

THE DETECTION OF SUBCLINICAL MASTITIS IN THE BACTRIAN CAMEL (*CAMELUS BACTRIANUS*) BY SOMATIC CELL COUNT AND CALIFORNIA MASTITIS TEST

O.A.Sh. ABDURAHMAN

Swedish University of Agricultural Sciences, Department of Obstetrics and Gynaecology, PO Box 7039, 750 07 Uppsala, Sweden

ABSTRACT

Abdurahman, O.A.Sh., 1996. The detection of subclinical mastitis in the bactrian camel (*Camelus bactrianus*) by somatic cell count and California mastitis test. *Veterinary Research Communications*, 20 (1), 9–14

Milk samples ($n = 160$) from 7 clinically healthy bactrian camels were cultured to detect subclinical udder infection. The samples were assessed by the Californian mastitis test (CMT) and somatic cell count (SCC). Bacteria were recovered from 36 (22.5%) of the milk samples. *Staphylococcus aureus* and coagulase-negative staphylococci (CNS) were the main organisms found.

Infected quarters had significantly higher mean values for the SCC ($p < 0.01$) and CMT ($p < 0.001$) than non-infected quarters. All 7 camels were infected with CNS but only 4 with *S. aureus*. CMT values for *S. aureus*-infected camels were significantly higher than for those only infected with CNS. The values for SCC and CMT were significantly influenced by the stage of lactation ($p < 0.05$). No significant difference was found from the effect of the quarters. Both SCC and CMT were of value in predicting the infection status of the udder.

Keywords: camel, mammary gland, milk, somatic cells, *Staphylococcus aureus*, udder

Abbreviations: CMT, California mastitis test; SCC, somatic cell count; CNS, coagulase-negative staphylococci

INTRODUCTION

Camels are kept for milk production in many arid lands of Africa and Asia. They produce milk for a long period (8–18 months) and also under unfavourable conditions. However, information on their udder health and milk hygiene is scanty. During the past decade there have been several reports on subclinical mastitis in dromedary camels (Obeid, 1983; Arush *et al.*, 1984; Quandil and Oudar, 1984; Barbour *et al.*, 1985; Mostafa *et al.*, 1987) and a few bactrian camels (Kospakov, 1976a,b), but little work has been done on subclinical mastitis and the udder's response to bacterial invasion. Barbour and colleagues (1985) applied CMT to composite milk samples from the dromedary camels and concluded that the test was useful for screening subclinically infected udders. Obeid (1983) found a good correlation between the milk leukocyte count and the 'rapid mastitis test'. In a previous study Abdurahman and colleagues (1992) have found that bactrian milk contains not only leukocytes but also large numbers of anuclear cell-like particles, so-called 'cell fragments'. The present

work was carried out to assess the relationships between udder infection, somatic cell counts and CMT and the value of SCC in predicting the infection status of the camel udder.

MATERIALS AND METHODS

Animals

Seven lactating camels (*Camelus bactrianus*) kept at the zoological park of Kolmården, Sweden, were used. The camels were of varying ages and lactation stages (1–8 months), and were suckling their calves. They were housed individually and fed with concentrates and hay. All the camels were free from clinical mastitis during the sampling period.

Sampling

The camels were restrained as described earlier (Abdurahman *et al.*, 1992). The udder and the teats were washed and cleaned with 70% alcohol. The first few streams of milk from each quarter were discarded. About 5–10 ml of milk was then collected into sterile polyethylene tubes. The samples were kept on ice during transportation. The seven camels were sampled at 4–6-week intervals for 8 months. A total of 160 milk samples were obtained. Of these, 124 and 159 samples were tested for SCC and CMT, respectively.

Bacteriological examination

Milk samples (0.01 ml) from each quarter were streaked on blood agar plates and the plates were incubated for 24–48 h at 37°C. The plates were then examined for growth, colony morphology, haemolysis, catalase and coagulase tests, according to the Scandinavian recommendations on examination of bovine quarter milk samples (Klastrup, 1975).

California mastitis test (CMT)

CMT was carried out using the method described by Schalm and Noorlander (1957). An equal volume of CMT reagent and milk was mixed and the reaction was graded 1, 2, 3, 4 or 5, according to the Scandinavian recommendations, corresponding to 0, trace, 1, 2 and 3 (Klastrup and Schmidt Madsen, 1974). The test was performed by a trained technician. The reactions were interpreted as follows: score 1 = no reaction; score 2 = slight slime which tends to disappear with continued swirling; score 3 = distinct slime but without gel formation; score 4 = immediate formation of gel which

moves as a mass during swirling; score 5 = gel develops a convex surface and adheres to the bottom of the paddle.

Microscopic somatic cell count (SCC)

A modification of the method described by Prescott and Breed (1910) was used. Quarter milk samples (0.01 ml) were spread over an area of 1 cm² of a glass slide with the aid of a microsyringe. Four such squares were prepared from each sample. The smears were stained with methylene blue and examined by microscopy under oil immersion. An attempt was made to count only the nucleated cells.

Statistical methods

The distribution of the SCC was skewed to the right. Logarithmic transformation (base 10) was used to bring the data closer to a normal distribution. Data were assessed by least-squares analysis of variance, using the general linear models procedure of the Statistical Analysis System (SAS Institute, 1989). The model included the effects of the animal, the quarter, the stage of lactation (8 classes, 1–8 months) and the bacteriological findings (non-infected, CNS and *S. aureus*). The results are presented as means \pm SEM. The correlation coefficient between the mastitis indicators CMT and SCC was calculated using Spearman rank correlation of the residual obtained after correcting for the effects included in the statistical model.

RESULTS

Intramammary infections were present in 36 (22.5%) of the 160 quarter milk samples examined (Table I). CNS and *S. aureus* represented 61.1% and 38.9% of the isolates, respectively. All 7 camels were infected with CNS but only 4 were infected with *S. aureus*. The mean values for SCC and CMT from the infected and non-infected quarters are shown in Table II. Infected udder quarters had significantly higher mean values for both SCC and CMT.

Table III shows separately the mean values for non-infected quarters and quarters infected by CNS and *S. aureus*. There were no significant differences in mean values between quarters infected by CNS and *S. aureus*. CMT values for *S. aureus*-infected camels were significantly higher ($p < 0.05$) than for those only infected with CNS. Mean SCC values were numerically but not significantly higher in *S. aureus*-infected camels. The correlations between SCC and CMT in all quarters, in non-infected and infected quarters were 0.39 ($n = 124$), 0.25 ($n = 99$) and 0.32 ($n = 25$), respectively. There was thus an overall poor but significant ($p < 0.001$) correlation between the mastitis indicators.

The effect of lactation stage had a significant ($p < 0.05$) influence on the SCC and CMT values. The values were high shortly after parturition, fell within the first 2 months and then showed a slight increase towards the end of lactation.

TABLE I
Bacteriological findings in quarter milk samples from bactrian camels

Microorganisms isolated	Number	Percentage
Non-infected	124	77.5
Coagulase-negative staphylococci	22	13.8
<i>Staphylococcus aureus</i>	14	8.7
Total	160	100

TABLE II
Mean \pm SEM of SCC and CMT for infected and non-infected quarters of bactrian camels

Component	<i>n</i>	Non-infected	Infected	<i>p</i> -value
SCC cells/ml	124	12.16 \pm 0.16 (<i>n</i> = 99)	13.37 \pm 0.26 (<i>n</i> = 25)	<i>p</i> < 0.01
CMT score	159	1.32 \pm 0.09 (<i>n</i> = 123)	2.33 \pm 0.19 (<i>n</i> = 36)	<i>p</i> < 0.001

CMT = California mastitis test; SCC = somatic cell count; *n* = number of observations

TABLE III
Mean \pm SEM of milk SCC and CMT in non-infected and in *S. aureus*- and CNS-infected quarters of bactrian camels

Component	<i>n</i>	Non-infected	<i>S. aureus</i>	CNS
SCC cells/ml	124	12.16 \pm 0.16 (<i>n</i> = 99)	13.02 \pm 0.39 (<i>n</i> = 10)	12.98 \pm 0.32 (<i>n</i> = 15)
CMT score	159	1.32 \pm 0.09 (<i>n</i> = 123)	2.07 \pm 0.24 (<i>n</i> = 14)	2.25 \pm 0.20 (<i>n</i> = 22)

CMT = California mastitis test; SCC = somatic cell count; CNS = coagulase-negative staphylococci; *n* = number of observations

DISCUSSION

The data presented in this study were derived from only 7 camels from a zoological garden and the results must be interpreted with caution. Despite these limitations, the bacteriological findings are comparable with results from both bactrian and dromedary camels (Kospakov, 1976a; Bakhiet *et al.*, 1992) and using such animals offered the possibility of follow-up and continued sampling. The latter is difficult under nomadic conditions.

CNS and *S. aureus* were the main bacteria recovered. Kospakov (1976b) isolated staphylococcal strains from udder tissue, bulk milk and udder skin of bactrian camels. The microorganisms found in the present study were regarded as important pathogens causing mastitis in dromedary camels (Bakhiet *et al.*, 1992), cows (Schalm *et al.*, 1971), ewes (Maisi *et al.*, 1987) and goats (Pettersson, 1981).

Very few reports are available regarding the cellular content of camel milk. Kospakov (1976a) reported an average of 1.3×10^6 cells/ml in non-infected bactrian milk during normal lactation, which was elevated to 7.9×10^6 cells/ml at the start of the dry period. In infected milk samples, total cell counts between 7.4×10^6 and 11.7×10^6 cells/ml were observed. In another study Mostafa and colleagues (1987) found an increase in the number of somatic cells in infected milk samples from dromedary camels.

The present study showed significant differences in the mean values for SCC and CMT between infected quarters and those free from infection. Camels infected with *S. aureus* had higher SCC and CMT values than those infected with CNS. *S. aureus* is regarded as a primary cause of mastitis in dairy animals, including camels (Schalm *et al.*, 1971; Obeid, 1983), and has been implicated in both clinical and subclinical mastitis (Barbour *et al.*, 1985; Kospakov, 1976a). However, the camel has not been the subject of experimental mastitis studies, and the epidemiology and pathogenicity of mastitis-causing organisms in these animals remains unclear.

As well as somatic cells, mastitis-free camel milk contains large numbers of non-nucleated cell fragments (Abdurahman *et al.*, 1992). Similar cell fragments in goat's milk did not react with CMT reagents (Schalm *et al.*, 1971). However, the cell fragments may be counted as cells in microscopic counting. Although an elevated cell count is not a constant feature, camels free of infection had a higher basal SCC than cows. This was also reported by Kospakov (1976a).

The observed influence of the stage of lactation is consistent with the finding of Kospakov (1976a) that the cell content in camel's milk increases as lactation progresses and can be regarded as similar to that described in the cow (Schalm *et al.*, 1971). In the present study SCC and CMT have both been shown to be valuable indicators of udder infection, but the number of animals used was small. Further investigations using larger numbers are necessary to understand the dynamics of mastitis in camels before such indicators could be routinely used in predicting udder infections in the camel.

ACKNOWLEDGEMENTS

This study was supported by the Swedish Agency for Research and Cooperation with developing countries (SAREC). The author thanks Professor G. Aström for advice and Agr. Dr Nils Lundeheim at the Department of Animal Breeding and Genetics, Uppsala, for help with statistical analysis. The invaluable help given by Dr B.O. Röken and the staff at Kolmården is gratefully acknowledged.

REFERENCES

- Abdurahman, O.S., Cooray, R. and Bornstein, S., 1992. The ultrastructure of cells and cell fragments in mammary secretions of *Camelus bactrianus*. *Journal of Veterinary Medicine A*, **39**, 648–655
- Arush, M.A., Valente, C., Compugnuci, M. and Hussein, H., 1984. Indagine sulla diffusione delle mastite del dromedario (*Camelus dromedarius*) in Somalia (Prevalence of mastitis in camels (*Camelus dromedarius*) in Somalia). *Bulletino Scientifico della Facolta di Zootechnia e Veterinaria*, **4**, 99–104
- Bakhiet, M.R., Aqab, H. and Mamoun, I.E., 1992. Camel mastitis in western Sudan. *Sudanese Journal of Veterinary Science and Animal Husbandry*, **31**, 58–59
- Barbour, E.K., Nabbut, N.H., Frerichs, W.M., Al-Nakhil, H.M. and Al-Mukayel, H.M., 1985. Mastitis in *Camelus dromedarius* in Saudi Arabia. *Tropical Animal Health and Production*, **17**, 173–179
- Klastrup, O., 1975. Nordic recommendations of quarter milk samples. *Annual Bulletin of International Dairy Federation*, **85**, 41–52
- Klastrup, O. and Schmidt Madsen, P., 1974. Nordiske rekommendationer vedrorende mastitisundersogelser af kirtelprover (Nordic recommendations concerning mastitis control of quarter samples). *Nordic Veterinar-Medicin*, **26**, 197–204
- Kospakov, Z.K., 1976a. Cell content in the milk of bactrian camels, depending on stage of lactation and condition of the udder. *Problemy Veterinarnoi Sanitarii*, **55**, 21–25 (in Russian). (*Veterinary Bulletin*, **48**(11) No. 1566 (1978))
- Kospakov, Z.K., 1976b. Phage typing of pathogenic Staphylococci isolated from milk and the environment of camel breeding farm. *Problemy Veterinarnoi Sanitarii*, **55**, 32–35 (in Russian). (*Veterinary Bulletin*, **48**(11) No. 6559 (1978))
- Maisi, P., Junttila, J. and Seppänen, S., 1987. Detection of subclinical mastitis in ewes. *British Veterinary Journal*, **143**, 402–409
- Mostafa, A.S., Ragab, A.M., Safwat, E.E., El-Sayed, Z., Abd-el-Rahman, M., El-Danaf, N.A. and Shouman, M.T., 1987. Examination of raw she-camel milk for detection of subclinical mastitis. *Journal of the Egyptian Veterinary Medical Association*, **47**, 117–128
- Obeid, A.I., 1983. *Field investigation, clinical and laboratory findings of camel mastitis*. (MSc Thesis, University of Khartoum)
- Petterson, K.-E., 1981. Cell content in goat's milk. *Acta Veterinaria Scandinavica*, **22**, 226–237
- Prescott, S.C. and Breed, R.S., 1910. The determination of the number of body cells in milk by a direct method. *Journal of Infectious Diseases*, **7**, 632–640
- Quandil, S.S. and Oudar, J., 1984. Bacteriological study of some cases of mastitis in the dromedary (*Camelus dromedarius*) in the United Arab Emirates, preliminary report. *Revue de Médecine Veterinaire*, **135**, 705–707
- SAS Institute, 1989. SAS User's Guide: Statistics Version 5, Edition 1989, (SAS Institute Inc., Cary, NC)
- Schalm, O.W. and Noorlander, D.O., 1957. Experiments and observations leading to development of California Mastitis Test. *Journal of American Veterinary Medical Association*, **130**, 199–204
- Schalm, O.W., Carroll, E.J. and Jain, N.C., 1971. *Bovine Mastitis*, (Lea and Febiger, Philadelphia)