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Acquisition of immunity in cattle against the blue tick, *Boophilus decoloratus*

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ABSTRACT

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It is well known that ixodid ticks have the ability to induce immunity in their host. We demonstrate, for the first time, that the tick *Boophilus decoloratus* induced immunity in its bovine host, since the mean weight of engorged females fed on naive animals dropped from 201.5 mg, to 173.7 mg and 155.3 mg, for females fed on calves previously exposed once and twice, respectively, to *B. decoloratus* infestations. Ticks which had been transferred from one individual host to another one were able to complete their feeding period on a sensitive host. Such ticks were significantly heavier ($\bar{x}$ 245.2 mg) than those fed on a naive ($\bar{x}$ 201.5 mg) host for the entire normal feeding period. A negative correlation between the mean weight of the engorged female ticks and the level of serum gamma globulins in the host was also demonstrated.

INTRODUCTION

The tick *Boophilus decoloratus*, restricted to the savannas of Africa South of the Sahara, is a vector of *Babesia bigemina* and *Anaplasma marginale* (Walker et al., 1978). The economic importance of this tick, the need for control, and the remarkably difficult and expensive requirements in achieving such control are also well documented (Walker et al., 1978).

A one-host tick (larval, nymphal and adult stages feed in sequence on the same individual host), *B. decoloratus* spends about 21 days feeding on its host prior to drop-off of engorged females (Walker et al., 1978). Eight days after the commencement of this feeding period, the level of the serum gamma globulin in the host is increased (Clarke, unpublished data, 1989). These facts indicate that the adults of *B. decoloratus* which are released as larvae on naive animals might complete their blood meal on an already immune host. The first objective of this study was to find out, if a transfer of ticks from one individual host to another, seven days after the commencement of feeding

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and before an increase in the level of gamma globulin in the host occurred, would produce heavier ticks than when fed for the entire 21 days on naive calves. The second enquiry was whether the mean weight of the engorged *B. decoloratus* female ticks and the level of gamma globulins in the host's serum could serve as indicators for evaluating the immunity status of the bovine host.

MATERIALS AND METHODS

Experimental ticks were collected from cattle and reared in the laboratory. Twenty-thousand, five-week-old unfed larvae were released into bags glued to the backs of each of eight two-month-old Jersey calves kept in experimental cages. Blood from the jugular vein of each calf was collected and analysed. Serum protein electrophoresis (SPE) was done on cellulose acetate membranes (Helena Laboratories, Beaumont, Texas) using a barbital/sodium buffer with a pH of 8.6 (Helena Laboratories). The membranes were scanned with an Appraise Densitometer (Beckman Instruments Fullerton, California) and were separated into the following fractions: albumin, alpha 1 (α_1), alpha 2 (α_2), beta (β) and gamma (γ) globulins. The densitometer automatically calculated the albumin and globulin ratios. Total protein levels were quantitated with an Astra 8 automated analyzer (Beckman Instruments) using the Biuret rate method. (Rechav et al., 1980, 1989; Rechav and Dauth, 1987). Ferrated ticks still attached and prior to moulting were scraped off the backs of the hosts with a pair of watchmakers' forceps and, together with epidermal tissue, transferred to similar bags placed on the secondary hosts. After moulting, the ticks attached immediately to their new hosts, thus allowing constant feeding on sensitive animals. Any feeding ticks that might also have been removed together with the ferrated ticks, failed to re-attach. The ticks were divided into four groups: Group A were fed for seven days on one calf, followed by seven days on a second calf, and then transferred to a third calf for completion of the normal 21-day feeding period; Group B were fed seven days on one calf and then completed the other 14 days on a second calf; Group C fed 14 days on one calf and then completed the remaining seven days on a second calf; and Group D were fed to the entire 21 days on the same individual calf (Table 1). Completion of the first infestation in Group D was followed by two more consecutive infestations on the same calf. In addition, a control group was established which consisted of uninfested calves from which blood samples were taken for comparison with calves on which Groups A, B, C and D were fed. Ticks from each group fed on two calves from which blood samples were taken at various intervals. Blood samples were also obtained from a control group, not infested with ticks for the duration of the experiment. The engorged females were weighed on an electronic balance (Sartorius 1800).

TABLE 1

Feeding periods (days on host) of *Boophilus decoloratus* on individual calf hosts from the various groups tested during the experiments

Group	Host		
	1	2	3
A	7	7	7
B	7	14	–
C	14	7	–
D	21	–	–

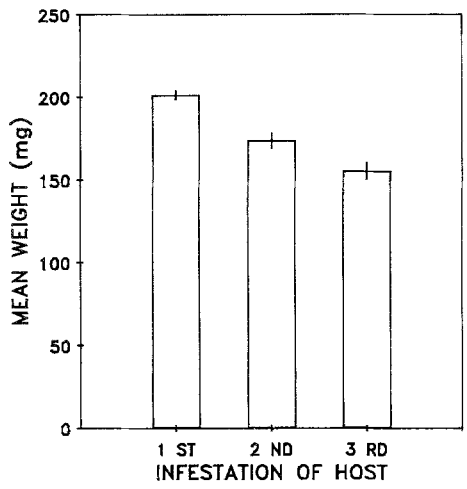

Fig. 1. Weight (mg) of *Boophilus decoloratus* engorged females (mean ± SE) fed on naive calves, and on calves previously exposed once and twice to *B. decoloratus* ticks.

Weights of engorged female ticks and levels of the serum gamma globulin fraction are presented as mean ± standard error. Anova was used for analysing the results, followed by the Newmans-Keuls multiple-range test.

RESULTS

We have shown that cattle acquired immunity to the blue tick *B. decoloratus* following a series of infestations (Fig. 1) (Group D). The mean weight of the engorged females decreased significantly ($P<0.001$), from 201.5 mg for females which fed on naive animals to 173.7 mg (a decrease of 14.8%) and 155.3 mg (a decrease of 30.0%) for females fed on calves previously exposed, once and twice respectively, to *B. decoloratus* infestations (Fig. 1) (Group D). Also, a negative correlation between the level of gamma globulins in the

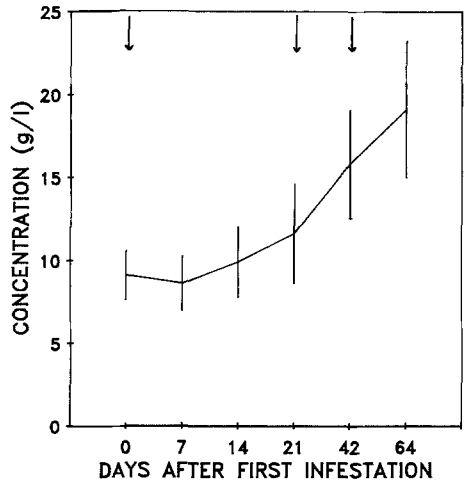

Fig. 2. Serum gamma globulin levels (g/l) (mean ± SE) of calves at various intervals after the first infestation with larvae of *Boophilus decoloratus*. Arrows represent days of repeated infestations after the day of the first infestation (day 0).

TABLE 2

Mean weights[1] of *Boophilus decoloratus* engorged females (mg ± SE) and changes in the concentration of gamma globulins[1] and total proteins[1] (g/l) in the sera of calves from the various groups at the end of the feeding period

Parameter	Group				
	A	B	C	D	Control
Mean weight	245.2 ± 5.19^a	222.2 ± 6.27^b	202.8 ± 3.22^c	201.5 ± 2.67^c	–
Gamma globulin	$+0.1^a$	$+0.7^a$	$+3.6^b$	$+7.0^c$	$+0.2^a$
Total proteins	-0.3^a	$+1.1^a$	$+3.9^b$	$+7.5^c$	$+0.1^a$

[1]Figures with differing superscripts are significantly different (Anova followed by Newmans-Keuls multiple-range test).

sera of the calves (Fig. 2) and the mean weight of the female ticks was evident (Fig. 1). The mean weight of engorged females and the changes in the level of the gamma globulins in the sera of the hosts on which ticks in Groups A, B, C and D fed are presented in Table 2. Ticks fed on calves from Group A were significantly ($P < 0.005$), heavier than ticks fed on animals from Groups B or C, or of those from first infestation (Group D). Furthermore, a high mean weight in engorged females (245.2 ± 5.19 mg, an increase of 17.9%) was correlated with a low level of gamma globulins similar to that which existed

in cattle prior to their exposure to ticks (9.4 ± 0.96 g/l) (Fig. 1). Changes in levels of total proteins (Table 2) reflected the changes in the concentration of gamma globulins.

DISCUSSION

We have shown, for the first time, that the blue tick *B. decoloratus* can induce immunity in its bovine host. This immunity was expressed by a significant decrease in the mean weight of the engorged female ticks fed on resistant calves when compared with that of female ticks fed on naive animals – an accepted indicator of an increase in host immunity. In addition, an increase in the level of the serum gamma globulins in resistant hosts was demonstrated.

It seems that the feeding performance of ticks infesting calves was affected by substances present in the gamma globulin fraction. These substances, probably protective antibodies, are produced by the host as a response to the antigens introduced by the feeding ticks. This humoral response probably complements that of the cellular (Ribeiro, 1989a). Together, these two contribute to the total immune response of the host. It is also clear that the time required by the calves to produce antibodies against the blue tick (8 days) is shorter than the feeding period of the ticks (21 days). Success in producing heavier females on three successive sensitive hosts, compared to females produced by a normal 21-day feeding period on a single host, could contribute to the understanding of the mechanisms involved in host immunity. It might also, together with research into the purification of the antigens from the ticks (Kemp et al., 1986, 1989; Opdebeeck et al., 1988; Willadsen et al., 1988), stimulate the pharmacological characteristics of the substances found in tick saliva (Ribeiro, 1989b) and the time required by the host to produce antibodies (Rechav et al., 1989) to the stimulation of the production of vaccines for immunising cattle.

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