# SUSCEPTIBILITY OF BACTERIA ISOLATED FROM CAMEL (Camelus dromedarius) MASTITIS TO COMMONLY USED ANTIMICROBIALS

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### ABSTRACT

A total of 219 isolates of bacterial pathogens from intramammary infection of lactating camel (*Camelus dromedarius*) were subjected to *in vitro* antimicrobial susceptibility test using single disc diffusion methods. The result showed that *Staphylococcus aureus* isolates were found to be 100% susceptible to tetracycline, kanamycin, chloramphenicol, gentamicin and streptomycin. Oxytetracycline and tetracycline were drugs of choice for *Streptococcus agalactiae*. *E. coli* isolates were highly sensitive to kanamycin and gentamicin. Oxytetracycline, tetracycline and chloramphenicol were effective drugs against camel mastitis pathogens with exception of *Pasteurella haemolytica* and *Enterobacter aerogenes*. A high level of resistance of mastitis pathogens of camels was recorded against nalidixic acid and erythromycin.

Key words : Antimicrobials, camel, mastitis, pathogens, susceptibility test

Most camels are kept by nomadic pastoralists, and play an essential role in their subsistence economy and camel milk is the main food for the nomads in the pastoral production system of the east Africa (Schwartz and Dioli, 1992).

Mastitis is a complex disease occurring worldwide among dairy animals, with heavy economic losses largely due to sub-clinical mastitis. In Ethiopia, Almaw and Molla (2000), Salah (2000), and Bekele and Molla (2001) investigated the udder infections in the dromedary camels. This study was aimed to record the susceptibility of bacteria isolated from camel mastitis to commonly used antimicrobials.

# Materials and Methods

#### Study area

The study was conducted in three different districts namely, Negele (Borena Region), Dire Dawa and Gewane (Afar Region) in the southern, eastern, and north eastern Ethiopia.

#### Sampling

The animals in this study belonged to different herds with similar traditional management practices. The non-probability sampling methods were used

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to collect the quarter milk samples based on the willingness of camel herd owners (Thrusfield, 1995). Before the collection, the teats were disinfected with cotton wool moistened with 70% ethyl alcohol. The first few squirts of milk were discarded and then about 5 - 20 ml milk was collected in a sterile universal bottle. The quarter milk samples were kept in an ice-box and transported as soon as possible to the laboratory and were kept at -20°C before the application of the California Mastitis Test (Schalm *et al*, 1971 and Quinn *et al*, 1994) and microbiological investi-gations.

Frozen quarter milk samples were thawed at room temperature for microbiological investi-gations (Quinn *et al*, 1994).

#### Anti-microbial susceptibility test

The bacterial isolates were subjected to single disc diffusion susceptibility test (Kirby-Bauer Method) (NCCLS, 1990 and 1997; Quinn *et al*, 1994). Briefly, this was performed as follows:

The isolates were transferred to a tube containing 5 ml tryptic soy broth. The mixture was incubated at 37°C until a slight visible turbidity appeared. This was compared with the McFarland 0.5

turbidity standard. McFarland 0.5 turbidity standard was prepared by mixing 0.5 ml of solution A (0.048 M BaCl<sub>2</sub>) and 99.5 ml of solution B (0.36 N H<sub>2</sub>SO<sub>4</sub>).

**Solution A :** 1.75 gram  $BaCl_2X2H_2O$  was diluted in 100 ml distilled water.

**Solution B :** 1 ml  $H_2SO_4$  was mixed with 100 ml distilled water.

The broth cultures were streaked onto Mueller-Hinton agar containing 5% defibrinated blood and were incubated at 37°C for 18 to 24 hours and the diameters of zone of growth-inhibition were measured in millimetres and reported as susceptible, moderately susceptible, intermediate and resistant (Table 1).

# Results

A total of 219 isolates of bacteria from intramammary infection of lactating camel (*Camelus dromedarius*) were subjected to *in vitro* antimicrobial susceptibility test with kanamycin, oxytetracycline, tetracycline, penicillin G, gentamicin, ampicillin, chloramphenicol, streptomycin, erythromycin, and nalidixic acid (Table 1). Staphylococcus aureus isolates were found 100% susceptible to kanamycin, tetracycline, chloramphenicol, gentamicin, and streptomycin (Table 1). They were also highly susceptible to oxytetracycline (97.1%), ampicillin (94.3%) and penicillin G (91.4%). *S. hyicus, S. intermedius* and *Micrococcus* spp. isolates were very sensitive (100%) to oxytetracycline, kanamycin, tetracycline, chloramphenicol, gentamicin, and streptomycin. Similar percentage was recorded for *S. epidermidis* against chloramphenicol and gentamicin. *S. aureus, S. epidermidis, S. intermedius,* and *Micrococcus* spp. isolates showed complete resistance (100%) against nalidixic acid.

Oxytetracycline and tetracycline were drugs of choice against 10 isolates of *Streptococcus agalactiae* followed by chloramphenicol (90%) and penicillin G (80%). *S. uberis* isolates were found sensitive to gentamicin, chloramphenicol, oxytetracycline, and tetracycline (92.9, 85.7, 78.6 and 71.4%, respectively.

Gentamicin was an effective antibiotic (100%) against *Corynebacterium bovis* and *Actinomyces pyogenes*. High sensitivity of the two isolates was also recorded for chloramphenicol (87.5 and 100%,

Table 1. Antimicrobial susceptibility test of bacteria isolated from camel mastitis.

Isolates	No. tested	Per cent susceptibility of isates aganst different antibotics									
		Κ 30 μg	ОТ 30 µg	С 30 µg	Т 30 µg	GM 30 μg	S 10 μg	PG 10 U	ΑΡ 10 μg	Е 5 µg	NA 30 μg
S. aureus	35	100.0	97.1	100.0	100.0	100.0	100.0	91.4	94.3	68.6	0.0
S. epidermidis	35	97.1	71.4	100.0	74.3	100.0	85.7	54.3	42.9	77.1	0.0
S. hyicus	35	100.0	100.0	100.0	100.0	100.0	100.0	97.1	97.1	88.6	25.7
S. intermedius	5	100.0	100.0	100.0	100.0	100.0	100.0	80.0	80.0	100.0	0.0
Micrococcus spp.	2	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	0.0
S. agalactiae	10	10.0	100.0	90.0	100.0	30.0	10.0	80.0	50.0	60.0	0.0
S. uberis	14	50.0	78.6	85.7	71.4	92.9	50.0	57.1	14.3	42.9	14.3
S. pyogenes	5	40.0	80.0	80.0	80.0	60.0	20.0	60.0	60.0	40.0	20.0
S. Pneumoniae	2	50.0	100.0	100.0	100.0	100.0	50.0	50.0	50.0	50.0	0.0
Enterococcus faecalis	2	100.0	100.0	100.0	100.0	100.0	100.0	50.0	100.0	50.0	0.0
Past. haemolytica	8	87.5	12.5	25.0	12.5	100.0	37.5	0.0	0.0	0.0	12.5
C. bovis	8	50.0	50.0	87.5	50.0	100.0	50.0	25.0	25.0	12.5	0.0
A. pyogenes	3	66.7	100.0	100.0	100.0	100.0	66.7	100.0	100.0	100.0	0.0
B. cereus	10	80.0	80.0	100.0	70.0	90.0	80.0	20.0	20.0	50.0	50.0
E. coli	19	100.0	84.2	94.7	78.9	100.0	47.4	21.1	84.2	21.1	42.1
Enterobacter aerogenes	10	80.0	30.0	60.0	30.0	90.0	40.0	10.0	10.0	0.0	90.0
Klebsiella pneumoniae	5	80.0	80.0	100.0	40.0	100.0	80.0	0.0	20.0	0.0	80.0
Serratia marcescens	3	66.7	66.7	100.0	66.7	66.7	33.3	0.0	33.3	0.0	0.0
Aeromonas hydrophila	5	100.0	100.0	60.0	20.0	100.0	40.0	0.0	0.0	0.0	40.0
Proteus mirabilis	3	100.0	100.0	100.0	66.7	100.0	100.0	66.7	66.7	66.7	33.3

K = Kanamycin; OT = Oxytetracycline; C = Chloramphenicol; T = Tetracycline; GM = Gentamicin; S = Streptomycin; PG = Penicillin G; AP = Ampicillin; E = Erythromycin; NA = Nalidixic acid;  $\mu$ g = Microgram and U = Units.

respectively). *Pasteurella haemolytica* and *Bacillus cereus* isolates were found sensitive to gentamicin (100 and 90%, respectively) and kanamycin (87.5 and 80%, respectively). Enterobacteriaceae were found susceptible to gentamicin, chloramphenicol, kanamycin and oxytetracycline. However, *Enterobacter aerogenes* and *Serratia marcescens* showed low susceptibility to oxytetracycline (30 and 66%, respectively). Erythromycin and penicillin G were found ineffective against *Enterobacter aerogenes*, *Klebsiella pneumoniae* and *Serratia marcescens*.

## Discussion

The results of *in vitro* antimicrobial susceptibility test revealed that oxytetracycline, tetracycline and chloramphenicol were effective drugs against camel mastitis pathogens with exception of *Pasteurella haemolytica* and *Enterobacter aerogenes*. These findings are in agreement with those of Salah (2000). A reasonably high level of resistance of mastitis pathogens of camels was recorded against erythromycin.

The less sensitivity of environmental streptococci such as *Streptococcus uberis* to most antimicrobials used may be due to epithelial cell invasion and movement of the bacteria into epithelial layers, possibly reducing the effectiveness of the antimicrobial (Radostits *et al*, 2000).

*Pasteurella haemolytica* was resistant to the most antimicrobials used. Quinn *et al* (1994) stated that antibiotic susceptibility tests should be carried out on strains of *Pasteurella haemolytica* as plasmid mediated resistance to some used antibiotic has been widely encountered.

Many types of antibiotics have been used for treatment of infectious diseases in camels for long times (Younan *et al*, 2001) which may be the cause of resistance patterns of some mastitis pathogens of camel against some of the commonly used antimicrobial agents.

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#### References

- Almaw G and Molla B (2000). Prevalence and etiology of mastitis in camels (*Camelus dromedarius*) in Eastern Ethiopia. Journal of Camel Practice and Research 7(1): 97-100.
- Bekele T and Molla B (2001). Mastitis in lactating camels (*Camelus dromedarus*) in Afar Region, north-east Ethiopia. Berl. Münch. Tierärztl. Wochenschr 114(5-6): 169-172.
- National Committee for Clinical Laboratory Standards (NCCLS) (1990). Performance Standards for Antimicrobial Disc Susceptibility Tests. 3rd Ed. Approved Standard M2-A3, 771 East Lancaster Avenue, Villanova, PA.
- National Committee for Clinical Laboratory Standards (NCCLS) (1997). Performance standards for antimicrobial disc and dilution susceptibility tests for bacteria isolated from animals; Tentatine Standard. 17(11):1-25.
- Quinn PJ, Carter ME, Markey B and Carter GR (1994). In: Clinical Veterinary Microbiology. I<sup>st</sup> Ed. Wolfe Publishing, London.
- Radostits OM, Gay CC, Blood DC and Hincheliff KW (2000): Mastitis. In: Veterinary Medicine: A textbook of the disease of cattle, sheep, pigs, goats and horses. 9th ed. London: Harcourt Publishers Ltd. pp 603-700.
- Salah W (2000). Camel milk bacteriology with special reference to mastitis in Borena lowland pastoral area, southwestern Ethiopia. DVM Thesis. Faculty of Veterinary Medicine. Addis Ababa University, Debre Zeit, Ethiopia.
- Schalm DW, Caroll EJ and Jain NC (1971). In: Bovine Mastitis. Lea and Febiger, Philadelphia.
- Schwartz HJ and Dioli M (1992). Introduction: The Camel (*Camelus dromedarius*) in Eastern Africa. In: The One-Humped Camel in Eastern Africa. A Pictorial Guide to Diseases, Healthcare and Management. Schwartz HJ and Dioli M (Eds). Verlag Josef Margraf, Weikersheim FR, Germany. pp 1-9.
- Thrusfield M (1995). Veterinary Epidemiology. 2nd ed. Blackwell Science Ltd. UK.
- Younan M, Ali Z, Bornstein S and Müller W (2001). Application of the California Mastitis Test in intramammary *Streptococcus agalactiae* and *Staphylococcus aureus* infections of camels (*Camelus dromedarius*) in Kenya. Preventive Veterinary Medicine 51(3-4):307-316.