BACTERIOLOGIC SURVEY OF BLACK RHINOCEROS *(Diceros bicornis)*[1]

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Abstract: A bacteriological survey was carried out on 30 black rhinoceros *(Diceros bicornis)* of which 23 were newly captured and seven were captive. A beta haemolytic *Streptococcus,* group L was found in skin lesions and various wounds, causing septicaemia and death in two animals. *Staphylococcus aureus* was found in 3 rhinoceros, and caused the death of one. The bacteria isolated often proved resistant to penicillin. Streptomycin is recommended for treatment. Sixteen other bacteria sp. were isolated, and apart from a *Salmonella* sp. none were considered to be specific pathogens.

INTRODUCTION

Free-ranging and newly-captured black rhinoceros *(Diceros bicornis)* are very susceptible to bacterial infections, and many of these infections have been very difficult to treat, according to the experiences of the authors. Furthermore, most adult rhinoceros in East Africa suffer from ulcerating skin lesions typically found on the ventral side of the neck, on the lower thorax, and behind the forelegs. A filarioid nematode *(Stephanofilaria dinniki)* is believed to be the primary cause of the lesions[4,5] but the role of secondary bacterial infections is unknown.

A survey was undertaken to determine the normal and pathogenic bacteria of the black rhinoceros.

MATERIAL AND METHODS

During translocation of rhinoceros in East Africa (Kenya and Tanzania) from 1969 to 1970, materials for bacteriologic culture were taken from 23 animals. Swabs were taken a few minutes after the animals were captured; they were transferred immediately into Stewart's transport medium and kept refrigerated for a maximum of 6 days before bacterial culture was attempted. Swabs taken from seven other rhinoceros kept in captivity have been treated in the same way, but culture was attempted within 48 h.

The areas examined were: normal skin, normal saliva and nasal secretions, external genitalia, typical *Stephanofilaria* lesions, and other lesions, i.e., arrow wounds, abscesses, septicaemia, etc.

Bacteriologic culture was carried out in the Diagnosis Section, Veterinary Research Laboratory, Kabete. Swabs were plated on blood agar and McConkey agar, incubated at 37 C and examined after 18 h. Colonies on the blood agar were characterized according to morphology, gram-stained smears, and biochemical reactions.

Alpha, Beta, and nonhaemolytic streptococci were tested biochemically, transferred into dextrose broth and typed serologically by the Lancefield system. Staphylococci and the *Corynebacterium* sp. were typed biochemically and serologically. *Bacillus* spp. were identified morphologically from gram-stained smears.

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Salmonella, *Escherichia coli*, *Klebsiella*, *Aeromonas*, *Pseudomonas* and *Proteus* were isolated from McConkey agar, transferred into peptone broth and typed biochemically. Bacteriologic identifications were made in accordance with Cowan and Steel[2] and Breed *et al.*[1]

Sensitivity tests were carried out on *Staphylococcus aureus* and the Beta-haemolytic *Streptococcus* group L, using disks containing chlortetracycline, chloramphenicol, furazolidone, oxytetracycline, penicillin G, streptomycin and sulphafuranzole.

In accordance with appearance and size, the animals were divided into the following five age groups: Calf, half-grown juvenile (½ grown), young adult (y.ad.) and adult (ad.).

RESULTS AND CONCLUSIONS

The bacteria isolated and sensitivity tests are listed in Tables 1 and 2. For convenience, the bacterial isolates were given code figures.

Three of the animals in the survey died of infection. Of these, only No. 29 was examined only after death. The animal had been transported on its side, devellopped pneumonia and died 10 days later. A group L *Streptococcus* was isolated from the lungs, heart blood and spleen, and *Salmonella weltevreden* was isolated from the heart blood, spleen and intestine. Susceptibility to the pneumonia was possibly increased by the stress of transport, resulting in terminal septicaemia.

Numbers 7 and 11 died a few weeks after they were captured. They had multiple skin abscesses. No pathogenic bacteria were found on rhinoceros 7 when captured, but after death *S. aureus* was isolated from liver, spleen and kidney. Group L *Streptococcus* was isolated from various lesions on rhinoceros 11 at the time of capture and *S. aureus* was isolated from an abscess which developed 8 days before the animal died. After death group L *Streptococcus* was isolated from liver, spleen and kidney.

Rhinoceros 30 became ill 3 weeks after capture. Group L *Streptococcus* and *Pseudomonas fluorescens* was cultured from the penis when the animal was caught, while *S. aureus* was cultured from one unopened abscess, and a group L *Streptococcus* from other open as well as unopened abscesses which developed in the skin. During captivity the animal survived an extremely radical surgical treatment involving the complete removal of more than 20 abscesses from the skin.

The filarial lesions were considered to be caused by *S. dinniki* and this was confirmed by demonstration of microfilariae in Giemsa-stained smears from skin lesions of the animals listed in Table 1 and, in one case (7), also in histologic sections of a biopsy.

Smears from these lesions also were stained by the Ziehl-Neelsen method for *Mycobacteria*, acid-fast organisms were never seen. Histologic examination of the damaged skin from numbers 7, 11 and 30 indicate a fungus as the primary cause, and various bacteria as secondary invaders in the *locus minoris resistentia* so created.

The results here presented show that a beta-haemolytic *Streptococcus* belonging to the Lancefield group L was the most frequently occurring pathogenic bacteria for the rhinoceros. It was isolated from filarial lesions from 17 of 19 rhinoceros, and in 17 of 25 other lesions, from 8 of 11 rhinoceros.

The *Streptococcus* usually was found together with other bacteria, except in unruptured abscesses, i.e., numbers 25 and 30. In only one animal, number 10, the group L *Streptococcus* was isolated from normal skin. However, it was also found on the penis (number 30), and seemed to be a regular inhabitant of the vagina of juvenile as well as of adult animals. Similarly, in domestic animals, the group L *Streptococci* are mainly

TABLE 1. Bacteria isolated from 30 black rhinoceros.

	No.	Age	Sex	Normal skin	Saliva	External genitalia	Filaria lesions	Other lesions
Recently captured	1	Ad.	F				A Q	
	2	Ad.	M			A Q	A Q	
	3	Ad.	M				A Q	
	4	Ad.	M				A Q	
	5	Ad.	M				A Q	
	6	Ad.	M				A C D J L Q	
	7	½ grown	M	J L Q	J L Q		J L Q	I
	8	y.Ad.	M	J L Q	H		A J K Q	
	9	Ad.	F	H L	J Q		A F J L Q	
	10	½ grown	F	A J P Q				
	11	½ grown	M	J Q			A J Q	A I Q
	12	Ad.	F			A J	A J	
	13	Baby	F	J				J Q Tick bite
	14	y.Ad.	M	J			A J	
	15	½ grown					A J	
	16	Ad.	F	J P		Sterile	A J Q	A J Q Arrow wound and abscess
	17	y.Ad.	F			A	A J K	
	18	Baby	F			E		
	19	½ grown				A J P	A O P	
	20	½ grown				A J	A J O	
	21	Ad.	M	J P			A J P	
	22	Ad.	F	J P				A J Arrow wound
	23	Ad.	F			A J		A J O Arrow wound
Captive animals	24	y.Ad.	M		P			A B G J Ulcer and abscess
	25	y.Ad.	F					A Unopened abscess
	26	Ad.	F				P	
	27	Baby	M	J P Q	Q			
	28	Baby	M	J P Q	B J			J Tick bite

TABLE 1. (continued)

No.	Age	Sex	Normal skin	Saliva	External genitalia	Filaria lesions	Other lesions
29	y.Ad.	M					A M
30	Ad.	M			A N		A I R

Bacterial code No.	
A	Beta-haemolytic *Streptococcus* Lancefield group L
B	" " C
C	Alpha-haemolytic " " D
D	" " H
E*	" " L
F	" " N
G	*Streptococcus dysgalactiae*
H	" *viridans*
I	" *aureus*
J	" *epidermis*
K	*Escherichia coli*
L	*Klebsiella ozonae*
M	*Salmonella weltevreden*
N	*Pseudomonas fluorescens*
O	*Proteus sp.*
P	*Bacillus sp.*
Q**	*Corynebacterium sp.*
R	*Aeromonas sp.*

*No. E is biochemically different from No. A.
**No. Q does not include any of the species known to be pathogenic.

TABLE 2. Results of sensitivity tests.

Bacteria:	*Staphylococcus aureus*			*Beta-haemolytic streptococcus group L*					
Animal ref. No.:	7	11	30	16	17	19	20	21	30
Chloramphenicol:	S	S	S	S	S	S	S	S	S
Furazolidone:	R	R	S	S	S	S	S	S	S
Neomycin:	S	R	S	S	S	S	S	S	S
Penicillin D:	S	R	R	R	R	R	R	R	S
Streptomycin:	S	S	R	S	S	S	S	S	S
Sulphafurazole:	R	R	R	R	R	R	R	R	R
Oxytetracycline:	S	S	S	S	S	S	S	S	S

S: Sensitive
R: Resistant

found in pigs,[3] in the throat and in the genital tract, sometimes causing lesions.

The other haemolytic Streptococci were isolated rarely in normal and diseased locations, and usually together with the group L *Streptococcus.*

S. aureus was found only in lesions on three animals, and caused septicaemia and death in one of them (number 7).

The history of the three animals which died of septicaemia (numbers 7, 11, and 29) and of number 30 indicate that both the group L *Streptococcus* and *S. aureus* exert their pathogenicity and even cause fatal infection when the animals lose resistance.

Most of the strains of the group L *Streptococcus* and *S. aureus* tested for sensitivity proved resistant to penicillin (Table 2).

The broad spectrum antibiotics often produce large local reactions when injected intramuscularly, and streptomycin is in our experience, the drug of choice for treatment of bacterial infections in black rhinoceros.

Streptococcus epidermidis was cultured from normal skin from 11 of 12 rhinoceros, but less frequently from filaria lesions and other lesions (23 of 47 cultures from 17 of 27 rhinoceros), indicating that this organism probably is a normal inhabitant of rhinoceros skin and often is present in skin lesions.

It is not possible to state whether *S. weltevreden* was present in the intestine when Rhino 29 was caught or whether it was introduced during captivity. Mixed infections in open wounds are to be expected when large animals like rhinoceros are handled, and the presence of *Streptococcus viridans, Escherichia coli, Klebsiella ozonae, Proteus* sp., *Corynebacterium* sp., *Bacillus* sp., and *Aeromonas* sp. on normal and diseased skin are believed to be due to contact with dust and soil, especially during capture. Bacterial flora was similar in filarial skin lesions and in the lesions of other origin. All bacteria found in the filarial lesions are considered secondary to the filarial worms but probably contribute to ulceration.

From Table 1 it can be seen that the group L *Streptococcus* can be isolated from apparently healthy rhinoceros just after capture indicating that this bacteria is a regularly inhabitant on rhinoceros. The frequent isolation of this bacteria from lesions primary caused by filaria nematodes and various injuries indicate the capability of this bacteria for developing in *Loci minoris resistentia* from which it can even cause septicaemia. *S. aureus* can be lethal, it does

not seem to be a regular pathogen on free-living black rhinoceros. Of the 16 other bacteria sp. isolated, only *S. weltevreden* have spread to the organs, while the rest seems to be without major importance for the black rhinoceros examined.

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LITERATURE CITED

1. BREED, R.S. 1957. *Bergey's Manual of Determinative Bacteriology*. Bailliere, Tindell & Cox Ltd., London.
2. COWAN, S.T. and K.J. STEEL. 1965. *Manual for Identification of Medical Bacteria*, Cambridge, Cambridge U.P.
3. JEPSEN, A. and H.E. ERREBO LARSEN. 1972. Veterinary Bacteriology and Microbiology. *Diagnostic Bacteriology*. C.F. Mortensen, Copenhagen.
4. SCHULZ, K.C.A. and E.B. KLUGE. 1960. Dermatitis in the black rhinoceros due to filariasis. J. S. Afr. vet. med. Ass. 31: 265-269.
5. TREMLETT, J.G. 1964. Observations on the pathology of lesions associated with *Stephanofilaria dinniki* Round 1964 from the black rhinoceros *(Diceros bicornis)*. J. Helminthol. 38: 171-174.

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