

Case Report:

The First Isolation of *Histophilus somni* from the Preputial Cavity of a Normal Bull in the Sudan.

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ملخص البحث

فحصت مائة مسحة من غلفة ثيران سليمه وذلك في محاولة لعزل الباكترية محبة الانسجة ولدم المنومة *Histophilus somni*. عزلت معزوله واحدة فقط من هذه البكتيريا. تم التعرف عليها حسب شكل البكتيريا وخواص المنابت والخواص الكيموحيوية. توضح هذه الدراسة أن هذه البكتيريا يمكن أن تكون جزءا من النبتة الطبيعية لهذا الجزء، كما يمكن لهذا التواجد أن يمثل عنصر عدوى. وكذلك عزلت بعض البكتيريا المصاحبة من نفس الموضع وهي المكورات العنقودية الدقيقة والسبحية وأنواع من العصوية والروتيا.

Summary

One hundred normal bulls' preputial cavity swabs were examined for the presence of *Histophilus somni* (*H. somni*). Only one *H. somni* isolate was isolated. Identification of the isolate was based on morphological, cultural and biochemical characteristics. This study indicated that *H. somni* may normally form part of the normal flora of the bovine prepuce and that may represent a possible source of *H. somni* infection. Other bacteria isolated were; *Staphylococci*, *Micrococci*, *Streptococci*, and *Bacillus*, *Proteus* and *Rothia* species.

Introduction

Thrombotic meningoencephalitis (formally known as throm-boembolic meningoencephalitis) is a fatal septicaemia-like disease of young cattle. The disease is caused by the small pleomorphic Gram-negative coccobacillus *Histophilus somni* (previously known as *Haemophilus somnus*) which is also the aetiological agent of other bovine disease conditions viz. pneumonia, abortion, infertility, arthritis, myocarditis and mastitis (Humphreys *et al*, 1983; Wu *et al.*, 2000; Sykte *et al*, 2001; Inzana *et al*, 2002; Kuckleburg *et al*, 2005).

Isolation of *H. somni* from the healthy or sick bovine reproductive tracts has been reported in the United States (Gossling, 1966), Canada (Humphreys *et al.*, 1982) and Switzerland (Lunginbühl and Küpfer, 1980). In the Sudan, *H. somni*- like organism was isolated by Eltom *et al*, (2003) from a Dairy farm in Khartoum North. *H. somni* was isolated from 77% (24/31) of urogenital tracts of clinically healthy bulls in the U.S.A. (Humphreys *et al.*, 1982); a high prevalence rate of infection was found in young bull's preputial cavity.

The objective of the present study was to investigate the presence of *H. somni* in flora of the healthy bull's preputial cavity.

Materials and Methods

Animals and Sampling:

Preputial swabs were randomly collected from 100 bulls slaughtered at Al-Huda and Ganawa slaughterhouses (Sudan), 30 minutes following slaughter. Swabs were then stored at 4°C till cultured.

Bacteriological examination:

Swabs were remoistened in sterile normal saline, cultured onto Blood Agar and then incubated for 4 days at 37°C in 10% CO₂ using the candle jar to give 2.5% CO₂.

Identification of *H. somni* and other organisms:

Gram's stained smears (Barrow and Feltham, 1993) were examined microscopically. Colonial morphology and cultural characteristics on Blood Agar were studied. Round colonies with a diameter between 0.3 and 3 mm after 48 hours incubation that comprised gram-negative pleomorphic coccobacilli consistent with *H. somni* were compared morphologically and biochemically with the five known isolates of *H. somni*-like organism (2, 10, 12, 13a, 13b) which were previously isolated by Eltom *et al.* (2003). *H. somni* suspect colonies were examined serologically against known hyper immune sera against known isolates using slide agglutination test. Other organisms isolated from the same swabs were biochemically identified up to genus level.

Results

One isolate of *H. somni* was obtained from one of the 100 samples of the prepuces of healthy bulls examined (1%). Colonial morphology of the isolated organism was typical of *H. somni* and was indistinguishable from that of the isolates 2, 10, 12, 13a and 13b previously identified as *H. somnus*-organism. The colony was creamy pink in colour and butyrous in consistency. The isolate was Gram-negative coccobacillus or short rod found singly or in clumps. It gave identical biochemical test reactions when compared with the *H. somni*-like organisms 2, 10, 12, 13a and 13b. The new isolate was named "Hadeel".

Gram-positive organisms isolated from the same prepuces prepare specimens were: *Staphylococcus*, *Micrococcus*, *Bacillus*, *Streptococcus* and *Rothia* genera while the only Gram-negative organism isolated was *Proteus* genus.

Discussion

In this study, *H. somni* was isolated from a bull's prepuce, even in the absence of overt clinico-pathological manifestations, indicating that *H. somni* may form a part of the normal bacterial flora of the bull's preputal cavity.

Only one positive sample was encountered (1%) and this may be attributed to the fact that *H. somni* is a feeble grower which can be easily overgrown by other bacteria (Stephens *et al*, 1981). It is suggested that low isolation rates of *H. somni* might have been due to the insufficiency of microbiological techniques used (Janzen *et al*, 1981).

Identification of our isolate was based on morphological, cultural characteristics and biochemical reactions, which are indistinguishable from those of the five strains of *H. somni*-like organism previously isolated by Eltom *et al* (2003).

Other organisms isolated were mostly Gram-positive bacteria, and only Gram-negative bacterium. It could be concluded that *H. somni* may represent a part of the bull's external genitalia flora. It could constitute a potential source of sexually transmissible genital tract infection. However, the presence of the organism as an environmental contaminant is to be considered.

Acknowledgements

The authors are grateful to the staff members of the Department of Microbiology, Faculty of Veterinary Medicine, University of Khartoum, for their technical help.

References

- Barrow, G. L. and Feltham, R. K. A. (1993). Cowan and Steel's manual for the identification of 3rd edn. Cambridge University press, Cambridge, UK.
- Eltom, K.; Aradaib, I. and El Sanousi, S. M. (2003). *Vet. Archiv.*, **73** (6): 315-321.
- Gossling, J. (1966). *Illinois Vet.*, **9**: 14-18.
- Humphreys, J. D.; Little, P. B.; Stephens, L. R.; Bamum, D. A.; Doig, P. A. and Thorsen, J. (1982). *Am. J. Vet. Res.*, **43**: 791-795.
- Humphreys, J. D.; Little, P. B.; Stephens, L. R.; Bamum, D. .; Doig, P. A. and Thorsen, J. (1982). *Vet. Bull.*, **53**: 987-1003.
- Inzana, T. J.; Glindemann, G. A. D.; Wakarchuk, W. and Howard, M. D. (2002). *Infect Immun.*, **70**: 6512.
- Janzen, E. D.; Cates, W. F.; Barth, A. N.; Echala, L.; Pawlyshyn, V.; Saunders, J. R. and Sbone, A. D. (1981). *Can. Vet. J.*, **22**: 361-262.

- Kuckleburg, C. J.; Sylte, M. J.; Inzana, T. J. Corbeil, L. B.; Darien, B. and Czuprynski, C. J. (2005).** *Microb. Pathol.*, **38**: 23-32.
- Lunginbühl, A. and Küpfer, U. (1980).** *Schweizer Arch. Tierheilk.*, **122**: 27-434.
- Stephens, L. R.; Little, P. B.; Wilkie, B. N. and Bamum, D. A. (1981).** *Am. J. Vet. Res.*, **42**: 468-473.
- Sykte, M.; Corbel, L. B.; Inzana, T. J. and Czuprynski, C. J. (2001).** *Infect. Immun.*, **69**: 1650-60.
- Wu, Y.; McQuiston, J. H.; Cox, A.; Pack, T. D. and Inzana, T. J. (2000).** *Infect. Immun.*, **68**: 310-319.