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THE TRANSOVARIAL TRANSMISSION OF *BABESIA CABALLI* BY *HYALOMMA TRUNCATUM*

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ABSTRACT

DE WAAL, D. T., 1990. The transovarial transmission of *Babesia caballi* by *Hyalomma truncatum*. *Onderstepoort Journal of Veterinary Research, 57,* 99–100 (1990).

Babesia caballi, isolated from a horse that originated from South West Africa/Namibia, was transmitted transovarially by adult *Hyalomma truncatum*. *B. caballi* proved to be highly infective for adult *H. truncatum*. Forty-five per cent of ticks feeding on a reacting animal with an extremely low parasitaemia became infected. In spite of a low parasitaemia, the ticks were severely affected by the parasite. Seventy per cent of the infected ticks either died during oviposition or after laying only a few eggs.

The features of the infection in horses were: a prepatent period of 10 days, very low parasitaemias with low pathogenicity and spontaneous recovery of the infected animals.

INTRODUCTION

Babesia equi is apparently more commonly diagnosed in clinical cases of equine babesiosis in the Republic of South Africa (RSA) than *Babesia caballi*. Although it is believed that *B. equi* is more pathogenic than *B. caballi,* the importance of *B. caballi* as a pathogen remains obscure. De Kock (1918) made a casual reference to the presence of *B. caballi* in some of his experimental horses. Apart from his research, no investigations have been undertaken to study the epidemiology of, and possible role that *B. caballi* might play in equine babesiosis in this country. A variety of tick species has reportedly transmitted *B. caballi* (Neitz, 1956; Stiller & Frerichs, 1979; Stiller, Frerichs, Leatch & Kuttler, 1980), but none of these species occur in this country.

Rhipicephalus evertsi evertsi was recently identified as a vector of *B. caballi* in the RSA (De Waal & Potgieter, 1987). Further studies on the transmission of equine babesiosis have led to the identification of a 2nd vector of this parasite, namely *Hyalomma truncatum*. This paper reports on the transovarial transmission of *B. caballi* by adults of *H. truncatum*, a two-host tick which has a wide distribution.

MATERIALS AND METHODS

Experimental animals

Two susceptible intact horses (451 and 42), respectively 10 and 12 months of age, born and reared under strict tick-free conditions at the laboratory of the Veterinary Research Institute, Onderstepoort (VRI), were used in this study.

Babesia caballi *isolate*

Seven horses were obtained for breeding purposes from a stock of "wild" horses from Garub in the Namib Desert, Namibia/SWA. These horses have been in the area for a number of generations, probably since the 1st World War. On arrival at the VRI, the horses were bled and screened serologically with the indirect fluorescent antibody test, for antibodies against *B. equi* and *B. caballi* (Madden & Holbrook, 1968). None of these horses had a positive *B. equi* titre, and only 1 tested positive for *B. caballi* antibodies. All blood smears examined from these horses were negative.

Two-hundred and fifty mℓ of blood was collected in heparin from the *B. caballi* seropositive horse and subinoculated intravenously into Horse 451 in an attempt to isolate the parasite.

Hyalomma truncatum *feeding and maintenance*

The Warrenton strain of *H. truncatum* used in this study was obtained from the Section of Entomology (VRI), where it had been maintained for several generations by feeding the larval-nymphal stage on rabbits and the adults on sheep. All non-feeding stages were maintained in an acaridarium at 25 °C and 85 % relative humidity.

For the purpose of this study, the adult stages were fed on the shoulder region of the horses (De Waal & Potgieter, 1987) and the larval-nymphal stages in back pockets on rabbits (Heyne, Elliott & Bezuidenhout, 1987).

Babesia caballi *reactions*

Parasitaemia: Blood smears, thick and thin, were made daily and quantified as described by De Waal & Potgieter (1987).

Clinical signs: Rectal temperatures and haematocrit determination were taken daily between 08:00 and 10:00.

Infection and transmission of B. caballi

Infection of ticks: On Day 11 post-infection when the first *B. caballi* parasites were detected in a thick blood smear, 100 *H. truncatum* adults (50 males and 50 females) were used to infest Horse 451.

Forty engorged female ticks were collected from Day 8 to 16 post-tick infestation from Horse 451, placed individually in small glass tubes (25 × 10 mm) and allowed to lay eggs in the acaridarium.

Detection of infection in ticks: Twenty-two days after engorgement the female ticks were screened for *B. caballi* infection. A leg of each tick was amputated and haemolymph smears were prepared, as described by Burgdorfer (1970). The smears were air-dried, fixed in methanol, stained in a 10 % Giemsa's stain for 35 min and examined for kinetes of *B. caballi.*

Transovarial transmission by H. truncatum *from adult to adult:* Thirty days after hatching, 2 tubes of larvae from infected females were fed on a rabbit. The engorged nymphae were collected 16–24 days later and allowed to moult in the acaridarium. Forty-eight days after moulting to adults, 20 males and 20 females were used to infest Horse 42.

Seventy kinetes in the haemolymph of engorged female *H. truncatum* were measured with a Kontron, Videoplan, computerized image analiser[1] using the standard measuring programme.

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[1] Kontron, Image Analysis Systems, West Germany

RESULTS

Horse 451 developed a patent *B. caballi* parasitaemia on Day 11 after subinoculation of 250 mℓ of blood collected from the horse that orginated from Namibia/SWA.

A patent parasitaemia was recorded for 8 days. Throughout this period parasites were visible on thick blood smears only. The 1st temperature reaction was recorded on Day 10, with a peak of 41 °C on Day 20. Forty engorged female ticks were collected from this horse.

B. caballi proved to be highly infective for *H. truncatum*, since 55 % of the female ticks became infected while engorging on the horse showing these very low parasitaemias. Tick infection rates as high as 98 % were recorded in a subsequent transmission experiment, where ticks were fed on a horse showing an equally low parasitaemia (D. T. de Waal, unpublished data, 1987–88).

During this investigation it was quite obvious that the ticks were severely affected by the parasite. Seventy per cent of the ticks showing positive haemolymph smears either died during oviposition or after laying only a few eggs. Less than 50 % of these eggs hatched. It is known that *B. caballi* has similar detrimental effects on *Dermacentor nitens* (Anthony, Johnson & Holbrook, 1970). The kinetes of *B. caballi* observed in the haemolymph smears measured 11,66 µm ± 1,08 µm in length with a diameter (as measured over the nucleus) of 2,40 µm ± 0,29 µm.

Transovarial transmission by H. truncatum

Out of a total of 20 female and 20 male *H. truncatum* ticks fed on Horse 42, 18 engorged female ticks were recovered. The first parasites were seen in thick blood smears on Day 10 post-infestation. A febrile reaction, that persisted for one day only, was recorded on Day 13 post-infection.

No marked drop in the haematocrit of Horse 42 was recorded during the reaction period. The maximum parasitaemia recorded was a score of 2 (1–5 parasites in 100 microscope fields with approximately 350 erythrocytes) on Day 17 post-infestation. The horse recovered without treatment.

DISCUSSION

Theiler (1906) in his experiments excluded *Hyalomma* spp. as possible vectors of equine babesiosis, because the immature stages of these two-host ticks occur only on small rodents and birds. The sero-conversion of horses during a sero-epidemiological survey and the large number of *Hyalomma* spp. found on horses during this survey (D. T. de Waal, unpublished observations, 1986–87) led to the investigation on *H. truncatum* as a possible vector of *B. caballi*. The first attempt to transmit *B. caballi* with *H. truncatum* was successful, whereas numerous attempts to transmit *B. caballi* with *H. m. rufipes* failed (D. T. de Waal, unpublished observations, 1985–1987).

H. truncatum is widely distributed in southern Africa, and is only absent from the higher parts of the South African plateau with its higher rainfall. It is present in the cooler winter rainfall areas of the Cape Province (Theiler, 1956). The wide distribution of this tick species would indicate that *B. caballi* should have a much wider distribution than was previously believed by earlier workers (Henning, 1949; Littlejohn, 1963).

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