



Preputial Bacterial Flora and antibacterial Susceptibility in Camels in Maiduguri, North-eastern, Nigeria

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ABSTRACT

This study aimed to investigate preputial aerobic bacterial flora from Camels in the arid zone of Northern Nigeria. Preputial swabs were randomly collected from twenty (20) clinically healthy adult Camels (*Camelus dromedarius*). Using standard bacteriological techniques, the following bacteria were identified; *Escherichia coli* (22.9%), *Streptococcus* species (22.9%), *Staphylococcus aureus* (21.4%), *Klebsiella* Species (17.1%) and *Shigella* (15.7%). These isolated bacteria were found to be susceptible to Amoxicillin, Ampiclox, Ciprofloxacin, Gentamycin and Levofloxacin. However, *Klebsiella* and *Shigella* species showed intermediate susceptibility to streptomycin and *Shigella* further showed resistance to chloramphenicol. The results of this study showed that some bacteria colonized the prepuce of camel and there is a need for preputial washing before breeding to reduce the transmission of venereal disease. Amoxicillin, Ampiclox, Ciprofloxacin, Gentamycin and Levofloxacin could be used as therapeutic agents for venereal diseases that could be caused by the aforementioned bacteria in Camelids.

KeyWords: Anti-microbial resistance, camel, infertility, prepuce

INTRODUCTION

Camels are an important animal resource in many parts of the world (Shokri *et al.*, 2010). In northern Nigeria, they are usually seen herded with other ruminants mingling freely on pasture, at watering points and marketplaces (Mshelia *et al.*, 2012). They are less affected by heat stress, poor feed or lack of water and are generally believed to be a better source of meat, milk and leather than cattle (Srikandakumar *et al.*, 2001; Peter *et al.*, 2015; Qamar, 2018; Purohit *et al.*, 2023). Camels are hardy animals and quite resistant to common ruminant diseases (Jenberie *et al.*, 2012), however, poor reproductive performance has been identified as a major problem in Camelids with pregnancy losses and infertility as the major problems (Tibary *et al.*, 2006; Shokri *et al.*, 2010; Mshelia *et al.*, 2013). Preputial microflora has been extensively studied in other species of animals such as Bulls, Rams, and Horses (Humphrey *et al.*, 1982; Hurtgen, 1983; Gouletsou *et al.*, 2006) but scanty information exists on the Camel bull. In camels, the prepuce is flattened and appears triangular when viewed laterally. The preputial opening is directed caudally when there is no sexual arousal. However, with penile erection, the cranial preputial muscles

pull both penis and prepuce cranially (Serin *et al.*, 2010). During coitus, the penis penetrates the cervical canal and enters deep into the uterine cavity (Tibary & Vaughan 2006). The prepuce has earlier been demonstrated to act as a reservoir of microorganisms (Agartan *et al.*, 2005). Notable sources of these preputial microorganisms are the soil, faeces and the female genital tract (Waheed *et al.*, 2022). Some of these microorganisms can be pathogenic and therefore might cause infertility thus allowing the passing of venereal infections from infected bulls to she-camels and vice versa. An infected bull camel usually does not show clinical signs but infection is traced back through symptoms shown by she-camels he had mated (AL-Qarawi, 2005).

To the best of our knowledge, there is little data on preputial bacterial flora in Camels in north-eastern, Nigeria. This study aimed to determine the type of bacteria that colonize the prepuce of Camel and to study the susceptibility of the isolated bacteria to some antibiotics.

MATERIALS AND METHODS

STUDY AREA AND ANIMALS

The study was conducted between October and December 2021 in Maiduguri (Maiduguri Metropolitan abattoir) using the facilities of the Microbiology Laboratory, Faculty of Veterinary Medicine, University of Maiduguri. Clinically healthy male Camels (n=20) aged between 8 and 12 years and between 350-500 Kg/bw were randomly selected for this study. These animals are used as trade animals and brought for slaughter from the surrounding States of Yobe and Adamawa States as well as neighbouring countries of Niger and Chad.

SAMPLE COLLECTION AND IDENTIFICATION OF BACTERIA

The Camels were restrained using ropes and turned to lateral recumbency. Preputial swabs were randomly collected from the Camels under strict aseptic conditions using sterile swab sticks (Evepon Sterile Swab Stick ®, Evepon Industries Limited, Anambra, Nigeria). The samples were transported to the laboratory on ice within 30 minutes of collection.

ISOLATION AND IDENTIFICATION

The preputial swab samples were initially inoculated on an enrichment media (nutrient broth and peptone water). A loop was taken from this media and streaked on Blood agar, Eosin Methylene Blue, Mannitol Salt Agar, Salmonella Shigella Agar, McConkey Agar. These were incubated at 37°C for 24h aerobically. Growing colonies were examined with gram staining techniques. Suspicious colonies were identified using biochemical tests (Lassen, 1975, Holt *et al.*, 1994).

ANTIMICROBIAL SUSCEPTIBILITY TEST

The antibiotic susceptibility test for the identified bacteria were applied with multi discs containing Amoxycillin (30 µg), Ampicillin (30 µg), Levofloxacin (10 µg), Chloramphenicol (20 µg), Ciprofloxacin (10 µg), Gentamycin (10 µg) and Streptomycin (30 µg). The antimicrobial susceptibility test was performed according to the Kirby Bauer disc diffusion method (Bauer *et al.*, 1966).

DATA ANALYSIS

All data generated were compiled and managed in Microsoft excel 2011 spreadsheet and presented in tables using descriptive statistics.

RESULTS

In this study, no gross preputial abnormality was observed in any of the sampled from Camel bulls. Cultures of preputial swabs from the twenty camels yield 70 bacterial isolates. The number of isolates/males ranged from two (2) to five (5) with means of 3.5 per male. Five (5) distinct bacterial species were identified, out of which two were gram-positive

(40%) while three (60%) were gram-negative. They were *E. coli* (22.9%), *Streptococcus* species (22.9%), *Staphylococcus aureus* (21.4%), *Klebsiella* Species (17.1%) and *Shigella* (15.7%) as shown in Table I. Antibiotic susceptibility test results presented in table Table II shows that *E. coli*, *Streptococcus* and *Staphylococcus aureus* were susceptible to Amoxicillin, Ampiclox, Chloramphenicol, Ciprofloxacin, Gentamycin, Levofloxacin and streptomycin. Isolated *Klebsiella* species was found to be susceptible to Amoxicillin, Ampiclox, Chloramphenicol, Ciprofloxacin, Gentamycin and Levofloxacin but had intermediate susceptibility to Streptomycin. The isolated *Shigella* species were susceptible to Amoxicillin, Ampiclox, Ciprofloxacin, Gentamycin and Levofloxacin but had intermediate susceptibility to Streptomycin and resistant to Chloramphenicol.

Table I. Table I: Bacteria isolated from prepuce of Camel in Maiduguri, north-eastern Nigeria.

Bacteria	Number of isolates n (%)
<i>Escherichia coli</i>	16 (22.9)
<i>Streptococcus</i>	16 (22.9)
<i>Staphylococcus aureus</i>	15 (21.4)
<i>Klebsiella spp</i>	12 (17.1)
<i>Shigella spp</i>	11 (15.7)
Total	70 (100)

DISCUSSION

Escherichia coli, *Streptococcus* spp, *Staphylococcus aureus*, *Klebsiella* spp and *Shigella* Spp were found to be common preputial bacteria in all the Camels samples (Table I). Gross preputial abnormality was not observed in any of the animals sampled for this study. This is expected as only apparently healthy Camels were selected for this study. Published data on the presence and distribution of bacteria in the prepuce of Camels are scarce. However, the results of this study agree with Serin *et al.*, (2010), with isolation of similar bacterial species and others from the prepuce of camels in Turkey. The results of this study further agree with a recent study by Waheed *et al.* (2022) who reported *Staphylococcus aureus*, *Streptococcus* and *Escherichia coli* as some of the commonly occurring bacteria in Camel prepuce in Saudi Arabia. *Streptococcus*, *Staphylococcus aureus* appear to be common bacteria isolated from prepuce of Camels irrespective of geographical location. The bacteria isolated from the prepuce of Camels in this study may or may not be necessarily associated with any type of an active disease process. However, these bacterial agents could easily

Table II: Antibiotic susceptibility of bacteria isolated from prepuce of camels in Maiduguri, North-eastern Nigeria.

Isolated bacteria	Antibacterial agent																				
	Amoxicillin			Ampiclox			Chloramphenicol			Ciprofloxacin	Gentamycin			Levofloxacin			Streptomycin				
	R	I	S	R	I	S	R	I	S		R	I	S	R	I	S	R	I	S		
<i>Escherichia coli</i>	-	-	16	-	-	16	-	-	16	-	-	16	-	-	16	-	-	16	-	-	16
<i>Streptococcus</i>	-	-	16	-	-	16	-	-	16	-	-	16	-	-	16	-	-	16	-	-	16
<i>Staphylococcus aureus</i>	-	-	15	-	-	15	-	-	15	-	-	15	-	-	15	-	-	15	-	-	15
<i>Klebsiella</i>	-	-	12	-	-	12	-	-	12	-	-	12	-	-	12	-	-	12	-	12	-
<i>Shigella</i>	-	-	11	-	-	11	11	-	-	-	-	11	-	-	11	-	-	11	-	11	-

S= Sensitive, I= Intermediate, R= Resistant

contaminate semen either during natural mating or collection for assisted reproduction. Bacteria contamination of semen leads to a sequence of changes that may include reduced sperm motility, sperm clumping, an increased proportion of altered acrosome and changes in semen pH (Orgtega-Ferrusola *et al.*, 2009; Ghoneim *et al.*, 2014). Furthermore, these bacteria have been incriminated as the cause of infertility in female Camels following their frequent isolation from the uterus of infertile camels (Wernery and Wernery, 1992; Ali *et al.*, 2010; Almohasen, 2011; Ghoneim *et al.*, 2014). For example, Umaru *et al.* (1999) isolated *Staphylococcus aureus*, *Streptococcus pyogenes*, *Escherichia coli* and *Klebsiella spp* from the uterus of infected uteri of Camels. These bacteria are also the same type of bacteria that were isolated from prepuce of Camels in this present study. This could mean that these bacteria exist in Camel population and may or may not affect reproduction in this species.

Bacteria species that colonize the prepuce such as those isolated from the prepuce of Camels in this study are generally commensals, but a host of them are potentially opportunistic pathogens and have the ability to cause venereal diseases (Tibary & Anouassi, 2001). Venereal diseases are mainly spread through coitus from an infected male mating with a susceptible female and this could impair their fertility. An uninfected male can acquire the infection after mating with an infected female and begins to spread the infection to susceptible females within a short period of time. In an unusual case, the use of infected semen and a failure to test such semen for venereal disease could lead to transmission of venereal disease during artificial insemination among animals. Immuno-competent female animals clear such infections through natural uterine defence mechanisms. However, in a proportion of females, the uterine defence mechanism may fail (partially or completely) thereby allowing the establishment of an infection (Tibary *et al.*, 2006), which may lead to infertility or sterility.

The *in-vitro* antibiotic sensitivity using some selected antibiotics indicated that the bacteria isolated in this study were found to be susceptible to Amoxicillin, Ampiclox, Ciprofloxacin, Gentamycin and Levofloxacin. However,

Klebsiella and *Shigella* species showed intermediate susceptibility to streptomycin and *Shigella* further showed resistance to chloramphenicol (Table II). Mshelia *et al.* (2014) have shown that Gentamycin was highly effective against genital *Staphylococcus aureus* and *Escherichia coli* isolated from ewes within the same study area. Ciprofloxacin was also reported to be effective against all bacteria that are associated with caprine vulvovaginitis (Ajala *et al.*, 2011) as well as from the vagina of cows and ewes (Martins *et al.*, 2009; Gongcuoglu *et al.*, 2012). *Shigella* spp isolated in this study showed resistance to Chloramphenicol and while *Klebsiella* and *Shigella* spp showed intermediate resistance to Streptomycin in this study. This finding suggests that Chloramphenicol and Streptomycin should not be used in treating venereal diseases in Camels. Antimicrobial agents have been used to successfully reverse the infections caused by microbes. Nonetheless, microbes have also developed resistance to antimicrobial agents (Chander & Raza, 2013). This resistance could have been developed through the acquisition of resistant genes and or modifying enzymes (Poole, 2005; Economou & Ghuosia, 2015) and could be a problem while addressing microbial agents with antimicrobials in animals and humans as well.

CONCLUSION

This study shows that *Escherichia coli*, *Streptococcus* species, *Staphylococcus aureus*, *Klebsiella* Species and *Shigella* are frequently isolated from the prepuce of Camels. These bacteria are a potential source of infection and could lead to infertility in Camel bulls. More detailed studies with larger sample size are required to elucidate the diversity of bacterial species that colonize the prepuce in livestock in this geo-location. Furthermore, studies will be required on these individual bacteria to understand their molecular characteristics and their inherent potential in causing gross and or microscopic pathologies of the reproductive tract in camel bulls in Maiduguri, Northeastern Nigeria. Preputial washing is strongly recommended before breeding in Camels in Maiduguri, North eastern Nigeria.

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CONFLICT OF INTEREST

The authors have none to declare.

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