

THE RESERVOIR STATUS OF GOATS RECOVERED FROM HEARTWATER

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ABSTRACT

BARRÉ, & N. CAMUS, E., 1987. The reservoir status of goats recovered from heartwater. *Onderstepoort Journal of Veterinary Research*, 54, 435–437 (1987).

Experiments were conducted with Creole goats and *Amblyomma variegatum* ticks in Guadeloupe to investigate whether it is possible to transmit *Cowdria ruminantium* to susceptible hosts with nymphs fed in the larval stage on recovered goats.

Of 88 batches of larvae fed after the return of the goats' temperature to normal, or after challenge or immunosuppression, only the 9 batches of larvae fed during the febrile reaction, 2 batches applied 2 and 3 days respectively after recovery, and a single batch applied 5 days after challenge, became infective.

On average, blood appears to be infective for *A. variegatum* larvae for only an 8-day period associated with the temperature reaction following a primary infection. Recovered goats are not reservoirs of *Cowdria*, even if—with one exception in our experiment—they are reinfected after recovery.

INTRODUCTION

In the epidemiology of heartwater, an explanation is needed for the low infection rate in tick populations (Alexander, 1931; Bonsma, 1944; Neitz & Alexander, 1945; Uilenberg, 1971). The usual absence of transovarial transmission (Bezuidenhout & Jacobsz, 1986) and, as stated by Alexander (1931), the fact that animals which recover from the disease do not retain *Cowdria* in their blood indefinitely could be among the reasons. According to Uilenberg (1981, 1983), blood is infective for ticks for a limited period only, i.e. from the start of the febrile reaction until, usually, a few weeks after recovery.

The answer to the question whether it is possible to transmit *Cowdria ruminantium* to susceptible hosts with nymphs fed in the larval stage on recovered goats has the following important implication: the risk of contaminating areas free of heartwater, but where vectors exist, by the introduction of recovered animals.

Most of the researchers who have studied the infectivity of blood did so by inoculating blood from recovered animals into susceptible ones. Marked differences have been reported following the use of this method. Uilenberg (1971) did not find the blood of sheep infective 1, 4 and 5 months after recovery, while Ilemobade (1978) found it infective 40 days after recovery in 1 case and 50 days after challenge in another case. In only 1 out of 12 cattle, challenged by Du Plessis & Bezuidenhout (1979) 2 years after artificial immunization, was blood found to be infective for sheep 15 days after challenge. Alexander (1931) stated that the period of infectivity did not exceed 35 days, while Neitz (1939) and Neitz, Alexander & Adelaar (1947) found that after primary or re-infection the maximum was 2 months.

These results cannot be accepted unchallenged to explain certain epidemiological features of heartwater. Inoculation of blood is not the natural mode of transmission and the amount injected by the different authors (5 to 10 ml) is greater than that ingested by larvae or nymphs during their blood meal. The amount of blood absorbed by these stages may be insufficient to infect them if the *Cowdria* concentration in the blood stream is very low. Or, on the contrary, ticks may multiply in their bodies very small numbers of *Cowdria* taken up in the blood.

For these reasons, we attempted transtadial transmission with a natural vector of heartwater. In the literature, we could find only the results obtained by Alexander (1931) with 2 batches of larvae fed on a recovered animal.

MATERIALS AND METHODS

Animals used

All the experiments have been carried out with Guadeloupean goats, of a breed called "Creole", introduced from Africa centuries ago.

The goats that were used originated from islands or farms free of both *Amblyomma* and heartwater and between 2 months and 1 year old. All of them were tested for heartwater antibodies by the indirect fluorescent antibody test (IFAT) with the Kümm strain (Du Plessis, 1981a) before the experiment. The goats used in attempts to infest larvae are called "donors" and those on which the infectivity of the resulting nymphs was tested "receivers".

Twenty-two goats which had been inoculated with virulent blood or infected with ticks, and had then shown typical fever and serological reactions before spontaneously recovering from heartwater, were used as donors. Receiver goats tested with nymphs were monitored daily for temperature and symptoms and once a week for antibodies. Recovered receivers were challenged with virulent blood to test their immune status. Three batches of male ticks, not infective in the nymphal stage, were also tested on receivers. Most of the experiments were done after a primary infection, but we also challenged 10 donor goats with 8–35 infected nymphs 45 to 100 days after recovery and tested the infectivity of larvae fed during the post challenge period. In a final experiment 3 donor goats were inoculated with an immunosuppressive compound (cyclophosphamid 30 mg/kg i.v.) 110 days after recovery, then infested with larvae which were subsequently tested as above on a receiver.

Ticks used

The only *Amblyomma* of ruminants present in the central Lesser Antilles is *A. variegatum*, the species we used in the experiment. A local strain has been reared in our laboratory for 3 years; after feeding on goats all stages were kept in incubators at 22–26 °C and 92 % RH for moulting and laying eggs.

Larvae were fed in ear bags; nymphs and adults in flank bags.

In this experiment, nymphs were used 1–6 weeks after moulting. The minimum in a batch of nymphs was 5 ticks, but most batches consisted of 10–100 or more specimens.

Nymphs were considered to be infective, and therefore larvae to have been infected, when receivers either showed a febrile reaction and other symptoms and died of heartwater with *Cowdria* colonies in their brain, or recovered, showed antibodies in their sera and were resistant to challenge with a homologous strain.

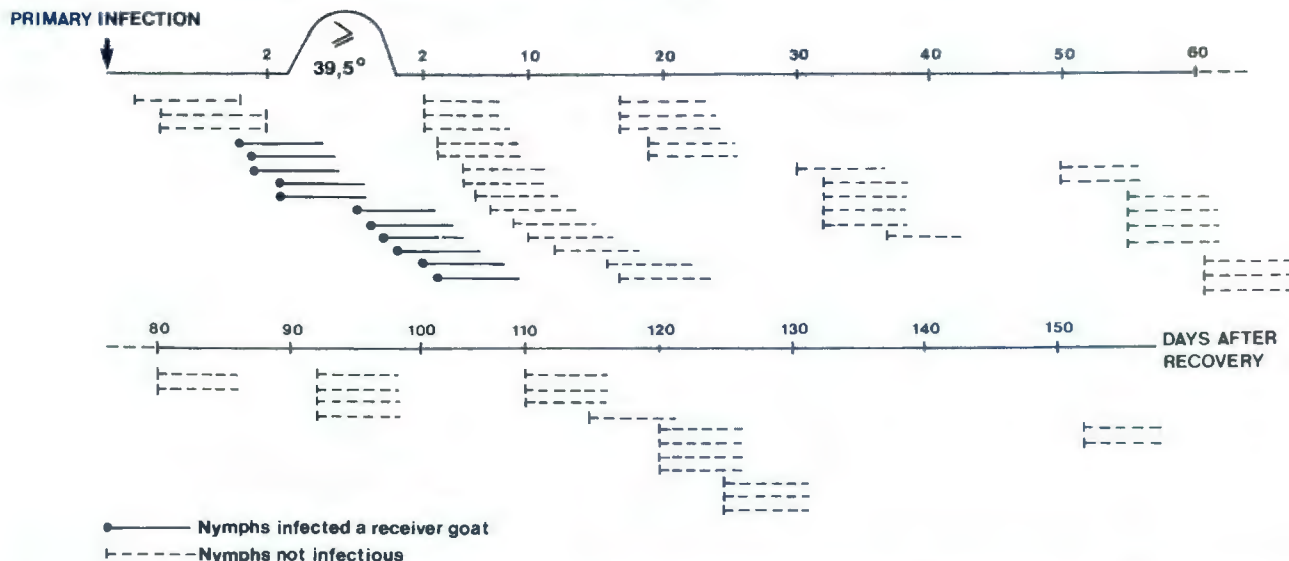


FIG. 1 Primary infection = infectivity of *A. variegatum* nymphs fed in larval stage on donor goats during the incubation period, the temperature reaction or at increasing intervals after recovery

TABLE 1 Summary of the experiments on the infectivity of *A. variegatum* nymphs fed in the larval stage on goats after primary infection, immunosuppression or challenge

Expt.	Feeding of larvae	Batches of nymphs infectious/tested	Total number of nymphs tested
Primary infection	During incubation, detached 2 days before temp. rise	0/3	372
	During temp. rise 39,5 °C	9/9	151
	2-18 days after temp. returns to normal	2/13	576
	12-160 days after temp. returns to normal	0/42	1 742
Immunosuppression	1-8 days after injection	0/2	35
	10-18 days after injection	0/3	271
Challenge	5-19 days after challenge	1/13	494
	26-49 days after challenge	0/3	173

Cowdria strain

The strain used, called the "Gardel strain", was isolated in December 1982 from adult ticks collected from cattle on a farm in the east of Guadeloupe (Uilenberg, Camus & Barré, 1985).

RESULTS

1. Infectivity of nymphs fed in the larval stage after a primary infection (Fig. 1, Table 1)

Larvae fed during the incubation period

In the 3 batches tested, larvae detached from the donor at the latest 2 days before the temperature rose (up to 39,5 °C). Transmission in the nymphal stage failed.

Larvae fed during the febrile reaction

The 9 batches of ticks fed during this period became infected even if larvae started to feed only the day the temperature returned to normal.

Larvae fed after the febrile reaction

Among 55 batches of larvae applied from 2 days to 5 months after recovery only 2, that started to feed 2 and 3 days respectively after the temperature returned to normal, became infected.

The batches of larval ticks fed 10, 30 and 80 days after recovery of the goat were not found to be infective as either nymphs or adult males.

2. Infectivity of nymphs fed in the larval stage after injection of goats with cyclophosphamid or after a challenge (Fig. 2, 3; Table 1)

Immunosuppression of recovered goats did not enhance the rickettsaemia. None of the 5 batches of nymphs fed during the larval stage 0 and 10 days after the injection of cyclophosphamid were infective.

In one case out of 16 a batch of larvae became infective when applied to a goat 5 days after a challenge infection given 45 days after recovery.

DISCUSSION

These results confirm the opinions of Alexander (1931) and Uilenberg (1981, 1983) regarding the limited period during which larvae, and probably also nymphs, can themselves become infected by feeding on an animal infected with heartwater.

From our experiments we can assume that the maximum duration of the infective period is from 2 days before the temperature begins to increase until 3 days after the temperature returns to normal. If we consider a mean fever reaction of 3,7 days (1-9 days in 103 goats in different experiments carried out in Guadeloupe) we can hypothesize that ticks become infected themselves on an animal only during the average period of 8 days.

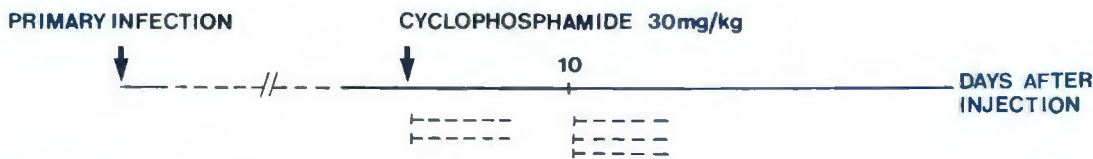


FIG. 2 Immunosuppression

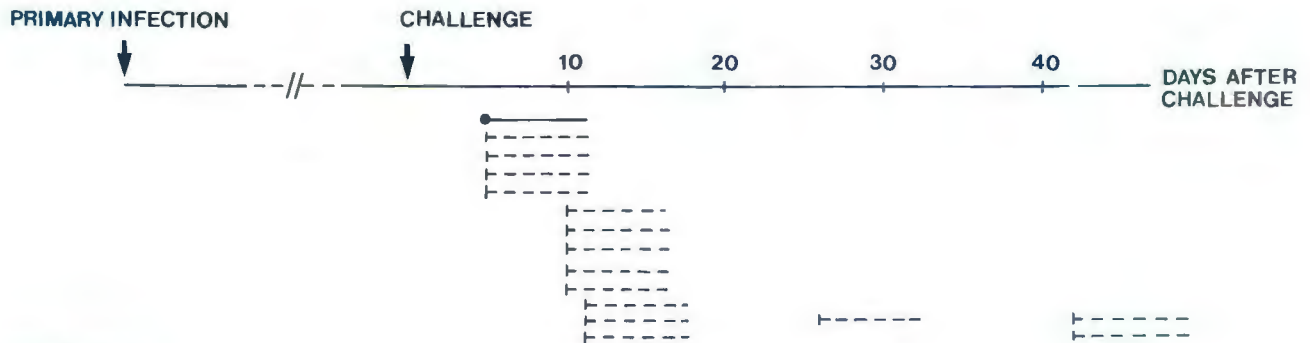


FIG. 3 Challenge

In an endemic area with repeated reinfestations that maintain the immunized status of goats, and considering that the blood of challenged goats is usually not infectious for ticks (with 1 exception only in our experiments), we may hypothesize that the infection of ticks generally occurs only once in the life of a host, during the short period that follows its primary infection.

This may explain in part why the infectious rate of ticks in heartwater endemic areas is so low.

It is possible to conclude that the risk of spreading the disease to heartwater-free areas where vectors are present by introducing recovered animals appears to be low. Recovered goats are possible carriers, but it is doubtful if they are significant reservoirs of infection.

Finally, we should add that our observations may be valid only for the combination of the Creole goat and *A. variegatum*. This tick may not be the most efficient vector of heartwater and this host may not be the most susceptible. Alexander (1931) succeeded in transmitting infection with 2 batches of *A. hebraeum* nymphs (of 2 tested) from larvae fed 7 and 14 days respectively after the recovery of a donor ox. Also, in other ruminant species or breeds, more or less severe relapses may occur after recovery (Neitz, 1968; Neitz, *et al.*, 1947; Du Plessis, 1981b; Uilenberg, 1983; Uilenberg *et al.*, 1985). This could make the infection of tick populations on some recovered animals a possibility. We never observed such a phenomenon in Guadeloupean goats.

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