoum State.

| Measurements | Farm 1 | Farm 2 | Farm 3 |
|----------------------------------|---|--|---|
| Farming systems | Intensive system | Semi-intensive system | Grazing + supplement system |
| Purpose of production | Commercial | Commercial and genetic improvement | Commercial and research objective |
| Camel breed | Kenani, Anafi | Kenani, Anafi, Bishari | Arabi |
| Herd size | 71 | 146 | 74 |
| Number of females | 25 | 62 | 20 |
| Number of lactating females | 14 | 17 | 14 |
| Number of calves | 20 | 18 | 33 |
| Number of mature males | 1 | 4 | 1 |
| No. of dry she camel | 6 | 20 | 3 |
| No. of pregnant she camel | 5 | 25 | 3 |
| Rearing other animals | Cows, goats, chickens | Cows, goats, sheep, chickens and horses | Non |
| Buildings and design of the farm | | | |
| Barn area/m ² | 360 m ² | 2160 m ² | 150 m ² |
| Type of fence | Steel angles | Steel angles | Steel angles |
| Type of roof | Zinc | No roof | Traditional |
| The area covered by shadow | 96 m ² | Non | 24 m ² |
| System of feeding | | | |
| System of feeding | at farm | at farm | grazing and at farm |
| Type of feeds | groundnut cake, Alfalfa, <i>Sorghum bicolor</i> (Abu70) | groundnut cake, <i>Sorghum bicolor</i> (Feterita), <i>Sorghum bicolor</i> (Abu 70) | grazing plants, <i>Sorghum bicolor</i> (Abu 70) |
| Water supply | 3 wells | 6 wells | Domestic Supply |

Collection of milk samples

A total of 220 camel milk samples from 43 healthy she-camels (with different lactation stages and parity numbers) from the three selected camel farms were collected. One sample of 50 ml from each she-camel was taken every 15 days for 3 months. The raw camel milk samples were collected in the early morning and immediately labeled, stored in an ice box and transferred within 2-3 hours to the laboratory of the Department of Dairy Production, Faculty of Animal Production, University of Khartoum for the chemical analysis.

Chemical analysis of milk

Chemical analysis of camel milk samples were determined by using LactoScan Milk Analyzer (Milkotronic LTD, Europe) according to the manufacturer's instructions. The instrument was first calibrated as illustrated in the accompanied technical manual for the measurement of camel milk constituents. The content of fat, protein, lactose and SNF and the density were obtained as follow: Twenty five ml of the samples were taken in the sample holder after mixed gently 4- 5 times. The sample holder was put in the analyzer in the recess position and the analyzer sucks the milk and makes the measurement. When the measurement is finished, the sample returns in the sample-holder and the digital indicator shows the specified result.

Statistical analysis

Data were analyzed using SPSS software (Statistical Package for Social Sciences, V.13). Differences between means were separated by LSD.

Results and Discussion

Reproduction, milk production and health management practiced in camel farms from Khartoum State

According to the questionnaire, the gestation period was 12 months in each of the three farming systems. The calving intervals were about 25 months for semi-intensive system and 24 months for both farms that adopted intensive and grazing +supplement farming system (Table 2). The length of the dry period was estimated as 2-3 months, 3-4 months and 4 months for intensive farming system, semi-intensive system and for grazing +supplement farming system, respectively. This result agreed with Musa et al. (2006b) who mentioned that gestation length was 370.28 ± 19.06 days. Similarly Musaad et al. (2013a) found that the overall mean for the lactation length for she camels kept in the intensive system was 12.5 months and the values differed according to season of calving. On the other hand, diseases, age and production problems were the main reasons for culling at the three farms. Calves were reared in small groups and fed by the same types of food as their parents (Table 2).

Table 2. Reproduction management in camel farms at Khartoum State.

| Farm management | Intensive system | Semi-intensive system | Grazing + supplement |
|-------------------------------|--|---|------------------------------------|
| Gestation length | 12 months | 12 months | 12 months |
| Length of the dry period | 2-3 months | 3-4 months | 4 months |
| Period of colostrums | 7 days | 7 days | 7 days |
| Culling practices | disease, age | disease, age | production problems, age |
| Calf rearing | | | |
| Calf rearing | at small groups | at small groups | at small groups |
| Age of weaning | 12 months | 1 month | 4 months |
| Using milk replacer | No | No | No |
| Milking procedure | in the presence of calf | in the presence of calf | in the presence of calf |
| Types of nutrition | groundnut cake, Alfalfa, <i>Sorghum bicolor</i> (Abu70) | groundnut cake, <i>Sorghum bicolor</i> (Feterita), <i>Sorghum bicolor</i> (Abu70) | <i>Sorghum bicolor</i> (Abu 70) |
| She camel | | | |
| Breed of milk production | Anafi | Kenani | Arabi |
| Source / origin | East of Sudan - Al Gadarif | East of Sudan and Kordufan | East and West of Sudan |
| Concentrates supplementation: | Yes | Yes | No |
| Mating system | Natural system | Natural system | Natural system |
| Calving interval /month | 24 months | 25 months | 25 months |

The daily milk yield of she camels were 40-60, 40-80 and 50 liters / day in the intensive system, semi-intensive system and grazing +supplement system, respectively (Table 3). The lactation length for camel included in this study was 9-10 months, 8-9 months and 8 months in intensive system, semi-intensive system and grazing +supplement system, respectively. However Musaad et al. (2013b) reported an average total milk production of 1207 L for 11 months range between 875 and 1616 L in Saudi Arabia. Milking in all farms was practiced in the presence of the calves. Al-Haj and Al-Kanhal (2010) mentioned that the factors affecting milk yields are those, which are common to all dairy animals such as nutrient supply, health status, genetic potential for milk production, number of previous lactations or age of the animal and adequate water supply. Camel herders in the selected farm are using hired labor for milking, which was done three times per day at intensive system and twice per day for semi-intensive system and grazing +supplement. Cooling facilities were available at the three systems, which disagreed with Shuiet et al. (2007) as they viewed no cooling was applied for camel milk. All these newly introduced practices indicated transitional stage towards modern dairy camel farming at the commercial basis. The type of milk containers were plastic in the intensive system and aluminum containers in the semi-intensive and grazing +supplement

system. The milk is sold fresh at the farms except for the semi-intensive system which is sold at the market.

The effect of husbandry practices on milk yield

The mean daily milk yield of the she camels kept in the intensive, semi intensive and grazing+supplement farming systems were 3.49 ± 0.89 , 2.76 ± 1.24 and 2.08 ± 0.87 L, respectively (Table 4). Milk yield was significantly ($P \leq 0.05$) affected by husbandry practices, however the milk yield from individual animal over a period of 3 months revealed non-significant variations. The mean daily milk yield of the camels reared under semi intensive farming system was higher than that reared under grazing+ supplement farming system (Table 4). Similarly Bakheit et al. (2008) found that camels raised under semi-intensive management were able to produce significantly more milk than the other reared under traditional system. This could be attributed to the forage availability and the supplementary diets, water availability and health care that oriented to the camels in the semi intensive system (Table 2 and 3). This mainly might be because of the current trend towards commercialization of camel milk in the adopted new semi intensive camel system that has been established in Khartoum (Shuiet and El Zubeir, 2012).

Table 3. Milk production, general hygiene and health care practiced at the selected camel farms in Khartoum State.

| Milk production | Intensive system | Semi-intensive system | Grazing + supplement |
|---|-------------------|-----------------------|----------------------|
| Average production of milk/day/farm (L) | 40 - 60 | 40 – 80 | 50 |
| No. of milking | three times / day | twice times / day | twice times / day |
| Length of lactation | 9-10 months | 8-9 months | 8 months |
| Selling milk | in the farm | in the market | in the farm |
| Price of camel milk per liter | 7 SDG | 8 SDG | 6 SDG |
| Milk processing | No | No | No |
| Type of milk containers | Plastic | Aluminum | Aluminum |
| Cooling facilities | Yes | Yes | Yes |
| Cleaning the udder before milking | no | Yes | No |
| Hygiene of milkers | Yes | Yes | Yes |
| Dung removal | every 2 week | Weekly | more than 2 weeks |
| Using disinfectants | Yes | Yes | Yes |
| Vaccination program | No | No | Yes |
| Veterinary visits | on call | on call | Daily |

Table 4. Effect of husbandry practices on milk yield and chemical composition of camel milk.

| Production system | Intensive system | | Semi-intensive system | | Grazing+ Supplement | |
|-------------------|------------------|---------|-----------------------|---------|---------------------|---------|
| | Minimum | Maximum | Minimum | Maximum | Minimum | Maximum |
| Milk yield L/day | 1.77 | 5.33 | 0.44 | 6.22 | 0.88 | 4.44 |
| Fat (%) | 1.39 | 6.55 | 1.81 | 6.34 | 1.05 | 5.52 |
| Protein (%) | 2.54 | 4.58 | 2.28 | 4.08 | 2.64 | 4.00 |
| Lactose (%) | 3.18 | 6.02 | 3.71 | 5.67 | 3.71 | 5.71 |
| SNF (%) | 6.15 | 11.36 | 7.02 | 10.56 | 6.83 | 10.23 |
| Acidity (%) | 0.12 | 0.25 | 0.13 | 0.25 | 0.1 | 0.26 |
| Density (%) | 1.023 | 1.038 | 1.023 | 1.037 | 1.023 | 1.036 |

The effect of husbandry practices on milk composition

The milk composition from she camels managed in the different farming systems revealed non-significant variations over a period of 3 months. Camel milk composition was significantly ($P<0.05$) affected by the husbandry practices (Table 5). The highest means of fat ($4.05\pm1.5\%$), SNF ($8.78\pm0.74\%$), protein ($3.41\pm0.3\%$) and lactose ($4.67\pm0.42\%$) were recorded for the camels kept at semi-intensive farming system in comparison with the other two farming systems. This might suggested the importance of grazing in rearing the camel. Variations observed in camel milk composition could be attributed to several factors including management systems (Bakheit et al., 2008; Shuiet al., 2008; Riyadh et al., 2012), geographical locations, feeding conditions (Khaskheli et al., 2005; Bakheit et al., 2008), seasons (Shuiet al., 2008; Riyadh et al., 2012), stage of lactation and calving number (El-Amin et al., 2006; Zeleke, 2007; Riyadh et al., 2012). Moreover Musaad et al. (2013b) reported significantly negative correlation between milk production and percentage of the different milk components due to dilution effect. The lower mean of fat content was found for the camel milk samples collected from the grazing+ supplement farming system ($3.29\pm1.06\%$). This result was higher than result reported by Shuiet al. (2008) in Sudan and Riyadh et al. (2012) in Saudi Arabia. However the maximum fat content of camel milk (6.55%) was found in the samples collected from the intensive farming system (Table 4). This result agreed with Riyadh et al. (2012) who reported that the fat content of camel milk was higher in the settled system (intensive) than nomadic and semi nomadic production system. This might be due to the feeding of concentrate. Similarly Shuiet al. (2008) attributed the variations of fat content to season which is affected by the availability of the grasses.

The average total protein content of camel milk samples collected from intensive, semi-intensive and grazing+ supplement farming systems were $3.28\pm0.38\%$, $3.41\pm0.3\%$ and $3.26\pm0.31\%$, respectively (Table 5). There were significant ($P<0.05$) differences between the semi intensive system and both intensive and grazing +supplement systems (Table 5). The result was higher than that reported by Haddadin et al. (2008) and Konuspayeva et al. (2009). However Shuiet al. (2008) reported non-significant differences in protein content for camel milk samples collected from semi-intensive and traditional systems.

Lactose content of camel milk were $4.43\pm0.48\%$, $4.05\pm1.5\%$ and $4.47\pm0.43\%$ in the intensive, semi-intensive and grazing+ supplement systems, respectively (Table 5). This result was higher than the result reported by Shuiet al. (2008), they reported that the lactose content of camel milk samples collected from traditional system and semi-intensive system were 2.90% and 3.12%.

The average titratable acidity of camel milk (Table 5) were $0.19\pm0.02\%$, $0.19\pm0.03\%$ and $0.18\pm0.03\%$ in the intensive, semi-intensive and grazing + supplement farming systems, respectively. The result disagreed with result reported by Shuiet al. (2008) who reported highly significant differences ($P\leq0.01$) in the titratable acidity between camel milk samples from semi-intensive system ($0.15\pm0.02\%$) and traditional system ($0.14\pm0.02\%$). Lower acidity of milk was reported for the grazing camel which supported Mohamed and El Zubeir (2012).

Table 5: Variations of milk yield and chemical composition of the she-camels kept at different husbandry systems

| Production system | Milk yield L/day | Fat (%) | Protein (%) | Lactose (%) | SNF (%) | Acidity (%) | Density (gm cm ³) |
|-----------------------|-------------------------|-------------------------|-------------------------|-------------------------|-------------------------|-------------------------|-------------------------------------|
| Intensive system | 3.49 ^b ±0.89 | 3.72 ^a ±1.2 | 3.28 ^b ±0.38 | 4.43 ^b ±0.48 | 8.26 ^b ±0.97 | 0.19 ^a ±0.02 | 1.028 ^a ±0.0030 |
| Semi-intensive system | 2.76 ^a ±1.24 | 4.05 ^a ±1.5 | 3.41 ^a ±0.3 | 4.67 ^a ±0.42 | 8.78 ^a ±0.74 | 0.19 ^a ±0.03 | 1.03 ^a ±0.0031 |
| Grazing+ Supplement | 2.08 ^c ±0.87 | 3.29 ^b ±1.06 | 3.26 ^b ±0.31 | 4.47 ^b ±0.43 | 8.39 ^b ±0.8 | 0.18 ^a ±0.03 | 1.032 ^a ±0.0029 |
| Average | 2.73±1.16 | 3.69±1.31 | 3.32±0.33 | 4.59±0.45 | 8.49±0.86 | 0.19±0.03 | 1.030±0.017 |

Different letters in same column indicates significant difference (P≤ 0.05)

Table 6: Effect of stage of lactation on yield and chemical composition of camel milk

| Stage of lactation | Milk yield | Fat (%) | Protein (%) | Lactose (%) | SNF (%) | Acidity (%) | Density (%) |
|--------------------|-------------------------|-------------------------|-------------------------|--------------------------|--------------------------|-------------------------|--------------|
| 1 - 3 months | 2.96 ^a ±1.28 | 4.46 ^a ±1.62 | 3.5 ^a ±0.27 | 4.75 ^a ±0.42 | 8.88 ^a ±0.89 | 0.2 ^a ±0.02 | 1.030±.0035 |
| 4 - 6 months | 2.47 ^a ±1.28 | 3.86 ^b ±1.01 | 3.39 ^{ab} ±0.4 | 4.61 ^{ab} ±0.48 | 8.64 ^{ab} ±0.92 | 0.19 ^a ±0.02 | 1.029±0.0032 |
| 7 - 9 months | 2.68 ^a ±1.08 | 3.43 ^b ±1.15 | 3.3 ^{bc} ±0.31 | 4.53 ^{bc} ±0.46 | 8.49 ^{bc} ±0.79 | 0.19 ^a ±0.02 | 1.029±.0028 |
| ≥ 9 months | 2.11 ^b ±0.99 | 3.49 ^b ±1.37 | 3.22 ^c ±0.29 | 4.4 ^c ±0.4 | 8.25 ^c ±0.81 | 0.19 ^a ±0.04 | 1.031±.029 |
| Average | 2.56±1.16 | 3.69±1.31 | 3.32±0.33 | 4.59±0.45 | 8.49±0.86 | 0.19±0.03 | 1.032±.017 |

Different letters in same column indicates significant difference (P≤ 0.05).

Effect of stages of lactation on milk yield and milk composition of camel

The highest milk yield in the present study was obtained for camels at first three months of lactation (2.96 ± 1.28 L) and the lower milk yield was found for camels at late lactation (2.11 ± 0.99 L) as shown in Table 6. Although the she camels were from different production systems are grouped together to calculate the average lactations the result agreed with Al-Saiady et al. (2012). The seasons, stage of lactation and calving number (El-Amin et al., 2006; Zeleke, 2007; Riyadh et al., 2012) and the management conditions (Musa et al., 2006b; Bakheit et al., 2008; Riyadh et al., 2012) were found to affect camel milk yield.

Significant ($P\leq0.05$) differences for stages of lactation on SNF, protein and lactose content of camel milk were observed (Table 6). The higher fat content of milk was observed (Table 6) for camels in the first three months of lactation compared to those in latter stages of lactation (4.46% and 3.49% respectively). The variations of this result from those obtained by El-Amin et al. (2006), Zeleke (2007) and Haddadin et al. (2008) could be because they follow the same animals, while this study examined the milk from different animals. Moreover Konuspayeva et al. (2010) reported that the fat content decreased all along the lactation period and the fat content varied from 4.34% to 7.81%.

Higher protein content in milk (Table 6) was found for camels at the first lactation period (3.5%) and the lower protein content was reported for camels at the end of lactation (3.22%). This result agreed with El-Amin et al. (2006), Zeleke, (2007) and Riyadh et al. (2012) who mentioned that the highest percentage of protein of camel milk were at the first lactation and then decreased along the lactation period. Significantly higher content of lactose in milk was found for camels at the first three months of lactation ($4.75\pm0.42\%$) compared to those at later stages of lactation. This result agreed with Zeleke (2007) and Riyadh et al. (2012) who found that the higher lactose content was at first months of lactation and then decreased significantly at the end of lactation period. However the result disagreed with El-Amin et al. (2006) who found non-significant differences in lactose content between stages of lactation. The variations of chemical composition of camel milk at the end of lactation period might be due to the

increase in the milk water content during the last stage of lactation (Riyadh et al., 2012).

Effect of parity number on milk yield and milk composition of camel

Slight differences for parities number on camel milk yield, SNF, protein and lactose was observed (Table 7). The highest milk yield was estimated for the camels in the second parity and the lowest milk yield was reported for camel at the last three parities (Table 7). This result disagreed with Al-Saiady et al. (2012) who reported that the lowest milk yield was at the 1st, 2nd, and 4th parity. The Higher milk productivity was at the 3rd and 6th season of lactation (Table 7), which agreed with Raziq et al. (2008) who reported that she-camel has higher milk production at the 3rd season and longer and Musaad et al. (2013a) who reported that the highest average yield recorded was for camels at sixth parity. These could be due to the increased in growth and number of secretary cells in the udder or increased secretary activity of the mammary tissue or both (Herndez et al., 2008). The result showed non-significant differences between the she camels in the different parities for fat content of milk. The percentages of fat content vary between 3.5 and 3.95% (Table 7). This result agreed with El-Amin et al. (2006) and higher than that reported by Riyadh et al. (2012). Lactose content of camel milk varies between 4.71% and 4.32% (Table 7), which were lower than the result reported by Riyadh et al. (2012). The highest level of lactose content of milk in the present study (4.71%) was reported for camels in the 5th parity, which disagreed with Zeleke (2007) who reported that the highest lactose content of camel milk was recorded in the first lactation. Lactose level was viewed to be high for camels in the 2nd, 4th and 5th parities and higher than those at the 6th and 7th parities. This result disagreed with El-Amin et al. (2006) who mentioned that the lactose content was decreased from the first parity (3.75%) to the second parity (3.48%) then increase significantly ($P<0.05\%$) in the third parity (4.24%). The differences could be due to the variations in lactose content obtained by different camels and the type of plants eaten by the camel (Khaskheli et al., 2005).

The statistical model did not take in account the co-variance due to the farming system and some results regarding the effect of parity and physiological stage could be influenced by the methodology used. It was the main limit of the present study.

Table 7. Effect of parity number on milk yield and chemical composition of camel milk.

| Parity No | Milk yield Lb/day | FAT (%) | Protein (%) | Lactose (%) | SNF (%) | Acidity (%) | Density (%) |
|-----------|-------------------------|-------------------------|--------------------------|--------------------------|--------------------------|-------------------------|----------------------------|
| 1 | 2.60 ^b ±0.99 | 3.81 ^a ±1.56 | 3.28 ^a ±0.38 | 4.48 ^a ±0.52 | 8.35 ^a ±1.04 | 0.2 ^a ±0.03 | 1.035 ^a ±0.041 |
| 2 | 4.06 ^a ±1.85 | 3.79 ^a ±1.42 | 3.31 ^a ±0.39 | 4.56 ^{ab} ±0.52 | 8.5 ^{ab} ±1.01 | 0.19 ^a ±0.02 | 1.09 ^a ±0.003 |
| 3 | 2.75 ^b ±1.04 | 3.61 ^a ±1.31 | 3.27 ^{ab} ±0.3 | 4.48 ^a ±0.43 | 8.33 ^{ac} ±0.78 | 0.19 ^a ±0.03 | 1.029 ^a ±0.0026 |
| 4 | 2.59 ^b ±1.04 | 3.75 ^a ±1.21 | 3.36 ^a ±0.35 | 4.54 ^{ab} ±0.41 | 8.62 ^{ab} ±0.82 | 0.19 ^a ±0.03 | 1.029 ^a ±0.003 |
| 5 | 1.95 ^c ±0.90 | 3.5 ^a ±1.32 | 3.42 ^{ac} ±0.33 | 4.71 ^b ±0.52 | 8.83 ^b ±0.86 | 0.19 ^a ±0.03 | 1.03 ^a ±0.0033 |
| 6 | 1.82 ^c ±0.89 | 3.95 ^a ±0.76 | 3.3 ^a ±0.25 | 4.53 ^{ab} ±0.33 | 8.48 ^{ab} ±0.62 | 0.19 ^a ±0.03 | 1.029 ^a ±0.0020 |
| 7 | 1.78 ^c ±0.00 | 3.25 ^a ±1.22 | 3.17 ^a ±0.18 | 4.32 ^a ±0.27 | 8.11 ^a ±0.44 | 0.19 ^a ±0.04 | 1.028 ^a ±0.0022 |
| Average | 2.52±1.11 | 3.69±1.31 | 3.32±0.33 | 4.59±0.45 | 8.49±0.86 | 0.19±0.03 | 1.030±0.017 |

Different letters in same column indicate significant difference (P≤ 0.05).

Conclusion

The present study confirmed that the husbandry practice, production system and the physiological status of camels have impact on milk yield and milk gross composition. The performance of she camels at semi-intensive system was better in comparison to the other management systems; therefore initiations of the semi-intensive system should be encouraged at the different states of Sudan. For future prospects, more research should be conducted to delineate management and nutrition requirements for the camel to improve the milk yield and composition in order to make camel rearing an economical proposition.

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REGULAR ARTICLE

Effect of Selenium injection in pregnant camels on selenium status of their new-born and milk

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Abstract

The effect of inoculation of selenium solution to pregnant camels was investigated to assess the impact on selenium status of the new-born and on the selenium concentration in milk. In the trial included 2 groups of 8 camels, the treated one receiving a single injection of selenium solution at the end of pregnancy. In blood, no difference was observed between control and treated group before injection. A significant difference was observed at delivery as well in dam (33.3 vs 44.7 ng/mL respectively) as in calf (28.5 vs 47.6 ng/mL respectively). In milk, the selenium was also significantly in higher concentration in treated group (93 ± 49 ng/mL) than in control one (59 ± 19 ng/mL) at the delivery time. Zinc concentration in milk was positively correlated to selenium content. The improvement of selenium status by a single injection was slight and more efficient supplementation ways could be proposed to the camel farmers.

Key words: Camel, Milk, Colostrum, Selenium, Copper, Zinc

Introduction

The selenium (Se) in milk was regularly investigated in cattle (Ceballos et al., 2009) or in ewe (Davis et al., 2006), but in camel, the references remain scarce. Previous studies were mainly limited to blood status (Faye and Seboussi, 2009) and only 3 references are available on the selenium quantity transferred through milk to the camel calf (Al-Qarawi et al., 2001; Seboussi et al., 2009a; Faye et al., 2011). These publications showed a high variability of the Se content in milk according to the Se status of the mother before calving and to lactation stage after calving. They stated also on the specific Se metabolism in camel regarding the toxicity threshold (Seboussi et al., 2009b) and the supplementation (Faye and

Seboussi, 2009) which cannot be applied directly from cattle requirements. Moreover, those former results obtained in Emirates (Seboussi et al., 2009a) were reported in a context of Se deficiency widely observed in the field with numerous cases of white muscle disease or heart failure due to lack of Se in the mother's diet, and furthermore partly with dams receiving before calving an oral Se supplementation under selenite form. In Saudi Arabia, where selenium status in human population was regarded as low (Al-Saleh, 2000), selenium deficiency was regularly incriminated also in grazing livestock. However, most of the camel farmers did not distribute oral selenium supplementation to their animals, but rather used non-organic selenium solution by injection in pregnant or new-born camels. However, the effect of an unique injection at the end of pregnancy on the selenium status of the new-born and especially on the level of selenium in milk which is the unique source of selenium for the calf, was never studied in camel.

In the present study, the selenium transfer through milk from the dam to the camel calf was analyzed after Se supplementation by injection

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before delivery in order to assess the impact on the selenium content in milk and selenium status of the new-born calves. Elsewhere, the interactions with other trace elements as copper and zinc were investigated.

Materials and Methods

Location and animals

This study was carried out in the camel farm of Al -Jouf "Camel & Range Research Center" located in north-west Saudi Arabia, 950 km from Riyadh. Average annual temperature was 20°C, ranging from 12°C to 27°C, and average annual rainfall was 55 mm. The herd was composed by camels of four ecotypes (Malhah, Wadhah, Hamrah and Safrah) but belonging to very close genotype (Abdallah and Faye, 2012; Almathen et al., 2012). The weight of the animals was on average 620 ± 101 kg. Camels were kept in-door throughout the year and housed in pens. Their normal diet was composed of alfalfa (*ad-libitum*), barley (3 kg/day/animal), salt, wheat bran (1kg/day/animal). As the calving season occurred between December and February, all the camels were approximately at the same stage of reproductive cycle. The milk production not including part drunken by camel calves was recorded every day.

Selenium treatment

For the experiment, 16 adult lactating camels 5-18 years old only were available. They were divided randomly into two groups of eight. In spite of the heterogeneous composition of the herd, the groups' composition was comparable (no significant difference) as well for mean age (10.6 ± 6.4 vs 8.8 ± 3.3 years for treated and control group respectively) as mean weight (624 ± 78 vs 616 ± 121 kg). The camels were in good health all along the experiment. The control group did not receive any selenium supplementation. The treated group was submitted to unique injection of Selepheral[®] from Vetoquinol Co as preventive dose for selenium deficiency. Selenopheral[®] contained sodium selenite (23 mg/100ml) and vitamin E as acetate (3.82 g/100ml). The pregnant she-camels received 75 ml (i.e. 17.25 mg Se) by deep IM route at different injection sites to avoid local reaction. The injection was done 3-weeks approximately before the delivery. This level of supplementation corresponded to what was locally practiced by the camel owners to prevent selenium deficiency in camel calf.

Sampling agenda and laboratory analysis

Milk was sampled at the morning milking at the delivery then at day 30 and 60 post-partum in a

plastic bottle. Blood samples were collected in the dams just before injection then at the delivery. Blood samples were collected also on camel calves after parturition at the same time of their dam. Samples (blood and milk) were stored in deep freezer at -80°C until laboratory analysis. In blood and milk samples, copper and zinc were determined by Atomic Absorption Spectrophotometer (AA-6650, Shimadzu, Japan) at the IDAC laboratory, Kharij (Saudi Arabia). Selenium was determined in the same laboratory with Hybrid Vapor Generator (HVG-1, Shimadzu, Japan). The data are reported as ng/mL for selenium, and $\mu\text{g}/100\text{ml}$ for copper and zinc.

Statistical analysis

The mean and standard deviation was calculated for each parameter and for each group. The variance analysis (ANOVA) for time series was applied to evaluate the difference between control and treated groups all along the experiment. Pearson correlation was determined to assess the relationships between the mineral statuses.

The software XLSTAT (Addinsoft[®]) was used for the data analysis.

Results and Discussion

Selenium in blood

The effect of selenium injection, yet widely used for preventing selenium deficiency in camel was not studied in this species. Moreover, it is difficult to compare the types of Se supplementation reported in the literature only on the quantitative basis as the form of Se administrated to the animals could differ strongly (injection or oral, organic or non-organic, different doses). So, the effect of the supplementation is essentially assessed by comparing the Se concentration in serum. This concentration is generally regarded as a good short-term indicator of the selenium status in animal. Due to this relatively long apparent terminal half-life, the concentration of Se in serum should be widely independent of small daily variations in Se intake (Haldimann et al., 1996).

In our study, the mean value of selenium concentration in serum was 37.9 ± 0.83 ng/mL in dams and 40.7 ± 1.25 ng/mL in camel calves. There was no difference between control and treated group at the time of injection (36.3 vs 37.2 ng/mL respectively), but a significant difference ($P < 0.01$) was observed at delivery as well in dam (33.3 vs 44.7 ng/mL respectively) as in calf (28.5 vs 47.6 ng/mL respectively) (Table 1). Those values corresponded globally to low level of selenium status. Indeed, on average, normal serum selenium

concentration in camel was regarded as about 100ng/mL (see review of Faye and Seboussi, 2009). For example, in Morocco, Hamliri et al. (1990) observed in whole blood, values varying according to age and sex, between 109.1 and 117.8 ng/mL. Similar figures were recorded by Liu et al. (1994) in China on Bactrian camel with concentrations varying from 97 to 112 ng/mL. However, in Sudan, Abdel Rahim (2005) reported values in whole blood varying between 25 and 53 ng/mL.

In serum from Moroccan dromedaries receiving probably a low Se basal diet, the plasma selenium concentration was quite lower, about 21 ng/mL (Bengoumi et al., 1998a). Recently, in male adult camels in healthy conditions from Iran, the selenium concentration reported in serum was 12.6 ng/mL only (Nazifi et al., 2011). In Saudi Arabia, serum Se values reported in young camels at the slaughterhouse varied between 5.3 and 131 ng/mL with 30% of samples higher than 100 ng/mL (Barri and Al-Sultan, 2007). In the same area than the present study, the serum Se was 50.5 ± 31.5 ng/mL, whatever the physiological stage of the camels (Althamma et al., 2012). In the United Arab Emirates (UAE), the mean value was 200 ± 90 ng/mL in animals with no Se supplementation (Seboussi et al., 2004). In recent experiments with different levels of Se supplementation, selenium content in serum for non-supplemented animals was on average 137.6 ± 18.7 ng/mL in non-pregnant, non-lactating camels (Seboussi et al., 2008), 109.3 ± 33.1 ng/mL in pregnant females, and 103.4 ± 28.7 ng/mL at milking period (Seboussi et al., 2009a). The variability was thus high and the range between 12 and 200 ng/mL with

an average of 100 ng/mL. However, in most of the reported values, the selenium status of the diet was unknown even if Se supplementation was not distributed to the animals. In Saudi Arabia, the basal diet could be very low in natural selenium.

The single Se injection improved slightly the Se status of the camel, appreciated by the increase in serum concentration. However, with daily oral supplementation, a most important effect was reported. In two groups of pregnant females receiving 0 and 2 mg Se respectively under sodium selenite form at the end of their gestation (last three months) and at the beginning of their lactation up to one month (Seboussi et al., 2009a), the mean value of selenium content in serum was significantly higher in supplemented group (2 mg) and was three-fold higher than the concentration compared to the control group (305.9 ± 103.3 ng/mL and 109.3 ± 33.1 ng/mL respectively). The selenium level at parturition was still significantly higher in the treated group in spite of a slight decrease around the calving period. In the trial of Al-Qarawi et al. (2001) involving selenodeficient camels with muscular dystrophy, treatment involving selenium – vitamin E (Bo-SE, Schering – Plough Animal health, 2.19 mg sodium selenite + 50 mg vitamin E) by IM injection at a dose rate of 0.5 mg/kg body weight for two consecutive days allowed getting an increase of selenium concentration on average 2.3 ng/mL up to 23.7 ng/mL, i.e. with a similar trend to that observed by Bengoumi et al. (1998a) who reported a multiplication by 10 of the serum Se after supplementation.

Table 1. Selenium (Se) concentrations in camel serum and milk (mean and S.D.) in Control (C) and Treated (T) groups in mother (at injection and delivery time) and calf for serum, and at delivery and every month for milk

| | Serum (ng/mL) | | | Milk (ng/mL) | | |
|------|-------------------|------------------|------------------|-------------------|-------------------|-------------------|
| | Injection | Delivery | Calf | Delivery | D30 | D90 |
| Se-C | 36.3 ± 6.0^a | 33.3 ± 2.3^a | 28.5 ± 7.8^a | 59.1 ± 19.2^a | 50.0 ± 29.3^a | 58.9 ± 13.2^a |
| Se-T | 37.2 ± 10.7^a | 44.7 ± 8.2^b | 47.6 ± 8.7^b | 93.2 ± 49.0^b | 69.1 ± 30.0^a | 72.2 ± 17.2^b |

^{a,b} Means in column with a different letter in superscript differ ($P < 0.05$)

Table 2. Copper (Cu) and zinc (Zn) concentrations in camel serum and milk (mean and S.D.) in Control (C) and Treated (T) groups in mother (at injection and delivery time) and calf for serum, and at delivery and every month for milk.

| | Serum (μ g/100mL) | | | Milk (ppm) | | |
|------|------------------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| | Injection | Delivery | Calf | Delivery | D30 | D90 |
| Cu-C | 70.1 ± 18.9 | 59.0 ± 24.1 | 78.2 ± 13.0 | 0.08 ± 0.01 | 0.08 ± 0.02 | 0.06 ± 0.02 |
| Cu-T | 81.8 ± 25.2 | 80.3 ± 29.9 | 63.0 ± 25.4 | 0.14 ± 0.07 | 0.07 ± 0.02 | 0.07 ± 0.02 |
| Zn-C | 77.7 ± 13.9 | 71.8 ± 12.2 | 77.6 ± 23.9 | 15.96 ± 3.2 | 3.59 ± 1.6 | 3.35 ± 0.8 |
| Zn-T | 71.5 ± 15.0 | 68.3 ± 18.9 | 47.6 ± 20.4 | 8.60 ± 14.8 | 2.54 ± 0.8 | 3.09 ± 0.7 |

In new-born animals, the serum selenium values reflected generally the Se status of the dam, with a positive correlation between the serum concentration in dam and in new-born ($r = 0.622$; $P > 0.01$). In the same area than our study, Athamma et al. (2012) found 37.2 and 46.1 ng/mL in dam and new-born respectively. In Emirates, with camel receiving 2mg/day oral Se supplementation, the Se serum concentrations in camel calf at parturition were 273.2 ± 48.0 and 106.3 ± 26.5 ng/mL in the treated and control groups respectively (Seboussi et al., 2009a) i.e. a similar proportion than in dams.

In our study, the breed composition of each group was composite and not similar. However, the breed effect on the selenium status of animal was not clearly stated. In non-pregnant sheep, Ramirez-Perez et al. (2000) did not report a significant difference between Rambouillet and Suffolk breed. At our knowledge, a genetic variability of selenium status was never reported on camel. Moreover, the camel ecotypes participating to the present experiment were regarded as very close genotypes (Al-Swailem et al., 2007). The age variability of camels in our trial was high. It was not stated from the literature an age effect on selenium status. In human, for example, no significant association was found between selenium and age (Akbaraly et al., 2010). Similar observations could be done for other trace-elements as copper and zinc in camel for which the age effect was not clearly stated in the

literature (see review of Faye and Bengoumi, 1994).

Selenium in milk

The selenium content in milk was significantly higher in treated group (93 ± 49 ng/mL) than in control one (59 ± 19 ng/mL) at the delivery time ($P > 0.05$). The difference was not significant one month later, but again slightly higher in treated group ($P > 0.05$) at the second month of lactation (figure 1). Those values appeared low compared to the results of Faye et al., (2011) in Emirates where the Se concentration in milk varied from 39.5 to 482.6 ng/mL with an average of 167.1 ± 97.3 ng/mL in treated group receiving 2 mg daily in oral supplementation before the delivery, and 86.4 ± 39.1 ng/mL in the control group. In this last study, both in control and treated groups, Se milk concentration decreased and difference was observed after one month as in our study. In their study on camel milk, but without specification on the lactation stage, Al-Awadi and Srikumar (2001) reported quite lower values (13.9 ± 2.4 ng/mL). In dairy cow, the milk Se concentration varied from 19.4 to 53.7 ng/mL with Se dietary selenium between 0.15 and 0.40 ppm (Juniper et al., 2006). According to the meta-analysis of Caballos et al. (2009), the selenium concentration in cow milk varied between 9.2 and 16.3 ng/mL with a maximum of 29.2 ng/mL observed in cattle supplemented with Se yeast. In ewe milk, the values varied from 32 to 81 ng/mL in non-supplemented animals (Davis et al., 2006).

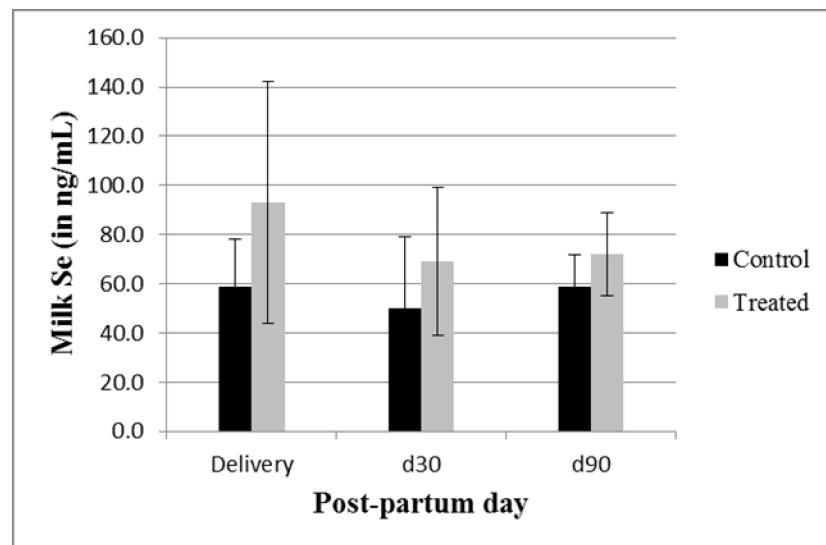


Figure 1. Changes in selenium concentration in camel milk in control and treated groups receiving Se injection.

Contrary to the results reported previously (Seboussi et al., 2009a), the colostrum Se concentration was not a clear reflect of the serum Se of the dam. In our results, the correlation was not significant ($P=0.06$), even if a tendency was observed. However, by comparing to the literature data in other dairy species (Ceballos et al., 2009; Davis et al., 2006), and in spite of the relative low Se status in serum, camel milk seems richer in selenium (Faye et al., 2011).

Interactions with other trace elements

The interactions between trace elements were reported in many publications. For example, for long time, studies have revealed an inverse relationship between zinc and selenium in human milk, and maternal selenium status was found to influence the protein binding pattern of zinc in human milk (Brätter et al. 1997). Zinc and copper have been found to be bound partially to the same proteins, e.g. lactalbumin, in colostrum and transitional milk (Kantola and Vartiainen, 2001), and a direct correlation has been found between copper and selenium in human milk (Perrone et al., 1994).

The interaction between selenium metal ions and other trace elements can alter their respective availability and cause deficiencies, with unforeseen consequences for the activity of enzymes requiring these trace elements as cofactors. Most studies have reported that, in different situations, the level of one element is (or is not) affected by the presence of the other one. The presence of selenium could reduce the availability of metal ions blocking them in insoluble compounds. On the other hand, selenium deficiency has been reported to cause an overload of iron and unbalanced in vivo distributions of other elements, such as magnesium, calcium, copper and zinc (Chareonpong-Kawamoto and Yasumoto, 1995).

In our study, copper concentrations in serum were in the normal range for camel (Table 2), with values between 31 and 121 $\mu\text{g}/100\text{mL}$. Similar values were reported by Athamma et al. (2012) in the same area: 70.3 ± 19.8 and 58.6 ± 13.9 $\mu\text{g}/100\text{ ml}$ for copper in female camels and their new-born respectively.

The range for zinc concentrations (38 to 112 $\mu\text{g}/100\text{ mL}$) was in the upper range than reported in the review of Faye and Bengoumi (1994). There was no difference between control and treated groups both for copper and zinc.

Regarding milk, few data were available. Our results for copper (Table 2), i.e. $85\pm42\mu\text{g}/\text{L}$ on average was comparable to the findings of Bengoumi et al. (1998b) in Morocco ($113\pm49\mu\text{g}/\text{L}$), but lower

than the values reported by Dell'Orto et al. (2000) in camel from the Horn of Africa (370 to 400 $\mu\text{g}/\text{L}$ on average according to the mineral supplementation) and those published in Saudi Arabia by Mehia et al. (1995). There was no difference between the groups in the copper concentration in milk whatever the date of sampling. In our study the average of zinc concentration in the milk was $7.5\pm9.6\text{ mg}/\text{L}$ which was quite higher than the values reported by Dell'Orto et al. (2000) and Bengoumi et al. (1998b) respectively 2.52 to $3.16\text{ mg}/\text{L}$, and $2.87\pm0.8\text{ mg}/\text{L}$. Contrary to copper, a slight significant difference ($P<0.05$) was observed at the delivery with a higher value in control group ($15.9\pm3.2\text{ mg}/\text{L}$) than in treated one ($8.6\pm14.8\text{ mg}/\text{L}$).

Contrary to the minerals in serum which had no correlations, the minerals' (Cu, Zn and Se) concentrations in milk were positively correlated: copper concentration was correlated to zinc ($r=0.537$; $P<0.01$) and zinc was correlated to selenium ($r=0.415$; $P<0.05$). In a previous study (Faye et al., 2009), a negative correlation was observed between Zn and Se in camel serum, but the analysis included animals with selenosis which provoked inflammation process leading to a drastic decrease of zinc and iron in serum. Probably, the negative interaction between selenium and zinc in milk reported by some authors (Brätter et al., 1997) could be observable within a certain range of concentration when one element saturated the binding sites as it was observed between zinc and copper in camel serum (Bengoumi et al., 1998c).

Conclusion

The selenium supplementation by a single injection in pregnant camel at the end of the gestation was commonly used by the camel farmers in Saudi Arabia, in a context of a wide deficiency of the soil and forages in selenium. It appeared that this practice could improve slightly the selenium status of the new-born calves by increasing Se in milk at least in the colostrum. But the improvement seemed to have short effect. Other ways for selenium supplementation, as organic selenium distributed in the diet which was never tested in camel, could be applied and proposed to the camel farmers. A new experiment is currently testing the effect of organic selenium on the status of camel in this essential element.

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REGULAR ARTICLE

Some lipid components of the camel milk and blood in intensive farm in Saudi Arabia

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Abstract

Eight lactating camels in intensive dairy farm were sampled for the determination of some lipid components of milk and serum. The gross composition of camel milk samples was close that was shown in literature. The main milk fatty acids (FA) were represented by long chain FA. The proportion of polyunsaturated FA was 3.4%, of monounsaturated 30.3% and of saturated was 66.4% with a ratio saturated/unsaturated FA of 1.97:1. The total cholesterol was on average 118.5 ± 13.0 mg/L, while vitamin A was 419.9 ± 80.9 IU/L, vitamin E 20.2 ± 1.05 μ g/100mL and vitamin C, 26.1 ± 3.5 mg/L. Vitamin D3 was below the detection limit. In serum, four FA were mostly present: C16:0, C18:0, C18:1 n-9 and C18:2 n-6 representing 89.1% of the whole FA. Total cholesterol was on average 130.0 ± 18.7 mg/L. According to global FA status, saturated FAs were 59.1%, monounsaturated 16.2% and polyunsaturated 24.1% with a ratio saturated/unsaturated of 1.5 only. There was no significant correlation between cholesterol content in milk and in blood samples, also between the main FA in milk and blood. Under in-door system, the camel receiving intensive diet did not change significantly the main composition of its milk and serum except low level in vitamins.

Key words: Camel, Lipid, Milk, Serum, Intensive System

Introduction

The composition of the camel milk is widely described in the literature, especially regarding its gross composition for long time, the first publication on the camel milk composition dating from 1905 (see the meta-analysis of Konuspayeva et al., 2009). Recent advances in fine milk composition are also available, notably regarding the protein (Al-Haj and Al-Kanhal, 2010) or lipid composition (Konuspayeva et al., 2008). However, the observed variability is high and linked to the nutritional and physiological status of the animals.

It is known that main components of milk are coming from serum. But few data are available in the literature, especially on camel regarding parallel study on fatty acid and cholesterol content in milk and serum blood as well as regarding the level of

the vitamin content.

Moreover, the current intensification of the farming system in camel growing countries like Saudi Arabia could also have an effect on the milk composition, notably because the intensive in-door feeding (alfalfa hay with barley or wheat bran or other concentrates) leads to a monotonous diet far away from the variability of the desert plants.

In the present study, only female camels at similar lactation stage and receiving the same diet in an intensive dairy farm were taking in account in order to analyzed the variability of some gross (fat, protein, lactose and ash) and fine components (fatty acids, vitamins, cholesterol) of the camel milk, as well as in blood with the aim to compare the results to those of camel reared in other contexts.

Material and Methods

Location and animals

This study was carried out in the camel farm of Al -Jouf "Camel & Range Research Center" located in north-west Saudi Arabia, 950 km from Riyadh. Average annual temperature was 20°C, ranging from 12°C to 27°C, and average annual rainfall was 55 mm. The 8 sampled camels 5 to 11 years old belonged to four ecotype breeds: Malhah, Waddah,

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Hamrah and Safrah. The range of their live weights was 552 to 831 kg. Camels were kept in-door throughout the year and housed in pens. Their normal diet was composed of alfalfa (*ad-libitum*), barley (3 kg/day/animal), salt, wheat bran (1 kg/day/animal). As the calving season occurred between December and February, the milk sampling was achieved at different time for each animal according to their lactation stage in order to get milk samples at the same stage, i.e. at the third month of lactation.

Sampling

The individual milk production not including part drunken by camel calves was recorded routinely. The milk sampling was achieved at the morning milking time (6:00) in clean plastic bottles (40 mL) in each camel included in the monitoring. Approximately 20 mL of blood was collected at the mammary vein in vacutainer dry tube, then centrifuged (15 min, 8000 rpm) for getting serum.

Laboratory analysis

In milk, the gross composition was determined (fat, total protein, lactose and ash) by automatic milk analyzer (lactoscan MCC) calibrated for camel milk. Density and conductivity were also reported. The fatty acid (FA) composition was determined at the UMR IATE-lipotechnie (CIRAD, France) by using the method already described by Konuspayeva et al. (2008). In addition to that, cholesterol and fat soluble vitamins (A,D and E) and vitamin C were analyzed at the IDAC laboratory (Al-Kharj, KSA).

In serum, cholesterol, triglycerides were determined by Kenza-Max biochemistry analyzer (Biolabo, France). The fatty acid composition of the serum was determined by using capillary gas-liquid chromatography at IDAC laboratory (Al-Kharj, KSA).

Statistical analysis

The different parameters were described by their mean \pm standard-deviation and the correlations by using Pearson coefficient. The test of Mann-Whitney was used to compare the distribution of fatty acids between milk and serum samples. The software XLSTAT (Addinsoft[®]) was used for the data analysis.

Results

Milk components

The gross composition of camel milk samples was in g/L 29.4 \pm 0.99 fat matter, 28.7 \pm 2.0 proteins, 40.9 \pm 2.8 lactose, 0.72 \pm 0.05% ash, 1027 \pm 1 kg/m³ density, and 76.8 \pm 5.4 g/L solid non fat. The main milk fatty acids were myristic acid

(C14:0), palmitic acid (C16:0), palmitoleic acid (C16:1 n-7), stearic acid (c18:0) and oleic acid (C18:1 n-9) representing as the whole 86.7 % of the milk fatty acids (Table 1). The proportion of polyunsaturated fatty acids (PUFA) was 3.4%, of monounsaturated (MUFA) 30.3% and of saturated (SAT) was 66.4% with a ratio SAT/unsaturated fatty acid of 1.97. The total cholesterol in our camel milk samples was on average 118.5 \pm 13.0 mg/L (table 1) while vitamin A was 419.9 \pm 80.9 IU/L, vitamin E 20.2 \pm 1.05 μ g/100mL and vitamin C, 26.1 \pm 3.5 mg/L. Vitamin D3 was below the detection limit.

Table 1. Composition on some lipid components and vitamins of the camel milk and blood in intensive farming system in Saudi Arabia.

| Components | Milk | Serum |
|----------------------------|------------------|------------------|
| Total fat (%) | 2.94 \pm 0.99 | nd |
| Cholesterol (mg/L) | 118.5 \pm 13.0 | 13.0 \pm 1.8 |
| Triglycerides (g/L) | nd | 0.5 \pm 0.2 |
| C4:0 | 0.11 \pm 0.08 | - |
| C6:0 | 0.9 \pm 0.06 | - |
| C8:0 | 0.22 \pm 0.05 | - |
| C10:0 | 0.23 \pm 0.08 | - |
| C12:0 | 1.54 \pm 0.72 | 0.32 \pm 0.04 |
| C14:0 | 15.89 \pm 2.66 | 2.29 \pm 0.25 |
| C15:0 ante iso | 0.56 \pm 0.08 | - |
| C15:0 | 1.39 \pm 0.16 | 0.76 \pm 0.14 |
| C16:0 iso | 0.50 \pm 0.09 | - |
| C16:0 | 34.65 \pm 3.91 | 30.09 \pm 3.48 |
| C16:0 isom | 0.73 \pm 0.34 | - |
| C16:1 (n-7) | 11.87 \pm 1.54 | - |
| C17:0 iso | 0.89 \pm 0.17 | - |
| C17:0 | 0.58 \pm 0.09 | - |
| C17:1 | 0.64 \pm 0.14 | - |
| C18:0 | 8.88 \pm 1.49 | 0.52 \pm 0.09 |
| C18:1 iso | 0.82 \pm 0.94 | - |
| C18:1 (n-9) | 15.44 \pm 2.64 | 23.38 \pm 1.85 |
| C18:1 (n-7) | 1.24 \pm 0.37 | - |
| C18:2iso | 0.23 \pm 0.06 | |
| C18:2(n-6) | 2.14 \pm 0.17 | 16.13 \pm 1.55 |
| C18:3 (n-6) | 0.28 \pm 0.12 | 19.51 \pm 1.63 |
| C18:3 (n-3) | 0.51 \pm 0.06 | - |
| C20:1 (n-9) | 0.23 \pm 0.04 | 1.05 \pm 0.34 |
| C20:4 | - | 3.45 \pm 0.68 |
| C20:5 (n-3) | 0.05 \pm 0.02 | - |
| C22:0 | - | 0.53 \pm 0.15 |
| C22:6 (n-3) | 0.19 \pm 0.06 | - |
| C23:0 | - | 0.65 \pm 0.2 |
| C24:0 | - | 0.51 \pm 0.13 |
| Vitamin A (μ g/100mL) | 12.6 \pm 2,4 | nd |
| Vitamin E (μ g/100mL) | 20.2 \pm 1.05 | nd |
| Vitamin C (mg/L) | 26.1 \pm 3.5 | nd |
| Vitamin D3 (IU/L) | BLD | BLD |

nd : non determined, BLD: below limit of detection

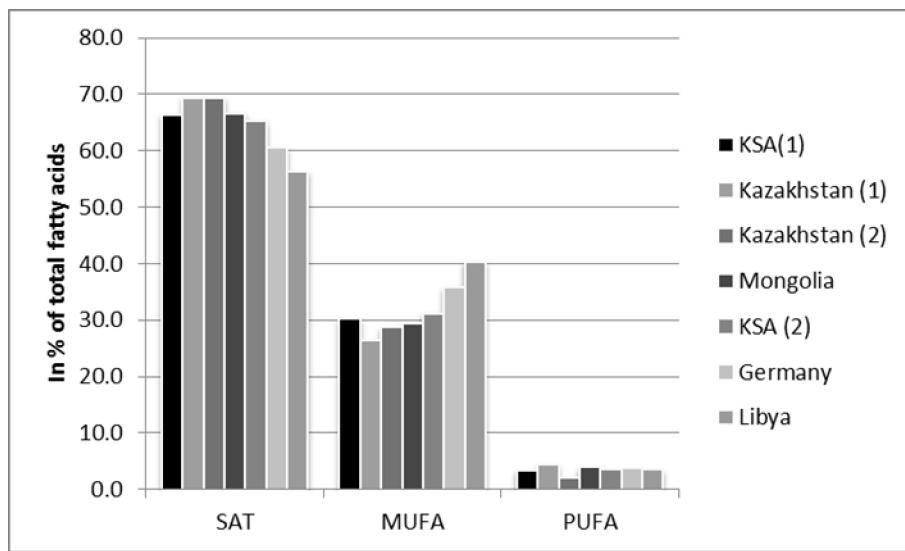


Figure 1. Comparison between fatty acid composition of camel milk according to Narmuratova et al, 2006 (Kazakhstan 1), Konuspayeva et al., 2008 (Kazakhstan 2), Jirimutu et al., 2010 (Mongolia), Dreiucker and Vetter, 2011 (Germany), Shibani et al., 2011 (Libya), Faye et al., 2013 (KSA1) and our results (KSA2).

Serum components

In serum, four acids only were widely present. Palmitic acid (C16:0), stearic acid (C18:0), oleic acid (C18:1 n-9) and linoleic acid (C18:2 n-6) representing 89.1 of the whole fatty acids (Table 1). Total cholesterol was on average 130.0 ± 18.7 mg/L (Table 1). According to saturated status of FA, SAT was 59.1%, MUFA 16.2% and PUFA 24.1% with a ratio saturated/unsaturated of 1.5 only.

There was no significant correlation between cholesterol content in milk and in blood samples. There was no correlation also between the main fatty acids in milk and blood (palmitic, stearic, oleic and linoleic acids). However, the distribution of FA groups (SAT, MUFA and PUFA) were comparable in milk and blood (test of Mann-Whitney not significant).

Discussion

On average, the fatty acid composition of dromedary milk in our study was in the range of the values reported in the recent references in very various conditions (FIGURE 1): Samples mixing Bactrian and dromedary camels (Konuspayeva et al., 2008; Narmuratova et al., 2005), Maghrebi camel from Saudi Arabia (Shibani et al., 2011) or dromedaries reared in Germany (Dreiucker and Vetter, 2011).

The proportion of unsaturated fatty acids in camel milk (33.6%) was higher than in cow milk (24.1% on average) as well as short-chain fatty acids (Attia et al., 2000; Karray et al., 2005). The camel milk was poor in short-chain fatty acids (C4:0 = 0.11%) compared to cow milk, which contains more than 3.0% of butyric acid (Schroeder et al., 2003). This confers upon camel milk some interesting nutritional properties; in particular, if we refer to some papers classifying short-chain fatty acids as promoters of atherosclerosis. The sum of short chain fatty acids C4 to C8 was only 0.52% in our camel milk samples, and 8.99% in the milk of cows fed with a nutritionally balanced diet (Palmquist et al., 1993). The long chain fatty acids C15 to C22 were much higher (81.8%) in our samples than in cow's milk (66.1%) (Palmquist et al., 1993). Content in C18:3 were 10 times more in camel's milk (0.79) than in cow's milk (0.07).

No reference was available on fatty acid composition of camel serum. In human, similar proportions of unsaturated and saturated fatty acids in serum and milk were reported with no significant changes along the lactation (Spear et al., 1992). The same fatty acids were in higher proportion (C16:0, C18:0, C18:1 n-9, C18:2 n-6, and C20:4) both in human and camel serum.

Regarding, cholesterol, the content in our camel milk samples appeared comparable to that in cow milk, (12-17 mg/100mL) (Sieber, 2005) and lower

than in ovine milk (28.8 mg/100 mL) (Goudjil et al., 2003). However, our result was quite lower than the value reported in camel milk from Kazakhstan (37.1 ± 7.73 mg/100mL) by Konuspayeva et al. (2008). According to Gorban and Izzeldin (1999), camel milk had a higher content of total cholesterol (31.3 mg/100mL) compared to cow milk (25.6 mg/100mL). However, the higher value observed by some authors could be due to the total fat content of camel milk (for example 6.4% on average in samples from Kazakhstan) which was nearly twice that in cow milk (3.4% on average) contrary to our results where fat content in camel milk appeared rather low (2.9% only on average).

The total cholesterol in camel serum was reported to be 235 ± 20 mg/100mL in Saudi Arabia (Ali et al., 2010), 35.4 to 48.7 mg/100mL in India (Gupta et al., 2012), 40.2 ± 12.4 mg/100mL in Iran (Mohri et al., 2008). Such wide values could be due to the analytical procedures. Our results were in the mean of those reported data. Regarding triglycerides, our results (50.0 mg/100 mL) were higher than those of Gupta et al. (2012) (22.8 - 27.9 mg/100mL), but quite lower than those of Ali et al. (2010) (173 ± 13 mg/100mL).

The content in vitamin A in our milk samples appeared in low quantity compared to the reported results of Stahl et al. (2006) (20.1 ± 1.0 μ g/100mL) and quite less than the retinol content in cow milk (for example 60.9 μ g/100mL for Stahl et al., 2006). Vitamin E appeared also in our milk samples lower than the 32.7 ± 12.8 μ g/100mL reported by the same authors. The vitamin C content was also lower in our samples than the values reported by Stahl et al. (2006) in Emirates (52.5 ± 15.8 mg/L) and overall by Konuspayeva et al. (2011) in Kazakhstan (150.4 ± 105 mg/L). Thus, globally, our camel milk samples appeared poor in vitamins.

Conclusion

Under in-door system, the camel receiving intensive diet did not change drastically the main composition of its milk and serum. However, due to the nature of the grass (mainly hay of alfalfa from irrigated field) and of the concentrates (cereals), the vitamins in milk appeared relatively low compared to camel grazing out-door. Further researches have to be implemented to deepen the risk of impoverishment of dietetic and nutritive value of camel milk in case of intensification of the camel production.

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REGULAR ARTICLE

Some parameters to process camel milk into cheese

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Abstract

Cheese from camel milk was never produced by traditional way. However, Hansen[®] (Denmark) delivered recently new coagulant agent named "Chy-Max M" containing transgenic camel chymosine. In the present study, impact of calcium, lactation stage and curd acidification were investigated. Camel milk was shared into 6 samples (100g each) submitted to 3 types of treatment (1. calcium chloride solution (500 g/L diluted 1/10 water); 2. powder of calcium phosphate; 3. no calcium) and 2 temperatures (20°C/36°C). Rennet 50 µL/L (Chy-Max) was added in all samples. Milk coagulation was faster at 36°C and renneting pH lower. No difference in clotting time and curd firmness after calcium addition was observed. The curd firmness at 36°C was stronger than at 20°C. For measuring impact of lactation stage, coagulation capacity and curd yield on milk was tested in milk provided by one camel from 12th to 25th day postpartum. Milk was coagulated by Chy-Max (50 µL/L/20°C). No coagulation was observed in the first days of experiment. Then curd start to be formed, but with low yield. The curd was correct and ready to use for cheese making only from the 20th day post-partum. Acidification of camel cheese curd without starters was measured at 20°C and 36°C during 10 hours. Milk pH and curd pH were measured during all cheese processing. At the beginning, milk pH was 6.38 whatever the temperature. Acidification was faster at 36°C than at 20°C. At the time of coagulation, pH of 20°C curd was 5.80 vs 5.08 at 36°C.

Key words: Camel Cheese, Fermentation, Calcium, Lactation stage

Introduction

In the world, camel milk is better known for its fermented products: *shubat* – in Kazakhstan; *chal* – in Turkmenistan; *khoormog* – in Mongolia; *gariss* – in Sudan; *suusac* – in Kenya, *zrig* -in Mauritania, rather than for its types of cheeses: *chuku* – in Niger or *caravan* – in Mauritania, fresh camel cheese – in Morocco (Bengoumi et al., 2002; Konuspayeva and Faye, 2010; Benkerroum et al., 2011). In the literature, there are some data on the use of bovine rennet, or rennet agent coming from vegetal sources for camel cheese making (Ramet, 1989; Boudjenah-Haroun et al., 2011; Boudjenah-Haroun et al., 2012; Ahmed and El Zubeir, 2011). Regarding bovine rennet, a lot of parameters

(rennet quantity, time of coagulation, curd description, pH value) for technological production of cheese from camel milk were studied by Ramet (1985).

However, HansenTM (Denmark) delivered recently new coagulant agent named "Chy-Max M" containing camel chymosine (Sorensen et al., 2011). With such camel rennet, no data about power and time of coagulation, acidification of curd, impact of physiological and environmental factors to coagulation of camel milk, was available. In the present study, impact of calcium and of lactation stage on coagulation to produce cheese, and then curd acidification of coagulated camel milk were investigated.

Material and Methods

Camel milk and early milk were sampled from healthy dromedary camels from Camel and Range Research Center, Al-Jouf, KSA at mid of lactation stage and between 12th to 25th days of lactation respectively. Percentage of fat and total protein was determined by automatic milk analyzer device

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(Lactoscan MCC) calibrated for camel milk. The ultrasonic technology used by Lactoscan allowed direct measurement of fat, proteins, lactose and salts. Lactoscan determined also the freezing point of each sample and the quantity of added water. The freezing point was calculated automatically from the components it depends on.

For clotting camel milk, specific liquid chymosin for camel milk – ChyMax M (Hansen[®], Denmark) was used. Dose 50 μ L/L was added with preliminary dilution 1/20.

Coagulation properties

Camel milk was shared into 6 samples of 100 g each submitted to 3 treatment: (i) calcium chloride solution (500 g/L diluted 1/10 water); (ii) powder of calcium phosphate; and (iii) control with no calcium. Two temperatures 20°C and 36°C were tested. After 30 min of heating or not, 50 μ L/L of rennet Chy-Max M (strength 1000 IMCU, international milk coagulating units) was added in all samples. The pH value was measured. Then visual determination of clotting time was done and after 60 min, the curd was cut and filtration through cloth was achieved. The weight of the curd (gross yield) was measured 1h 30 after clotting. Corrected yield was calculated as:

Corrected yield (DM curd=30% and DM whey= 6). Gross yield = [(DM curd- DM whey) / (30 – 6)], with DM = dry matter.

Impact of lactation period

For measuring impact of lactation stage, coagulation capacity and curd yield on milk was

tested in milk provided by one camel from 12th to 27th day *postpartum*. Milk was coagulated by Chy-Max M (50 μ L/L/20°C).

Natural acidification of camel cheese curd

The pH value was measured at 20°C and 36°C during 10 hours with Ph-meter Hanna Instruments HI221 pH/mV/ORP

Results

Coagulation properties

Before testing the milk, its gross physico-chemical composition was analyzed (Table 1) and its microbiological status was assessed (total flora and coliforms).

Table 1. Global composition of camel milk (g/100g).

| Parameters | Mean and SD |
|---------------|-----------------|
| Fat | 2.72 \pm 0.17 |
| Solid non-fat | 9.37 \pm 0.12 |
| Protein | 2.83 \pm 0.04 |

Milk coagulation was faster at 36°C and pH renneting lower (Table 2). No difference in clotting time and curd firmness between calcium treatments was observed. The curd firmness at 36°C was stronger. The molding was more effective with the curd obtained at 36°C.

The effect of calcium salt quantity was also tested at 36°C (Table 3). There was no effect on type of calcium source and of the dose on the time of coagulation, pH value and on curd yield comparatively to control.

Table 2. Coagulation characteristics as function of type of calcium added.

| Parameters | 20°C | | | 36°C | | |
|------------------------|-----------------|----------------------|------------------------|-----------------|----------------------|------------------------|
| | Control | Ca phosphate (1g/kg) | Ca chloride (0.1mL/kg) | Control | Ca phosphate (1g/kg) | Ca chloride (0.1mL/kg) |
| pH renneting | 6.26 \pm 0.05 | 6.25 \pm 0.05 | 6.25 \pm 0.05 | 5.78 \pm 0.11 | 5.75 \pm 0.11 | 5.75 \pm 0.07 |
| Coagulation time (min) | 14 \pm 0 | 14 \pm 0 | 14 \pm 0 | 6 \pm 0 | 6 \pm 0 | 6 \pm 0 |

Table 3. The dose of different calcium source on coagulation of camel milk at 36°C.

| | pH renneting 36°C | Coagulation Time (min) | Yield(g/100g) 1h30after moulding | Dry matter | Corrected yield |
|---------------------------|-------------------|------------------------|----------------------------------|---------------|-----------------|
| | | | | Curd (g/100g) | (g/100g) |
| Control | 6.40 | 8 | 14.80 | 27.46 | 13.07 |
| Phosphate Ca 2g/L | 6.40 | 8 | 12.91 | 31.10 | 13.45 |
| Phosphate Ca 4 g/L | 6.40 | 8 | 13.26 | 31.33 | 13.97 |
| CaCl ₂ 0.2mL/L | 6.37 | 8 | 14.35 | 28.94 | 13.65 |

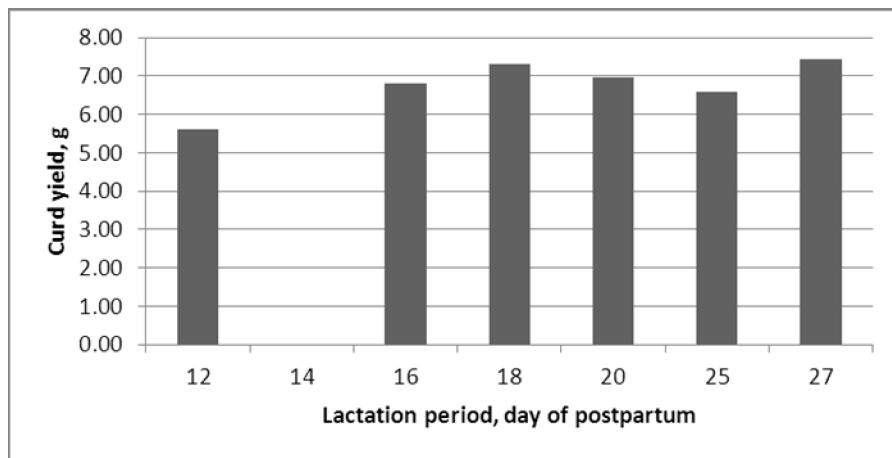


Figure 1. Curd yield according to stage of lactation of camel from 12th *postpartum* day.

Impact of lactation stage on coagulation capacity and curd yield

No coagulation was observed before 12th day of lactation (Figure 1). At 12th *postpartum* day, first coagulation induced the formation of a very weak curd and low yield. At 14th day no coagulation was observed and consequently, and no curd was obtained. Then curd became better, with increase of

curd yield. The milk at 25-27th *postpartum* day was acceptable to get curd and was ready to use for cheese making.

Natural acidification of camel cheese curd

At the beginning, milk pH was 6.38 whatever the temperature (Figure 2). Acidification was faster at 36°C. At the end (when coagulation occurred), pH of 20°C milk was 5.80 vs 5.08 at 36°C.

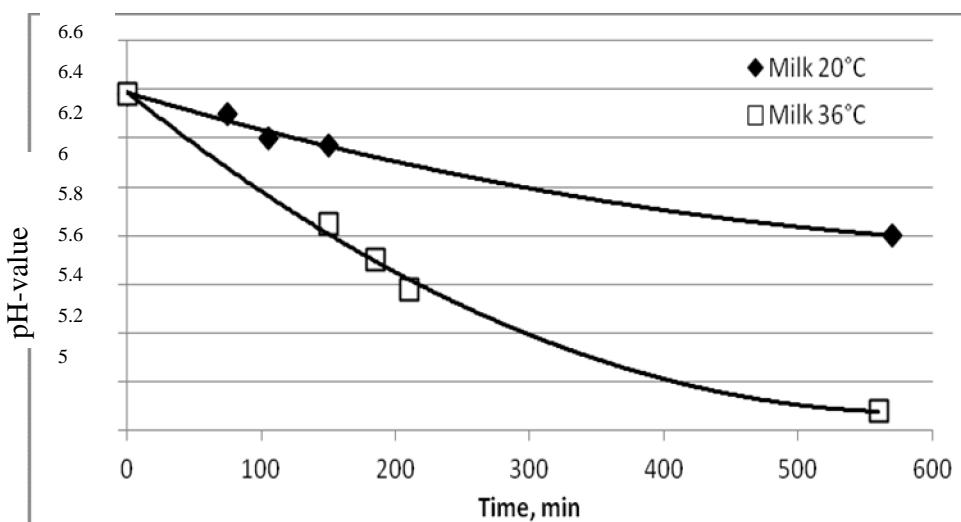


Figure 2. Acidification curves of camel milk at 20°C and 36°C.

Discussion

The physico-chemical composition of camel milk was analyzed before starting the experimentation. The fat and protein contents of our camel milk were in the range of the normal values reported in the literature (Farah, 1993; Konuspayeva et al., 2009).

To transform milk into cheese, the gel obtained after coagulation play important role. For cow milk, calcium ions help to attend this gel stable in all types of milk. Usually calcium phosphate or calcium chloride is used, mainly on milk after heat treatment. It is stated that to get firmness curd of camel milk, 10-15 g of calcium chloride per 100 kg of milk have to be added when bovine rennet is used (Ramet, 1985, Benkerroum et al., 2011). Indeed, in our trial, camel milk was not heated. In such conditions, camel milk showed no effect of adding of calcium ions, whatever the form, phosphate or chloride on clotting time and yield. In all published data, the described trials used heat treated camel milk.

The effect of lactation stage on cheese making is known mainly with cow, goat or ewe milk. With camel milk no data was available in the literature. It is stated that at the first month *post-partum*, the quality of protein in milk undergoes important changes: immunoglobulins and some other whey proteins decreased and proteins from complex casein increased. For cheese making, only casein proteins are of main interest. The optimal time for cheese making will be after 25 days *post-partum*.

In the case of preparation of different types of cheese from camel milk, it is necessary to know the acidification patterns, how many times it takes before attend determined pH value. For coagulation of milk, 3 types of coagulation are described: rennet-coagulation, lactic coagulation and mixed coagulation (Goudedranche et al., 2001). For camel milk, only bovine rennet was tested or extract of young camel stomach (Boudjenah-Haroun et al., 2011). In the literature, no data on coagulation with pure camel rennet is available.

Only one reference using Chymax of Hansen company were used, but it was bovine one (Benkerroum et al., 2011). In our trial, only 50 µl was used (Chymax M strength 1000 IMCU) per liter of raw milk instead 170 µl (Chymax-bovine strength 600IMCU) per liter of pasteurized milk by Benkerroum et al. (2011). Also, regarding the type of coagulation, these authors used lactic coagulation for preparing soft cheese from camel milk. Milk acidification in their trial was faster in the presence of *Streptococcus thermophilus* and

Lactobacillus bulgaricus. The pH value decreased below 5 after 240 minutes in room temperature. In our trial, such decreasing needed more than 500 minutes, because no starters were used and the acidification was natural.

These technological parameters of camel milk processing into cheese by camel rennet represent informative steps for further trials and could be useful for industrial scale cheese processing of camel milk.

Acknowledgment

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REGULAR ARTICLE

Camel milk value chain in Northern Saudi Arabia

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Abstract

In Saudi Arabia, the increasing demand in camel milk by a growing urbanized population is stimulating the development of camel dairy farms, especially around the towns. The average per capita consumption in the country is about 33 L / year. It was reported that the production of camel milk is potentially higher than that of the cow in the same farming and climatic conditions. With an individual production between 5 to 20 l/day, the production potential of camel is far away from negligible. However, the dairy value chain is not well known except for the biggest dairy farms. In the present study, a survey including 119 camel farms belonging to all kind of farming system was achieved in the northern part of the country. It showed that only 16 farms contributed to the camel milk market, the other ones producing milk only for self-consumption. The market integrated sector is weakly organized, except for the industrial farms. Indeed, it is represented by two subsystems: (i) an informal one based on suburban farming with traditional mini-dairy plants and delivering milk in local shops and retail outlets; (ii) a formal system represented by large modern dairy farms and dairy plants approved by Ministry of Agriculture. These two subsystems produced 1176.44 t/year, while the volume self-consumed was estimated to be 1854t/year. Such, the market potential for camel milk could be highly developed in the future.

Key words: Saudi Arabia, Camel milk, Milk value-chain, Dairy system

Introduction

In the Kingdom of Saudi Arabia (KSA), camel milk is consumed in relatively high quantity, especially during different celebrations. On the base of FAO statistics (FAO Stat, 2012), the consumption per inhabitant in KSA is approximately 33 l/hab/year which places the country among the large-scale consumers in the world (Map. 1). Elsewhere, as camel milk demand is higher than the offer, the market price is high, almost twice the cow milk price (Ismail and Al-Mutairi, 1994). Yet, in spite of the modernization of camel dairy farms (milking machine, in-door feeding, genetic selection, intensification etc.,), in spite of the high demand for cultural and health reasons, the camel milk sector appears weakly organized compared to cow milk sector.

In order to understand the added value chain of the camel milk sector in KSA and to estimate the

production potential for the camel milk sector, a survey was achieved among producers, processors and distributors. The survey was limited to the Northern part of the country.

Material and Methods

Place of the study

The present study was achieved in the northern part of KSA around Nafud desert, most precisely in the neighborhood of the towns of Sakaka, Doumat-al-Jandal, Gurayat, Hail, Ar'ar and Tabarjal (Map 2). It was supported by the Camel and Range Research Center based at Sakakah (Al-Jouf province).

Added value chain approach

The added value approach for a determined product as camel milk allows identifying the relationships between the different segments of the commodity chain, their complementarity and their pathway between the different stages of process within the systems (Duteurtre et al., 2000). Three aspects have to be taken in account (Boutonnet, 2010): (i) the height of the channel including the different activities or functions (production, processing, distribution, and consumption), (ii) the width involving the different modalities of the

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channel within the different sub-system, and (iii) the thickness corresponding to the diversification of the products and their geographical expansion.

To achieve this approach, data on the quantification of the flow (production, marketing, purchasing, consumption) and on the strategies of production and marketing are necessary.

Survey design

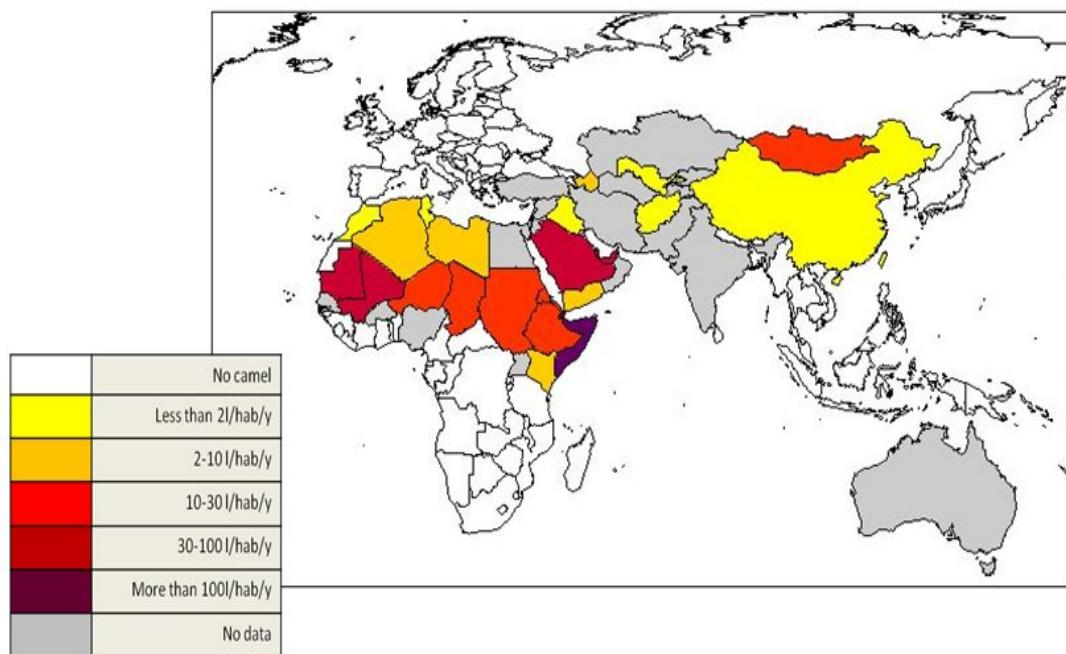
The study included two different methods: (i) the collect of indirect information from bibliography and available local statistics, and (ii) the collect of direct data among the different stakeholders of the camel milk sector (producers, carriers, processors, distributors) based on questionnaire adapted to each. The questionnaire for the producers included data on their status (place, age, tribe), the herd composition (species, breed, age), and the milk (production, price, market integration). Regarding the distributors and the shops, data on the sold and purchased quantity of milk, the prices and the benefit were collected. For dairy plants, the data involved the owner status, the processed volume, the organization of the service, the milk prices and the perspectives for the region.

Sampling procedure

The camel milk producers were selected randomly, except for the big farms processing their milk which were exhaustively interviewed. The selection of the shops was based on the knowledge of the producers. As the whole, 119 camel milk producers and 16 sale points in the main towns or along the roads were inquired.

Statistical analysis

The data were managed in Excel table, then analyzed by XLstat software (Addinsoft[®]). In order to obtain homogenous table including qualitative data only, the quantitative data were analyzed by Principal components analysis (PCA) followed by Ascending Hierarchical Classification (AHC), and the convenient classes were used as modalities of synthetic qualitative variables used in the final analysis. The qualitative data were analyzed by Multiple Correspondence analysis (MCA) and the types of stakeholders were identified after cluster analysis (Jobson, 1992). The variance analysis was used to determine the significant differences in quantitative data (milk production, number of camels) between modalities of qualitative variables. Chi square test was used for contingency tables crossing the qualitative variables two by two.



Map 1. Camel milk consumption in l/hab/year in 2009 (according to Faye and Bonnet, 2012).



Map 2. Localization of the study zone (Source Wikipedia).

Results and discussion

The producers

Among the 119 camel producers, 35% only were pure breeders. The sample included also retired people (24%), civil servants with the government (21%), security agents as policeman or military (13%), and education workers (7%). Thus, the multi-activity of the camel producers is highly underlined and is in accordance with the observations of Abdallah and Faye (2013): in a survey including 218 camel owners from Northern KSA, 37% were pure Bedouins living in desert, 9% were civil servants including education field and living mainly in town, 30% were agents working in security field, 17% were retired people and the remain being of different origins. Regarding the camelstock system, the producers were classified in extensive system (mainly bedouin and representing 36% of the producers), semi-intensive system (feeding supplementation, sedentarisation) representing 24% of the camel farmers, periurban system located around the towns, 35%, and intensive systems (with irrigated fodders, modern camel housing, in-door feeding), 5% only. In spite of higher mean daily production in intensive

system, no significant difference was observed between the systems (Figure 1). On average in our sample, the mean daily milk production was 5.04 ± 2.46 l/camel/day with a herd range of 3 to 14 l/day. However, few of the camel farms ($n=16$) were selling milk on the market. In the remaining farms, the milk was self-consumed.

The herd size was on average 70 ± 227 heads with a high variability within each system, explaining the lack of statistical difference between extensive (61 ± 33), semi-intensive (47 ± 45), periurban (96 ± 381) and intensive (72 ± 32). By considering all the qualitative variables describing the camel farms (multi-activity, farming system, seniority of the owner, modality of herd size, milk production level, type of milking, milk marketing, breed composition of the camel herd, choice of the reproducers and strategy for increasing milk production), the multivariate analysis (MCA + AHC) allowed identifying 3 types of camel producers (Figure 2) explaining 55% of the variance.

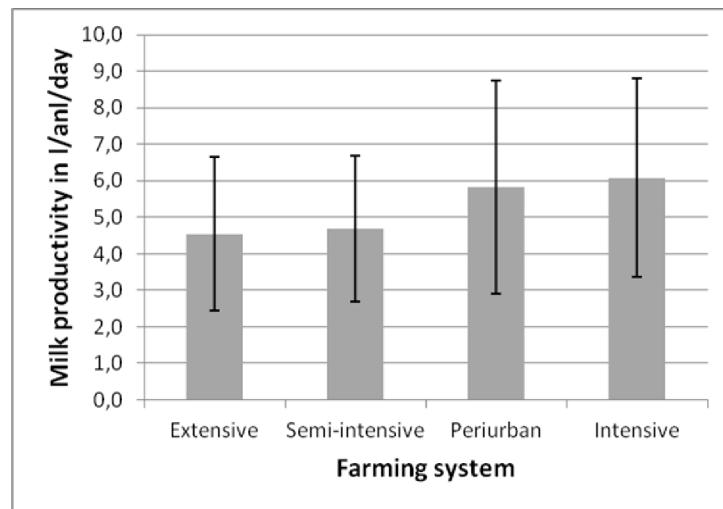


Figure 1. Mean daily milk production in camel from different farming systems.

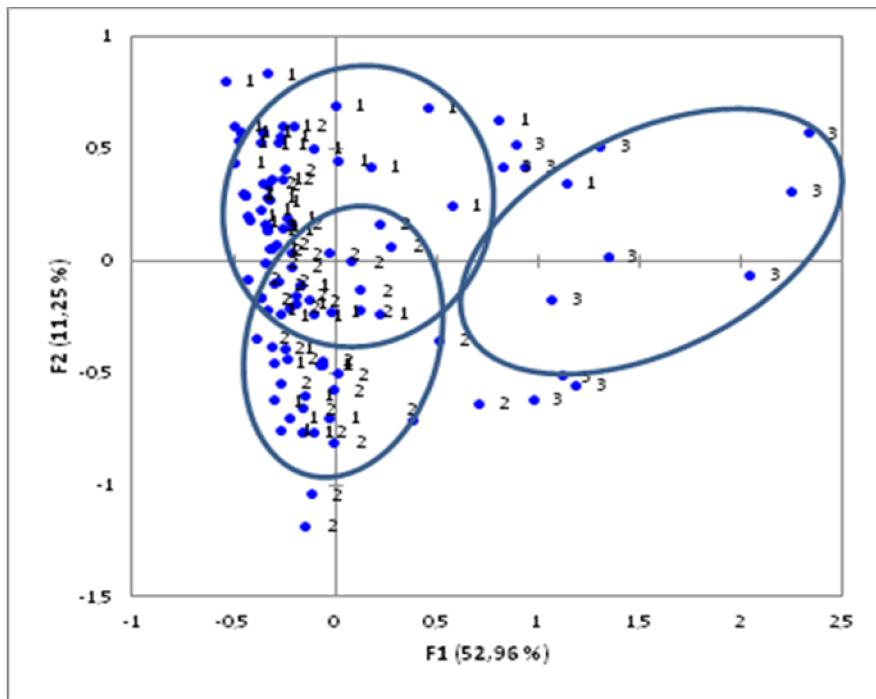


Figure 2. Projection of the 3 classes (ellipse of inertia) obtained after cluster analysis on the main factorial plan (1,2) of the MCA.

The type 1 (n=53) corresponded to farms without milk marketing, mainly in extensive system, small or medium herd size. The type 2 (n=44), did not sell milk in majority also and corresponded mainly to extensive or periurban system with small or medium herd size. The type 3 (n=12) was all farms integrated into milk market,

using milking machine, corresponding mainly to intensive system with small or big herd size. The total milk production was significantly higher in this type compared to the others (Figure 3).

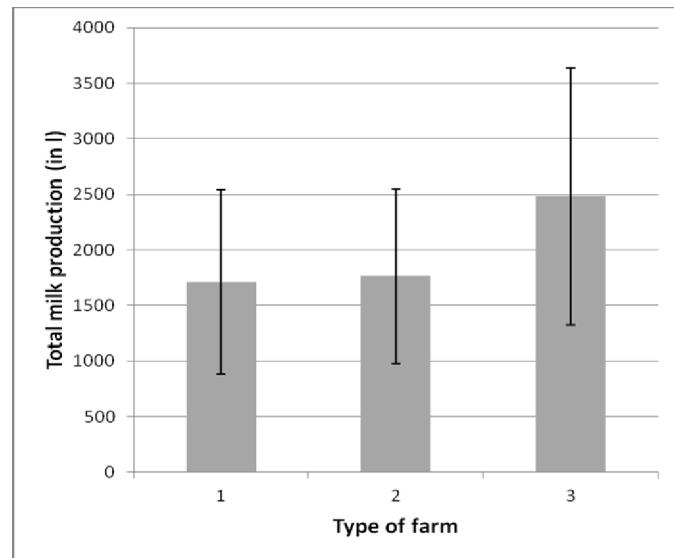


Figure 3. Total milk production per lactation/camel (in l) according to the type of farm.

Regarding the milk marketing, 2 subsystems could be described:

- The formal sub-system including two big integrated intensive farms (Watania and Turath), the farm of the Camel and Range Research Center and one producer having agreement for camel milk selling. These farms have big herd size, milking machine and dairy plant processing pasteurized milk, packaged in plastic bottles. The camel herd is under veterinary control and a part of the feed is produced on-farm. The milk productivity was 2240 l/lactation.
- The informal sub-system including periurban producers having small-scale traditional dairy plant producing raw or fermented milk, packaged in plastic bag, usually without agreement. The feed is produced out of the farm, but the non-productive part of the herd could be maintained in desert pasture. The milk productivity was 2090 l/lactation

In addition, the remaining producers were classified into “out milk market system”. The productivity was estimated to 1659 l/lactation. The separation into formal and informal sub-system in dairy sector is usual in many countries, notably in Africa (Corniaux et al., 2007; Sow Dia et al., 2007). The camel milk processing in Saudi Arabia, contrary to Mauritania for example (Abeiderrahmane, 1997), was characterized by a poor diversification of the products. Only fresh, fermented or pasteurized was proposed to the

consumers. The cheese processing was only experimental for the moment (Konuspayeva et al., 2012).

The milk marketing

As mentioned above, the sold milk was packaged either in plastic bag (in 56% of the farmers selling milk) or in plastic bottles (44% of the farmers). All the camel milk producers managed the packaging themselves. There was no dairy plant out of the camel farms. Three market chains were used by the farmers: (i) producers having traditional dairy workshop selling milk to local small shops and mini-markets and a lower part directly to the consumers; (ii) producers mainly in Ar’ar region, having medium herd size in settled enclosures (*chabek*) and selling all the milk directly to the consumers in bulk, along the roads; (iii) producers selling all the milk to distributors or having their own distribution network, notably the big integrated dairy farms like Watania.

The camel milk price was 7 to 12 Saudi Rials (SAR) per liter according to the type of packaging and the type of milk (fresh, fermented or pasteurized). The milk bottle (1l) produced by small scale dairy plant was sold 8 SAR. It was 10 SAR for pasteurized milk from big dairy companies. The margin between production price and consumption price was around one SAR/l (0.21 €).

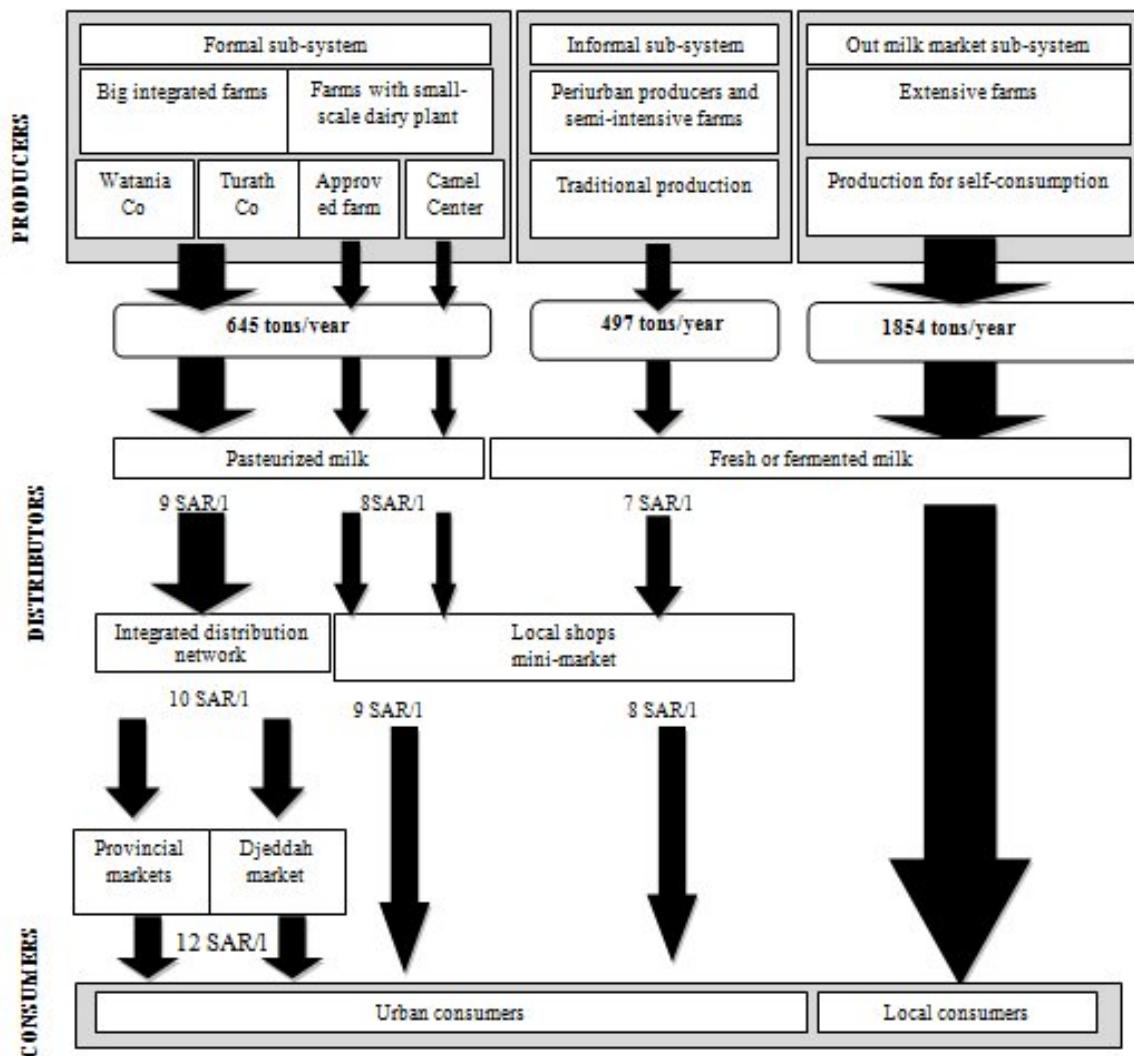


Figure 4. Conceptual model of the camel milk value chain in Northern Saudi-Arabia.

The milk flow

The total milk flow was estimated according to the number of lactating animal in formal and informal sector and of their mean milk productivity. The data were checked beside the selling point. Regarding the camel farmers no selling milk ($n=103$), ten of them did not milk the lactating animals at the time of the survey (all milk was given to the camel calf). In the remaining producers, self-consumption was estimated according to the number of milked animals and to their productivity.

Finally, the camel milk quantity available for consumers was estimated to 654 tons/year in formal sector, 497 tons/year for informal sector, and the self-consumption was estimated to 1854

tons/year, probably under-estimated. Based on these data, a conceptual model of the camel milk value chain in Northern KSA could be proposed (Figure 4).

The added value chain analysis was already applied to study the camel milk commodity channel in Mauritania (Kouassi, 1998).

Conclusion

Traditionally regarded as a gift for the visitors, the camel milk was recently integrated in the market in many countries of the camel world. The urbanization and the modernization of the farming systems had contributed to the development of a camel milk commodity channel although, the organization of this value chain is just beginning. In Saudi Arabia, the potential for high development

of a camel milk sector is existing, but is still dominated by informal sector (not only in volume, but mainly in number of stakeholders) and by self-consumption. The distribution network, except for the big integrated farms, is limited to small shops in the towns. For example, it is noticeable that camel milk is very rarely available in the main chain of supermarkets in the northern part of the country. Yet, the demand is increasing in spite of the high price of the camel milk. The development of the camel milk value chain requires a better selection of the best dairy animals, a better access to the urban market, an efficient quality control and a distribution network fleshed out.

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REGULAR ARTICLE

Comparison of dairy performances between dromedaries, bactrian and crossbred camels in the conditions of South Kazakhstan

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Abstract

The aims of the work compare similarly the yield and the composition. In this work determined the Camel milk composition (fat content, dry matter, density) and milk yield of Dromedaries, Bactrians and Hybrids in South-Kazakhstan condition in same farm, same time and repeated same animals. The milk sampled of 20 camel's milk, where 6 Bactrians (B), 5 dromedaries (D), 2 hybrids F1 *Iner* (I), 4 hybrids F1¹ *Nar* (N), and finally 3 hybrids F2 *Kospak* (K) with repeated 3 times (days). The milk of Bactrian camels contained significantly more DM and the same tendency was noted for the fat content. In the same time, the milk yield tended to be lower even if no signification threshold was reached. Contrarily, the milk of dromedaries was not so rich in absence of any significant difference to F1 and F2 hybrids except an increased density. F1 hybrids (*Nar-maya* and *Iner-maya*) had a slight but not significant tendency of increased milk yield but a more or less reduced contents and density. This difference seems to be extenuated for F2 (*Kospak*) animals. The effect of calving year was illustrated by significantly lower milk yields in the second year of lactation (3.8 versus 2.8 L/d, $P<0.05$), slightly increased contents of fat (4.9 versus 4.2 g/L, $P<0.10$) and Dry matter (14.0 and 13.8 g/L, NS) and also density (1030.0 versus 1032.3 g/L).

Key words: Milk yield, Composition, Camel species, Kazakhstan

Introduction

The Republic of Kazakhstan is an original area of camel breeding as different populations of old-world camels cohabit on its territory. There are 186.6 thousands of heads camels (Agency of the Republic of Kazakhstan on Statistics, 2013). Indeed, there are double-humped (*Camelus bactrianus*) and one-humped (*Camelus dromedarius*) camels as well as hybrids at different levels of hybridization (Faye and Konuspayeva, 2012a). Depending on their geographical location Kazakh Bactrian camels were described in detail and proved in the form of genetic types:

- Uralo-Bukeyev type: most large animals, common in the north of the Caspian Sea (living in Atyraou, West Kazakhstan and Aktobe regions);

- Kyzylorda type: a smaller-sized animals, spread around the Aral Sea and along the course of the Syr Darya River (South part of Aktobe and Kyzylorda);

- Ontustik-Kazakhstan type (the South Kazakhstan): Kazakh Bactrian camels are small, but have all the productive characteristics of the breed, common in the South (South Kazakhstan, Zhambyl and Almaty region) (Terentyev, 1975).

The Bactrian camel is the species historically present in the colder part of Central Asia (Mongolia, NW-China and Kazakhstan) as these animals are better adapted to the strong winter by developing a thick woolen coat and their higher milk fat content to nourish the calf. Moreover, the more productive dromedary population which is widespread in the southern part of Asia and especially the Turkmen Arvana breed is present in the overlapping zone of both populations on the territory of Kazakhstan. Therefore, Kazakh camel breeder can hybridize these species to produce fertile off spring for dairy purposes (Skidmore et al., 2001) which would cohabit in the same herd (Faye and Konuspayeva, 2012b).

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The dairy production by a herd composed by different species raised the question of the differences in milk yield and composition. Generally, Bactrian camels are known to be less productive. A comparison of milk composition between both species in different Kazakh herds (Faye et al., 2008) showed increased fat and protein content in milk of Bactrian camels in comparison to dromedaries and lower milk density in Bactrian compared to this density in hybrids. Nevertheless, the main product of Kazakh camel breeder is *shubat*, a fermented product based on the whole milk what make the breeder sensitive to improve especially the milk yield of their animals.

Therefore, the present work aims to compare similarly the yield and the composition in milk of both Old World species as well as hybrids in a Kazakh production system.

Materials and Methods

The trial was carried out in the village Aigene (43°20' N, 79°58' E) in South Kazakhstan (Suzak region) situated on the borderline between steppe and the desert Moyumkum (Figure 1). This zone is characterized by few rainfall (<150 mm per year) and huge variations between summer (average of 28°C with some peaks over 40°C) and winter temperature (average of -17°C with some peaks under -30°C). According to Faye et al (2008), the following definition was used to identify the different genetic variants of camels: Bactrian and Dromedary are pure animals of the species *Camelus bactrianus* and *Camelus dromedarius* respectively. *Iner* is a F1 hybrid produced by a female dromedary and a Bactrian male, *Nar* is a F1 hybrid of Bactrian female and a dromedary male and finally F2 hybrids (*Kospak*). The herd of lactating females was composed of six Bactrians (B), five dromedaries (D), two hybrids F1 *Iner* (I), four hybrids F1 *Nar* (N), and finally three hybrids F2 *Kospak* (K). The herd went on pasture on steppe vegetation approximately 5-7 km around the village but came back for watering. The vegetation of this area was composed by low gramineae (*Bromus inermis*, *Zostaqzostis splendens*) and some shrubs (*Haloxylon ammodendron*, *Alhagi maurorum* or camelthorn, *Artemisia*, *Climacoptera lanata*, *Salsola arbuscula*). No supplementary feed was distributed to the animals.

Milking routine consists in milking shared between the calf and the farmer. The milk ejection was initiated by the presence of the calf. After the colostral phase, the calf emptied one teat and the three others were milked simultaneously by the farmer. The animals were milked 2 times daily. The

first milking time was at 6 am in the morning. Then the adults went to pasture in the steppe without the calf and came back around 11 am for drinking and a second milking time. Afterwards, they returned to the steppe with their calves but they stayed close to the farm due to the heat, then after 5 or 6 pm they went away again for grazing. Approximately at 9.30 pm, they came back to farm and were separated from calves and spent the night without the calves.

Milk yield and composition have been determined the 21st, 24th and 26th of June 2013, each time on the first morning milking. The yielded milk of the three milked teats were measured in a graduated measuring cup, the recorded yield was divided by 0,75 as one teat has been emptied by the calf and this morning milking has been multiplied by two in order to estimate the milk yield of 24h. The yielded milk was gently homogenized and a sample was taken in order to determine the contents of fat (FC), non fat dry matter (NFDM) and the density of milk (De) using a mid-infrared spectrophotometer equipment (Lactan 1-4 MINI[®], Sibagropribor, Krasnoobsk, Russia). The total DM of milk was calculated by the sum of fat content and SNF and the fat yield corresponded to the multiplication of fat content and milk yield.

An analysis of variance was performed to compare all determined variables using the MIXED procedure of SAS[®] (version 9.3 2009, SAS Inst., Inc., Cary, NC) with the repeated time option. The model includes the fixed effects species (Bactrian, Dromedary, *Iner-maya*: F1 Dromedary female x Bactrian Male, *Nar-maya*: F1 Bactrian Female x Dromedary Male or F2-Kospak: *Iner-maya* female x Bactrian Male), parity (primiparous or multiparous), calving year (2012 or 2013), and the interaction between the species and the calving year. The experimental unit was the camel repeated at three sampling times. The covariance structure between the different sampling times was defined in the model as being auto-regressive after verification of Akaike and Schwarz-Bayesian criterions (Littell et al., 1996). Significance was declared at $P < 0.05$ using the error of the sum of square type III. The values of the analyzed variables were presented as least square means (i.e. adjusted for the effects of the other factors in the model) and were compared by Tukey t-test.

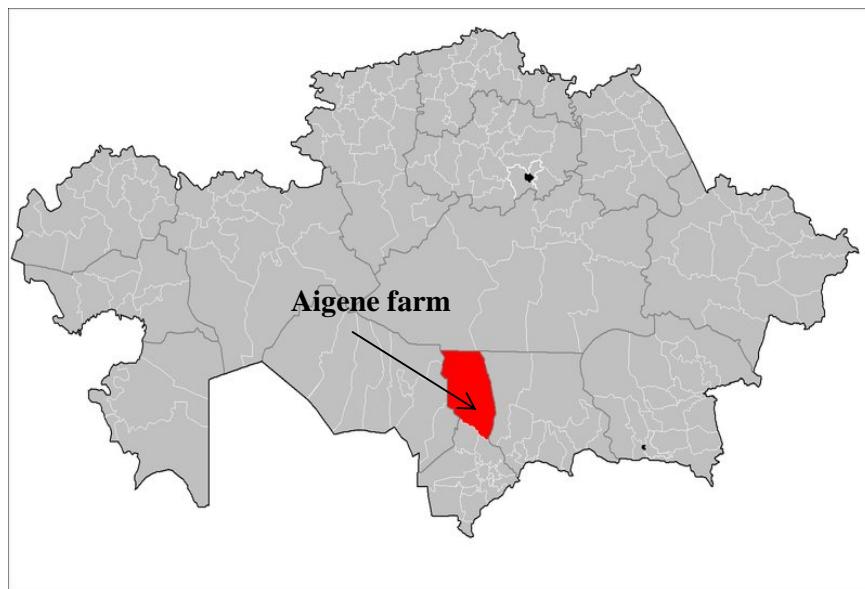


Figure 1. Localisation of Aigene farm.

Table 1. Effect of species, parity and calving year on milk yield and composition.

| Effects | species | parity | Calv. yr | Interaction Sp x Cy | Root MSE | Least Square Means | | | | |
|------------------------|---------|--------|-------------|------------------------|-------------|--------------------|---------|--------|---------|---------|
| | | | | | | B | D | F1-N | F1-I | F2-K |
| n | | | | | | 6 | 5 | 4 | 2 | 3 |
| Milk yield (L/d) | NS | <0.10 | <0.05 | NS | 1.1 | 2.9 | 3.2 | 4.0 | 3.5 | 2.9 |
| Fat content (g/L) | NS | NS | <0.10 | NS | 2.2 | 5.3 a | 4.2 b | 4.2b | 4.7 ab | 4.4 ab |
| Fat free DM (g/L) | <0.001 | NS | <0.01 | <0.10 | 0.9 | 9.8 a | 9.6 ab | 8.6 d | 9.2 c | 9.3 bc |
| DM content (g/L) | <0.05 | NS | NS | NS | 2.8 | 15.1 a | 13.8 ab | 12.8 b | 13.9 ab | 13.8 ab |
| Density (-1000 g/L) | <0.001 | NS | <0.001 | <0.05 | 0.9 | 32.5 a | 32.5 a | 28.9 c | 30.5 b | 31.3 ab |

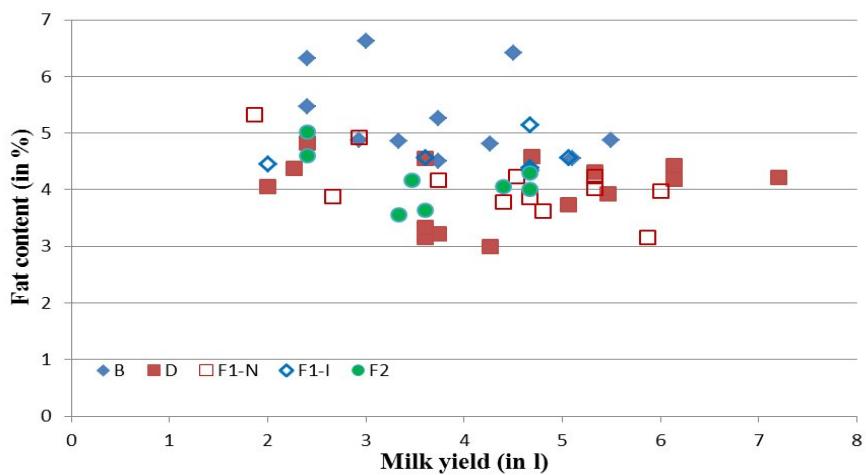


Figure 2. Fat content depending on milk yield and camel species.

Results and Discussions

The species affected significantly the DM content (fat free and total) and the density of the milk. Indeed milk of Bactrian camels contained significantly more DM than this of dromedaries and the same tendency ($P>0.1$) was noted for the fat content (Table 1). In the same time, the milk yield tended to be lower even if no signification threshold was reached. The observed values (fat, DM, density) in the context of Kazakhstan appeared higher than those reported in dromedary in Saudi Arabia (Musaad et al., 2013b) and the milk yield quite lower (Musaad et al., 2013a). Indeed, the milk of dromedaries was not so rich compared du Bactrian. Elsewhere, no significant difference was observed between dromedary and F1 or F2 hybrids except an increased density. F1 hybrids (*Nar-maya* and *Iner-maya*) had a slight but not significant tendency of increased milk yield but a more or less reduced contents and density in comparison to Bactrians. This difference seems to be extenuated for F2 (*Kospak*) animals.

The effect of calving year was illustrated by significantly lower milk yields in the second year of lactation (2.8 versus 3.8 L/d, $P<0.05$), slightly increased contents of fat (4,9 versus 4,2 g/L, $P<0.10$) and dry matter (14,0 and 13,8 g/L, NS) and also density (1030,0 versus 1032,3 g/L), $P<0.01$.

Although the small number of animals would weaken the statistical power of our comparisons and the use of a conservative test to analyze multiple comparisons (t of Tukey), it seems that F1 hybrids would be more productive but with a lower milk yield. This effect tended to disappear in the F2 generation. The Figure 2 illustrated these relationships at the example of relationship between milk yield and fat content. Indeed, Bactrians did not reach so high milk yields but had the highest fat content in confirmation to the observations of Faye et al. (2008). Nevertheless, this work did not mention the milk yield of the studied animals. The concentration of milk in less productive animals has been reported in cows (Boland et al., 2013) or goats (Koop et al., 2010). This phenomenon has not only genetic reasons but mainly physiologic although selection ruminants in the Northern countries aimed to improve milk yield and to mitigate the decrease of contents. However, our results seem to confirm this phenomenon in lactating camels.

Conclusions

This comparison of milk and composition between different camel species at the same time in the same herd showed no difference of milk yield

between Bactrians and dromedary but increased yield in F1 animals. The fat content in Bactrian camels is significantly higher than in all other species. Therefore Fat yield and DM content of Bactrians are not lower in our experimental conditions contrarily of what has been reported in the literature. Thus, Bactrian camels seem as productive in dairy performances as dromedary or F1 camels but better adapted to strong winter conditions in Kazakhstan. Contrarily, F2 animals have lowest dairy performances what would limit their interest for dairy purposes.

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REGULAR ARTICLE

Morphological diversities and eco-geographical structuring of Ethiopian camel (*Camelus dromedarius*) populations

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Abstract

The objectives of this study were to identify and characterize indigenous camel ecotypes and to assess phenotypic diversity and relationship of camel populations in Ethiopia. A total of 494 heads of camels were investigated for phenotypic characterization. The study involved Jijiga, Liben, Gelleb, Hoor and Shinille from Somali as well as Amibara and Mille camel populations from Afar national regional states, which are the major camel rearing areas. The results showed that average barrel and heart girths of Liben camel population were significantly ($p<0.05$) larger than the remaining camel populations. Gelleb camels were significantly ($p<0.05$) superior for morphological variables particularly height at shoulder, chest depth, chest width and hip width to other camel populations examined. Females of Amibara camel population recorded significantly ($p<0.05$) lower values for traits mentioned above as compared to other camel populations. The greatest morphological divergence was observed between Mille and Shinille followed by the difference between Amibara and Shinille camel populations. The least morphological divergence was detected between Hoor and Gelleb followed by that between Amibara and Mille camels in aggregate gender. Quantitative and qualitative study indicated that Jijiga and Hoor camel populations are milk type whereas Liben and Gelleb camel populations are meat type. The principal component analysis showed that body height traits and body height together with body shape traits explained most of the shared variability in female and male camel populations, respectively. The canonical analysis identified two canonical variables to be significant ($p<0.0001$) and sufficient to classify all camels studied. Combined differences among all morphological variables categorized these seven Ethiopian camel populations into five major camel groups. Therefore the findings from this study can be used for the description of body conformation, characterization, improvement and conservation of various camel populations in the country.

Key words: Body measurement, Camel population, Diversity, Morphology

Introduction

Camels are the most capable animals in utilizing marginal areas because they can survive under harsh environmental conditions. Many pastoral groups and communities in diverse eco-zones throughout the world are depending on camels for their livelihoods. The world camel population is estimated to be around 25 million, of which 11 million are present in arid and semi-arid

regions, particularly in the arid lowlands of East Africa (FAOSTAT, 2011). Even though the exact number is not known, approximately 2,400,000 camels are reported to prevail in Ethiopia (FAOSTAT, 2011), of which the Somali and Afar regional states keep around 92% of the total camel population (LDMPS, 2006).

Utilization of camel in Ethiopia is basically traditional and no camel ecotype is specialized for milk, meat, draft or racing purpose except for the pastoralists' traditional classification of camel types in Somali regional state. In this region, pastoralists classify camel population based on some phenotypic descriptors. According to their perception, some of the camel ecotypes are taller while others have a wider hip. They also distinguish different camel ecotypes for milk, meat

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and dual purposes. Moreover, they have the opinion that some of the camel ecotypes are more adaptive to harsh environment than others (Ahmed, 2002). According to FAO (2011), the traditional classification should be used as a basis for phenotypic and genetic characterization studies.

However, study on camel production system, phenotypic and genetic characterization is scanty (Yohannes et al., 2007) and there is a serious lack of information on camel genetic diversity in East Africa (Gifford-Gonzalez and Hanotte, 2011). This hindered the design of appropriate strategy for utilization of existing potential of camel genetic resources and establishment of breeding programs. Given the current importance of camels in contributing to the livelihoods of large human population in marginal areas, and the role it plays towards resilience to present climate change, it is imperative to identify and differentiate the phenotypic characteristics of camel populations in Ethiopia based on FAO guidelines. Therefore the present study was undertaken with the objectives to identify and characterize indigenous camel ecotypes of south, east and northeastern Ethiopia and to describe the relationship of these camel populations.

Materials and Methods

Study area

The study involved two major camel rearing geographical locations viz. Somali and Afar national regional states (Figure 1). The two regional states accounted for about 92% of the camel population in Ethiopia and were purposively selected for the study. The specific study sites from Somali national regional state included three rural localities (RLs) from Jijiga District (representing Jijiga camel population), four RLs from Gode District (two RLs each for Hoor and Gelleb camel populations), four RLs from Moyale District (Liben camel population) and two RLs from Shinille District (Shinille camel population). The sampling area from Afar national regional state involved two RLs from Mille District (Mille camel population), and two and one RLs from Amibara and Dulessa Districts, respectively (Amibara camel population). The study sites were purposively selected based on traditional classification of camel populations while households were selected randomly. Exploratory approach (undertaken in situations in which no reliable background information on the existence of recognized breeds in the study area was available) was used in the absence of traditional classification.

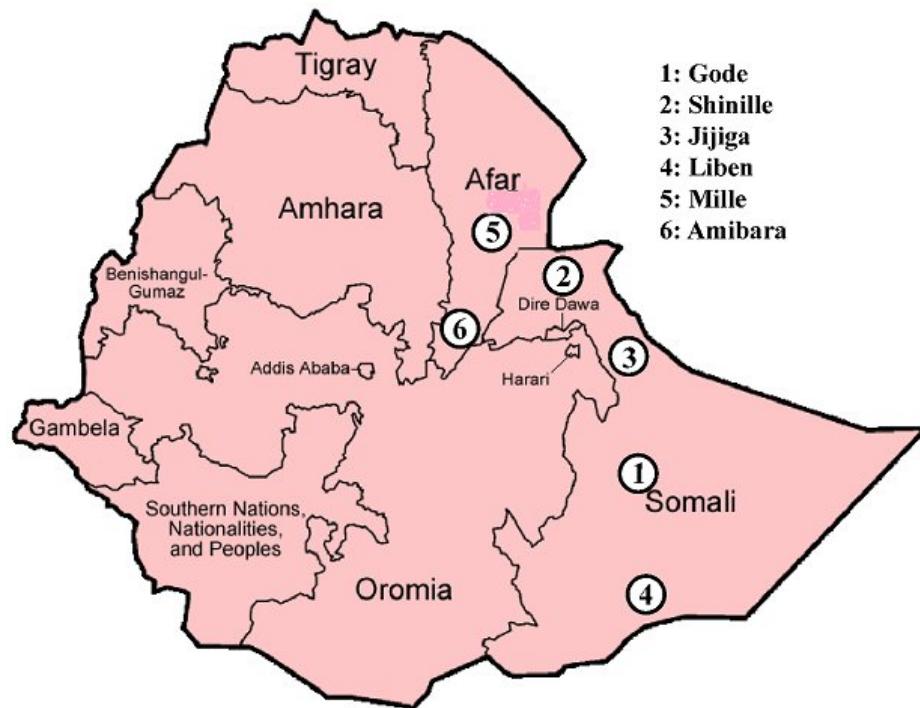


Figure 1. Map of study areas in Afar and Somali regional states, Ethiopia.

Methods for data collection and description of morphological variables

A rapid rural appraisal technique was applied to collect data. Structured questionnaires were used to gather information from pastoral households so that to generate relevant information on husbandry practices of camels, historical perspectives and people's perception of camel rearing, and traditional ways of classifying and describing the differences among and within camel populations as well as of understanding breed characteristics in terms of milk yield, resistance to drought and related environmental hazards, selection criteria, and qualitative descriptions of camels such as body color, hair length and distribution, hump, ear size, ear orientation, tail length, and udder size. Moreover, relevant information was generated and physical data was obtained through informal group discussion held with key informants (elders, community leaders and development agents) at all study sites and at various levels. Information collected during group discussion was supported by personal observation during a transect walk where critical environmental observation was done. Camels above eight years of age were used for linear measurement. Age was determined based on dentition and also information obtained from the owners.

Data collection formats for discrete/qualitative, quantitative, herd level data, and origin and development of camels were adapted from FAO guidelines on phenotypic characterization (FAO, 2011). In this study, a total of 103 male and 391 female mature (full mouth) and unrelated camels were randomly selected from the identified populations (Table 1). The populations were identified during the exploratory assessment in reference to the traditionally recognized types, the geographical differences among the populations, and the ethnic nomenclature. A total of 18 different body measurements were recorded for each of the sampled individuals within the population. Measurements were taken using a measuring tape while the animals were standing on level ground. The types and anatomical positions of different linear measurements taken are indicated in Table 2 and Figure 2. Body weight estimation was done using Barymetric weight estimation formula of Yagil (1994):

$$Y = SH \times TG \times BG \times 50$$

Where, Y = The weight in kg.

SH = The height at shoulder in meters.

TG = The chest girth behind the chest pad in meters.

BG = The barrel girth over the highest part of the hump in meters.

Table 1. Number of males, females and total number of camels sampled per population.

| Populations | Females | Males | Total | Percentage | Cumulative percentage |
|-------------|---------|-------|-------|------------|-----------------------|
| Amibara | 57 | 14 | 71 | 14.37 | 14.37 |
| Gelleb | 57 | 14 | 71 | 14.37 | 28.74 |
| Hoor | 56 | 14 | 70 | 14.17 | 42.91 |
| Jijiga | 58 | 15 | 73 | 14.77 | 57.68 |
| Liben | 53 | 15 | 68 | 13.77 | 71.46 |
| Mille | 58 | 14 | 72 | 14.57 | 86.03 |
| Shinille | 52 | 17 | 69 | 13.97 | 100.00 |
| Total | 391 | 103 | 394 | | |

Table 2. Definition of morphological variables measured on Ethiopian camels.

Morphological variables^a

1. Heart or Chest girth (cm): the circumference of the body immediately behind the shoulder blades in a vertical plane, perpendicular to the long axis of the body as quantified using a measuring tape (F).
2. Height at shoulder/wither (cm): the height (vertical) from the bottom of the front foot to the highest point of the withers measured using a measuring stick (C-G).
3. Barrel girth (cm): the measurement of the distance around the abdomen over the highest part of the hump measured by a measuring tape (E).
4. Body length (cm): the horizontal distance from the point of shoulder to the pin bone measured using a measuring stick (A-D).
5. Depth of chest (cm): distance from wither to sternum measured using a measuring tape (G-H).

Table 2. Contd..

6. Width of chest (cm): distance from left to the right upper arm measured using a measuring tape (M-N).
7. Width of hip (cm): distance from the left to the right point of hip measured using a measuring tape (K-L).
8. Length of forelimb (cm): distance from the surface of the ground level to front of sternum measured using a measuring stick (C-D).
9. Length of hind limb (cm): distance from the bottom of the leg to the pin bone of hip measured using a measuring stick (A-B).
10. Tail length: distance from the tail base to the tip of tail measured by a measuring tape (I-J).
11. Hind leg hoof circumference: circumference of hind leg hoof around the wider part measured using a measuring tape (V).
12. Foreleg hoof circumference: circumference of foreleg hoof around the wider part measured using a measuring tape (U).
13. Hump circumference: the perimeter of the hump from a point at the anterior end of the hump to a point at its posterior end measured using a measuring tape (Z1).
14. Hump length: length from the bottom to the tip of the hump measured using a measuring tape (Y-Z).
15. Neck length: distance from the lower part of mandible to the sternum measured using a measuring tape (O-P).
16. Face length: distance from the midpoint of the two ears to the mouth measured using a measuring tape (Q-R).
17. Ear length: length of the external ear from its root on the base to the tip measured using a measuring tape (X-W).
18. Distance between eyes: distance between the two eyes measured using a measuring tape (S-T).

^aLetters in parenthesis indicate positions of measurements as illustrated in Figure 2.

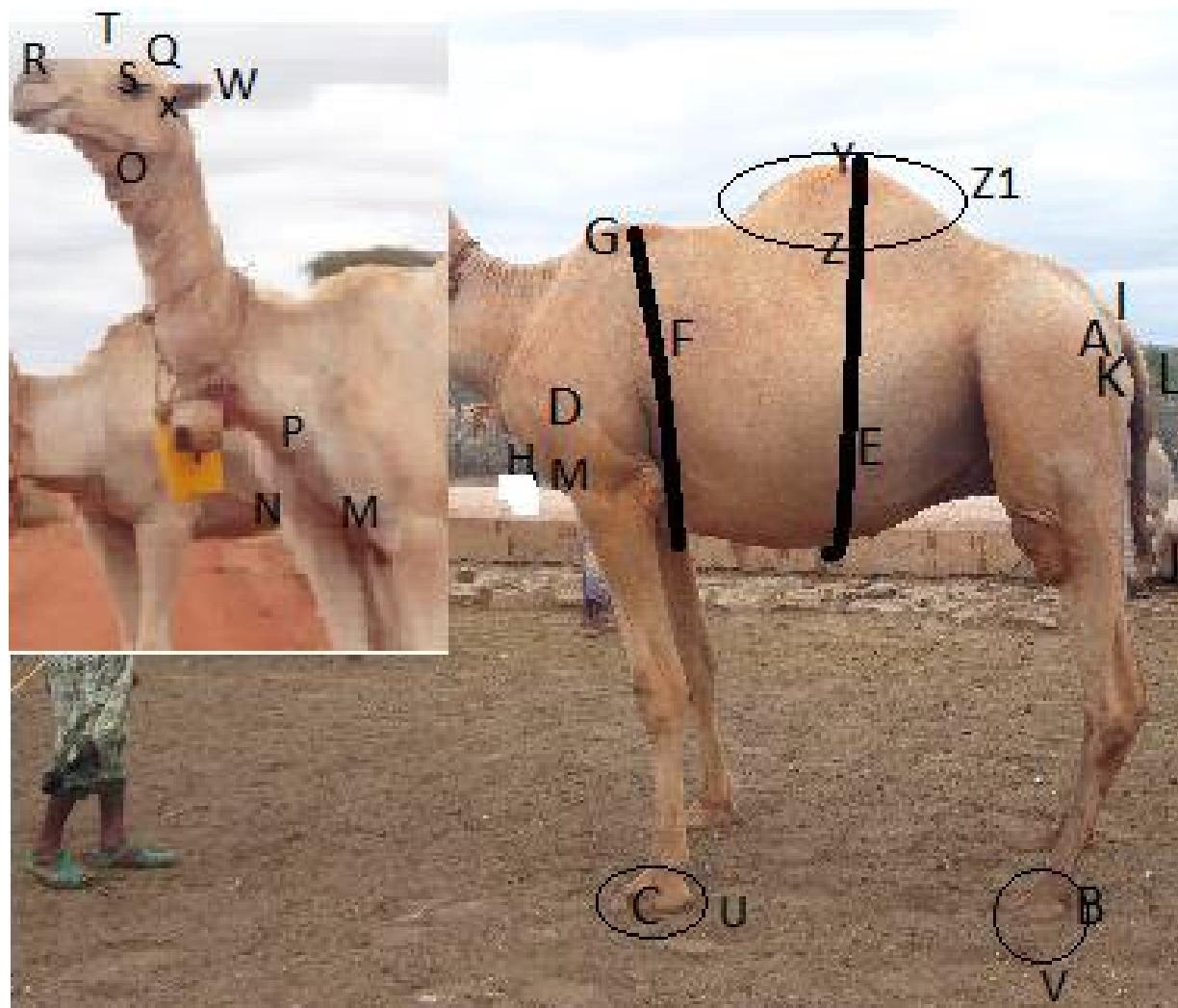


Figure 2. Positions of the various morphological variables measured on a camel.

Data analysis

Data were analyzed using the GLM procedure of SAS (2008). Descriptive statistics, univariate and multivariate analyses were employed. Cluster analysis was undertaken to identify groups of individuals that are similar to each other but different from individuals in other groups. Discriminant analysis was employed to define the relationship between independent and dependent variables on data sets for which pre-specified and well defined groups already exist.

Principal component analysis (PCA) was carried out for the two genders separately to determine different variables or parameters for differentiation of camel populations into different groups that were mutually exclusive, and to summarize the variables into few meaningful ones that accounted for most of the variations in the population. Cross validation for proper classification of different camel groups in the original population and tolerance evaluation were undertaken for each sex separately and for aggregate gender. In addition, Eigen values greater than one was described in the principal component analysis. After tolerance evaluation, some variables that did not reveal significant difference among male camel populations were removed.

Canonical discriminant function analysis was also performed to find out linear combination of quantitative variables that gave maximal separations between populations. The scored canonical variables were used to plot pairs of canonical variables to aid visual interpretation of group differences. In order to know the relationship of hump length and barrel girth with other variables, both traits were measured separately. To avoid redundancy, hump length was removed from all analyses except for mean comparison and PCA.

A stepwise procedure was used to determine the relationship among different populations. In the stepwise procedure, discriminant analysis with forward selection procedure was carried out to find out variables that best showed differences among populations and to identify important discriminating variables. Some variables that had below 0.1 tolerance values were not described but variables with wilks' lambda values close to zero or one were described. Squared Mahalanobis distance was computed between populations as:

$$D_{ij}^2 = (\bar{X}_i - \bar{X}_j)' \text{COV}^{-1} (\bar{X}_i - \bar{X}_j)$$

Where D_{ij}^2 is the distance between populations i and j , COV^{-1} is the inverse of the covariance matrix of measured variables, y and \bar{X}_i and \bar{X}_j are the means of variable y in i^{th} and j^{th} populations, respectively. Squared Mahalanobis distance matrix was used via agglomerative hierarchical cluster procedure to build a dendrogram using unweighted pair group method with arithmetic mean (UPGMA) employing tree procedure in SAS (2008). Thus distance between populations based on Mahalanobis distance procedure (Mahalanobis, 1936) was used.

Results

Breed means and mean comparisons

Mean values of the 18 morphological variables and body weight of the seven Ethiopian camel populations are presented for male, female and aggregate gender in Tables 3, 4 and 5, respectively. Pair wise mean comparison showed significant differences for most of the morphological variables among male camel populations. Height at shoulder (HS), body length (BL), heart girth (HG), barrel girth (BG) and body weight (BW) were significantly ($p<0.05$) higher for Liben male camels than other male camel populations. Hoor and Gelleb male camels had significantly ($p<0.05$) higher chest depth (CD), chest width (CW) and hip width (HW) than other male camel populations. But Gelleb and Hoor male camel populations recorded a significantly ($p<0.05$) lower HG than males from other camel populations. Males of Mille and Liben camel populations were superior ($p<0.05$) in length of hind (LHL) and forelegs (LFL) to other male camel populations studied. Shinille male camels were significantly ($p<0.05$) superior in hind (HLHC) and forelegs (FLHC) hoof circumferences to males of other camel populations. Males of Gelleb and Liben camel populations were significantly ($p<0.05$) superior in hump circumference (HC) to males of other camel populations studied (Table 3).

Table 3. Mean and pair wise comparison of morphological variables (cm) with their standard errors in each camel population: Male.

| Traits ^b | Camel populations | | | | | | |
|---------------------|-----------------------------|----------------------------|----------------------------|----------------------------|----------------------------|----------------------------|---------------------------|
| | Jijiga | Hoor | Gelleb | Amibara | Mille | Liben | Shinille |
| No. | 15 | 14 | 14 | 14 | 14 | 15 | 17 |
| HG | 198.20(3.33) ^{bcd} | 194.00(1.09) ^d | 196.85(0.52) ^{cd} | 200.71(1.12) ^{bc} | 202.57(1.36) ^b | 219.86(1.58) ^a | 185.52(0.66) ^e |
| BG | 240.33(3.78) ^b | 236.35(0.99) ^{bc} | 238.21(0.59) ^{bc} | 233.14(0.99) ^{cd} | 237.07(0.98) ^{bc} | 265.26(1.83) ^a | 230.64(1.15) ^d |
| HS | 184.26(2.74) ^c | 201.64(1.13) ^a | 205.78(0.58) ^a | 194.71(0.69) ^b | 196.71(0.81) ^b | 205.13(2.60) ^a | 184.52(1.23) ^c |
| BW | 443.13(20.57) ^c | 462.64(6.50) ^{bc} | 482.61(3.57) ^b | 455.83(5.54) ^{bc} | 477.08(7.49) ^b | 599.58(14.23) ^a | 407.59(3.76) ^d |
| BL | 134.20(2.32) ^b | 149.71(0.39) ^a | 150.07(0.47) ^a | 129.42(1.34) ^c | 130.14(1.13) ^c | 149.26(2.67) ^a | 146.70(0.83) ^a |
| CD | 67.26(2.93) ^b | 82.00(0.65) ^a | 80.57(0.40) ^a | 56.14(1.74) ^d | 55.35(0.78) ^d | 64.26(1.46) ^{bc} | 61.05(0.77) ^c |
| CW | 40.26(1.92) ^c | 52.28(1.18) ^a | 54.14(0.55) ^a | 39.85(0.83) ^c | 48.07(0.65) ^b | 52.66(2.08) ^a | 47.58(0.35) ^b |
| HW | 41.73(1.16) ^c | 46.50(0.85) ^a | 44.64(0.57) ^{ab} | 36.64(0.42) ^d | 42.71(0.62) ^{bc} | 44.40(0.82) ^{ab} | 42.47(0.44) ^{bc} |
| LHL | 155.20(1.48) ^d | 161.35(0.74) ^c | 162.71(0.56) ^{bc} | 164.50(1.44) ^{bc} | 165.92(2.05) ^{ab} | 169.33(1.86) ^a | 147.11(0.74) ^e |
| LFL | 147.06(1.08) ^d | 155.64(0.45) ^c | 156.35(0.67) ^{bc} | 154.35(0.89) ^c | 158.92(1.31) ^{ab} | 160.20(1.75) ^a | 142.11(0.67) ^e |
| TL | 63.13(3.06) ^{bc} | 69.00(0.55) ^a | 70.21(0.48) ^a | 61.21(0.53) ^c | 67.07(0.67) ^{ab} | 59.80(2.41) ^c | 54.88(0.42) ^d |
| FLHC | 66.26(1.96) ^d | 75.57(0.85) ^b | 71.85(0.43) ^c | 66.07(0.67) ^d | 63.42(0.76) ^d | 76.73(1.03) ^b | 95.64(1.10) ^a |
| HLHC | 60.00(1.14) ^d | 70.71(0.80) ^c | 72.00(1.52) ^c | 58.42(0.40) ^d | 57.78(0.57) ^d | 78.66(2.59) ^b | 87.82(0.90) ^a |
| HC | 108.40(8.05) ^c | 137.28(0.78) ^b | 141.42(0.73) ^{ab} | 88.35(1.92) ^d | 95.35(0.76) ^d | 153.06(6.31) ^a | 91.41(3.01) ^d |
| HL | 31.66(2.04) ^b | 33.85(0.65) ^b | 33.57(0.38) ^b | 21.71(0.26) ^c | 22.57(0.30) ^c | 37.66(1.55) ^a | 22.11(0.34) ^c |
| NL | 93.20(3.33) ^d | 120.00(0.93) ^a | 122.57(0.57) ^a | 101.85(1.37) ^c | 101.92(0.72) ^c | 108.20(3.70) ^b | 99.52(0.64) ^c |
| FCL | 51.33(0.31) ^c | 58.28(0.80) ^b | 60.92(0.70) ^a | 52.71(0.42) ^c | 53.07(0.48) ^c | 57.80(0.92) ^b | 45.17(0.29) ^d |
| EL | 11.80(0.14) ^b | 11.57(0.13) ^b | 12.00(0.14) ^{ab} | 12.07(0.16) ^{ab} | 12.00(0.18) ^{ab} | 12.06(0.26) ^{ab} | 12.47(0.12) ^a |
| DES | 24.40(0.48) ^b | 22.28(0.22) ^c | 24.50(0.17) ^b | 21.28(0.33) ^d | 22.14(0.25) ^{cd} | 24.26(0.35) ^b | 25.47(0.19) ^a |

^b HG = Heart girth, BG = Barrel girth, HS = Height at shoulder/wither, BW = Body weight, BL = Body length, CD = Chest depth, CW = Chest width, HW = Hip width, LHL = Length of hind leg, LFL = Length of foreleg, TL = Tail length, FLHC = Foreleg hoof circumference, HLHC = Hind leg hoof circumference, HC = Hump circumference, HL = Hump length, NL = Neck length, FCL = Face length, EL = Ear length, DE = Distance between eyes. Figures in parentheses = s.e. Different superscripts labeled for values in the same raw indicate their statistical significances at p<0.05. The same abbreviations and rules are also applied to all relevant tables and figures.

Table 4. Mean and pair wise comparison of morphological variables (cm) with their standard errors in each camel population: Female.

| Traits | Camel populations | | | | | | |
|--------|----------------------------|----------------------------|---------------------------|---------------------------|---------------------------|---------------------------|---------------------------|
| | Jijiga | Hoor | Gelleb | Amibara | Mille | Liben | Shinille |
| No. | 58 | 56 | 57 | 57 | 58 | 53 | 52 |
| HG | 198.89(1.68) ^c | 210.35(0.62) ^b | 214.67(1.33) ^a | 181.89(0.75) ^e | 185.25(0.45) ^d | 209.64(1.45) ^b | 185.24(0.60) ^d |
| BG | 248.86(1.56) ^b | 260.49(0.85) ^a | 261.91(1.10) ^a | 219.25(0.84) ^d | 229.96(0.53) ^c | 263.25(1.02) ^a | 230.50(0.71) ^c |
| HS | 176.71(0.83) ^d | 194.73(0.90) ^b | 201.31(0.59) ^a | 181.84(0.72) ^c | 180.42(0.37) ^c | 193.94(2.01) ^b | 175.47(0.54) ^d |
| BW | 439.76(7.50) ^c | 533.95(4.46) ^b | 567.00(6.45) ^a | 362.80(3.59) ^e | 384.47(2.07) ^d | 532.18(7.71) ^b | 375.14(3.11) ^e |
| BL | 142.20(0.88) ^c | 144.98(0.53) ^b | 141.08(0.68) ^c | 126.14(1.52) ^e | 124.91(0.39) ^e | 148.04(1.41) ^a | 137.77(0.87) ^d |
| CD | 69.54(0.94) ^c | 78.57(0.31) ^b | 80.63(0.43) ^a | 54.67(0.55) ^f | 53.13(0.39) ^f | 62.66(0.77) ^d | 57.32(0.29) ^e |
| CW | 39.77(0.55) ^d | 45.00(0.79) ^c | 51.13(0.37) ^a | 37.87(0.53) ^e | 36.72(0.36) ^e | 47.68(0.64) ^b | 39.88(0.40) ^d |
| HW | 37.56(0.50) ^d | 43.57(0.45) ^b | 47.69(0.26) ^a | 34.00(0.33) ^f | 39.90(0.27) ^c | 43.13(0.56) ^b | 35.58(0.31) ^e |
| LHL | 150.14(1.03) ^c | 157.73(0.35) ^b | 156.43(0.39) ^b | 149.00(1.08) ^c | 150.61(0.50) ^c | 160.76(1.02) ^a | 143.13(0.48) ^d |
| LFL | 139.88(1.26) ^{ef} | 149.93(0.45) ^b | 146.76(0.40) ^c | 140.55(0.91) ^e | 143.46(0.53) ^d | 153.46(0.87) ^a | 137.79(0.39) ^f |
| TL | 59.55(0.46) ^b | 63.17(0.39) ^a | 63.25(0.29) ^a | 56.24(0.84) ^c | 58.37(0.38) ^b | 56.05(0.71) ^c | 48.24(0.64) ^d |
| FLHC | 65.07(0.41) ^c | 72.75(0.36) ^a | 67.84(0.89) ^b | 53.60(0.61) ^e | 53.00(0.42) ^e | 68.92(0.90) ^b | 62.66(0.84) ^d |
| HLHC | 61.34(0.84) ^c | 67.00(0.38) ^b | 64.79(0.88) ^b | 49.77(0.49) ^e | 46.36(0.47) ^f | 69.87(1.38) ^a | 56.86(0.78) ^d |
| HC | 124.42(3.16) ^b | 127.89(1.46) ^{ab} | 130.83(0.88) ^a | 79.74(1.12) ^e | 96.45(0.83) ^c | 131.79(2.31) ^a | 85.35(0.62) ^d |
| HL | 36.50(0.81) ^a | 29.24(0.58) ^c | 30.71(0.52) ^b | 19.25(0.25) ^e | 21.29(0.25) ^d | 35.33(0.52) ^a | 20.32(0.22) ^e |
| NL | 94.71(0.68) ^c | 104.42(0.60) ^a | 103.84(0.34) ^a | 91.80(0.56) ^d | 91.79(0.95) ^d | 100.35(0.94) ^b | 83.62(1.04) ^e |
| FCL | 50.18(0.29) ^d | 53.92(0.61) ^b | 56.13(0.39) ^a | 48.82(0.62d) ^e | 47.55(0.32) ^e | 52.39(0.65) ^c | 41.30(0.25) ^f |
| EL | 11.86(0.08) ^a | 11.26(0.11) ^b | 11.87(0.08) ^a | 11.22(0.13) ^b | 11.40(0.11) ^b | 12.13(0.09) ^a | 12.11(0.09) ^a |
| DE | 22.91(0.17) ^c | 22.91(0.19) ^c | 25.10(0.17) ^b | 20.48(0.30) ^d | 20.08(0.21) ^d | 22.83(0.23) ^c | 26.09(0.13) ^a |

Table 5. Mean and pair wise comparison of morphological variables (cm) with their standard errors in each camel population: Aggregate gender.

| Traits | Camel populations | | | | | | |
|--------|----------------------------|---------------------------|----------------------------|----------------------------|---------------------------|---------------------------|---------------------------|
| | Jijiga | Hoor | Gelleb | Amibara | Mille | Liben | Shinille |
| No. | 73 | 70 | 71 | 71 | 72 | 68 | 69 |
| HG | 198.75(1.49) ^c | 207.12(0.94) ^b | 211.20(1.36) ^a | 185.55(1.09) ^d | 188.57(0.92) ^d | 211.87(1.28) ^a | 185.31(0.48) ^d |
| BG | 247.13(1.50) ^c | 255.73(1.35) ^b | 257.30(1.42) ^b | 221.95(0.95) ^e | 231.32(0.57) ^d | 263.69(0.89) ^a | 230.54(0.60) ^d |
| HS | 178.24(0.92) ^d | 196.09(0.82) ^b | 202.18(0.53) ^a | 184.34(0.85) ^c | 183.54(0.82) ^c | 196.37(1.76) ^b | 177.67(0.68) ^d |
| BW | 440.44(7.27) ^c | 519.91(5.08) ^b | 550.59(6.56) ^a | 380.88(5.34) ^e | 402.23(4.81) ^d | 546.83(7.53) ^a | 383.02(3.02) ^e |
| BL | 140.58(0.92) ^{bc} | 145.91(0.49) ^a | 142.83(0.69) ^b | 126.77(1.25) ^d | 127.49(0.54) ^d | 148.30(1.24) ^a | 139.94(0.82) ^c |
| CD | 69.08(0.95) ^b | 79.25(0.32) ^a | 80.62(0.35) ^a | 54.95(0.56) ^e | 53.56(0.36) ^e | 63.01(0.68) ^c | 58.22(0.34) ^d |
| CW | 39.87(0.58) ^e | 46.43(0.76) ^c | 51.72(0.34) ^a | 38.26(0.46) ^e | 38.90(0.61) ^e | 48.76(0.71) ^b | 41.75(0.50) ^d |
| HW | 38.40(0.50) ^d | 44.15(0.42) ^b | 47.09(0.27) ^a | 34.51(0.30) ^f | 40.43(0.27) ^c | 43.40(0.47) ^b | 37.25(0.43) ^e |
| LHL | 151.16(0.90) ^d | 158.45(0.35) ^b | 157.65(0.44) ^b | 152.01(1.16) ^{cd} | 153.54(0.90) ^c | 162.62(0.98) ^a | 144.10(0.45) ^e |
| LFL | 141.33(1.08) ^d | 151.05(0.46) ^b | 148.62(0.57) ^c | 143.23(0.99) ^d | 146.42(0.87) ^c | 154.92(0.84) ^a | 138.84(0.40) ^e |
| TL | 60.28(0.72) ^b | 64.32(0.43) ^a | 64.61(0.41) ^a | 57.20(0.72) ^c | 60.04(0.52) ^b | 56.86(0.77) ^c | 49.85(0.60) ^d |
| FLHC | 65.31(0.51) ^c | 73.30(0.35) ^a | 68.62(0.75) ^b | 56.02(0.77) ^d | 55.00(0.60) ^d | 70.62(0.83) ^b | 70.67(1.83) ^b |
| HLHC | 61.06(0.71) ^d | 67.73(0.38) ^b | 66.19(0.83) ^c | 51.45(0.57) ^e | 48.54(0.66) ^f | 71.78(1.29) ^a | 64.38(1.71) ^c |
| HC | 121.17(3.06) ^c | 129.74(1.26) ^b | 132.88(0.87) ^{ab} | 81.41(1.05) ^f | 96.24(0.68) ^d | 136.42(2.48) ^a | 86.82(0.91) ^e |
| HL | 35.52(0.79) ^a | 30.15(0.52) ^b | 31.26(0.44) ^b | 19.73(0.23) ^d | 21.53(0.21) ^c | 35.84(0.53) ^a | 20.75(0.20) ^{cd} |
| NL | 94.40(0.85) ^c | 107.49(0.90) ^a | 107.48(0.92) ^a | 93.59(0.71) ^c | 93.74(0.91) ^c | 102.05(1.14) ^b | 87.48(1.15) ^d |
| FCL | 50.41(0.37) ^c | 54.78(0.55) ^b | 57.05(0.41) ^a | 49.58(0.53) ^{cd} | 48.61(0.37) ^d | 53.56(0.60) ^b | 42.24(0.28) ^e |
| EL | 11.85(0.07) ^b | 11.85(0.07) ^c | 11.90(0.06) ^b | 11.38(0.11) ^c | 11.52(0.10) ^c | 12.11(0.09) ^{ab} | 12.20(0.07) ^a |
| DE | 23.21(0.17) ^c | 22.78(0.16) ^c | 24.98(0.14) ^b | 20.63(0.25) ^d | 20.47(0.19) ^d | 23.14(0.21) ^c | 25.94(0.11) ^a |

With regard to female morphological variables, females of Gelleb camel population were significantly superior ($p<0.05$) in HG, HS, BW, CD, CW and HW to females of other camel populations (Table 4). Females of Liben and Hoor camel populations also showed higher values in HG, HS and BW than the remaining populations. Females of Shinille and Amibara camel populations recorded significantly ($p<0.05$) the lowest values as compared with other populations for HG, HS and BW. Jijiga female camel population had higher HG and BW than Amibara, Mille and Shinille female camel populations which are found in the sparse vegetation cover and high temperature environment. Females of Gelleb camel population recorded significantly ($p<0.05$) higher CD, CW and HW than females of other camel populations studied. Hump length (HL) of Gelleb female camel population was significantly larger than Hoor female camel population but both of them had a similar BG within the same environment. Hoor and Liben followed that of Gelleb female camels in all the preceding morphological variables. Female camels of Amibara and Mille populations recorded the lowest ($p<0.05$) values for CD and CW.

Mean comparison for aggregate gender (Table 5) revealed that Hoor and Liben camel populations exhibited a significantly ($p<0.05$) longer BL than other camel populations studied. BG and HL had a positive relationship in both Hoor and Gelleb camel populations which are distributed within the same environment. Mille and Amibara camels recorded a significantly ($p<0.5$) shorter BL than other camel populations. Gelleb followed by Liben and Hoor camel populations had significantly ($p<0.05$)

superior morphological variables of HS, CD, CW and HW to the remaining camel populations.

Canonical and discriminant analysis

The discriminate function correctly classified 99.61% of all camels investigated. Classification of cross-validation (Table 6) indicated an average success rate at 93.05%. About 83.78%, 87.32%, 95.83%, 94.44%, 98.63%, 91.30% and 100 % for Jijiga, Hoor, Gelleb, Amibara, Mille, Liben and Shinille camels were correctly assigned into their distinct sources of origins, respectively.

All squared Mahalanobis distances within males (Table 7), females (Table 7) and aggregate gender (Table 8) of all camel populations studied were highly significant ($p<0.001$). Among the male camel populations, the largest distance was observed between Shinille and Amibara followed by the distance between Shinille and Gelleb. Males of Shinille camel population were significantly ($p<0.001$) distant from males of other camel populations. A relatively close Mahalanobis distance was recorded between Hoor and Gelleb followed by that between Amibara and Mille male camel populations. The greatest morphological divergences in female camel populations were observed between Shinille and Mille and between Mille and Gelleb. The least morphological divergence was observed between Hoor and Gelleb followed by that between Mille and Amibara female camel populations. The largest morphological divergence for aggregate gender was observed between Mille and Shinille followed by that between Gelleb and Mille camel populations while the least value was recorded between Hoor and Gelleb followed by that between Amibara and Mille camel populations (Table 8).

Table 6. Number of observations (before the bracket) and percentage classified (in bracket) in different camel populations using discriminant analysis.

| Populations | Jijiga | Hoor | Gelleb | Amibara | Mille | Liben | Shinille |
|-------------|----------|----------|----------|----------|----------|----------|-----------|
| Jijiga | 61(83.6) | 7(9.5) | 0(0.00) | 0(0.00) | 3(4.05) | 0(0.00) | 2(2.70) |
| Hoor | 2(2.8) | 61(87.1) | 7(9.86) | 0(0.00) | 0(0.00) | 0(0.00) | 0(0.00) |
| Gelleb | 0(0.00) | 3(4.17) | 68(95.8) | 0(0.00) | 0(0.00) | 0(0.00) | 0(0.00) |
| Amibara | 0(0.00) | 0(0.00) | 0(0.00) | 67(94.4) | 4(5.6) | 0(0.00) | 0(0.00) |
| Mille | 0(0.00) | 0(0.00) | 0(0.00) | 1(1.37) | 71(98.6) | 0(0.00) | 0(0.00) |
| Liben | 2(2.90) | 3(4.35) | 1(1.45) | 0(0.00) | 0(0.00) | 62(91.2) | 0(0.00) |
| Shinille | 0(0.00) | 0(0.00) | 0(0.00) | 0(0.00) | 0(0.00) | 0(0.00) | 69(100.0) |

Table 7. Squared Mahalanobis distances between Ethiopian camel populations (values for female camels are above the diagonal while those for male camels below the diagonal).

| Populations | Jijiga | Hoor | Gelleb | Amibara | Mille | Liben | Shinille |
|-------------|--------|--------|--------|---------|--------|--------|----------|
| Jijiga | 0 | 36.33 | 50.16 | 75.50 | 95.33 | 42.61 | 47.99 |
| Hoor | 87.17 | 0 | 12.12 | 90.30 | 95.20 | 38.93 | 70.29 |
| Gelleb | 119.86 | 13.80 | 0 | 87.84 | 96.58 | 44.62 | 77.61 |
| Amibara | 81.41 | 140.09 | 155.74 | 0 | 18.63 | 78.15 | 70.27 |
| Mille | 67.40 | 120.04 | 143.47 | 18.33 | 0 | 64.27 | 96.60 |
| Liben | 122.27 | 248.78 | 267.40 | 216.15 | 184.48 | 0 | 66.53 |
| Shinille | 495.71 | 504.11 | 620.80 | 621.03 | 561.10 | 510.59 | 0 |

Table 8. Squared Mahalanobis distances between Ethiopian camel populations (aggregate gender).

| Populations | Jijiga | Hoor | Gelleb | Amibara | Mille | Liben | Shinille |
|-------------|--------|-------|--------|---------|-------|-------|----------|
| Jijiga | 0 | | | | | | |
| Hoor | 25.89 | 0 | | | | | |
| Gelleb | 37.30 | 8.85 | 0 | | | | |
| Amibara | 52.48 | 61.02 | 68.27 | 0 | | | |
| Mille | 65.42 | 62.76 | 72.47 | 12.11 | 0 | | |
| Liben | 34.59 | 35.68 | 40.24 | 64.66 | 54.06 | 0 | |
| Shinille | 40.67 | 55.75 | 63.72 | 66.05 | 84.23 | 61.06 | 0 |

The first four most important morphometric variables for aggregate gender (Table 9) with higher Wilks' lambda and F-values (comparatively near to one) used for discriminating between camel diversity were CD, BL, distance between eyes (DE), and HS. The tolerance values obtained for these variables were greater than 0.1, indicating absence of collinearity problem among the nine most discriminating morphometric variables. The other variables such as HG, BW, HLHC, CW, ear length (EL), neck length (NL), LFL and LHL all had a Wilks' lambda relatively near to zero.

Stepwise discriminant analysis of the first five morphometric variables in females and the first six in males (Table 10) showed no collinearity problem among the variables. CW, BL and DE were important variables to differentiate the two genders. The most important traits in discriminating between females of all camel populations were CD and BG whereas FLHC and CD were the two most important traits in discriminating between male camel populations.

Table 9. Stepwise discriminant analysis for aggregate gender.

| Step | Variables entered | Partial R-square | F-values | Pr>F | Wilks' lambda | Tolerance |
|------|-------------------|------------------|----------|--------|---------------|-----------|
| 1 | CD | 0.8263 | 391.80 | <.0001 | 0.17365195 | 0.18 |
| 2 | BL | 0.6010 | 123.74 | <.0001 | 0.06929538 | 0.65 |
| 3 | DE | 0.5191 | 88.52 | <.0001 | 0.03332253 | 0.59 |
| 4 | HS | 0.5008 | 82.09 | <.0001 | 0.01663537 | 0.51 |
| 5 | BG | 0.4289 | 61.34 | <.0001 | 0.00949979 | 0.44 |
| 6 | FCL | 0.3233 | 38.94 | <.0001 | 0.00642843 | 0.40 |
| 7 | HW | 0.2968 | 34.32 | <.0001 | 0.00452067 | 0.38 |
| 8 | FLHC | 0.2432 | 26.08 | <.0001 | 0.00342132 | 0.36 |
| 9 | TL | 0.2026 | 20.58 | <.0001 | 0.00272828 | 0.34 |
| 10 | BW | 0.1982 | 19.98 | <.0001 | 0.00218758 | |
| 11 | LFL | 0.2218 | 22.99 | <.0001 | 0.00170234 | |
| 12 | HG | 0.1169 | 10.65 | <.0001 | 0.00150337 | |
| 13 | HLHC | 0.1129 | 10.22 | <.0001 | 0.00133364 | |
| 14 | CW | 0.0834 | 7.29 | <.0001 | 0.00122242 | |
| 15 | EL | 0.0833 | 7.27 | <.0001 | 0.00112061 | |
| 16 | NL | 0.0634 | 5.40 | <.0001 | 0.00104960 | |
| 17 | LHL | 0.0597 | 5.06 | <.0001 | 0.00098697 | |

Table 10. Stepwise discriminant analysis for female and male camel populations.

| Step | Stepwise selection summary | | | | | | | | | | | |
|-------------------|----------------------------|----------|------|---------------|-----------|-------------------|-------------------|----------|------|---------------|-----------|------|
| | Females | | | Males | | | | | | | | |
| Variables entered | Partial R-squared | F-values | Pr>F | Wilks' lambda | Tolerance | Variables entered | Partial R-squared | F-values | Pr>F | Wilks' lambda | Tolerance | |
| 1 | CD | 0.85 | 378 | <0.0001 | 0.14 | 0.15 | FLHC | 0.86 | 106 | <0.0001 | 0.1304 | 0.14 |
| 2 | BG | 0.66 | 128 | <0.0001 | 0.04 | 0.43 | CD | 0.87 | 109 | <0.0001 | 0.0164 | 0.97 |
| 3 | DE | 0.56 | 85 | <0.0001 | 0.02 | 0.40 | HG | 0.83 | 78 | <0.0001 | 0.0027 | 0.90 |
| 4 | BW | 0.55 | 80 | <0.0001 | 0.009 | 0.12 | HS | 0.69 | 35 | <0.0001 | 0.0008 | 0.38 |
| 5 | BL | 0.51 | 69 | <0.0001 | 0.004 | 0.11 | DE | 0.57 | 20 | <0.0001 | 0.0003 | 0.36 |
| 6 | HW | 0.43 | 48 | <0.0001 | 0.002 | | BL | 0.53 | 17 | <0.0001 | 0.0001 | 0.26 |
| 7 | FCL | 0.32 | 30 | <0.0001 | 0.001 | | BG | 0.41 | 10 | <0.0001 | 0.0001 | |
| 8 | LFL | 0.28 | 25 | <0.0001 | 0.001 | | TL | 0.44 | 11 | <0.0001 | 0.0001 | |
| 9 | TL | 0.22 | 18 | <0.0001 | 0.0009 | | CW | 0.35 | 7 | <0.0001 | 0.00003 | |
| 10 | HG | 0.16 | 12 | <0.0001 | 0.0008 | | LHL | 0.28 | 5 | <0.0001 | 0.00002 | |
| 11 | FLHC | 0.15 | 11 | <0.0001 | 0.0006 | | HW | 0.30 | 6 | <0.0001 | 0.00001 | |
| 12 | HC | 0.16 | 12 | <0.0001 | 0.0005 | | FCL | 0.26 | 5 | 0.0001 | 0.000012 | |
| 13 | HLHC | 0.13 | 9 | <0.0001 | 0.0005 | | LFL | 0.21 | 3 | 0.0016 | 0.000009 | |
| 14 | EL | 0.13 | 9 | <0.0001 | 0.0004 | | BW | 0.13 | 2 | 0.0513 | 0.000008 | |
| 15 | CW | 0.10 | 7 | <0.0001 | 0.0003 | | | | | | | |
| 16 | NL | 0.07 | 5 | <0.0001 | 0.0003 | | | | | | | |
| 17 | LHL | 0.06 | 4 | 0.0003 | 0.0003 | | | | | | | |
| 18 | HS | 0.04 | 3 | 0.0056 | 0.0003 | | | | | | | |

Principal component analysis

Principal components and correlation circles for morphological measurements of female and male camel populations are shown in Table 11 and Figure 3. The first two principal components expressed 78% of the total variation in both genders (Table 12). The first principal component in both male and female camel populations was positively correlated with all

variables. Most of the variation in female camel populations was accounted by body length variables (BG, HG, HS, LHL and LFL) whereas variation in male camel populations was mainly determined by both body length and width variables (BG, HS, BL, CD and HW). The first two components in female camel populations were closely associated with HS, LFL and LHL.

Table 11. Weighting of each trait in the PCA analysis. Values indicate the relative (negative and positive) contributions of traits to the first two principal components 1 and 2.

| Traits | Principal component 1 | | Principal component 2 | |
|--------|-----------------------|---------|-----------------------|---------|
| | Males | Females | Males | Females |
| HG | 0.345 | 0.363 | -0.297 | -0.123 |
| BG | 0.357 | 0.368 | -0.090 | -0.181 |
| HS | 0.369 | 0.302 | -0.007 | 0.319 |
| BW | 0.393 | 0.377 | -0.144 | -0.019 |
| BL | 0.184 | 0.276 | 0.530 | -0.303 |
| CD | 0.199 | 0.313 | 0.495 | -0.252 |
| HW | 0.237 | 0.297 | 0.371 | -0.072 |
| LHL | 0.328 | 0.293 | -0.303 | 0.503 |
| LFL | 0.335 | 0.270 | -0.239 | 0.567 |
| HL | 0.336 | 0.278 | 0.267 | -0.339 |

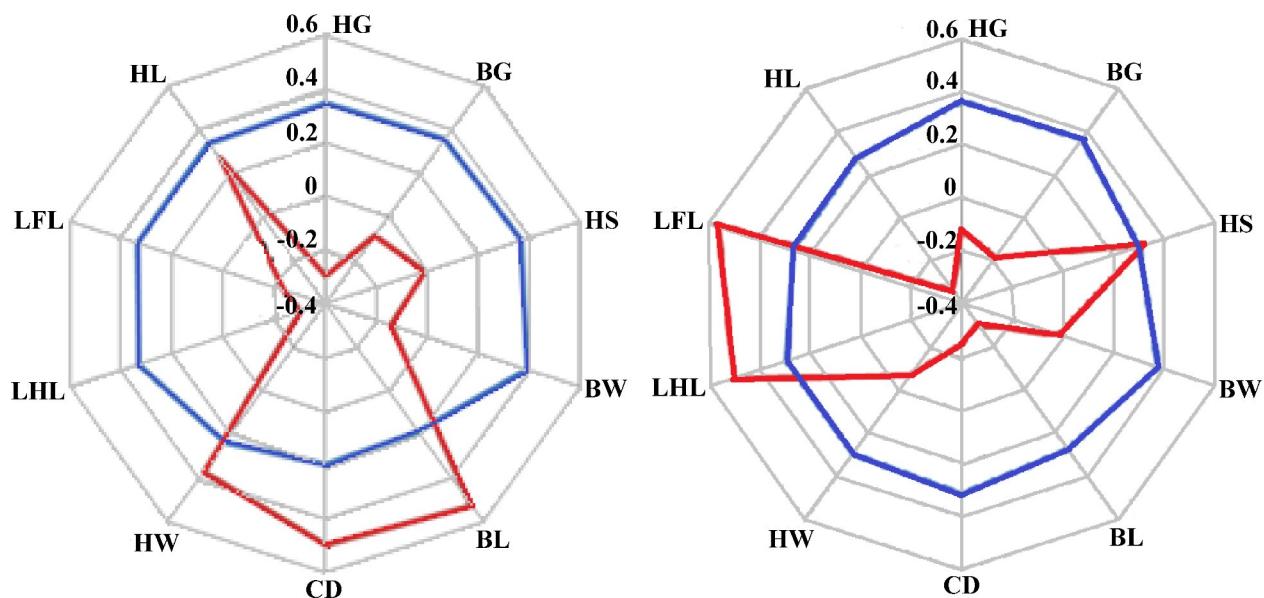


Figure 3. Correlation circles of morphological variables on the first two principal components (blue line for principal component 1 and red line principal component 2) (males on the right side and females the on left side).

Table 12. Eigen values and variance of the principal component analysis for body measurements.

| Female camel populations | | | | Male camel populations | | | |
|--|--------------|--------------|--|------------------------|--------------|--------------------|--|
| Eigen values of the correlation matrix | | | Eigen values of the correlation matrix | | | | |
| PCs | Eigen values | Variance (%) | Total variance (%) | Eigen values | Variance (%) | Total variance (%) | |
| PC1 | 6.613 | 66 | 66 | 5.651 | 56 | 56 | |
| PC2 | 1.219 | 12 | 78 | 2.179 | 22 | 78 | |

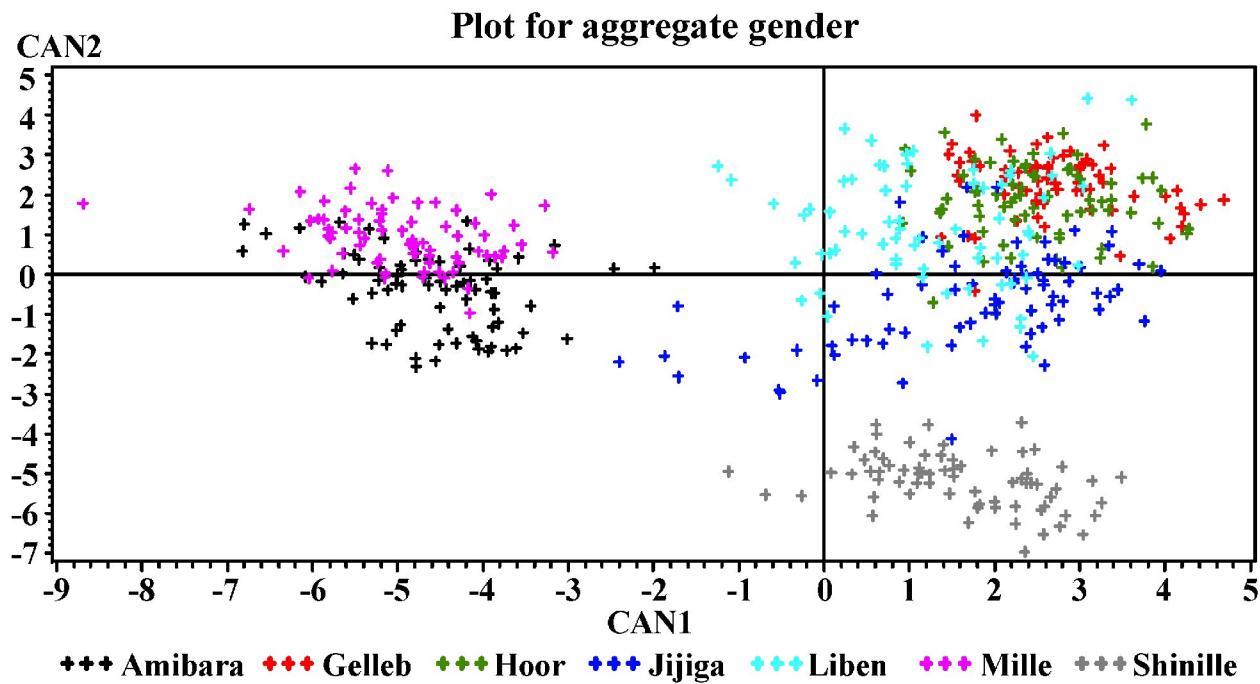


Figure 4. Plot of canonical discriminant analysis illustrating the first against the second canonical variable for all 494 Ethiopian camels.

The canonical analysis for all seven camel populations in aggregate gender allowed identifying two canonical variables (CAN1 and CAN2) which were statistically significant ($p<0.0001$). The CAN1 and CAN2 accounted for 49.2% and 27.5% of the total variation, respectively. Figure 4 shows the results of these two canonical variables that separate all 494 Ethiopian camels. CAN1 separated two camel groups: Amibara and Mille as one group and Shinille, Jijiga, Liben, Hoor and Gelleb as another group. CAN2 also divided two groups: (1) Shinille, Jijiga and Amibara; and (2) Mille, Hoor, Liben and Gelleb.

At the final stage of classification tree in aggregate gender, the seven Ethiopian camel populations were divided into two major groups (Figure 5). The first group contained the short,

light weight camel populations (Amibara, Mille, Shinille and Jijiga) observed in the lowland ecology. The second group included the long, heavy weight, long body sized Hoor, Gelleb and Liben camel populations. Then camel populations within each group were further divided into phenotypically distinct and agro-ecologically separated sub-groups. At a distance level of 0.4 and greater, three sub-groups can be distinguished. Jijiga camel population can be treated as a separate sub-group distinct from Amibara, Mille and Shinille camel populations which are distributed in arid and semi-arid ecology with sparse vegetation cover and high temperature while Jijiga area is characterized by low temperature, better vegetation cover and wet environment. The rather close relationship between Hoor and Gelleb camel populations,

both are present in Gode area, can be explained by the mating practice followed by the communities. According to Ogden pastoral communities, crossbreds between Hoor and Gelleb camel populations exist and are named as Aiden (Figure 6, No. 6). As indicated in Table 13 and Figure 6, Jijiga and Hoor camels

have large barrel girth and udder size. Similarly, Liben and Gelleb camels have tall height and wide body size. Besides, various colors of camels were also identified in this study, including a white camel as shown in Figure 6 (No. 3).

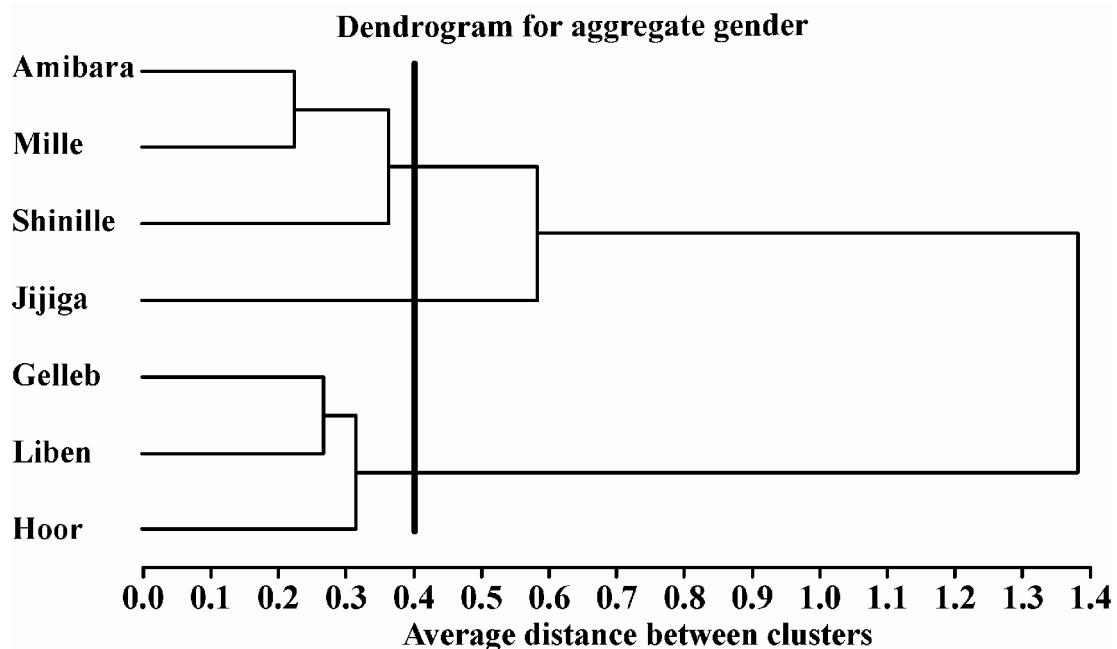


Figure 5. Hierarchical classification tree (dendrogram) of seven Ethiopian camel populations (vertical line indicates 0.4 dis-similarity).

Table 13. The five major camel groups among seven Ethiopian camel populations.

| No. | Camel groups | Features |
|-----|--------------------------|--|
| 1 | Hoor | Wide belly, long legs, Long body, tall height, small hip width |
| 2. | Gelleb and Liben | prominent hump, wide chest and hip, long neck and tail |
| 3. | Jijiga | Short length, medium body size and barrel girth |
| 4. | Shinille | Long ear with small body weight and heart girth, short height at shoulder, barrel girth, and short neck length |
| 5. | Amibara and Mille (Afar) | Small barrel and heart girth with small body weight, and long tail |

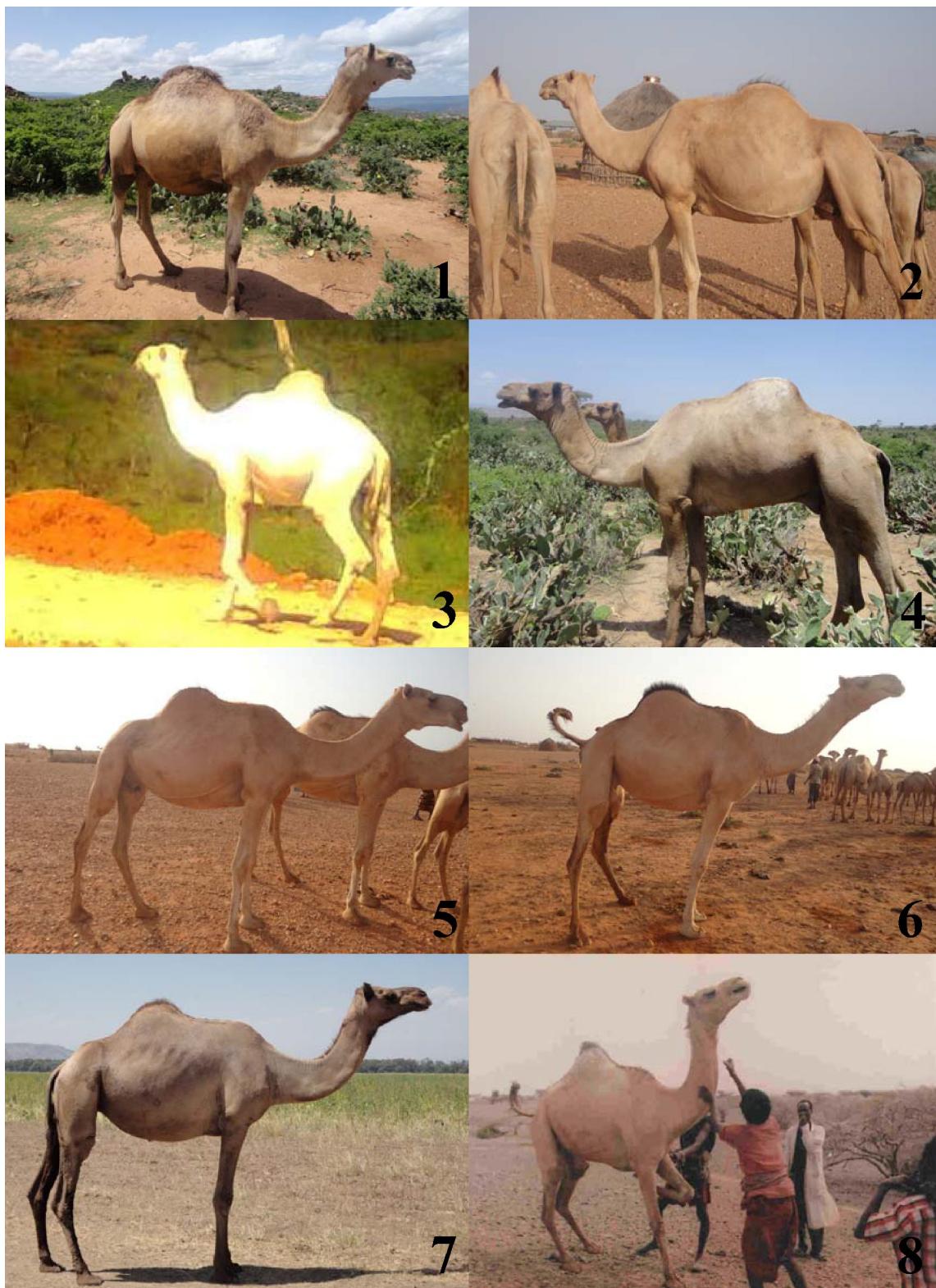


Figure 6. Camels in south, east and northeast in Ethiopia.
1 = Jijiga camel; 2 = Hoor camel; 3 = Liben camel; 4 = Shinille camel; 5 = Gelleb camel; 6 = Aiden camel; 7 = Amibara camel; 8 = Mille camel.

Discussion

The overall significantly ($p<0.05$) superior body and morphometric length (leg, neck, ear, face, and tail), height (height at shoulder and barrel girth) and width (chest width and hip width) traits in male to female camels indicate the presence of sexual dimorphisms among the camel populations, which were also reported by Yohannes et al. (2007) and Ishag et al. (2011) in Jijiga and Sudanese camel populations, respectively. The wide chest and hip and heavy weight exhibited by Gelleb and Liben camel populations show their potential for meat production. This result is in agreement with Abebe (1991) who reported that these camels have a greater potential in terms of meat production. On the other hand, the character features of large BG, small CW and HW as well as large udder size for Jijiga and Hoor camel populations may indicate their milk production potential. Previous study noted that milk production potential of these camels is higher than Issa (Shinille) and Afar types of camels (Abebe, 1991). The different HL but similar BG in Hoor and Gelleb camel populations may be due to their difference in milk production characteristics. Hoor camel population is more suitable and preferred in most of the time for milk production than Gelleb camel population in Gode pastoral communities. It may be related with utilization of stored energy in the hump for milk production during scarcity of feed or drought periods.

The calculated average BW of Hoor, Gelleb (Ogaden) and Liben camels are higher than values reported by Manayzewal (1987), Ishag et al. (2011) and Raziq et al. (2011) for Areho type of Erythrean camel, Sudanese camel and Raigi camel from Pashtoon nomads of Afghanistan and Pakistan, respectively, but lower than the value in Muhammed (2001). The lower values of BG, HG, BW, CW and HW recorded for Amibara, Mille and Shinille camels may be attributed to the high intensity of temperature and scarcity in feed availability of the environment of origin of these populations. The morphological body structures of these camels (e.g. small body size) are important attributes for adaptation to scarcity of feed and high temperature. Shinille camels are the smallest one in Ethiopia, but it has prominent shoulders,

a deep chest and well-muscled straight legs, an indication of their capacity for draft purpose. The HG, NL and HS of this camel population are much lower than the measurements taken on Saudi Arabian camel breeds. Amibara and Mille camels are comparable in almost all measurements with values for Saudi Arabian camel breeds (Abdallah and Faye, 2012).

Significantly long hind and forelegs for Mille and Liben camels may show their adaptive long leg traits to arid areas. Moreover, the small body size and long legs may indicate the riding character of Mille camels. The presence of significantly superior TL in Hoor and Gelleb camels may indicate their adaptive nature to protect themselves from biting flies, some of which are disease causing organisms. This can be supported by the fact that the natural environment for Hoor and Gelleb camel populations is Wabe Shebele River basin, where there is a favorable condition for breeding and multiplication of the biting flies. The study of Abebe (1991) indicated that trypanosomiasis is one of the major diseases and infection of *Trypanosoma evansi* was common in Ogaden (Hoor and Gelleb) camel populations.

Squared Mahalanobis distances differ between genders. The highest phenotypic distance was observed between Shinille male camels and males of other camel populations. As noted in this study, mean values of this population are exceptionally below the average means of other populations in BW, HG, HS, BG, which make the Shinille male camels distant from others. According to the group discussion with elders in Shinille District, male camels are used for transportation of fuel wood and other activities year round, and do not accompany other herds during migration in search of feed and water. But female camels migrate during dry season for three months to other places where better feeds are available. Thus the major feed resource for camels in this area is Cactus pear (*Opuntia ficus-indica*), which is available throughout the year. However, Cactus pear has low nutrient contents especially the protein which is even below the maintenance requirement, hence can affect growth of livestock (Tegegne, 2001). In addition, ratio of Ca:P level is not negligible for appropriate skeletal development. One study on

O. polyacantha revealed that phosphorus content was below livestock dietary requirement (Shoop et al., 1997). Other study explained that phosphorus (P) is one of the essential minerals for all animals. It plays a critical role in cellular metabolism as part of the energy currency of the cell, in cellular regulatory mechanisms and in bones. Through its involvement in these metabolic and structural processes, P is essential for animals to attain their optimum genetic potential in growth as well as skeletal development (Todd and Roselina, 2008). The low nutritional quality of Cactus pear might have therefore been the major factor that negatively hampered most body measurements of Shinille male and to some extent female camels. This implies the importance of supplementing camels with additional feeds especially having high protein content in addition to Cactus pear in this area.

Squared Mahalanobis distances between Mille and Amibara and between Hoor and Gelleb camels are small in comparison with those between other camel populations in aggregate gender. The differences among these camel populations can be justified from the relatedness of ecology, management and population history.

Stepwise discriminant analysis also indicates the existence of sexual dimorphisms in camels. This result is in agreement with Ishag et al. (2011) and Abdallah and Faye (2012) who reported the presence of sexual dimorphisms in Sudanese and Saudi Arabian camels. In this study, it was possible to discriminate female camel populations through CD, BG and DE whereas male camel populations can be discriminated by FLHC, CD and HG. For aggregate gender, morphometric variables of CD, BL, DE and HS were important variables to differentiate variability within camel populations. It shows that all these variables are not affected by environment and thus describe inherent size of the variables. This result was in agreement with Kefena et al. (2011) who reported body height and body length to be more important variables to discriminate between Ethiopian donkey populations. Variations in variables like HG, HLHC, CW,

EL, NL and LHL among camel populations were due to inherent population differences.

Body length traits (HG, HS, BG, LHL and LFL) in female camels and both body length and width traits in male camels can be used as selection indicators (strong effect on variation) in present camel populations. The result of correlation estimate is comparable with that reported by Abebe et al. (2002). The positive correlation indicates that simultaneous genetic improvement in some variables can be achieved when selection is applied to other variables. It is also useful to estimate the weight of camels from correlated linear measurements, where weighing scale is not easily available.

Combining both canonical discriminant analysis at individual level (Figure 4) and hierarchical classification tree built at population level (Figure 5) based on the differences among all morphological variables in aggregate gender, five major groups can be defined among the seven Ethiopian camel populations with major features as summarized in Table 13. These classifications are largely in agreement with the shared agro-ecological similarities under which these camels are distributed (e.g. the Amibara and Mille camels) and/or the unique management practice and population history of specific camel populations. For example, elders in Ogden note that a pastoral household who owns more number of the crossbreds between Hoor and Gelleb camels in the herd is considered as prestigious. This is because of the pastoralists' belief that Aiden camels are more tolerant to high temperature, scarcity of feed and water and resistant to disease than the two parental populations. Such practice certainly facilitates a regular gene flow between these two camel populations.

Conclusion

The extent of phenotypic variation is valuable to select and utilize different camel populations based on their specific characteristics and body conformation in breeding program. The presence of different camel populations in morphology, productive, adaptive and other characters in present study may provide a basis for selection and improvement. Thus attention should be given to

exploit the performance of all camel populations based on their specialization to fulfill the current demand of camel and camel by-products in the country and also in different parts of the world. The present study can be used to understand the camel resources of the country for future genetic improvement and conservation actions.

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