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## Identification of pathogen bacteria from camel (*Camelus dromedarius*) mastitis and investigation of antibiotic susceptibility

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### Abstract

The scope of this study was to investigate the presence of pathogenic bacteria in milk from female camels with mastitis and to select antibiotics for treatment with antibiotic susceptibility testing. A total of 40 milk samples taken from 20 dromedarian females, after application of CMT test and determination of SCC values, the camels were diagnosed with subclinical mastitis. Milk samples were inoculated into blood agar for identification of bacterial agents leading to mastitis. A total of 4 (12.5%) *Staphylococcus aureus*, 4 (12.5%) *S. auricularis*, 2 (6.25%) *S. pettenkoperi*, 2 (6.25%) *S. cohnii* spp. *cohnii*, 2 (6.25%) *S. equorum*, 2 (6.25%) *S. capitis*, 2 (6.25%) *Streptococcus agalactiae*, 2 (6.25%) *S. dysgalactiae*, 4 (12.5%) *Escherichia coli*, 2 (6.25%) *Pseudomonas pseudalcaligenes*, 2 (6.25%) *Corynebacterium pseudotuberculosis*, 2 (6.25%) *Aerococcus viridans* and 2 (6.25%) *Gemella morbillorum* were identified. Gram-positive bacteria were sensitive to Levofloxacin, Linezolid and Tetracycline and Daptomycin, resistant to Beta lactam-group antibiotics and macrolides. Vancomycin resistance was determined in *S. aureus* and *S. cohnii* spp. *cohnii* strains. Gram-negative strains are found generally susceptible to Cefepime and Pipersilin; resistant to Trimethoprim-sulfomethoxazole and Amoxicillin-Clavulanic acid. As a result, it is recommended to use antibiotic use to prevent the development of antimicrobial resistance as well as mastitis control methods such as the prevention of infection and monitoring the health status of the mammary of camels.

**Keywords:** Camel, Subclinical mastitis, Identification, Antimicrobial susceptibility.

### Introduction

Camel milk, meat and products have appeared in many cuisines around the world for centuries. Camel milk has beneficial effects on many biological processes such as digestion, absorption, growth and immunity [1]. Additionally, camel milk can be stored at room temperature longer than milk from other animals [2]. Camel whey protein contains a heterogeneous group of proteins, including serum albumin,  $\alpha$ -lactalbumin, immunoglobulin, lactoferrin, and peptidoglycan recognition protein [3]. Compared to milk, camel milk contains more antibacterial substances and has a high concentration of vitamin C, so it has valuable nutritional properties. Milk is characterized by a high percentage of lactoferrin, which can be considered a good source of minerals and vitamins. Also, because camel milk has the most important nutrients, camel milk can meet most of human's daily needs in these nutrients. Camel milk has a similar composition to human milk due to its protective proteins such as low cholesterol, low sugar, high mineral content (sodium, potassium, iron, copper, zinc and magnesium), vitamin C, lactoferrin, lactoperoxidase, immunoglobulins and lysozyme [4]. The present studies confirmed that camel milk is unique in terms of antioxidant, antibacterial, antiviral, anti-fungal, anti-hepatitis, tuberculosis, hypoglycemic, anti-cancer, anti-tumor, anti-aging, auto-immune disease, cosmetic and detergents [5-6]. Mastitis is a major global complex disease among dairy livestock with high economic losses. Camel mastitis is estimated to affect more than 25% of lactating camels. It is also known to cause about 70% loss from milk production. Mastitis has both extreme animal infections and economic significance. It is responsible for some harmful effects for human health and animal production. There is little published information about the pathogens associated with camel mastitis compared to bovine mastitis.

However, much detailed research still remains to be accomplished, especially in the mastery of breeding techniques, since these species is confronted with particular health issues, including mastitis, which can be significant. There has long been a lack of concern in camel mastitis on the grounds that clinical mastitis in this species is infrequent. Bacterial infection can be a major cause of mastitis in livestock. Several bacterial agents are associated with mastitis cases and their presence in milk can have a negative impact on consumer health [8-9]. Some research shows the genus *Staphylococcus* sp., *Streptococcus* sp., [10,11], *Micrococcus* sp., *Streptococcus agalactiae*, *Coagulase Negative Staphylococci* [12], *Staphylococcus epidermidis*, *Mannheimia haemolytica*, *Escherichia coli* and *Corynebacterium* sp. were isolated from camel mastitis cases. Camel has been linked to the cause of mastitis. Mammary infections (mastitis) are the leading risks of herd management in camel breeding as well as in cattle breeding. The scope of this study was to investigate the presence of pathogenic bacteria in milk from female camels with mastitis and to select antibiotics for treatment with antibiotic susceptibility testing.

## Materials and Methods

### Sample collection

Milk samples were taken from 20 female one-humped camels suspected of having mastitis during lactation. The study was carried out from August 2018 to April 2019 at private camel farms in Aydin province, Turkey. In this study, 40 camel milk samples were obtained from the right and left lobes of 20 dromedary camels at farms with a capacity of 5 to 20 camels. Animal material consists of female camels aged 4 to 10 years who are lactating without antibiotic treatment. Mastitis was diagnosed by applying CMT (California Mastitis Test DeLaval®, Sweden) and determining somatic cell counts. NucleoCounter™ SCC-100 (Chemometec®, Denmark) was used to determine the SCC value. The identification of bacterial isolates was determined by an automatic identification system Phoenix™ M50 (Becton, Dickinson U.K. Limited).

### Ethics of animal use

The authors of this research hereby declare that collection of specimens was carried out in accordance with the guidelines laid down by the US National Guide for the Care and Use of Laboratory Animals, National Academy Press, Washington, D.C. 2011 and with the European Communities Council Directive of 24 November 1986 (86/609/EEC).

### California mastitis test

Somatic cell number score in milk samples was determined qualitatively. Detergent containing bromocresol purple was used as the reagent to release the nucleic acid and rupture the somatic cell membrane, and it was observed whether a gel-like matrix was formed whose viscosity was proportional to the leukocyte number. Plastic shovels with four glasses were used. Equal amounts (3 ml) of milk and reagent were put into each glass of

the paddle and the contents were mixed in gentle circular motion in a horizontal plane. Astitis cases were determined according to the degree of agglutination. CMT is a highly sensitive and fast method to detect abnormally cell-rich milk. The principle consists of a mixture of milk and tea pools (detergent) in amounts intended to rupture the cells, so that the nuclear DNA of the cells gels when they come into contact with the latter [13].

### Determination of somatic cell count

Somatic cell count of the samples was determined via automatic device. The device is a counter based on the optical fluorescence principle. Ethidium bromide enters the structure of nuclear DNA, producing a fluorescent signal corresponding to SHS in milk. It is advantageous because it is an automatic and fast technique. The milk sample was brought to room temperature ( $21 \pm 1^\circ\text{C}$ ) and homogenized. Then, 0.75 ml of Reagan C at room temperature was added to the clean 1.5 ml ependorfs and 0.75 ml of the milk sample was added. The ependorfs prepared for analysis were homogenized by vortexing. For somatic cell counting, a pre-prepared eppendorf sample at room temperature was drawn into the cassette and loaded into the instrument. After loading, the result is automatically saved by the device within 2 minutes.

### Bacterial isolation and identification

Samples were brought to the laboratory by a cold chain and collected with sterile gauze from milk-filled tubes that were streaked in blood agar. The agar plates were incubated at  $37^\circ\text{C}$  for 24-48 h under aerobic conditions. Gram staining is performed on colonies that have grown as a result of incubation. Gram-positive and Gram-negative samples were inoculated onto Tryptic soy agar. TSA Petri dishes were incubated for 24 h at  $37^\circ\text{C}$ . Biochemical assays of pure colonies formed in TSA after incubation were verified using the BD Phoneix™ M50 device. This method is used for rapid identification and susceptibility testing of many aerobic and facultative anaerobic Gram positive and negative bacteria containing selected antimicrobial agents. 24-hour fresh cultures purified on triptic soy agar were suspended with ID broth in glass tubes according to McFarland 0.5 colony density. BD Phoneix™ PMIC / ID87 was used for gram positive bacterial isolates and BD Phoneix™ NMIC / ID-400 panel kit was used for gram negative bacterial isolates. Identification was performed on the instrument using separate panels for each sample. ID Broth suspension tubes prepared for each sample were placed in the device for bacterial diagnosis. Biochemical identification data obtained from the device were evaluated.

### Determination of antibiotic susceptibility

Antibiotic susceptibility tests were performed for the isolates that were identified with the automated device using the BD Phoneix™ PMIC / ID87 and NMIC / ID-400 kit. 24 h fresh cultures purified on triptic soy agar were prepared with AST Broth present in glass tubes according to McFarland 0.5 colony density. Panels containing bacterial suspensions were placed in the

device and MIC detection and sensitivity results were obtained from the electronic system. The antimicrobials and their MIC values of bacterial isolates (n=32) were determined according to CLSI standards [14].

## Results

### California mastitis test and somatic cell count numbers

A plastic shovel with four glasses was used. An equal amount (3 ml) of milk and reagent was placed in each glass of the shovel and the contents mixed in a horizontal plane with a slight circular motion. According to the degree of agglutination, the CMT score was determined as "Negative" (-), suspicious "+/-", Mild (+), Significant (++) and Severe (+++) [9]. The CMT score and somatic cell numbers of 40 milk samples determined by the NucleoCounter™ SCC-100 (Chemometec®, Denmark) device are shown in Table 1.

**Table 1.** CMT Score and Somatic cell counts of milk samples.

Milk samples (n=40)	Milk samples CMT scores	Somatic cell count standards
8	Negative (-)	100
16	Suspicious (+/-)	100-300
14	Mild (+)	300-900
2	Severe (+++)	900-2700

### Identification

Bacterial growth was not observed in 8 samples taken from 4 (20%) female camels. Bacterial growth was detected in 16 (80%) female camel milk samples (n = 32 samples). As a result of identification, 4 (12.5%) *Staphylococcus aureus*, 4 (12.5%) *Staphylococcus auricularis*, 2 (6.25%) *Staphylococcus pettenkoferi*, 2 (6.25%) *Staphylococcus cohnii* spp. *cohnii*, 2 (6.25%) *Staphylococcus equorum*, 2 (6.25%) *Staphylococcus capitis*, 2 (6.25%) *Streptococcus agalactiae*, 2 (6.25%) *Streptococcus dysgalactiae*, 4 (12.5%) *Escherichia coli*, 2 (6.25%) *Pseudomonas pseudalcaligenes*, 2 (6.25%) *Corynebacterium pseudotuberculosis*, 2 (6.25%) *Aerococcus viridans* and 2 (6.25%) *Gemella morbillorum* were identified (Table 2).

### Antibiotic susceptibility

The evaluation of MIC values indicating the antibiotic susceptibility of the strains identified in our study (n = 32) showed that *S. aureus* (n=4) strains were 100% resistant to Clindamycin and Trimethoprim-Sulfomethoxazole, and 75% resistant to Erythromycin, Penicillin G and Vancomycin. *S. auricularis* (n=4) strains were resistant to Daptomycin, Fusidic acid at a rate of 100%, and against Quinupristine dalfopristin and Trimethoprim-Sulfomethoxazole at a rate of 75%. *S. pettenkoferi* (n=2) strains were 100% resistant to Oxacillin and Penicillin G. *S. cohnii*spp. *cohnii* (n=2) strains were 100% resistant to Ampicillin, Clindamycin, Fosfomycin, Oxacillin, Penicillin G and Vancomycin. *S. equorum* (n=2) strains were 100% resistant to Ampicillin, Fusidic acid and Penicillin G. *S. capitis* (n=2) strains were exam-

ined, it was found that the strains were 100% resistant to Vancomycin, Amoxicillin-Clavulanic acid and Ampicillin. *S. agalactiae* (n=2) strains were 100% resistant to Ciprofloxacin, Erythromycin, Fusidic acid, Gentamicin, Penicillin G and Teicoplanin. *S. dysgalactiae* (n=2) strains were 100% resistant to Ampicillin, Cefoxacin, Ciprofloxacin, Daptomycin, Erythromycin, Fusidic Acid, Gentamicin, Nitrofurantoin, Penicillin G, Quinupristine dalfopristin, Rifampin and Teicoplanin. *C. pseudotuberculosis* (n=2) strains were 100% resistant to Ampicillin, Ciprofloxacin, Fusidic acid, Nitrofurantoin and Penicillin G. *A. viridans* (n=2) strains were 100% resistant to Ciprofloxacin, Erythromycin and Gentamicin. *G. morbillorum* (n=2) strains were 100% resistant to Ciprofloxacin, Erythromycin and Gentamicin. *E. coli* (n=4) strains were 100% resistant to Cefotaxime, Ciprofloxacin, Gentamicin, Meropenem, Netilmicin, Tigecycline and Trimethoprim-Sulfomethoxazole. *P. pseudalcaligenes* (n=2) strains were 100% resistant to Amoxicillin-Clavulanic acid, Aztreonam, Cefuroxime and Trimethoprim-Sulfomethoxazole (Table 3)

## Discussion

Subclinical camel mastitis has not been extensively studied and reported in prevalence studies. Subclinical mastitis, where macroscopic clinical symptoms cannot be seen and indirect tools are needed for diagnosis, remains a significant public health problem for camel populations and consumers. Consuming camel milk has effects on animal health and reduces the quantity and quality of milk. In studies on the relationship between the California mastitis test (CMT) and the number of bacteria in the camel milk test, it was shown that a high proportion of subclinical mastitis was CMT positive and that there was a significant difference between positive / negative CMT cases [15-17]. In another study conducted in Sudan, CMT, somatic cell count (SCC) and bacterial isolates were compared and showed that the mean values of CMT and SCC were higher in samples taken from an infected mammary gland [18]. In addition, 47.3% of 336 milk samples tested positive in CMT and somatic cell count of 757 samples were reported to have a count range of  $5 \times 10^5$  to  $7.5 \times 10^6$  cells / ml [19].

CMT has a sensitivity of about 70% and a specificity of 91% in camel mastitis [20]. Additionally, in another study, it was suggested that detection of SCC in camels was more sensitive in detecting subclinical mastitis in camels than the N-acetyl beta-D-glucosaminidase test [21]. The prevalence of subclinical mastitis in camels in Riyadh, Saudi Arabia was 33% in milk samples examined according to CMT [15]. In other studies, the overall prevalence of mastitis was 44.8% and the prevalence of subclinical mastitis was 46% [16,17]. In this study, the CMT test and digital CCS were used to investigate subclinical mastitis in female camels. Milk samples were obtained from 20 camels for the diagnosis of subclinical mastitis and the CMT score was negative, contaminated with +1, +2 and +3. The mean CCS obtained from the assessed healthy camel was 100,000 cells/ml. In our study, no bacterial growth was found in animals with so-

**Table 2.** The distribution of bacteria isolated from samples.

Milk samples with bacterial growth	Bacterial Identification	
	Right lobe	Left lobe
Sample 1	<i>Staphylococcus aureus</i>	<i>Staphylococcus aureus</i>
Sample 2	<i>Staphylococcus aureus</i>	<i>Escherichia coli</i>
Sample 3	<i>Staphylococcus auricularis</i>	<i>Staphylococcus auricularis</i>
Sample 4	<i>Staphylococcus aureus</i>	<i>Staphylococcus auricularis</i>
Sample 5	<i>Escherichia coli</i>	<i>Staphylococcus auricularis</i>
Sample 6	<i>Staphylococcus pettenkoferi</i>	<i>Staphylococcus pettenkoferi</i>
Sample 7	<i>Staphylococcus cohnii</i> spp. <i>cohnii</i>	<i>Staphylococcus cohnii</i> spp. <i>cohnii</i>
Sample 8	<i>Staphylococcus equorum</i>	<i>Staphylococcus equorum</i>
Sample 9	<i>Staphylococcus capitis</i>	<i>Staphylococcus capitis</i>
Sample 10	<i>Streptococcus agalactiae</i>	<i>Streptococcus agalactiae</i>
Sample 11	<i>Streptococcus dysgalactiae</i>	<i>Streptococcus dysgalactiae</i>
Sample 12	<i>Escherichia coli</i>	<i>Escherichia coli</i>
Sample 13	<i>Pseudomonas pseudalcaligenes</i>	<i>Pseudomonas pseudalcaligenes</i>
Sample 14	<i>Corynebacterium pseudotuberculosis</i>	<i>Corynebacterium pseudotuberculosis</i>
Sample 15	<i>Aerococcus viridans</i>	<i>Aerococcus viridans</i>
Sample 16	<i>Gemella morbillorum</i>	<i>Gemella morbillorum</i>

matic cell counts less than 100,000 cells/ml. In our study, the prevalence of subclinical mastitis was determined to be 80% in milk samples randomly selected from healthy-looking camels. However, previous studies have shown that CMT has a sensitivity of 70% and specificity of 91% in camel mastitis and there is a positive correlation between SCC/CMT scores [20-22]. Because early and effective treatment of mastitis is very important, accurate diagnosis of subclinical mastitis is always a priority. The technique in which the intranuclear dye penetrates into the DNA of the cell and the stained cells are detected by spectrophotometer has been shown to be applicable in the diagnosis of subclinical mastitis in camels. This study was confirmed by isolating and identifying the causative agent from milk samples from animals evaluated for subclinical mastitis by determining CMT and SCC. Epidemiological data collected in recent years show that the prevalence of clinical mastitis in camels is between 31-38% in Europe and 31% in Uruguay. While the prevalence of general mastitis in camels was 18.52%, subclinical mastitis was more common (24.7%) than clinical mastitis (11.67%). The major pathogens isolated were *Staphylococcus* sp. (41.67%) followed by *Streptococcus* sp. (21.67%), *Enterobacter* sp. (15.00%), *Corynebacterium pyogenes* (10.00%), *Micrococcus* sp. (5.00%), *Pasteurella* sp. (5.00%) and *Pseudomonas* sp. 1.66%. In North Kordofan, Sudan, in 2013, the incidence rate was found to be 25% subclinical mastitis, 13.3% and 15% using other techniques such as SCC and White Side Test. The isolated pathogens were *Staphylococcus* sp. (80.30%), *Bacillus* sp. (9.09%), *Pasteurella* sp. (6.06%), *Corynebacterium* sp. (3.03%) and *Streptococcus* sp. (1.52%) [23]. *Staphylococcus* sp. As a result of the study conducted on 90 female camels in Jordan to identify and generate data on camel mastitis and pathogens, 21% of the camels had clinical symptoms of mastitis and *Micrococcus* sp., *Staphy-*

*lococcus aureus*, *Streptococcus* sp. and *Corynebacterium* sp. identified. The isolates were susceptible to antibiotics such as Gentamicin, Ampicillin and Tetracycline. The study concluded that Gram-positive cocci are predominantly the result of mastitis [24]. The data obtained are in parallel with the findings of our research. In this study, 81% of Gram-positive bacteria were isolated. *Staphylococcus* sp., *Streptococcus* sp. and *Micrococcus* sp. isolated camels have been reported to be the dominant pathogen of these species [25,26].

The incidence and causes of mastitis in camels differ significantly due to geographical region and individual herd management [17]. The prevalence of camel mastitis in Sudan was 30.2%, of which 4.9% were clinically and 25.3% subclinically in the Jiggiga region. There was a high rate of chronic form (72.41%) followed by acute form (24.14%), and three types of clinical mastitis were diagnosed, the least seen gangrenous form (3.45%). Clinical mastitis occurred in animals older than 10 years of age and late in lactation (55%). In our study, subclinical mastitis cases were detected in animals younger than 10 years of age. The dominant isolated microorganisms are *Staphylococcus* sp. (37.8%), *E. coli* (18.9%), *Streptococcus* sp. (13.5%), *Bacillus* sp. (10.8%), *Micrococcus* sp. (8.1%), *Corynebacterium* sp. (5.4%) and *Salmonella* sp. (5.4%) [27]. Of the mastitis (76.0%) evaluated in Eastern Ethiopia and observed in camel herds, *Staphylococcus aureus* (4.2%), *Coagulase Negative Staphylococci* (39.6%), *Streptococcus agalactiae* (3.5%), *Streptococcus dysgalactiae* (22.2%), *Corynebacterium* sp. (9%), *Bacillus* sp. (7.6%), *Streptococcus uberis* (7.6%), *Escherichia coli* (6.37%) were identified [28]. Similar to these findings, *Staphylococcus* sp., *Streptococcus* sp., *E. coli*, *Corynebacterium pseudotuberculosis* and *Pseudomonas pseudalcaligenes* species were identified in addition to *A. viridans* and *Gemella morbillorum*.

**Table 3.** Antibacterial susceptibility results of identified bacterial strains according to MIC values. AMC: Amoxicillin-Clavulanic acid, AM: Ampicillin, FOX: Cefoxitin, CIP: Ciprofloxacin, CC: Clindamycin, DAP: Daptomycin, E: Erythromycin, FF: Fosfomycin, FA: Fusidic acid, GM: Gentamycin, LVX: Levofloxacin, LZD: Linezolid, FM: Nitrofurantoin, OX: Oxacillin, P: Penicillin G, SYN: Quinupristin-dalfopristin, RA: Rifampin, TEC: Teicoplanin, TE: Tetracycline, NN: Tobramycin, SXT: Trimethoprim-Sulfomethoxazole, VA: Vancomycin, AN: Amikacin, ATM: Aztreonam, FEP: Cefepime, CAZ: Ceftazidime, CRO: Ceftriaxone, CXM: Cefuroxime, CL: Colistin, ETP: Ertapenem, IPM: Imipenem, MEM: Meropenem, NET: Netilmicin, PIP: Piperacillin, TZP: Piperacillin-Tazobactam, TGC: Tigecycline.

	AMC		AM		FOX		CIP		CC		DAP		E		FF		FA		GM		LVX		LZD		
	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	
<i>Staph. aureus</i> (n=4)	2	1	1	1	1	2	2	2	1	2	1		4	4	1	3	1	3	2	2	2	2	3	1	4
<i>Staph. auricularis</i> (n=4)	3		1	4	4		3	1	3	1		4	3	1	4			4	4		3	1	2	2	
<i>Staph. pettenkoferi</i> (n=2)	2				2	2		2		2	2				2	2		2		2		2		2	
<i>Staph. cohnii</i> spp. <i>cohnii</i> (n=2)		2			2	1	1	2		2	2		2			2	2		2		2		2	2	
<i>Staph. equorum</i> (n=2)	2				2	2		2		2	2				2	2		2	2		2		2	2	
<i>Staph. capitis</i> (n=2)			2		2	2	2		2		2			2		2		2		2		2		2	
<i>Str. agalactiae</i> (n=2)	2			2	2			2	2		2			2	2		2		2	2		2		2	
<i>Str. dysgalactiae</i> (n=2)	1	1		2		2		2	2		2			2	2		2		2	2		2		2	
<i>C. pseudotuberculosis</i> (n=2)	2			2	2		2	2		2	2			2		2	2		2	2		2		2	
<i>A. viridans</i> (n=2)	2			2	2		2	2		2	2			2	2		2		2	2		2		2	
<i>G. morbillorum</i> (n=2)				2		2		2	2		2			2	2		2		2	2		2		2	
	AN		AMC		AM		ATM		FEP		CAZ		CRO		CXM		CIP		CL		ETP		GM		
	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	
<i>E. coli</i> (n=4)	3	1	1	3	4	4	4	4	4	4	4	4	4	4	4	2	2	3	1					4	
<i>P. pseudocaligenes</i> (n=2)	2			2	2		2	2		2	2		2		2	2		2		2		2		2	
	FM		OX		P		SYN		RA		TEC		TE		TGC		NN		SXT		VA				
	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	
<i>Staph. aureus</i> (n=4)	4		2	2	1	3	3	1	4	3	1	4	4	4	4		4		4	1	3				
<i>Staph. auricularis</i> (n=4)	3	1	2	1	1	3	1	1	3	3	1	4	4	4	4	4	1	3	4						
<i>Staph. pettenkoferi</i> (n=2)	2			2	2	1	1	2	2	2	2	2	2	2	2	2	2	2		2					
<i>Staph. cohnii</i> spp. <i>cohnii</i> (n=2)	2			2	2	2	2	2	2	2	2	2	2	2	2	2	2	2		2					
<i>Staph. equorum</i> (n=2)	2	1	1	2	2		1	1	2	2	2	2	2	2	2	2	2	2		2					
<i>Staph. capitis</i> (n=2)	2			2	2		2	2	2	2	2	2	2	2	2	2	2	2		2					
<i>Str. agalactiae</i> (n=2)	2		2		2	2		2		2	2		2	2		2		2		2					
<i>Str. dysgalactiae</i> (n=2)		2	2		2	2		2	2	2	2		2	2		2		2		2					
<i>C. pseudotuberculosis</i> (n=2)		2	2		2	2		2		2	2		2	2		2		2		2					
<i>A. viridans</i> (n=2)	2		2		2	2		2	2	2	2		2	2		2		2		2					
<i>G. morbillorum</i> (n=2)	2			2	2	2		2	2	2	2		2	2		2		2		2					
	IPM		MEM		NET		PIP		TZP		TGC		SXT												
	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R										
<i>E. coli</i> (n=4)	1	3		4	4	4	4	4	4	4	4	4	4	4											
<i>P. pseudocaligenes</i> (n=2)	2		2	2	2	2	2	2	2	2	2	2	2	2											

Besides, 23.8% of Gram-negative bacteria species have been reported in previous studies [29]. In our study, Gram negative species were determined as *Pseudomonas pseudocaligenes*, especially *E. coli* as a cause of circumferential mastitis. In our study, 12.5% of *Staphylococcus aureus* were isolated. *S. auricularis*, *S. pettenkoferi*, *S. cohnii* spp. *cohnii*, *S. equorum*, *S. capitis* were other Staphylococcal species isolated in this research. Our result (12.5%) was found to be lower than the results reported before [30] (44.82%). In the same study, *E. coli* (18.92%) was the second most isolated bacterium. In our study, *E. coli* was identified at a rate of 12.5%.

Previous antimicrobial susceptibility tests showed high susceptibility to commonly used antibiotics against bacteria. The

isolates were found to be highly sensitive to Gentamicin, Ciprofloxacin, Cloxacillin and Amikacin, moderately sensitive to Ampicillin / Sulbactam and Trimoxazole, and highly resistant to Tetracycline and Chloramphenicol [16, 29, 30, 31, 32].

In the study conducted by Abdelgadir [25], Oxytetracycline, Tetracycline and Chloramphenicol were determined as effective antibiotics against mastitis pathogens in camels. According to the antibiotic susceptibility findings obtained in our study, Staphylococcus species were found to be susceptible to Daptomycin, Tetracycline, Levofloxacin, Tigecycline and Tetracycline antibiotics and Trimetoprim-Sulfomethoxazole and Penicillin G resistant. As an important result of antibiotic susceptibility findings, Vancomycin resistance in *S. aureus* and *Staph. cohnii* spp.

*cohnii* strains is remarkable. *Streptococcus* species identified in our study were found to be susceptible to Clindamycin, Levofloxacin, Linezolid and Tetracycline and resistant to Penicillin G, Gentamicin, Erythromycin and Ciprofloxacin. Resistance to beta lactam group and macrolides was determined. Other Gram-positive strains identified as *C. pseudotuberculosis*, *A. viridans* and *G. morbillorum* were also susceptible to Amoxicillin-Clavulanic acid, Levofloxacin and Daptomycin, and resistant to Ampicillin and Penicillin G.

Antimicrobial susceptibility of *E. coli* strains was found to be susceptible to Aztreonam, Cefepime and Piperacillin, and resistant to Gentamycin, Meropenem, Tigecycline, Trimethoprim-Sulfometoxazole and Amoxicillin Clavulanic acid; *Pseudomonas pseudocaligenes* strains were susceptible to Cefepime, Ciprofloxacin, Piperacillin.

Gram-positive bacteria identified in our study were found to be susceptible to Levofloxacin, Linezolid, Tetracycline, Daptomycin, and resistant to beta lactam group antibiotics and macrolides, Vancomycin resistance was determined in *S. aureus* and *S. cohnii* spp. *cohnii* strains. Gram negative strains were generally susceptible to Cefepime and Piperacillin; resistant to Trimethoprim-Sulphomethoxazole and Amoxicillin-Clavulanic acid. As a result, it has been shown that multiple antibiotic resistance develops in bacteria causing mastitis in camels.

Our study suggests that subclinical mastitis in camels is more common than other forms of mastitis and that infected animals may be a source of contamination as a microorganism reservoir; identification of both infectious and environmental mastitis pathogens. Since the prevalence of mastitis in camels differs considerably depending on geographical area and individual herd management, it is recommended to use antibiotics to prevent the development of antimicrobial resistance as well as mastitis control methods; such as elimination of existing infection, prevention of new infection and monitoring the health status of the mammary. Considering that camel milk is used raw for human consumption without heat treatment in order not to affect the nutritional and immunological factors it contains, the pathogens causing subclinical mastitis can easily threaten public health, and it is essential to inform the farmers and consumers about the related pathogens.

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