AN OUTBREAK OF BABESIOSIS IN IMPORTED SABLE ANTELOPE *(HIPPOTRAGUS NIGER).*

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ABSTRACT

A complete necropsy performed on 2 sable antelope *(Hippotragus niger)*, revealed lesions concomitant with a massive haemolytic crisis. These included widespread oedema and anaemia of the carcass, severe oedema of the lungs, petechiae and echymoses of the epicardium, a moderate splenomegaly and a severe haemoglobinuria. The histopathological lesions included a moderate alveolar oedema, the presence of haemosiderin in the spleen and lymph nodes, and mild degenerative changes of the renal tubular epithelium. Peripheral blood and brain smears contained numerous parasitised red blood cells. The parasites were round or oval in shape containing a single or double area of purple-staining chromatin along a portion of the margin of the organism. It was identified as *Babesia irvinesmithi* Martinaglia, 1936, which is unique to sable. Seven sable antelope were subsequently treated with imidocarb diproprionate at a dose of 1,2 mg kg^{-1}. No adverse side-effects have been noted in these animals.

Key words: Sable, *Hippotragus niger,* babesiosis, *Babesia irvinesmithi*

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INTRODUCTION

Babesiosis in sable antelope *(Hippotragus niger)* has only been recorded on 3 occasions[5] [13] [14]. Martinaglia[5] reported the death, due to babesiosis, of a captive sable antelope, brought from the northern Transvaal to the Johannesburg Zoo. In 1936, Martinaglia named the parasite *Babesia irvinesmithi*[6]. Wilson et al[14] reported the presence of a *Babesia* sp., in the blood smears of 2 young sable that had died of other causes. Thomas et al[13] reported the presence of *Babesia* sp. in 7 out of 124 blood smears taken from sable in South Africa and Zimbabwe. Five of the positive smears were from animals that had been found dead in the veld.

Babesia sp. have been reported from various antelopes. Carmichael & Hobday[2] reported the presence of a small *Babesia* in a tsessebe *(Damaliscus lunatus)* and a large *Babesia* in a blue wildebeest *(Connochaetes taurinus)*. An occasional large *Babesia*-like ring form was seen in impala *(Aepyceros melampus)*[4], while Bigalke et al[1] reported a *Babesia* sp. in a bushbuck *(Tragelaphus scriptus)*.

There is no evidence that *B. bigemina* is harboured by antelope in South Africa[9]. Attempts to transmit the parasite to sable antelope[13], blesbok *(Damaliscus dorcas phillipsi)* and common duiker *(Sylvicarpra grimmia)*[10] were unsuccessful. However other attempts proved the susceptibility of a splenectomised Soemmering's gazelle *(Gazella soemmeringi)* from the Sudan to *B. bigemina* (Enigk & Friedhoff, as cited by Neitz[8]). Attempts to transmit *Babesia bovis* to intact and splenectomised sable antelope were unsuccessful[13].

Nine adult sable antelope were imported from a zoo in West Germany to South Africa. The animals were transported by ship to Cape Town and thence by road to a game farm near Brits in the Transvaal (25° 30' S 27° 48' E). This farm, used previously as a cattle ranch, had been used exclusively for game for the last 4 years. After their arrival, the imported animals were kept in pens on the farm for one month. They were fed silage and lucerne hay and had access to water ad lib. During this time they were exposed to a sable antelope that originated from the Gravelotte district in the eastern Transvaal, which was kept in an adjacent enclosure. This animal had been housed for 2 weeks in one of the pens used for the imported animals 2 months prior to their arrival. No tick control measures were implemented. The 2 imported sable antelope that succumbed, were in the enclosure in which the Gravelotte animal had previously been kept. The 7 surviving sable were housed in individual enclosures adjacent to the above-mentioned pens.

The 2 sable which died, succumbed to the disease approximately 2 months after their arrival in South Africa. Sable 1 was reported to be ill, displaying anorexia, depression and recumbency, approximately 24 h before it died. The carcass was kept at 4°C for 48 h before being submitted for necropsy. Sable 2 did not show clinical signs; it was found dead in the pen, 4 d after the death of the first animal and submitted for necropsy within 12 h. Several *Boophilus decoloratus* and *Rhipicephalus evertsi evertsi* were found on both animals at necropsy.

MATERIALS AND METHODS

A complete necropsy was performed on the 2 sable antelope. Selected tissues were fixed in 10% buffered formalin. Sections of the fixed tissues from these animals were prepared and stained with haematoxylin and eosin (HE). Urine from the bladder was tested for the presence of haemoglobin and protein (N-Multistix, Ames Division, Miles Laboratories Limited Stoke Poges, Slough SL2 4LY England). Peripheral blood smears were made, using blood obtained from the tail. Brain smears were made, using macerated grey matter of the cerbral cortex. An impression smear of the spleen was also pre-

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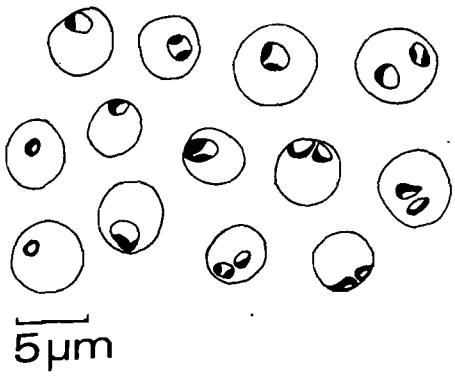

Fig. 1: **Drawings of infected blood cells from Sable 2, showing the different forms of the parasite**

pared. All of the slides were stained with RapiDiff (Clinical Sciences Diagnostics, Division of D.M.S.L. (Pty) Lty. P.O. Box 38939, 2016 Booysens), and examined under a light microscope.

Parasites (n = 100) were measured with an ocular micrometer and mean values were determined. Drawings of the parasites were made with the aid of a drawing tube.

Levels of parasitaemia were determined in the blood and brain smears by counting the number of erythrocytes in one microscopic field and then counting the number of parasites in 15 similar fields.

The 7 remaining imported sable antelope were immobilised using 4-5 mg etorphine hydrochloride (M.99, R & C Pharmaceuticals, Mobani, RSA) together with 40 to 60 mg azaperone (Azaperone, Janssen Pharmaceuticals, Olifantsfontein, RSA) delivered by dart, using Telinject equipment (Telinject SA, Randburg, RSA.). Blood smears were made from each animal with capillary blood obtained from the tail and these were stained and examined as described above. The body mass of each animal was estimated and each was treated with 1,2 mg kg^{-1} imidocarb diproprionate (Forray 65, Coopers Animal Health, Kempton Park, RSA.) administered subcutaneously. No un-toward side-effects were reported to the above treatment.

RESULTS

Post mortem examination revealed that Sable 1 was in a fair body condition, while the body condition of Sable 2 was good. Both the animals had a mild hydrothorax, hydropericardium, ascites and mild pulmonary oedema. In Sable 2, this was evident by the presence of white foam at the bifurcation of the trachea. Both the animals displayed pale buccal mucous membranes which indicated a severe anaemia. Multifocal echymoses and petechiae were present on the epicardial and endocardial surfaces of Sable 2. A moderate enlargement of the spleen was seen in both the animals. The spleen bulged on cut surface and there was an increase in the red pulp. The kidneys of Sable 1 were black in colour, while those of Sable 2 were pale, soft and slightly enlarged.

Both antelope displayed a severe haemoglobinuria and proteinuria. The bone marrow of Sable 2 was pale, yellow, fatty and gelatinous.

The tissues of Sable 1 were too autolysed for histopathological examination. Sable 2 showed severe subendocardial haemorrhage. The lymph nodes revealed severe oedema and focal congestion. Extensive medullary haemosiderosis was present, indicating severe intravascular haemolysis. In addition, moderate erythrophagocytosis was present.

The spleen showed severe congestion of the red pulp with the presence of many macrophages packed with haemosiderin. Despite the presence of parasites in the red blood cells of the brain (as seen in the brain smear), the cerebrum only showed congestion of the blood vessels. No signs of encephalomalacia or haemorrhage were seen. The kidney displayed a moderate interstitial congestion. Mild degenerative changes and small amounts of haemosiderin were present in the tubular epithelial cells.

The pulmonary tissue showed considerable congestion of the interstitial blood vessels. The number of alveolar macrophages was considerably increased and many contained large amounts of haemosiderin. Focal areas of alveolar oedema and interstitial collapse were occasionally seen. In these areas, neutrophils were often present in the interstitium.

The liver showed an increase in the number of neutrophils present within the sinusoids. Haemosiderosis was also evident. A mild periportal inflammatory infiltrate was seen.

The parasitaemia in Sable 1 was 4% and in Sable 2, it was 6%. Extracellular forms were present in both of the antelope. There was a tendency for parasitised erythrocytes to accumulate in the brain capillaries and approximately 40% of these were infected with *Babesia* parasites.

The parasites in the erythrocytes were round, oval, piriform or irregularly shaped, usually with a single or double area of purple staining chromatin situated along a portion of the margin of the organism (see Fig. 1 & 2). The paired forms were usually piriform or irregularly shaped. In Sable 2, there was a tendency for the parasites to occur on the margin of the cell. The round forms in Sable 1 were 0,8-1,8 µm (mean 1,1 µm) in diameter and the elongated parasites measured 2,3-1,0 µm x 1,5-0,8 µm (mean 1,4 x 1,1 µm) while those of Sable 1 measured 2,4-0,8 µm (mean 1,5 µm) and 2,4-1,0 x 1,8-0,8 µm (mean 1,7 x 1,3 µm) respectively. Paired forms were fairly rare.

Examination of the blood smears of the 7 surviving sable did not reveal the presence of any *Babesia* parasites. A *Theileria*-like piroplasm, however, was seen in one smear.

DISCUSSION

Neitz[8] suggested that the relationship between *B. irvinesmithi* Martinaglia, 1936 of the sable antelope and *B. bovis* should be determined. Thomas et al[13] made an attempt to isolate the sable *Babesia* sp. by the subinoculation of blood from sable to splenectomised and intact sable calves and splenectomised cattle as well as transmission with *B. decoloratus* from sable to a splenectomised bovine. Attempts to infect sable with *B. bigemina* and *B. bovis* failed. Thomas et al[13] concluded that *B. irvinesmithi* was a distinct and valid species of sable antelope. Although no transmission studies were completed, we identified the parasite as *B. irvinesmithi*, based on the parasite's morphological characteristics which appear to be identical with those described by Martinaglia[6].

The *Babesia* described in this study is similar in size to that described by Thomas et al[13] The morphology of the parasite from Sable 1 differed in some respects from that of Sable 2. The autolysis in Sable 1 appeared to cause enlargement of the parasite and there were many more round forms present, although oval, piriform and irregular forms were also seen. The mean diameter of the round forms increased from 1,1 µm in Sable 2 (the recently dead sable) to 1,5 µm in Sable 1 (the autolysed sable) and the elongated forms increased from 1,4 x 1,1 to 1,7 x 1,3 µm. Bigalke et al[1] also noted a rounding-off of *Babesia* parasites due to autolytic changes in a bushbuck. Thomas et al[13] observed the parasite to be randomly situated in the erythrocytes. This phenomenon was noted in Sable 1, but in contrast, in Sable 2, 80% of the parasites were situated on the margin of the erythrocytes. These findings suggest that both the size and the situation of the parasite can possibly vary between different specimens. Whether these morphological variations only occur after death cannot, as yet, be determined until more specimens from clinical sable babesiosis are examined.

A comparison of the size of the *B. irvinesmithi* with other cattle *Babesia* (Table 1) shows that the *B. irvinesmithi* is smaller. As these measurements are from relatively fresh material, the increase in size due to autolysis seen in Sable 1 should not be taken into consideration for comparative purposes. This would tend to contradict Thomas et al[13] who suggested a similarity in size to *B. bovis*.

The source of infection for these sable is a matter of speculation. A possibility

Table 1: **A comparison of the sizes of the sable *Babesia* and the bovine *Babesia* species**

Babesia sp.	Authors	Mean Length μm	Mean Width μm
B. irvinesmithi	This study	1,4	1,1
B. irvinesmithi	Thomas et al[13]	1,58	1,11
B. bovis	Gray & de Vos[3]	2,29	1,10
B. bovis	Riek[12]	1,8	1,2
B. bovis	Potgieter[11]	2,0	1,2
B. bovis	Neitz[7]	1,5	
B. occultans	Gray & de Vos[3]	2,88	1,22
B. bigemina	Gray & de Vos[3]	3,29	1,49

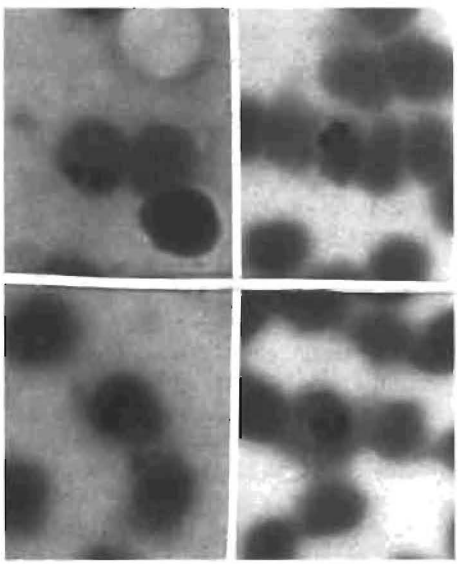

Fig. 2: **Different forms of *Babesia irvinesmithi* from Sable 2 (x100)**

exists that the imported sable relapsed due to the stress of importation. This seems unlikely as babesiosis has not been reported in sable kept in zoos. A less likely explanation is that the parasite was of cattle origin or had originated from another unknown host. Since cattle had not been kept on the game farm for 4 years, and the pens had been recently erected, this possibility seems very unlikely. The most likely source of infection was the Gravelotte sable which had had close contact with these animals.

Wilson et al.[14], reported the macroscopic lesions of anaemia, haemoglobinuria, mild hypoxic lesions, visceral haemosiderosis and sub-pial petechial haemorrhages of the cerebellum found in the necropsies of 2 sable, both of which had *Babesia* parasites in their blood smears. The 2 sable in this study also displayed anaemia and haemoglobinuria at the post mortem examination. Martinaglia[5] reported icterus, anaemia and a splenomegaly in a case of babesiosis in a sable. Moderate enlargement of the spleen was noted in both animals in this study.

The level of *Babesia* parasitaemia is reported to vary between 0,01 and 4,4%[13]. This is similar to the sable in this study which displayed parasitaemias of 4% and 6%, respectively.

Both *B. bovis* and the sable *Babesia* have a tendency to accumulate in the blood capillaries of the brain[13]. This phenomenon was also noted in the brain smears of the 2 sable antelope of this study.

The piroplasm seen in one of the blood smears of the remaining, healthy sable would appear to be a *Theileria* sp.[12]. One of the authors (BLP) found *Theileria* sp. but no *Babesia* sp. in 12/12 clinically normal sable in the Gravelotte area. These findings are in accordance with those of Wilson et al[14], who described theilerial parasitaemias varying from a few parasites to 14,4% in the blood smears of sable calves and adults that they examined; the theilerias did not appear to have an adverse effect on the sable. Similarily, the sable in this study did not display any untoward clinical signs. The findings of Carmichael & Hobday[2] regarding theilerial piroplasms in sable, appear to corroborate the above information.

At present, the importance of babesiosis as a cause of disease in sable is difficult to assess. Wilson et al[14] suggested that latent infections may relapse in animals weakened by other factors and thus contribute to the cause of death. He also speculated that babesiosis may play a role in the high mortality rate of sable calves under the age of 12 weeks. This study shows that if highly susceptible, adult animals, such as those in a zoological gardens situation, which are unlikely to have been exposed to tick infestations, are infected with *Babesia* parasites, then mortality is likely to occur.

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