The impact of 2 dipping systems on endemic stability to bovine babesiosis and anaplasmosis in cattle in 4 communally grazed areas in Limpopo Province, South Africa

B O Rikhotso[a,b], W H Stoltsz[a], N R Bryson[a*] and J E M Sommerville[c]

ABSTRACT

A 12-month study was conducted in 4 communal grazing areas in the Bushbuckridge region, Limpopo Province, South Africa. The main objective was to investigate the impact of reduced acaricide application on endemic stability to bovine babesiosis (*Babesia bigemina* and *Babesia bovis*) and anaplasmosis (*Anaplasma marginale*) in the local cattle population. To this end 60 cattle in each communal grazing area were bled at the beginning and the conclusion of the experimental period and their sera were assayed for *B. bovis, B. bigemina* and *Anaplasma* antibodies. Cattle in the intensively dipped group were dipped 26 times and maintained on a 14-day dipping interval throughout the study, whereas cattle in the strategically dipped group were dipped only 13 times. Three cattle, from which adult ticks were collected, were selected from each village, while immature ticks were collected by drag-sampling the surrounding vegetation. During the dipping process, a questionnaire aimed at assessing the prevalence of clinical cases of tick-borne disease, abscesses and mortalities was completed by an Animal Health Technician at each diptank. An increase in seroprevalence to *B. bovis* and *B. bigemina* and a decrease in seroprevalence to *Anaplasma* was detected in the strategically dipped group while in the intensively dipped group the converse was true. *Amblyomma hebraeum* was the most numerous tick species on the cattle, and *Rhipicephalus (Boophilus) microplus* was more plentiful than *Rhipicephalus (Boophilus) decoloratus.* Drag samples yielded more immature stages of *A. hebraeum* than of *Rhipicephalus (Boophilus)* spp. The incidence of clinical cases of tick-borne disease and of abscesses increased in the strategically dipped group at the start of the survey.

Key words: *Amblyomma hebraeum, Anaplasma marginale, Babesia bovis, Babesia bigemina,* cattle, communal grazing, endemic stability, *Rhipicephalus (Boophilus) decoloratus, Rhipicephalus (Boophilus) microplus,* strategic dipping.

Rikhotso B O, Stoltsz W H, Bryson N R, Sommerville J E M **The impact of 2 dipping systems on endemic stability to bovine babesiosis and anaplasmosis in cattle in 4 communally grazed areas in Limpopo Province, South Africa**. *Journal of the South African Veterinary Association* (2005) 76(4): 217–223 (En.). Department of Veterinary Tropical Diseases, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, Onderstepoort, 0110 South Africa.

INTRODUCTION

Tick infestation and the diseases transmitted by ticks are a major problem for farmers in the tropical and subtropical regions of the world and are widespread in Africa[12,26]. Ticks and tick-borne diseases (TBD) cause considerable economic losses to owners in those areas of southern Africa in which cattle are communally grazed, and the occurrence of tick worry, abscesses and even mortality is often high[31].

Three economically important TBD occur in the region, namely bovine babesiosis, caused by *Babesia bovis* and *Babesia bigemina,* bovine anaplasmosis caused by *Anaplasma marginale*, and heartwater or cowdriosis, caused by *Erhlichia (Cowdria) ruminatium. Babesia bovis* is transmitted by *Rhipicephalus (Boophilus) microplus*, the only known vector in southern Africa[38], *B. bigemina* by *Rhipicephalus (Boophilus) decoloratus* and *R. (B.) microplus*[30] and to a much lesser extent by *Rhipicephalus evertsi evertsi*[7] and *A. marginale* chiefly by *R. (B.) decoloratus* and *R. (B.) microplus,* and to a lesser extent by *Hyalomma marginatum rufipes, R. evertsi evertsi* and *Rhipicephalus simus.* Mechanical transmission by biting flies also occurs. In South Africa *E. (Cowdria) ruminantium* is transmitted by *Amblyomma hebraeum.*

Livestock production in southern Africa is heavily dependent on improved animal health and this entails good tick and TBD control. Many commercial and some rural subsistence farmers use regular short-interval dipping to keep their cattle virtually tick-free. Intensive dipping policies were first instituted during the East Coast fever (*Theileria parva*) era and this has led to endemic instability to many of the TBD[31]. More recently, however, there has been a shift towards strategic and threshold tick control with acaricides applied less frequently during periods of low tick abundance and more frequently during the critical times of the year to avoid the damaging effects of adult ticks[15,29].

In the Bushbuckridge region, farmers were previously dependent on the Department of Veterinary Services in Limpopo Province, South Africa, for the free provision of acaricide, maintenance of diptanks and for supplying diptank personnel and other labour. In extensive beef production systems, especially on commercial ranches in southern Africa, the gathering of cattle for dipping is labour-intensive and costly[31]. Furthermore, farmers may suffer production losses as a result of stress, abortions, drowning and physical injuries and may also lose a day's animal traction and labour on dipping days. Many commercial beef farmers would like to move away from intensive dipping, but are not convinced that alternative control strategies are cost-effective and do not carry a high risk of outbreaks of TBD[32].

The aforementioned are some of the important reasons for re-appraising current tick control strategies in Africa. The alternative approaches emphasise the maintenance of endemic stability (75 % of animals in a herd are seropositive), the use of vaccines against TBD, and the introduction of tick-resistant cattle. Where vector density is high, infection with TBD is common and usually occurs early in the host's life, accompanied by reduced morbidity and mortality[31,34]. Stability to bovine babesiosis, anaplasmosis and heartwater

[a]Department of Veterinary Tropical Diseases, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, Onderstepoort, 0110 South Africa.

[b]Department of Veterinary Services, Limpopo Provincial Government, Private Bag X1321, Thulamahashe, 1365 South Africa.

[c]Department of Statistics, Faculty of Agriculture and Life Sciences, University of Pretoria, Pretoria, 0002 South Africa.

Author for correspondence.
E-mail: nigel.bryson@up.ac.za

Received: July 2005. Accepted: October 2005.

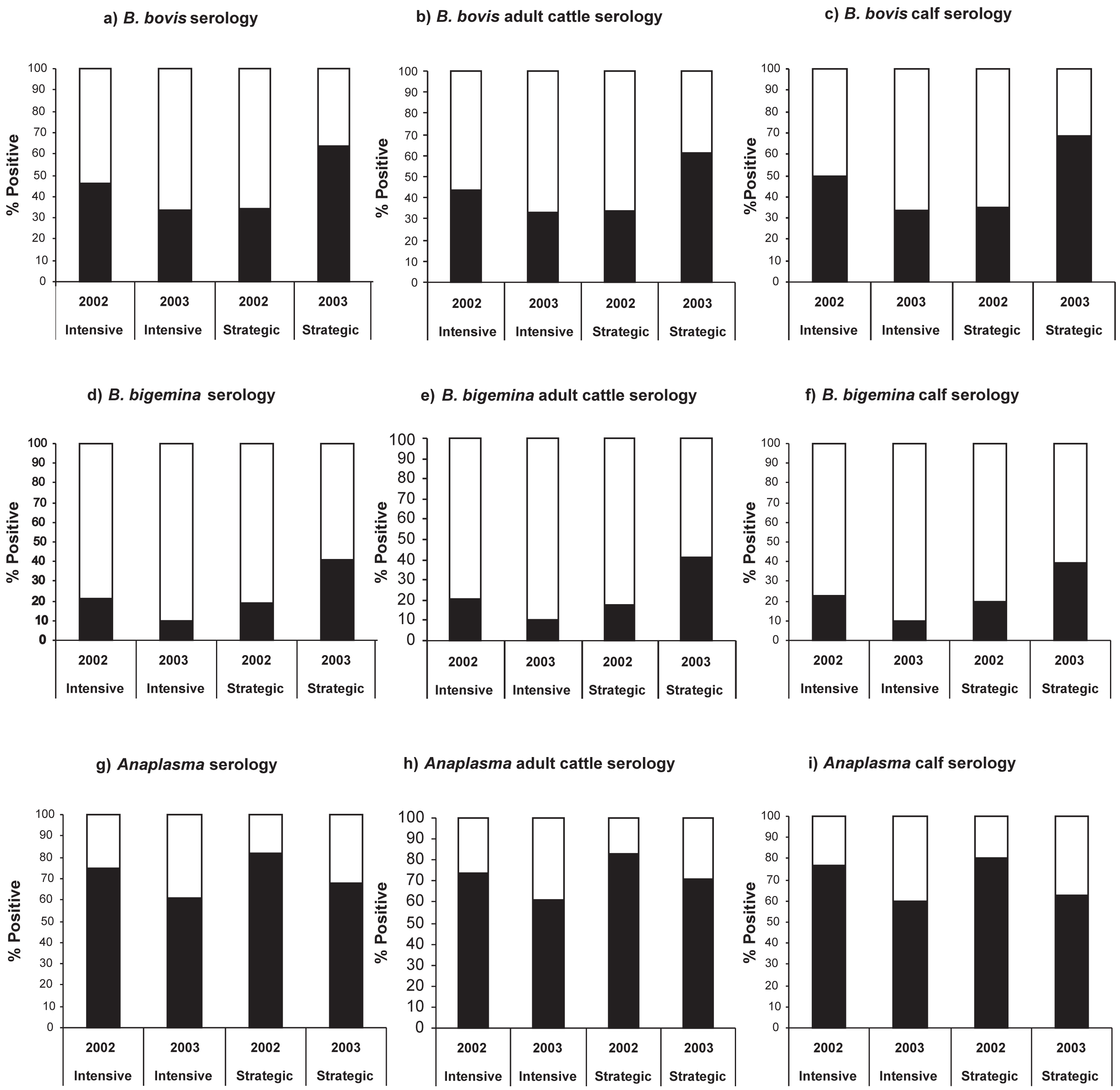

Fig. 1: **Comparison of the seroprevalence of *Babesia bovis, B. bigemina* and *Anaplasma* of adult cattle and calves dipped intensively and strategically between April 2002 and March 2003.** a: **Total *B. bovis* serology;** b: ***B. bovis* adult serology;** c: ***B. bovis* calf serology;** d: **total *B. bigemina* serology;** e: ***B. bigemina* adult serology;** f: ***B. bigemina* calf serology;** g: **total *Anaplasma* serology;** h: ***Anaplasma* adult serology;** i: ***Anaplasma* calf serology.**

is common in endemic areas in Africa[10,30]. Infestation with *R. (B.) decoloratus* usually indicates endemic stability to *B. bigemina*[11,30], thus reducing the risk of losses due to the parasite. In a study conducted in South Africa, cattle that were treated erratically with an acaricide or on which there was a reduction in the intensity of acaricide application, passed from endemic instability and low prevalence of seropositivity to endemic stability and high prevalence of seropositivity without outbreaks of clinical disease[42]. On the other hand, intensive dipping interferes with the development of endemic stability to TBD[8]. There are now many proponents of the view that it is advisable to aim for endemic stability to TBD in communally grazed areas in Africa because it is the more sustainable option[30,31].

Frozen, live, blood-based vaccines against bovine babesiosis and anaplasmosis are presently available[4,11] and a single vaccination should provide long-lasting protection. Research is now focused on the use of indigenous African cattle (*Bos indicus*), which are more resistant to ticks than European breeds[36,39]. This characteristic is attributed to both innate resistance[5,14,39] and the genetic ability to acquire resistance[4]. Tick-resistant cattle are able to attain a state of endemic stability to most TBD without tick control measures being applied[30]. Endemic stability is maintained by continuous exposure of the cattle to infected ticks; young calves become infected and are able to take advantage of an age-related resistance or colostral immunity, which minimises the effects of the disease[31]. A recombinant DNA vaccine against the 1-host tick *R. (B.) microplus* has also been developed[9,45] and registered for commercial use in Australia[45]. It targets the gut cells, destroying the tick's digestive tract and resulting in a 90 % reduction in its weight and egg production capacity[17,45]. Vaccination with recombinant gut antigens of *R. (B.) microplus* has controlled tick infestations in South America[17].

It is likely that in future communally-grazed indigenous cattle will probably require minimal tick control, ranging from none during the dry season to strategic and threshold control during the wet season[27]. Strategic control would be applied only at critical times of the year to minimise the seasonal damage caused by adult ticks. Most economically important tick species display a clear seasonal pattern of adult activity, which makes the notion of strategic control feasible[2,20,21]. Threshold tick control, which is applied only when the number of ticks per individual host exceeds a predetermined economic threshold is now also widely used[8]. It has been found that seasonal peaks in the numbers of immature *A. hebraeum* and *Rhipicephalus appendiculatus* occurred in spring and autumn, while all stages of *R. (B.) decoloratus* peaked in spring and late summer[2]. It was also found that immature *R. appendiculatus* peaked in spring and autumn and the adults of *Hyalomma* spp. in summer[40]. These studies indicate that dipping during peak tick activity should control those tick species prevalent in large parts of this region[13].

The primary objective of the present study was to investigate the impact of reduced acaricide application on endemic stability to bovine babesiosis and anaplasmosis in the local cattle population in the Bushbuckridge region of the Limpopo Province and to implement alternative measures to maintain endemic stability to TBD without resulting in a significant increase in their occurrence and of tick damage.

MATERIALS AND METHODS

The study was conducted in 4 communal grazing areas (CGA) at Bushbuckridge, namely Oakley (31°15′S, 24°58′E), Cunningmore (31°16′S, 24°56′E), Mkhuhlu (31°16′S, 25°00′E) and Ronaldsy (31°18′S, 24°55′E). Fences separated the farms but all 4 CGA were located in a single ward and had similar vegetation and climatological and ecological conditions. This is a summer rainfall area with high tick challenge during the summer months (November to February) and the grazing consists of natural sourveld[1]. Two hundred and forty, predominantly Nguni cattle, aged between 6 months to fully grown adults were selected from the 4 CGA (60 animals per CGA). One hundred and sixty of these were older than 2 years and 80 were calves less than 1 year old. The adult cattle and calves were selected for the experiment by a random sampling technique[41]. The 4 diptanks in the study region were the primary sampling units and the individual cattle/calves were the secondary sampling units[41]. The prevalence of bovine babesiosis and anaplasmosis in the study region was unknown, hence an estimation of a 25% prevalence with a 95% confidence level was made. The sample size was determined by assuming that the estimated prevalence was within 5 % of the true level[41].

At Cunningmore and Mkhuhlu, 120 cattle were dipped intensively, i.e. at 14-day intervals, while a similar number at Ronaldsy and Oakley were dipped strategically. All adult cattle and calves in the study area had been dipped at 14-day intervals prior to the start of the experiment. The 2 groups of the sample population were run in separate but similar grazing camps from April 2002 to March 2003. Blood samples were collected from 240 animals during April 2002 and again from 240 animals (not necessarily the same animals) during March 2003 and the sera were assayed using the indirect fluorescent antibody test (IFAT) for *B. bovis* and *B. bigemina*[23] and the competition inhibition enzyme-linked immunosorbent assay (CI-ELISA) for *A. marginale*[44].

Adult ticks were collected at monthly intervals prior to dipping from 3 animals (a calf and 2 adults) at each village. The animals were restrained in a crush-pen and adult ticks were collected from one half of the animal for identification and counting. The numbers recovered were multiplied by 2 to give the total number of ticks on each animal. Immature ticks were collected by drag sampling the vegetation of the 4 CGA, and 3 drag-samples per month were done per village[35].

The intensively treated group was dipped 26 times during the study period and the strategically treated group 13 times during the same period. The strategically treated group was allowed to acquire moderate to heavy tick burdens between acaricide treatments, especially during peak adult tick activity. A questionnaire aimed at estimating the damage caused by adult ticks, including abscesses, clinical disease and mortality was completed by the Animal Health Technician stationed at each diptank during dipping.

Statistical analyses of the results were performed at the Department of Statistics at the University of Pretoria using the Chi-square test to test for association between 2 variables (SAS v 8.2 programme). If the test gave a P-value of less than 0.05 the association between the 2 variables was significant at the 95 % confidence level. A low P-value also indicated that the association was not due to random error or to chance. The tests were performed using 2-way frequency tables and the serological data from the cattle and calves were compared for 2002 and 2003 taking the age of the animals and the dipping intervals into account.

RESULTS

Serology

The results of the serological tests for *B. bovis, B. bigemina* and *Anaplasma* for the strategically and intensively treated groups of cattle are illustrated in Fig. 1.

The percentage of sera positive for *B. bovis* in the strategically treated group increased significantly in 2003 compared with 2002 in both adult cattle and calves ($P < 0.05$). Seroprevalence for *B. bigemina* also increased significantly in the adult cattle ($P < 0.05$) in 2003, but the increase in the calves was not significant. Seroprevalence for both *B. bovis* and *B. bigemina* did not change significantly from 2002 to 2003 ($P > 0.05$) in the intensively treated group. There was a significant decrease in the seroprevalence of *Anaplasma* in 2003 when compared with 2002 ($P < 0.05$).

Ticks on cattle and the vegetation

The total counts of adult and immature ticks from cattle, calves and the vegeta-

Table 1: **Total number of adult ticks collected from adult cattle** (a + b) **and calves** (c + d) (2002/2003) **and immature** (Imm) **ticks collected from the vegetation.**

Treatment group	*R. (B.) microplus/ R. (B.) decoloratus*			*A. hebraeum*			*R. appendiculatus*			*H. m. rufipes*		
	Male	Female	Imm	Male	Female	Imm	Male	Female	Imm	Male	Female	Imm
a) Intensively treated group (adult cattle)	45	46	851	45	50	1243	43	51	639	45	46	678
b) Strategically treated group (adult cattle)	43	67	953	115	120	1630	65	66	893	43	62	899
c) Intensively treated group (calves)	43	39	–	46	45	–	57	55	–	48	72	–
d) Strategically treated group (calves)	51	68	–	102	123	–	63	64	–	40	51	–

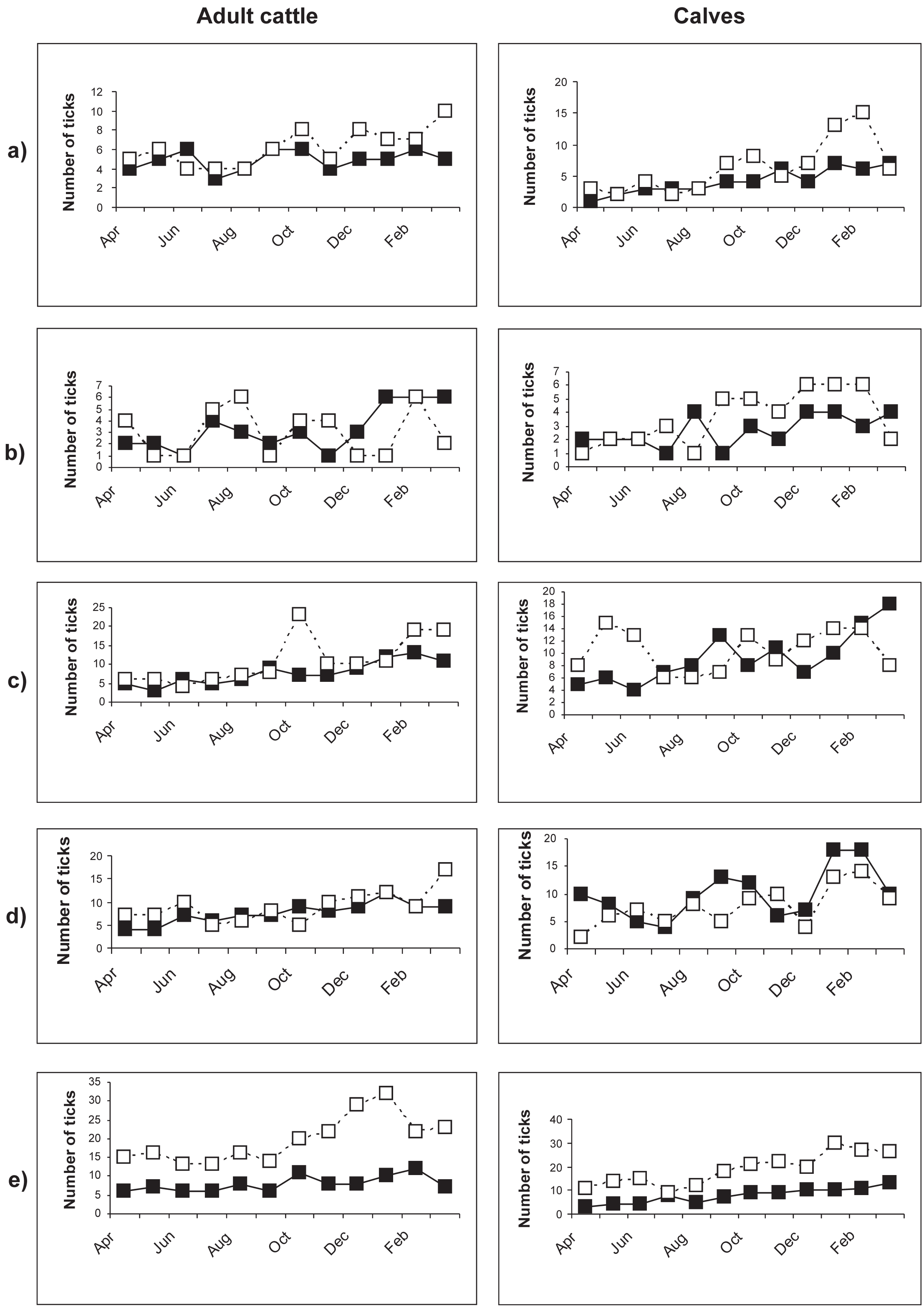

Fig. 2: **Adult ticks collected from intensively (–■–) and strategically (-- □--) dipped adult cattle and calves between April 2002 and March 2003.** a: ***Rhipicephalus (Boophilus) microplus;*** b: ***R. (B.) decoloratus;*** c: ***R. appendiculatus;*** d: ***Hyalomma marginatum rufipes;*** e: ***Amblyomma hebraeum.***

tion for the intensively and strategically treated groups had been pooled separately and are presented in Table 1 and Figs 2, 3 and 4. *A. hebraeum* was the most common tick species collected with *R. (B.) decoloratus, R. (B.) microplus, R. appendiculatus* and *H. m. rufipes* also present. Adult ticks peaked during spring and summer while immature ticks peaked during autumn and spring (*P*-value of less than 0.05).

The prevalence of clinical disease, mortality and abscesses

Three clinical cases of babesiosis (*B. bigemina*) and 9 cases of anaplasmosis were recorded in the strategically dipped group while only 1 case of anaplasmosis was reported in the intensively dipped group during the survey. A further 3 mortalities due to anaplasmosis were recorded in the strategically dipped group and 1 in the intensively dipped group. Seventeen abscesses were recorded in the strategically treated group and only 2 in the intensively treated group (Table 2). The TBD in affected animals were diagnosed by a veterinarian, who used a combination of clinical signs and the microscopic examination of blood smears.

DISCUSSION

It is generally accepted that endemic stability to TBD exists when the number of seropositive animals in a herd reaches 75 % by 9 months[25,30]. Mahoney and Ross's model was developed using serological results from calves up to 9 months of age. Older cattle were included in the present study to give a more realistic idea of the risk of disease outbreaks in the area[33]. The statistically significant increase in seroprevalence to *B. bovis* in both adult cattle and calves ($P < 0.05$) in 2003 at Oakley and Ronaldsy, where the strategically treated groups grazed, is similar to findings in other surveys in southern Africa in which *B. bovis* seroprevalence was high and where *R. (B.) microplus* was common and little tick control was practiced[30,33,37]. The main cause of the increase in seroprevalence was probably due to the reduced frequency of dipping which resulted in a greater number of ticks on the cattle, thus increasing the rate of transmission of *Babesia*[33,37]. The seroprevalence levels of *B. bovis* in both calves and adult cattle in the intensively treated group (Cunningmore and Mkhuhlu) declined during the study probably because this group remained on the same intensive dipping regimen that had been practiced previously[28]. In addition, the decline was probably due to a greater compliance by farmers in bringing their cattle for dipping compared with previous years.

The significant increase in seroprevalence to *B. bigemina* in adult cattle in 2003 is consistent with findings on farms with medium tick control in Zimbabwe[30]. The increase in seroprevalence to *B. bigemina* in the strategically treated group is also probably due to an increase in tick burdens on the cattle (Figs 3, 4) especially *R. (B.) decoloratus* and *R. (B.) microplus;* The overall seroprevalence to *B. bovis* was higher than that to *B. bigemina* in the strategically treated group than in the intensively treated group, whereas one would expect the converse to be true[10,11]. It is common knowledge that the sensitivity of IFAT to *B. bovis, B. bigemina* and *A. marginale* regresses with time after exposure but it flattens out after 98 days[19,24]. It has also been shown that a serological cross-reaction between *B. bovis* and *B. bigemina* exists[3,18,19]. At the Onderstepoort Veterinary Institute (OVI) positive IFAT control slides that are very specific for *B. bovis* and *B. bigemina* were used, therefore the positives were definite (O Matthee, pers. comm. 2002). However, several studies in Africa[30,33,34] have reported higher prevalence rates to *B. bovis* than to *B. bigemina* in communal herds where both *B. bovis* and *B. bigemina* co-exist. This could be due at least in part to the displacement of *R. (B.) decoloratus* by *R. (B.) microplus*[43] and the fact that *R. (B.) microplus* feeds more efficiently on cattle than *R. (B.) decoloratus.* If one uses the criteria suggested by Mahoney and Ross, then endemic stability to *B. bovis* and *B. bigemina* was not present in this cattle population[25].

Unfortunately an exact comparison between the 2002 and 2003 serological results for *Anaplasma* could not be made because the laboratory at the OVI , where the tests were done, used different antigenic kits for the 2002 and 2003 tests. Despite an increase in the number of *R. (Boophilus)* spp. ticks on the strategically treated group there was a sharp decline in the seroprevalence to *Anaplasma; Anaplasma marginale* is also mechanically transmitted by biting flies whose behaviour is greatly influenced by climatic conditions. However, the degree of endemic

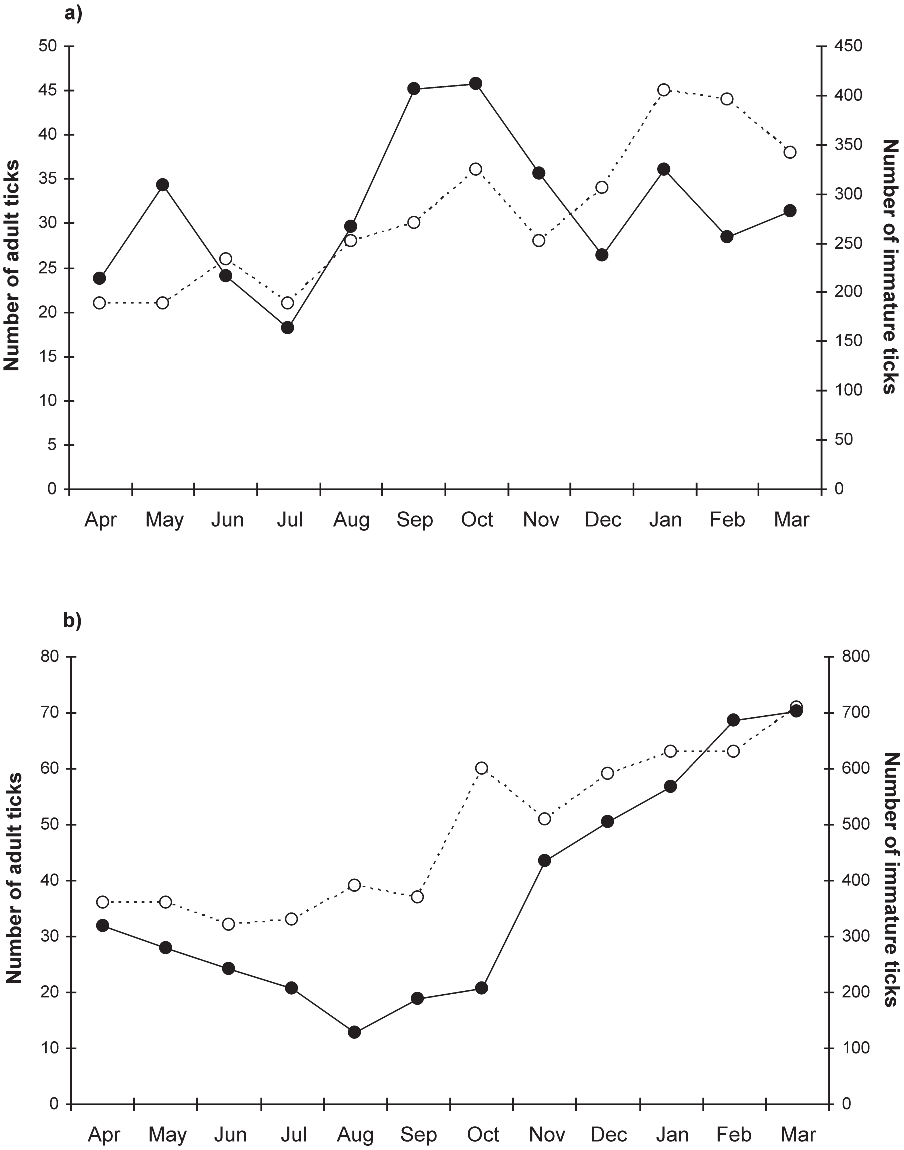

Fig. 3: **Comparison of the total number of adult ticks (all species) (--O--) collected off intensively** (a) **and strategically** (b) **dipped cattle and calves between April 2002 and March 2003.**

Table 2: **The prevalence of clinical disease, mortality and abscesses in cattle at Bushbuckridge; the number of clinical cases in each case is also indicated.**

Dipping regime	Clinical disease	Mortality	Abscesses
Intensive	Anaplasmosis (1)	Anaplasmosis (1)	2
Strategic	Anaplasmosis (9) Babesiosis (3)	Anaplasmosis (3)	17

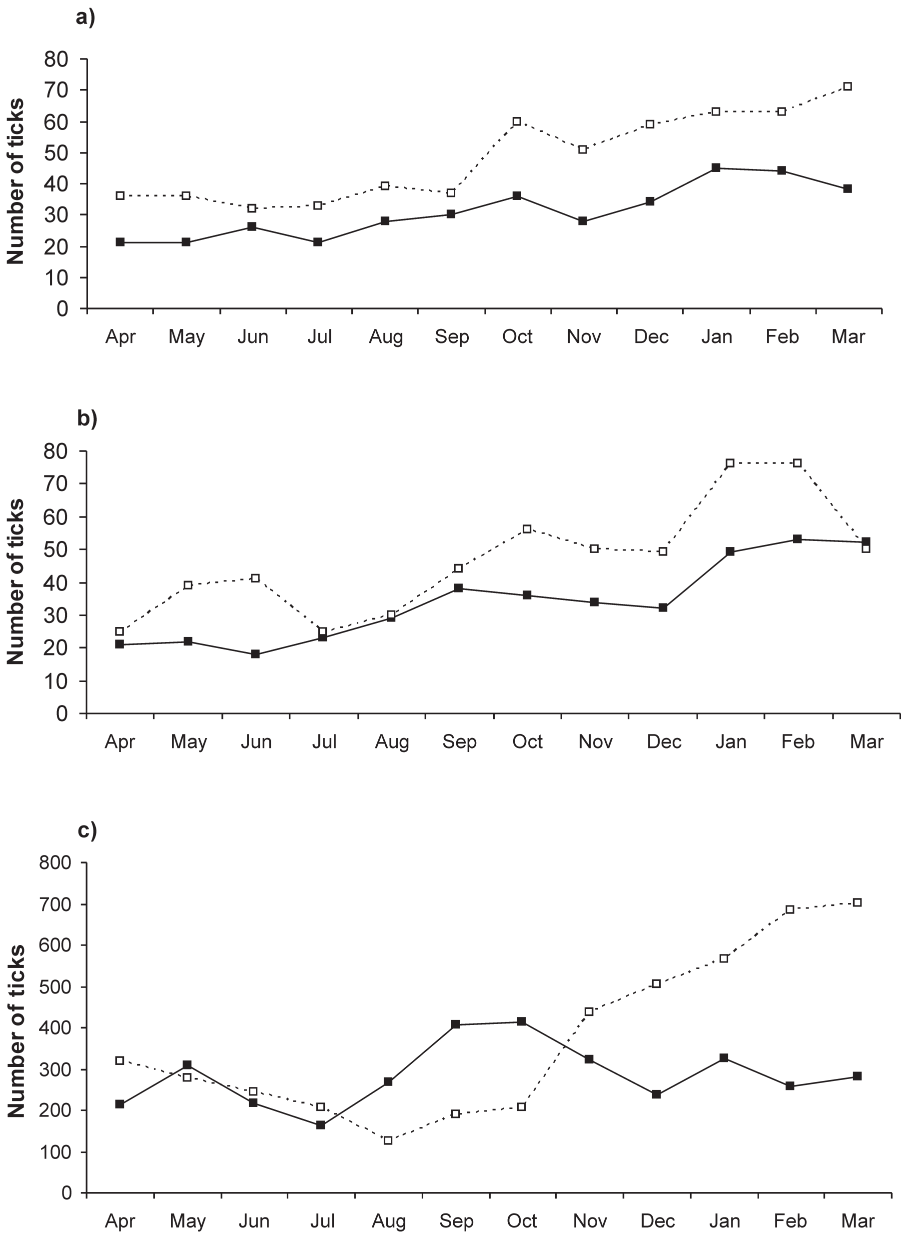

Fig. 4: **Total number of adult ticks collected from intensively (—■—) and strategically (--□--) dipped** (a) **adult cattle,** (b) **calves and immature ticks collected from the vegetation** (c) **between April 2002 and March 2003.**

stability to anaplasmosis does not necessarily correlate with dipping frequency and it has also been stated that dipping frequency does not reduce the seropositivity to *Anaplasma*[30]. The absence of outbreaks of clinical disease prior to the study could possibly be ascribed to the presence of non-pathogenic strains of the organism in the study region. Cattle in the villages may also have been resistant to TBD after years of exposure to these diseases.

During the study adult tick numbers peaked in spring and summer (Fig. 4), and statistically significant peaks in immature tick numbers occurred in autumn and in spring ($P < 0.05$). There was, however, no statistically significant correlation between the adult tick burdens of the adult cattle and those of the calves. The relationship between the seasonal counts of immature ticks and the 2 dipping regimens was, however, statistically significant ($P < 0.05$). The most prevalent tick species collected from the cattle was *A. hebraeum,* a finding similar to that of other studies done on cattle in the Eastern Cape Province[16,21]. After *R. (B.) decoloratus, A. hebraeum* was the most numerous tick species on kudus in the Kruger National Park, which is adjacent to the study sites[22]. *Rhipicephalus (B.) decoloratus, R. (B.) microplus, R. appendiculatus* and *H. m. rufipes,* were also collected in significant numbers. The results are also similar to those of another recent survey in this region[6]. The presence of adult *R. (B.) microplus* recorded in the present survey might also explain the increase in seroprevalence to *B. bovis,* a finding similar to the one done in the Limpopo Province[43]. The number of *R. (Boophilus)* spp. collected from the cattle peaked during spring and autumn, a common finding in other regions of South Africa[2,6,35].

Eleven cases of clinical TBD occurred in the strategically treated group during the 1st quarter of the study period. This group of cattle had been maintained on a 14-day dipping interval prior to the study. A single case of anaplasmosis was recorded in the intensively dipped group of cattle during the corresponding period. The low incidence of clinical anaplasmosis could be due to a greater degree of endemic stability as suggested by the 75 % positive seroprevalence. All 12 clinical cases were treated and 8 recovered.

It is concluded that it is unnecessary to dip cattle intensively at fortnightly intervals in this region especially when one considers the relatively low tick burdens on the cattle and on the vegetation. The increase in seroprevalence to *B. bovis* and *B. bigemina* in the strategically treated group of cattle implies that if a reduced dipping frequency could be maintained for long enough an endemically stable disease situation should result. Outbreaks of clinical cases of disease could be treated and vaccination could be used to supplement the natural tick challenge if it is not sufficient to maintain endemic stability. The increased seroprevalence to both *B. bovis* and *B. bigemina* in calves suggests that calf vaccination is unnecessary and that tick control should therefore be aimed mainly at preventing excessive tick worry.

ACKNOWLEDGEMENTS

Deepest gratitude is expressed to the Director of Veterinary Services, by B O Rikhotso, to Dr L. Mampane and the Regional Director, Mr M.B. Mboweni of the Limpopo Provincial Government, for allowing him to conduct the research and for funding the project. Professors J A W Coetzer, E H Venter, I G Horak and B L Penzhorn of the Department of Veterinary Tropical Diseases for funding the project and for academic administration. Thanks is also due to the farmers who agreed to participate in this project and the 4 communities where the project was conducted as well as the diptank committee members and the herdsmen who were involved in restraining the animals and who encouraged the community members to cooperate and bring their cattle to the diptanks. Thanks are also due to the Animal Health Technicians who helped with the bleeding of the cattle and the collection of ticks namely Messrs M V Mnisi, S Mkoni and C F Zwane and their teams who assisted them at the diptanks as well as Mr Mngomezulu and his work-

ers for assistance with drag-sampling the vegetation for immature ticks. The OVI and its staff are thanked for performing the serological tests.

REFERENCES

1. Acocks J P H 1988 *Veld types of South Africa. Memoirs of the Botanical Survey of South Africa* 57: 1–146
2. Baker J A F, Ducasse F B W, Sutherst R W, Maywald G F 1989 The seasonal tick populations on traditional and commercial cattle grazed at four altitudes in Natal. *Journal of South African Veterinary Association* 60: 95–101
3. Bessenger R, Schoeman J H 1983 Serological response of cattle to infection with *Babesia bigemina* and *Babesia bovis* in southern Africa. *Onderstepoort Journal of Veterinary Research* 50: 115–117
4. Bock R E, De Vos A J 2001 Immunity following use of Australian tick fever vaccine: a review of the evidence. *Australian Veterinary Journal* 79: 832–839
5. Bock R E, Kingston T G, De Vos A J 1999 Effect of breed of cattle on innate resistance to infection with *Anaplasma marginale* transmitted by *Boophilus microplus. Australian Veterinary Journal* 77: 748–751
6. Bryson N R, Tice G A, Horak I G, Stewart C G, Du Plessis B J A 2002 Ixodid ticks on cattle belonging to small-scale farmers at 4 communal grazing areas of South Africa. *Journal of the South African Veterinary Association* 73: 98–103
7. Buscher G 1988 The infection of various tick species with *Babesia bigemina,* its transmission and identification. *Parasitology Research* 74: 324–330
8. Cook A J C 1991 Communal farmers and tick control, a field study in Zimbabwe. *Tropical Animal Health and Production* 23: 161–166
9. Dayton L 1991 Anti-tick vaccines promise reduced costs for cattle farmers. *New Scientist* 130: 18
10. De Vos A J 1979 Epidemiology and control of bovine babesiosis in South Africa. *Journal of the South African Veterinary Association* 50: 357–362
11. De Vos A J, Potgieter F T 1983 The effect of tick control on the epidemiology of bovine babesiosis. *Onderstepoort Journal of Veterinary Research* 50: 3–6
12. Dipeolu O O, Mongi A O, Punyua D K, Latif A A, Amoo O A Odhiambo T R 1992 Current concepts and approach to control of livestock ticks in Africa. *Discovery Innovation* 4: 35–44
13. Dreyer K, Fourie L J, Kok D J 1998 Tick diversity, abundance and seasonal dynamics in a resource-poor urban environment in the Free State Province. *Onderstepoort Journal of Veterinary Research* 65: 305–316
14. Fivaz B H, De Waal D T, Lander K 1992 Indigenous and cross-bred cattle: comparison of resistance to ticks and implications for their strategic control in Zimbabwe. *Tropical Animal Health and Production* 24: 81–89
15. Fivaz B H, De Waal D T 1993 An evaluation of strategic and tactical tick control in indigenous, exotic and crossbred cattle. *Tropical Animal Health and Production* 25: 19–25
16. Fourie L J, Horak I G 1990 Parasites of cattle in the south western Orange Free State. *Journal of the South African Veterinary Association* 61: 27–28
17. Fuente J, Rodriguez M, Garcia-Garcia J C 2000 Immunological control through vaccination with *Boophilus microplus* gut antigens. *Annals of the New York Academy of Sciences* 916: 617–621
18. Gray J S, De Vos A J 1981 Studies on a bovine *Babesia* transmitted by *Hyalomma marginatum rufipes.; Onderstepoort Journal of Veterinary Research* 48: 215–233
19. Guglielmone A A, Lugaresi C I, Volpogni M M, Anziani O S, Vanzini V R 1997 Babesial antibody dynamics after cattle immunisation with live vaccines measured with an indirect immunofluorescence test. *Veterinary Parasitology* 70: 33–39
20. Horak I G 1982 Parasites of domestic and wild animals in South Africa. XV. The seasonal prevalence of ectoparasites on impala and cattle in the Northern Transvaal. *Onderstepoort Journal of Veterinary Research* 49: 85–93
21. Horak I G 1999 Parasites of domestic and wild animals in South Africa. XXXVII. Ixodid ticks on cattle on Kikuyu grass pastures and in Valley Bushveld in the Eastern Cape Province. *Onderstepoort Journal of Veterinary Research* 66: 175–184
22. Horak I G, Boomker J, Spickett A M, De Vos V 1992 Parasites of domestic and wild animals in South Africa. XXX. Ectoparasites of kudus in the Eastern Transvaal Lowveld and the Eastern Cape Province. *Onderstepoort Journal of Veterinary Research* 59: 259–273
23. Joyner L P, Donnelly J, Payne R, Brocklesby D W 1972 The Indirect Fluorescent Antibody Test for the differentiation of infections with *Babesia divergens* or *Babesia major. Research in Veterinary Science* 13: 515–518
24. Kuttler K L, Adams L G, Todorovic R A 1977 Comparisons of the complement-fixation and indirect fluorescent antibody reactions in the detection of bovine babesiosis. *American Journal of Veterinary Research* 38: 153–156
25. Mahoney D F, Ross D R 1972 Epizootiological factors in the control of bovine babesiosis. *Australian Veterinary Journal* 48: 292–298
26. Masika P J, Sonandi A, Van Averbeke A 1997 Tick control by small-scale farmers in the Central Eastern Cape Province, South Africa. *Journal of the South African Veterinary Association* 68: 45–49
27. Matthewson M D 1984 The future of tick control: a review of chemical and non-chemical control. *Preventative Veterinary Medicine* 2: 559–568
28. Meltzer M I, Norval R A I. Donachie P L 1995 Effects of tick infestation and tick-borne disease infections (heartwater, anaplasmosis and babesiosis) on the lactation and weight gain on Mashona cattle in south-eastern Zimbabwe. *Tropical Animal Health and Production* 27: 129–144
29. Norval R A I 1983 Arguments against intensive dipping. *Zimbabwe Veterinary Journal* 14: 19–25
30. Norval R A I, Fivaz B H, Lawrence J A, Brown A E 1983 Epidemiology of tick-borne diseases of cattle in Zimbabwe. 11 Anaplasmosis. *Tropical Animal Health and Production* 15: 87–94
31. Norval R A I, Perry B D, Hagreaves S K 1992 Tick and tick-borne disease control in Zimbabwe: what might the future hold? *Zimbabwe Veterinary Journal* 23: 1–15
32. Onen-okello J, Mukhebi A W, Tukahirwa E M, Musisi G, Bode E, Heinonen R, Perry B D, Opuda-asibo J 1998 Financial analysis of dipping strategies for indigenous cattle under ranch conditions in Uganda. *Preventative Veterinary Medicine* 33: 241–250
33. Perry B D, Musisi F L, Pegram R G, Schels H F 1989 Assessment of enzootic stability to tick-borne diseases. *World Animal Review* 56: 24–32
34. Perry B D, Young A S 1995 The past and future roles of epidemiology and economics in the control of tick-borne diseases of livestock in Africa. *Preventative Veterinary Medicine* 25: 107–120
35. Rechav Y 1982 Dynamics of tick populations (Acaricide: Ixodidae) in the Eastern Cape Province of South Africa. *Journal of Medical Entomology* 19: 679–700
36. Rechav Y, Kostrzewski M W 1991 Relative resistance of cattle breeds to the tick *Boophilus decoloratus* in South Africa. *Onderstepoort Journal of Veterinary Research* 58: 181–186
37. Regassa A, Penzhorn B L, Bryson N R 2003 Attainment of endemic stability to *Babesia bigemina* in cattle on a South African ranch where non-intensive tick control was applied. *Veterinary Parasitology* 116: 267–274
38. Riek R F 1966 The life cycle of *Babesia argentina* (Lignieres 1903) (Sporozoa: Piroplasmidea) in the tick vector *Boophilus microplus* (Canestrini). *Australian Journal of Agricultural Research* 177: 247–254
39. Spickett A M, De Klerk D, Enslin C B, Scholtz M M 1989 Resistance of Nguni, Bonsmara and Hereford cattle to ticks in the bushveld region of South Africa. *Onderstepoort Journal of Veterinary Research* 56: 245–250
40. Spickett A M, Fivaz B H 1992 A survey of cattle tick control practices in the Eastern Cape Province of South Africa. *Onderstepoort Journal of Veterinary Research* 59: 203–210
41. Thrusfield M 1995 *Veterinary Epidemiology* (2nd edn). Blackwell Science, London
42. Tice G A, Bryson N R, Stewart C G, Du Plessis J L, De Waal D T 1998 The absence of clinical disease in cattle in communal grazing areas where farmers are changing from an intensive dipping programme to one of endemic stability to tick-borne diseases. *Onderstepoort Journal of Veterinary Research* 65: 169–179
43. Tønnessen M H, Penzhorn B L, Bryson N R, Stoltsz W H, Masibigiri T 2004 Displacement of *Boophilus decoloratus* by *Boophilus microplus* in the Soutpansberg region, Limpopo Province, South Africa. *Experimental and Applied Acarology* 32: 199–208
44. Visser E S, McGuire T C, Palmer G H, Davis W C, Skhap V, Pipano E. & Knowles D P 1992. The *Anaplasma marginale msp5* gene encodes a 19 kilodalton protein conserved in all recognized *Anaplasma* species. *Infection and Immunity* 60: 5139–5144
45. Willadsen P, Bird P, Cobon G S, Hungerford J 1995 Commercialisation of a recombinant vaccine against *Boophilus microplus. Parasitology* 110: 843–850