

STAPHYLOCOCCAL SPECIES RESPONSIBLE FOR BOVINE MASTITIS IN KHARTOUM STATE, SUDAN

Reem Rabie Mohammed Salih ¹and A/ Basit Osman Ahmed

Department of Clinical Medicine, Faculty of Veterinary Medicine, University of
Khartoum.

المستخلص

هذه الدراسة أجريت في مناطق معينة من ولاية الخرطوم تشمل مناطق (التبنة، فلسطين، شمبات، حلة كوكو، الحلفايا، السامراب ومزرعة جامعة الخرطوم)، وذلك خلال الفترة من مايو 2006 وحتى 2008 وذلك لتحديد المكورات العنقودية التي تسبب التهاب الضرع في الأبقار. العدد الكلي للأبقار الحليب التي تم اختيارها في عدد 34 مزرعة تم اختيارها بلغ عدد 500 حيوان. الأبقار المصابة بالتهاب الضرع وجدت في كل المزارع التي تم إستقصائها. النسبة المئوية لالتهاب الضرع الحاد الذي تسببه المكورات العنقودية الذهبية والمكورات العنقودية الخنزيرية بلغة 24% في حين أن النسبة المئوية لالتهاب الضرع المزمن الذي تسببه المكورات العنقودية الذهبية بلغت 44% والذي تسببه المكورات العنقودية الخنزيرية بلغت 8%

Abstract

This study was conducted in certain area at Khartoum State (Eltebna, Falasteen, Shambat, Hilat Kuku, Elhalfaia, Elsamrab and The University of Khartoum farms) from May/2006 to April/2008 to determine the type of *Staphylococcus* spp which cause bovine mastitis. The total number of dairy cows, which were examined in 34 investigated farms, amounted to 500 animals. The infected cows with mastitis were found in all investigated farms. The percentages of acute mastitis caused by *Staph aureus* and *Staph hyicus* amounted to 24% and the percentage of chronic mastitis caused by *Staph aureus* was 44% and that caused by *Staph hyicus* was 8%.

Keywords: *Staph, cattle, mastitis*

Introduction

Bovine mastitis is caused mainly by certain *Staphylococcus* and *Streptococcus* species. These include: *Staphylococcus aureus*, *Staph. chromogenes*, *Staph. epidermidis*, *Staph. hyicus*, *Staph. simulans*, *Staph. xylosus*, *Streptococcus agalactiae*, *Strep. dysgalactiae* and *Strep. Uberis* (Forsman, 1997). One of the most common

¹ P.O. Box 32, Khartoum North, Sudan, E-mail: reemat7@yahoo.com

types of chronic mastitis is caused by the bacteria *Staphylococcus aureus* which is often significant subclinical and extremely difficult to control by treatment alone (Radostitis *et al.*, 2000). Successful control is gained only through prevention of new infections and cow culling (Roberson *et al.*, 1994). *Staph. aureus* colonizes abnormal teat ends or teat lesions. Milkers' hands, wash cloths, teat cup liners, and flies are ways in which the infection can be spread from cow to cow. The organisms probably penetrate the teat canal during milking. Irregular vacuum fluctuations impact milk droplets and bacteria against the teat end with sufficient force to cause teat canal penetration and possible development of new infection. *Staphylococcus hyicus* was isolated from mastitic cows from acute and chronic mastitis (Miedzobrodzki, *et al.*, 1989).

Clinical mastitis may flare-up in some cows, especially after calving. The *Staph* spp persist in mammary glands, teat canals, and teat lesions and are considered contagious. In some herds with somatic cell counts (SCC) below 200.000, dairy managers have not been able to eradicate *Staph. aureus*, even when they practiced standard milking time hygiene techniques (Roberson *et al.*, 1994). Many remain infected throughout the first lactation and are reservoirs for infecting other cows in the herd. Although as many as half of the cows with high (SCC) may be infected with *Staph. aureus*, somatic cell counts alone are not sensitive enough to positively diagnose *Staph. aureus* infection.

The aim of this work was to detect the types of *Staph* spp that causing bovine mastitis.

Materials and Methods

This study was conducted in certain area at Khartoum State (Eltebna, Falasteen, Shambat, Hilat Kuku, Elhalfaia, Elsamrab and The University of Khartoum farms) from May/2006 to April/2008 to determine the type of *Staphylococcus* spp which cause bovine mastitis. The total number of dairy cows, which were examined in 34 investigated farms, amounted to 500 animals.

The milk pH was determined by using of indicator paper made for determine pH (manufactured by Kruse Company in Denmark) before collection of milk samples. The test was applied by adding one drop of milk on yellow spot and observed for the change in colour.

Milk samples were collected under strict aseptic condition as stated by Barrow and Feltham (2003). Samples were immediately frozen in ice and submitted for bacteriological examinations within 24 hours. A sterile bacteriological loop was used

to spread each milk sample into a freshly prepared Blood agar plate (Oxoid) with sheep red-blood cells, and into MacConkey's agar (Oxoid).

The Primary tests were used for isolation and identification included: Gram's stain, motility, oxidase, catalase, oxidation fermentation and glucose fermentation and the secondary tests were production of acids from (lactose, maltose, mannitol, fructose, sucrose, trehalose, xylose, cellobiose, raffinose and mannose), phosphatase test, nitrate utilization, urea, protease and novobiocin test. (Barrow and Feltham, 2003).

Results

All samples were collected gave a positive reaction for pH test, positive samples for mastitis showed change in colour from yellow to bluish green, while negative sample showed no changes or change to yellowish green. Were shown in Table (1) *Staph aureus* and *Staph hyicus* were identified. Table (2) shows the species of *Staph* identified, number of infected quarters and type of inflammation. It is clear that only *Staph aureus* and *Staph hyicus* were isolated from acute and chronic mastitis in all areas of study except in Falasteen where *Staph aureus* only was isolated from chronic mastitis. Also one or more quarters were affected.

According to coagulase test and Christie Atkins Munch Petersen (CAMP) reaction together with primary and secondary tests 16 milk samples yielded *Staph aureus* and 8 samples yielded *Staph hyicus*. Figure (1) shows the percentage of *Staph* spp. and the type of mastitis caused by the isolated *Staph* spp. Both *Staph aureus* and *Staph hyicus* caused 24% of acute mastitis, but *Staph aureus* recorded the high percentage of chronic mastitis 44%.

Table (1): Tests used for identification of *Staphylococcus* species

Test	<i>Staph. aureus</i>	<i>Staph. hyicus</i>
Gram's stain	+	+
CAMP	+	+
Growth anaerobically	+	+
Oxidase	-	-
V.P	+	-
Coagulase	+	+
Lactose	+	+
Maltose	+	-
Mannitol	+	-
Fructose	+	+
Sucrose	+	+
Trehalose	+	+
Xylose	-	-
Cellobiose	-	-
Raffinose	-	-
Mannose	+	+
Phosphatase	+	+
Nitrate	+	+
Urea	+	+
Protease	+	+
Novobiocin	+	+

Table (2): The type of inflammation, spp. of Staph, number of infected quarters and places of milk samples collection

<i>Location</i>	<i>Isolated bacteria</i>	<i>Number of infected quarters</i>	<i>Type of inflammation</i>
Eltebna	<i>Staph hyicus</i>	1	Acute
	<i>Staph. aureus</i>	2	Acute
	<i>Staph. Aureus</i>	1	Acute
Falasteen	<i>Staph. aureus</i>	1	Chronic
	<i>Staph. aureus</i>	1	Chronic
Elhalfaia	2: <i>Staph. aureus</i>	4	Chronic
	<i>Staph.hyicus.</i>	4	2:chronic and 2:acute
	<i>Staph.hyicus</i>	4	Chronic
	<i>Staph.aureus</i>	2	Acute
Elsamrab	3: <i>Staph.hyicus</i>	4	Acute
	1: <i>Staph.aureus</i>	4	Chronic
	<i>Staph.aureus</i>	4	Acute
University of Khartoum	<i>Staph.aureus</i>	2	Chronic
	<i>Staph. aureus</i>	1	Chronic
	<i>Staph. aureus</i>	1	Chronic
	<i>Staph. aureus</i>	1	Chronic
	<i>Staph. aureus</i>	1	Chronic
	<i>Staph.aureus</i>	1	Chronic
	<i>Staph.aureus, Staph.hyicus</i>	4	Acute
	<i>Staph.hyicus</i>	4	Acute

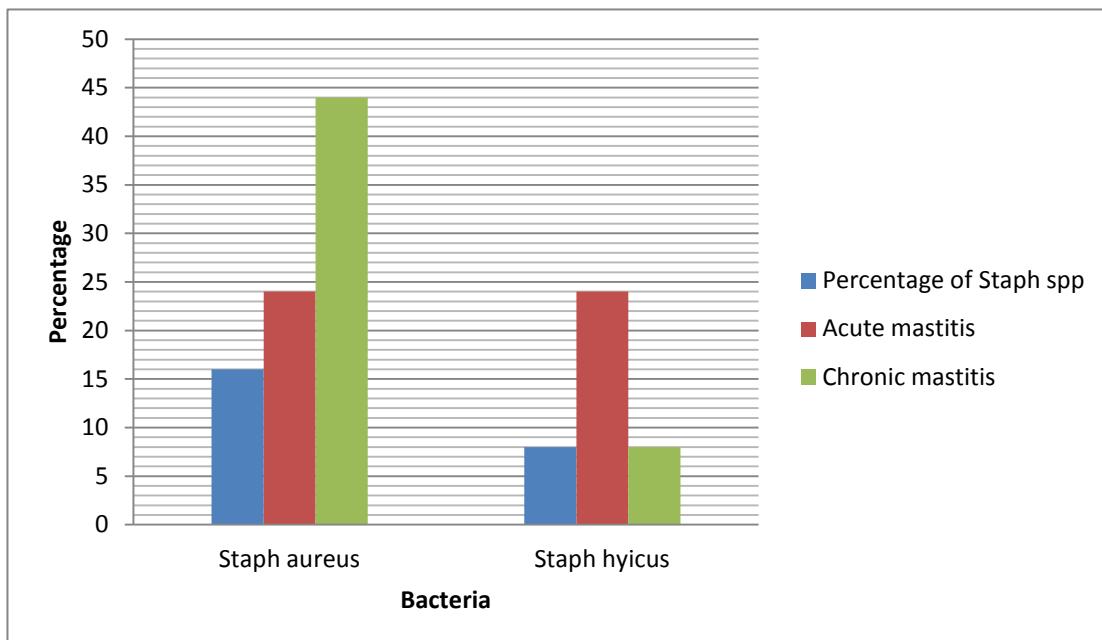


Figure (1): The percentage of Staph spp. and the type of mastitis caused by the isolated Staph spp.

Discussion

Mastitis is an inflammation of the mammary glands regardless of the cause (Blood *et al.*, 1983). It plays a very important role in human health and animal (Kromber and Grabowski, 2002). In this study the types of *Staph* spp. isolated from acute and chronic mastitis were *Staph aureus* and *Staph hyicus*. This agrees with the finding of DaRong *et al.*, (2010) and Jan *et al.*, (1998). Radostits *et al.*, (1994) mentioned that *Staph aureus* is the first microorganism incriminated in bovine mastitis. A predominance of *Staph aureus* mastitis in cows has been reported by Watts, (1988), Falade *et al.*, (1989) and Carlos (1990).

Elsayed, (2000) isolated *Staph aureus* and *Staph hyicus* from 499 milk samples from different domestic animals: cows, sheep, goat and camels. AlAyies, (2004) isolated *Staph aureus* (73.7%) and *Staph hyicus* (6%) from 100 bovine mastitic milk samples.

On conclusion this study confirmed the role of *Staphylococcus* spp. as a cause of mastitis in bovine. The percentage of incidence of bovine mastitis was high according to our findings after examinations.

References

AlAyis; A. A. M (2004). Studies on Staphylococci associated with bovine mastitis in Khartoum State. (M.V.SC). Thesis, University of Khartoum.

Barrow; G. I. and Feltham; R. K. A (2003). Cowan and Steel's manual for identification of medical bacteria. 3rd (ed) Cambridge press.

Blood; D. C, Radostits; O. M and Henderson; J. B (1983). Veterinary Medicine, 6th ed. Bailliere Tindal, London. pp. 451.

Carlos; A. D. M (1990). Characteristic of *Staphylococcus aureus* from subclinical bovine mastitis in Brazil. Br. Vet. J. **146**: pp. 443-448.

DaRong; C¹, Shan; Y. Z², Zhao; H. Y¹, Wen; W. D³, Zhi; X. M¹, Zhi; R. S¹ and Huai; C. S¹ (2010). Prevalence of bacterial infection responsible for bovine Mastitis. African Journal of Microbiology Research Vol. **4** (11). pp. 1110-1116.

Elsayed; N. I (2000). Staphylococcal species in normal and mastitic milk of some domestic animals. (M.V.Sc). Thesis, University of Khartoum.

Falade, S.; Nwanaza, L. and Wulaya, A (1989). The incidence of bovine mastitis in Kenya. Bull. Anim. Hlth. Prod. Africa. **26**: pp. 55-61.

Forsman; P, Tilsala-Timisjarvi; A and Alatossava; T (1997). Identification of staphylococcal and streptococcal causes of bovine mastitis using 16S-23S rRNA spacer regions. Microbiology, (143): PP: 3491-3500, Copyright © 1997 by Society for General Microbiology.

Jan; M, Sargeant; Morgan; S, Ken; E. L, Mary; J. I and Anna; B (1998). Clinical mastitis in dairy cattle in Ontario: Frequency of occurrence and bacteriological isolates. *Can Vet J* Volume **39**.

Kromber; V and Grabowski; N. T (2002). Risk factor analysis for mastitis caused by environmental pathogens in the environment of dairy herds. Abstract-xxii, world Buiatrics Congress, Germany [www.ncbi.nlm.nih.gov\Pubmet](http://www.ncbi.nlm.nih.gov/Pubmed).

Miedzobrodzki ; J, Naidu; A. S, Watts, J. L, Ciborowski ; P, Palm; K and Wadstorm; T (1989). Effect of Milk on Fibronectin and Collagen Type I Binding to *Staphylococcus aureus* and Coagulase-Negative *Staphylococci* Isolated from Bovine Mastitis. *Journal of Clinical Microbiology*. pp. 540-544.

Radostits; O. M, Blood; D. C and Gat (1994). Veterinary Medicine. 12th ed. Balliere. Tindall. London. pp. 510-560.

Radostits; O. M, Gay; C. C, Blood; D. C. and Hinch cliff, K. W (2000). Mastitis in Veterinary Medicine 9th ed. W. B. Saunders, London. pp. 690-720.

Roberson; J. R, Fox; L. K, Hancock; D. D, Gay; J. M. and Besser, T. E (1994). Ecology of *Staphylococcus aureus* isolated from various sites on dairy farms. *J. Dairy Sci.* (77). pp: 3354.

Watts; J. L (1988). Etiologic agents of bovine mastitis. *Vet. Microbiol.* **16**: pp. 41-66.