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PROBLEMS WITH THE INTERPRETATION OF EPIDEMIOLOGICAL DATA IN HEARTWATER: A STUDY ON 23 FARMS

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ABSTRACT

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In an epidemiological study undertaken on 23 farms where heartwater occurs endemically, it was found that on an overall average, antibodies to *Cowdria ruminantium* were detected in 64,3 % of the cattle, 6 adult *Amblyomma hebraeum* ticks were counted per animal and 7,0 % of ticks were infected with the heartwater agent. It was found that the seropositivity of the animals was determined largely by the tick loads to which they were subjected and that the influence of the tick *C. ruminantium* infection rate was less evident.

There was no parallel between the prevalence of heartwater on the farms and the immune status of the animals. In general, higher tick counts were recorded in herds where strategic tick control is practised than on farms with a total tick control programme. The method of tick control did not, however, appear to influence the immune status of the cattle, the tick infection rate, or the prevalence of heartwater.

INTRODUCTION

Since nymphal and adult stages of *Amblyomma hebraeum* are of cardinal importance in the transmission of heartwater (HW), their control plays a vital role in the epidemiology of the disease. Protagonists of total tick control on one hand maintain that a disease-free, or at least a minimal-disease situation, can be achieved by means of an intensive dipping programme aimed at tick eradication. Those in favour of strategic tick control, on the other hand, argue that tick eradication is not always feasible and warn that total tick control is bound to compromise the tick-mediated immunity of cattle and create susceptible herds.

In an earlier investigation to determine whether the immune status of cattle can be related to the tick control practised (Du Plessis, 1982) it was found that there was no correlation between the number of cattle that were serologically positive to *Cowdria ruminantium* and the severity of the tick control to which they were subjected. In this study information regarding tick control, tick loads and the prevalence of heartwater was thought to be inadequate. It was decided to obtain a better knowledge of the role played by ticks in the epidemiology of the disease by making tick counts and determining *C. ruminantium* infection rates of ticks on selected farms where good management and record keeping would facilitate more accurate information.

MATERIALS AND METHODS

Selection of farms

Twenty-three farms with closed herds were selected in HW endemic regions of the Transvaal and Natal. The selection was based on knowledge of the level of management on these farms and on the opinion that reliance could be placed on the information supplied by the owners. With the exception of 2 farms, herds where HW immunization was not practised, were selected.

On each farm 15 4–6-month-old calves and 15 16- to 18-month-old heifers or oxen were selected for the study. Four- to 6-month-old calves were selected because, on the one hand, they were old enough so that a positive indirect fluorescent antibody (IFA) test on their sera could be attributed to tick infection and not to passively transferred colostral antibodies (Du Plessis, 1984). On the other hand it was argued that at this young age a positive serological test would more likely reflect a single tick infection than in older animals, and could therefore be correlated more accurately with the tick numbers at the time of collection. The older age group was included to compare their serology and tick numbers with that of the calves.

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To ensure that the tick counts recorded would be a true reflection of the tick loads carried by the cattle subjected to the dipping programme practised on a particular farm, the visits were planned in such a way that the day on which the ticks were counted, and the samples collected coincided as closely as possible with the day on which the animals were due to be dipped in line with the dipping programme in force on each farm.

It must be pointed out that this investigation was completed during the course of February 1983 in the midst of one of the worst droughts ever experienced in southern Africa. Although the adult stage of *A. hebraeum* reaches its peak at this time of the year (Londt, Horak & De Villiers, 1979) the counts in all probability were much reduced. Since the same dry conditions prevailed throughout the time and localities of the investigation, the results should be comparable.

As each animal was cast for the tick count, serum and adult *A. hebraeum* ticks were collected from the animals.

Correlation between seropositivity and immunity

Since the IFA test is highly specific for the detection of antibodies to *C. ruminantium* (Du Plessis & Malan, 1987), it can be accepted that the seropositivity of unvaccinated cattle exposed to *A. hebraeum* in the field is due to infection through the tick. In the context of the role played by the tick in the epidemiology of HW, it is essential to determine the relationship between a positive serological reaction and the immunity of cattle. To ascertain whether seropositivity can be equated with resistance to challenge through the tick, the data obtained in two previous studies had to be taken into consideration in the present study.

In one of these (Du Plessis, Bezuidenhout & Lüdemann, 1984), 2 groups of cattle that had been immunized as calves were subsequently challenged artificially, the one after having been exposed to tick challenge and the other not. Equal numbers of unvaccinated control animals in each group were challenged at the same time and with the same challenge inoculum as the vaccinated animals. Serum antibody and conglutinin levels on the day of challenge were determined. The correlation between the serology of these animals, their conglutinin levels and their reactions to the challenge are relevant to the present study.

The effect of repeated challenge on antibody detectable with the IFA test is another aspect of the correlation between serology and immunity that must be taken into consideration in the present study. Knowledge on this aspect was gained from the results of experiments primarily conducted to study the role played by conglutinin in resistance to HW (Du Plessis, 1985b). Twenty 8-month-old HW-susceptible Bonsmara bull calves were

TABLE 1 Correlation between seropositivity and resistance to challenge

Tick challenge	No. of animals		Age in months at challenge	IFA positive		IFA negative	
	Immunized	Controls		No. resistant	No. susceptible	No. resistant	No. susceptible
Absent	9	—	6	6 (0)	0	2 (0)	1 (0)
	—	10	6	0	0	7 (0)	3 (0)
	10	—	12	2 (0)	0	5 (1)[1]	3 (0)
	—	10	12	0	0	1 (1)	9 (0)
	10	—	24	0	0	10 (9)	0
	—	9	24	0	0	6 (6)	3 (0)
Present	10	—	6	3 (0)	0	5 (0)	2 (0)
	—	10	6	7 (0)	0	2 (2)	1 (0)
	10	—	12	5 (3)	0	2 (2)	3 (0)
	—	10	12	3 (2)	0	2 (1)	5 (0)
	9	—	24	1 (1)	0	8 (7)	0
	—	9	24	4 (4)	0	5 (5)	0
Total	58	58		31 (10)	0	55 (34)	30 (0)

[1] 5 (1) = One out of 5 animals had a conglutinin titre of 1:640 or higher

inoculated with *C. ruminantium* infected sheep's blood. They were challenged 124 days later and re-challenged at intervals between the first and the second challenge that varied from 94 days to 323 days. Their sera were submitted to the IFA test at monthly intervals after the first challenge, and to the conglutinin test when they were infected and at each challenge. The day of the second challenge was chosen so that the period that elapsed between the first negative IFA test at a serum dilution of 1:20 and the 2nd challenge varied from 3–7 months.

Serology

The ratio of animals serologically positive to *C. ruminantium,* as an indication of the heartwater immune status of the cattle on each farm *(vide infra),* was determined by subjecting the sera, diluted to 1:20, to the IFA test, as previously described and modified (Du Plessis, 1982; 1985a).

Tick counts

The animals were cast and all the adult *A. hebraeum* males and females counted on the whole body of each of the 30 animals selected on a farm. To ascertain that the tick numbers recorded were a true reflection of the tick loads to which the animals were subjected, a second count was carried out on 7 of the farms at an interval after the first count which corresponded with the frequency of the dipping practised on the farm.

C. ruminantium *infection rate of ticks*

The ratio of ticks infected with the HW agent was determined for each farm according to the method described previously (Du Plessis, 1985a).

Additional data supplied by stock owner

The prevalence of HW was classified as low or moderate according to the number of deaths recorded by the stock owner over the past 3–5 years. Accurate mortality records were available for 2 of the farms only, while estimates by the stock owners were recorded for the rest. Based on the mortality rates recorded on these 2 farms, the prevalence of the disease was classified as low on those farms where less than 1 % mortalities in relation to the number of cattle on a farm had occurred and as moderate on those where more than 1 % mortalities had been recorded.

The tick control on a particular farm was rated "total" where weekly or fortnightly dipping was strictly practised throughout the year and "strategic" where animals were dipped only when some of the animals in the herd were seen to carry ticks. Some of the farms had switched from total to strategic control during the previous 12 months.

Results

Correlation between seropositivity and immunity

It can be seen from Table 1 that every one of 31 out of 116 cattle that tested serologically positive on the day when they were challenged, was resistant to the challenge, whereas only 30 out of 85 that were serologically negative were susceptible. The other 55 seronegative cattle were resistant to the challenge. It is important to note though that not a single seropositive animal reacted to the challenge.

It should be noted that 34 out of the 55 seronegative resistant animals had conglutinin titres of 1:640 or higher on the day of challenge, whereas none of the 30 seronegative susceptible and only 10 out of the 31 seropositive resistant animals had conglutinin levels of this magnitude.

The resistance to challenge of 20 serologically positive year-old cattle that had been infected artificially 4 months earlier (Table 2) lends further support to the finding that cattle with detectable levels of antibodies to *C. ruminantium* are consistently immune. On the other hand, a second challenge carried out on these animals 3–7 months after they had become seronegative, confirmed the finding reflected in Table 1 that serologically negative cattle are not necessarily susceptible, since they were all immune to the second challenge. The resistance of only some of these animals could be attributed to conglutinin, since only 9 of them had titres below 1:640.

Two further observations that were made on these 20 cattle also concern the significance of antibody levels detectable with the IFA test. First, it is evident from the IFA titres given in Table 2 that there was no dramatic rise in antibody levels when these animals were challenged the first time, and even less after they were challenged a second time. Secondly, although with one exception, antibody titres varying from 1:20 to 1:1280 were detectable in the serum of these animals 28 days after being challenged, they became seronegative within 4–7 months thereafter.

In the present study it can therefore be accepted that the seropositivity of the cattle on the 23 farms is a true reflection of their immunity to HW acquired through the tick, i.e. that the serologically positive animals had been infected with the HW agent through the tick and would be immune to further challenge. It must be borne in mind that such animals become seronegative in the absence of re-infection and remain resistant to challenge for as long as 6 months after testing negative.

TABLE 2 Effects of repeated challenge on antibody levels detectable with the IFA test

Animal No.	Reaction category Reciprocal of conglutinin titre in parenthesis			Reciprocal of IFA titre				Interval in days between		
	Infection	Challenge 1	Challenge 2	42 days after infection	At challenge 1	28 days after challenge 1	60 days after challenge 2	Challenge 1 and sero-negativity	Challenge 1 & 2	Seronega-tivity and challenge 2
1	III (320)[1]	IV (640)	IV (1280)	320	20	20	—	142	229	87
2	IV (160)	III (640)	IV (640)	> 320	80	320	20	180	260	80
3	III (320)	IV (640)	III (640)	320	20	20	20	170	94	141
4	III (320)	III (320)	IV (320)	320	80	20	20	113	260	92
5	III (160)	IV (160)	IV (160)	320	80	80	—	170	229	141
6	IV (640)	IV (160)	IV (640)	320	80	20	20	142	229	115
7	III (640)	IV (2560)	IV (2560)	320	80	320	20	113	260	92
8	III (320)	IV (640)	IV (640)	320	80	20	20	142	292	115
9	III (160)	IV (160)	III (160)	320	80	320	< 20	170	292	141
10	III (160)	IV (320)	IV (320)	320	20	80	< 20	170	292	141
11	IV (160)	IV (80)	IV (160)	320	20	80	20	170	323	141
12	III (320)	IV (320)	IV (320)	320	80	1280	20	180	323	175
13	III (320)	IV (640)	IV (2560)	320	80	< 20	—	< 30	210	< 30
14	IV (160)	IV (320)	IV (320)	80	20	80	20	210	292	200
15	III (80)	IV (320)	IV (1280)	320	80	20	20	180	260	175
16	IV (40)	IV (80)	IV (80)	320	80	80	20	180	260	175
17	III (320)	IV (160)	IV (320)	320	80	320	20	> 210	323	200
18	IV (320)	IV (1280)	IV (2560)	320	80	320	20	180	323	175
19	IV (320)	IV (320)	IV (1280)	320	80	80	20	113	260	92
20	III (640)	IV (2560)	IV (2560)	320	80	80	20	113	260	92

[1] III (320) = animal 1 showed a category III reaction and had a conglutinin titre of 1:320 when it was infected

TABLE 3 Percentage seropositivity, tick loads, percentage ticks infected, type of tick control practised and prevalence of heartwater on 23 farms listed in order of percentage seropositivity of calves

Farm No.	Cattle breed	% seropositivity		Average No. of ticks per animal			% infected ticks	Tick control		Prevalence of heartwater disease
		Animals aged		Animals aged		Per farm		Total	Strategic	
		4–6 months	16–18 months	4–6 months	16–18 months					
1	Friesian	100	100	50	15	32,5	10		+	+[1]
2[2]	Brahman ×	100	80	1,8	1,5	1,6	10		+	++
3	Brahman ×	100	80	0,5	3,5	2	30	+		++
4	Brahman ×	100	60	3	47	25	10		+	+
5	Brahman ×	100	50	2,1	18,5	10,3	20		+	+
6	Brahman ×	90	100	3	10	6,5	1,6		+	++
7	Simmentaler	90	60	0,3	0,9	0,6	10	+		+
8	Bonsmara ×	90	60	1,5	12,5	7	< 5		+	+
9	Brahman ×	80	90	0,3	1,8	1	< 5		+	+
10	Bonsmara	70	100	0,4	2,8	1,6	10	+		++
11	Bonsmara	70	60	17	34	25,5	5		+	+
12	Brahman ×	60	100	0,1	2,1	1,1	5	+		++
13	Brahman ×	60	90	0,9	3,1	2	5	+		++
14	Various breeds	60	70	0,7	3,9	2,3	5		+	+
15[2]	Bonsmara	50	70	10,5	9,7	10,1	2,5		+	+
16	Brahman ×	50	50	0,9	3,2	2,1	5	+		++
17	Drakensberger	40	100	1,1	4,3	2,7	10		+[3]	++
18	South Devon/Afrikander ×	40	40	0	0,2	0,1	< 5	+		+
19	Brahman ×	30	90	1,3	1,5	1,4	10	+		+
20	Bonsmara	15	25	0,9	1,4	1,2	2		+[3]	++
21	Brahman ×	10	70	0,1	0,1	0,1	< 5		+[3]	++
22	Brahman ×	10	0	0,1	2,7	1,4	2,5	+		++
23	Drakensberger	0	0	0	0	0	–	+		+
	Average	61,6	67,1	4,2	7,8	6	7,0[4]			

[1] Low (+) and moderate (++) prevalence of heartwater
[2] Calf immunization practised
[3] Had switched to strategic dipping during past 18 months
[4] % of farm 6 allotted to farms 8, 9, 18, 21 & 23 to calculate average

TABLE 4 Heartwater mortalities on farms 17 and 20

Farm No.	Year	Age of cattle in months				Total	% of herd
		<7	7–12	13–24	>24		
17	1977	1	2	1		4	0,5
	1978	7	7	3		17	1,7
	1979	5	5	2	1	13	1,2
	1980	15	6	5		26	2,4
	1981	4	6	4		14	1,3
	1982	3	10	6		19	1,6
	1983[1]	4	8	4		16	1,4
	1984	1	5	4		10	1,1
	Total	40	49	29	1	119	1,4[2]
20	1970	4	9	5		18	1,0
	1971	6	3	1		10	0,6
	1972		11	14		25	1,6
	1973	7	21	3		31	2,4
	1974	4	4	8	1	17	1,2
	1975	6		3		9	0,7
	1976	8	2	7	1	18	1,3
	1977			10		10	0,7
	1978		2	13	1	16	1,3
	1979	1	3	9	1	14	1,1
	1980	2	4	7	1	14	1,3
	1981	3		2		5	0,5
	1982	11	20	19	3	53	5,1
	1983[1]	7	2	3	1	13	1,6
	1984	6	3	1	2	12	2,0
	Total	65	84	105	11	265	1,5[2]

[1] Survey carried out in February 1983

[2] Average % for farm

Serology on 23 farms

The percentages of calves and older animals found to be serologically positive to *C. ruminantium* are given in Table 3. It can be seen that in the case of 11 herds (farms 1, 2, 3, 4, 5, 7, 8, 11, 16, 18 and 22) a percentage seropositivity was recorded in the 4- to 6-month-old calves that was equal to or higher than that detected in the older animals.

Tick counts

The average number of ticks per animals per farm, calculated from the averages of the 2 age groups, varied from 0 to 32,5 (Table 3). An overall average of 6 ticks per animal was recorded on the 23 farms. With rare exceptions (farms 1, 2 and 15) the older animals carried many more ticks than the calves. On farms 1, 3, 6, 7, 13, 16 and 22 where a second count was carried out, average counts of 16; 15,1; 6; 3,5; 1,7; 0,8 and 1,4 were recorded. Compared with the overall average for all the farms, which was used as the criterion in determining the relationship of the immune status of the herds to the tick counts, the differences between the 2 counts were insignificant.

C. ruminantium *infection rate of ticks*

It can be seen from Table 3 that the infection rate varied from 1,6 to 30 %. No ticks were found on the 30 animals examined on farm 23 and in an effort to ascertain whether the vector of HW did occur on this farm, additional animals were examined and one dead *A. herbraeum* male was found on a bull.

Only 20 ticks were available for examination in the case of each of 4 farms (8, 9, 18 and 21) and they were all found to be negative. In the case of farms 6, 15, 20 and 22 where the first 20 ticks examined were negative and sufficient numbers were available, additional batches of 10 were examined until a positive tick was identified.

Relationship between seropositivity, tick loads and tick infection rates

Taking the seropositivity of the 4- to 6-month-old calves on each farm as an indication of the percentage of animals that acquired an immunity to *C. ruminantium* through the tick on each farm, the results given in Table 3 suggest that this immunity is related to the number of ticks to which they were exposed. On all except one of the 12 farms where a seropositivity below 61,5 % (the average for the 4- to 6-month-old calves) was recorded, the average tick counts per farm were well below the overall average of 6 ticks per animal recorded on all 23 farms. The highest tick counts (farms 1, 4, 5, 6, 8 and 11) were all associated with seropositivity percentages above the average for the calves. There were, however, several farms (2, 3, 7, 9 and 10) on which low tick counts were associated with high percentages of seropositivity.

The relationship between the *C. ruminantium* infection rate of the ticks and the calf percentage seropositivity on each farm is also shown in Table 3. Some of the highest percentages of seropositivity (farms 1, 2, 3, 4, 5, 7 and 10) were associated with tick infection rates above the overall average of 7,0 % recorded in the study. With the exception of farms 17 and 19 a lower than average infection rate was recorded on all 13 farms (12–23) where the seropositivity was below the overall average of 61,5 %. A higher than average seropositivity on farms 6, 8, 9 and 11 was accompanied by a tick infection rate below 7,0 %.

Seropositivity, tick control and prevalence of heartwater

The relationship between the seropositivity of the cattle, the type of tick control practised and the prevalence of HW are reflected in Tables 3 and 4.

Out of the 10 farms where total tick control was practised, the seropositivity of the 4- to 6-month-old calves was below the average of 61,5 % on no less than 7 of them. On 3 further farms (17, 20 and 21) where seropositivity was low, tick control had been switched from total to strategic as recently as 18 months prior to the survey. Although a high seropositivity was recorded on 3 farms (3, 7 and 10) where total tick control was practised, these findings suggest that the low seropositivity on these 10 farms is associated with the total tick control practised.

Accurate records of the number of deaths due to HW prior and subsequent to the investigation were available for farms 17 and 20 only and are shown in Table 4. The annual incidence was calculated as a percentage of the total number of deaths per number of cattle in the herd. It can be seen that the incidence varied from 0,5 to 5,1 % with an overall prevalence of 1,4 and 1,5 % respectively. In the case of both farms the disease was much more prevalent in animals under a year old, markedly fewer deaths in cattle older than 2 years having been recorded. It is interesting to note that although both these farms had switched from a total to a strategic tick control programme 18 months prior to the investigation, i.e. in 1981, there was no noticeable decrease in the incidence of heartwater during the ensuing 3 years.

There appears to be no correlation between the prevalence of HW and any of the other parameters studied.

Except for the Friesland cattle on farm 1 where the heaviest tick load was recorded, none of the epidemiological parameters studied appeared to be related to any particular breed of cattle. Although Brahman-cross cattle constituted almost 50 % of the herds in this study, they showed no particular tendency with regard to any of the features studied.

The relatively low percentage seropositivity and tick infection rate on farm 15, one of the 2 farms where calf vaccination is practised, is noteworthy.

Discussion

The detection of antibodies to *C. ruminantium* by means of the IFA test in the present study was used to determine the proportion of animals in each herd that had been infected through ticks. The finding in earlier experiments that without a single exception all 31 serologically positive cattle infected either artificially or through the tick were fully resistant to challenge, justifies the conclusion that seropositivity can be correlated with immunity.

While all serologically positive cattle are immune to HW, all those that are serologically negative are not necessarily susceptible, and resistance to either artificial or natural challenge does therefore not always correlate with the serological reaction. It has previously been found (Du Plessis, 1982; Du Plessis *et al.*, 1984) and the results in the present study have confirmed that on the one hand significant numbers of serologically negative cattle are resistant to artificial challenge and on the other that serologically positive cattle become seronegative from 6 months after both experimental and natural infection. The percentages of seropositive animals on the 23 farms are therefore not necessarily a true reflection of the actual proportion of animals resistant to HW and more animals than those that were serologically positive would therefore have resisted future tick challenge.

To obviate the disappearance of antibodies detectable with the IFA test after infection, calves younger than 6 months were sampled and the presence of antibodies in their sera, rather than that of the older animals, taken as proof of infection through the tick. The data thus obtained were used to study the relationship between immunity and the other parameters.

The seropositivity of the calves as an indication of the rate at which they had been infected through the tick appeared to be related to the numbers of ticks to which they had been exposed. On all except one of the 12 farms where a seropositivity below the average for this age-group was recorded, tick counts were well below the average of all the animals examined. The highest tick counts recorded during the study were all associated with seropositivity percentages above the average for the calves. There were a few exceptions to this relationship, since there were several farms on which low tick counts were associated with high percentages of seropositivity.

For two reasons the conclusion that the immunity of cattle to HW is largely determined by the numbers of *Amblyomma* ticks to which they are exposed, must at this stage be drawn with caution. First, only one count was made on the majority of the farms and although a second count on 7 of them revealed no significant differences with the first, additional counts on the calves at earlier intervals during their lives might have given a more reliable indication of the tick loads to which they had been subjected. Second, only adult ticks were counted and since *Amblyomma* nymphae also transmit *C. ruminantium,* the numbers of ticks at this stage of development should perhaps also have been taken into consideration. It must, however, be pointed out that the contribution by nymphae to the seropositivity of the calves could only have been minimal compared to that of the adult ticks, since the calves were not exposed to peak nymphal activity from the beginning of May until the end of September (Baker & Ducasse, 1967), whereas adult ticks were at their peak at the time when the counts were made. The single count and the omission of nymphae should not detract much from the conclusion that the tick numbers to which cattle are exposed play an important role in their immunity to HW.

The average of 7,0 % of ticks infected with *C. ruminantium* that was recorded in this study is only slightly higher than the infection rate of 5 % reported in an earlier study (Du Plessis, 1985a), in which a portion of the ticks examined in the present study were used. The tick infection rate, believed to play an important role in the epidemiology of HW (Uilenberg, 1983) appears to have influenced the percentage of 4- to 6-month-old calves that were serologically positive. Some of the farms with the highest percentages of seropositivity were associated with tick infection rates above the 7,0 % and with only 2 exceptions a lower than average infection rate was recorded on all 13 farms where the seropositivity was below the average of 61,5 %. This relationship was, however, not absolute since on 4 farms high seropositivity was associated with tick infection rates below the average.

It was found that the type of tick control practised on the farms was reflected in the immune status of the herds, since low seropositivity was recorded on 10 of the farms where total tick control was practised and where below average numbers of ticks were counted. On all except 3 of these farms the prevalence of heartwater was moderate, whereas on the majority of the farms where strategic tick control was practised, the prevalence was low. While it can be concluded from this study that the tick load to which cattle are exposed is probably the most important single factor that determines the immune status of a herd, it was not possible to obtain an indication of the approximate number of ticks required to maintain herd immunity under a strategic tick control programme, a prerequisite if this type of control is to be advocated.

If the concept of enzootic stability, applied to the epidemiology of babesiosis (Mahoney, 1977) were used in HW to indicate a stable situation in a herd where a high percentage of serologically positive and therefore immune animals is associated with no or very few HW mortalities, the findings in this study present a problem. On some of the farms a moderate prevalence of HW was reported and on the rest only a mild prevalence, but this variation occurred across the entire spectrum of seropositivity. The disease was no less prevalent in some of the herds where 80–100 % of calves were serologically positive and on serveral farms where the seropositivity was 60 % or less, the prevalence was low. This lack of correspondence between disease and immunity was also observed on the 2 farms where accurate records of mortalities were available. Although 70 % of both the younger and older age group were serologically positive in the case of the one and only 20 % in the other, the disease prevalence on the farms was almost identical.

It seems that in HW an enzootically stable situation is perhaps never reached. This is probably because more than just one factor plays a role in the resistance of cattle to HW (Du Plessis, 1985b). The suckling-calf resistance, which declines with advancing age and has normally disappeared by 6–8 months of age, conglutinin which confers resistance in adulthood, and possibly even other factors are responsible for the non-specific component of resistance. Specific immunity, acquired either artificially or through the tick, accounts for the specific component. The absence or low prevalence of HW mortalities and clinical cases on a particular farm would therefore be determined by the presence of one or more of these resistance factors. Depending on the breed and age of cattle involved and the proportion of the herd infected through the tick, the level of resistance can vary considerably. This variation in conjunction with the fact that the percentage of animals positive to the IFA test only reflects the specific component of their resistance possibly explains the lack of correlation between seropositivity and disease prevalence.

This study entailed a considerable amount of work and much data was collected. The fact that few conclusions could be drawn with certainty, however, shows the complexity of the factors involved in the epidemiology of HW and underlines the necessity of pinpointing the shortcomings in the design of the experiments. It would appear that the nymphal stage of *Amblyomma* should also be taken into account, not only as far as tick counts are concerned but their *C. ruminantium* infection rate should also be determined. Regular tick counts accompanied by serological monitoring over the course of several years in a single herd under good management will ensure a more complete picture. It would also facilitate accurate records of mortalities as well as clinical cases of HW. By assaying the conglutinin levels of the experimental animals at regular intervals, the role of at least one of the 2 non-specific resistance factors would be monitored enabling a more accurate assessment of the relationship between the immunity of the herd and the rate of *C. ruminantium* infection through the tick as shown by the IFA test.

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