CHAPTER 1. INTRODUCTION

1. 1. Importance of bovine babesiosis worldwide.

Bovine babesiosis or redwater occurs worldwide, with the exception of a few countries where it is too cold for the tick vectors to survive (Hoyte, 1976; Callow *et al.*, 1976b; McCosker, 1981). *Babesia bigemina* and *Babesia bovis* historically caused the most widespread tick-borne diseases and continue to be among the greatest obstacles to the development of the livestock industry in tropical and subtropical regions of the world. They are still important causes of mortality in cattle (McCosker, 1981) and as many as one billion cattle worldwide are thought to be exposed to redwater (Mahoney, 1976, as cited by Smith and Kakoma, 1989).

1. 1. 1. The *Babesia* parasites. The *Babesias* are protozoan parasites belonging to the phylum Apicomplexa, class Sporozoasida, order Eucoccidiorida, suborder Piroplasmorina and family Babesiidae (Levine, 1971). The parasites are usually host-specific (Callow and Dalgliesh, 1982). The four *Babesia* species responsible for redwater in cattle are *Babesia bovis* (syn. *Babesia argentina*) (Riek, 1968; Hoyte, 1976; Callow *et al*., 1976b; Potgieter, 1977), *Babesia bigemina*, *Babesia divergens* and *Babesia major*, but the latter two species do not occur in South Africa. *Babesia bigemina* and *Babesia bovis*, the two important species found in cattle in South Africa, are transmitted by one-host ticks of the genus *Boophilus*. *Boophilus decoloratus* transmits only *Babesia bigemina*, whilst *Boophilus microplus* transmits both *Babesia* *bigemina* and *Babesia bovis* (Potgieter, 1977). *Babesia bigemina* can also be transmitted by *Rhipicephalus evertsi evertsi* (Büscher, 1988).

1. 2. The historical importance of bovine babesiosis in Africa.

When Europeans first colonized southern Africa they introduced highly susceptible cattle which soon suffered heavy losses due to redwater. *Babesia bigemina*, the cause of African redwater, appeared to have been present in the indigenous cattle, but caused few clinical problems (Lawrence and Norval, 1979). Fatal cases of the disease in the indigenous cattle only occasionally occurred (Theiler, 1975) as they seemed to acquire immunity as young calves (Norval *et al.*, 1992a). In 1893 Smith and Kilborne first demonstrated that *Babesia bigemina* was transmitted by a tick vector, *Boophilus annulatus,* in the USA (Levine, 1971; Hoyte, 1976; Smith and Kakoma, 1989; Brown *et al*., 1990). In 1898 Koch established that *Boophilus decoloratus* was the main vector of *Babesia bigemina* in Africa (Neitz, 1941).

The European settlers in Africa soon realized that young cattle were less susceptible to bovine babesiosis than adult cattle and that cattle from known redwater areas could survive when moved to new redwater endemic areas. Cattle could also be moved to new areas in winter, when tick worry was minimal (Lawrence and Norval, 1979). The blood parasite first encountered in these areas was *Babesia bigemina* which was indigenous to Africa. However, after the rinderpest epidemic in 1896, *Boophilus microplus*-infested cattle carrying *Babesia bovis* were imported from southern Asia via Madagascar to the

African continent (Theiler and Robinson, 1954; Callow, 1977; Lawrence and Norval, 1979). *Babesia bovis*, the cause of Asiatic redwater, then spread from the eastern coastline of South Africa and Mozambique into the interior of these countries and into Zimbabwe and Zambia. Its spread was dependent on the presence of the vector, *Boophilus microplus,* which has since continued to spread into new areas of southern Africa (Howell *et al.*, 1978; Norval and Short, 1984).

1. 3. Importance of bovine babesiosis in South Africa.

1. 3. 1. General. Tick-borne diseases (TBD) are economically important in southern Africa, and bovine babesiosis is one of the most prevalent and widespread in these regions. De Vos (1979) stated that 85 % of the cattle population in South Africa was potentially at risk to bovine babesiosis; in 1971 nearly 8000 cattle died in KwaZulu-Natal alone (De Vos and Every, 1981). In 1981 it was estimated that the cost of controlling TBD in South Africa could be as high as 70 million Rand (De Vos, 1981). When dipping broke down in Zimbabwe due to civil war (1973 – 1980), nearly a million cattle died of TBD. Many of the deaths were due to *Babesia* infections and in some areas cattle mortality due to TBD was nearly 95 % (Norval, 1979).

1. 3. 2. Importance of bovine babesiosis in the study area. The farming systems in the area, which is situated in the northern part of the Northern Province, are divided into commercial and subsistence (small-scale) farming. Historically there has been a dipping service for the subsistence farmers, subsidized by the state and provincial governments.

Dipping is undertaken to control the ticks that transmit East Coast fever, African redwater, Asiatic redwater, heartwater and anaplasmosis (Tice, 1997). In certain areas dipping has been carried out for over a century, making it difficult for cattle herds to reach endemic stability to TBD, especially as calves are also dipped (Norval *et al*., 1983). This prevents calves from developing a natural immunity by being exposed to the parasites when they are relatively resistant to *Babesia* infections (Norval *et al.*, 1983).

Gray and de Vos (1981) found no evidence of *Babesia bovis* during a serological survey in the Northern Province of South Africa. De Vos and Potgieter (1983) reported that *Babesia bigemina* was present on 26 farms in the Northern Transvaal but they found no evidence of *Babesia bovis* during their survey. However, Sutherst (1987a) used a climatic model, CLIMEX, in South Africa, and singled out Venda as a possible area where *Boophilus microplus* might successfully become established.

The study area is endemic for *Babesia bigemina*, but over the past 15 years *Babesia bovis* has established itself in the eastern part of the Soutpansberg and Venda Districts of the Northern Province. Clinical outbreaks of Asiatic redwater (*Babesia bovis* infection) were reported from Venda in the mid-1980s (Loock, 1999, personal communication; Gous, 1999, personal communication). In Venda, redwater is mainly transmitted during the rainy season (October to May) when *Boophilus* numbers are high (De Vos, 1979). Only 10 % of the 144 cases of redwater confirmed in Venda between 1997 and 1999 (Fig. 1. 1) were recorded during winter. Most of the confirmed cases (74 %) were due to *Babesia bovis* (Loock, 1999, personal communication). Other clinical cases were probably not reported by the farmers and it was presumed that the actual mortality rate due to redwater was much higher (Loock, 2000, personal communication).

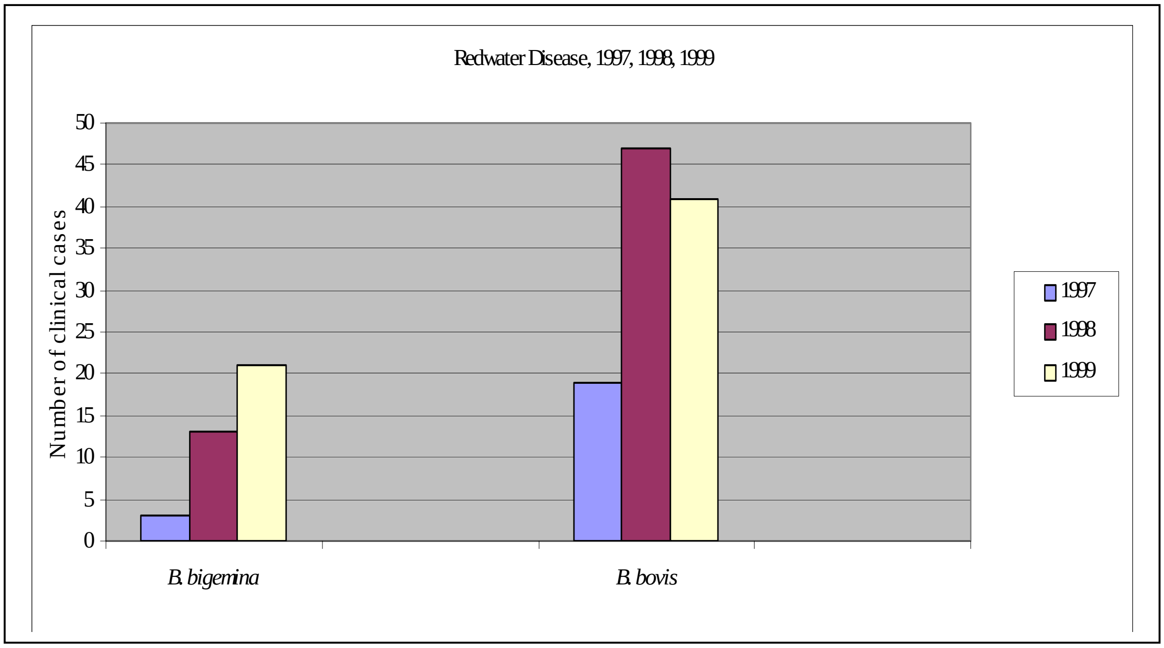


Fig. 1. 1. Confirmed cases of bovine babesiosis in the study area in 1997, 1998 and 1999.

1. 4. The main objectives of this study were:

- To confirm the presence of *Boophilus microplus* in the Soutpansberg, Dzanani, Mutale, Thohoyandou and Vuwani Districts in the Northern Province of South Africa and to determine its geographical distribution within this area.

- To determine the serological prevalence of *Babesia bovis* and *Babesia bigemina* in a representative sample from cattle in the same districts.

- To determine the relative number of *Boophilus microplus* in relation to *Boophilus decoloratus* and to establish if any displacement of *Boophilus decoloratus* by *Boophilus microplus* is taking place.

- To map the distribution of *Boophilus decoloratus* and *Boophilus microplus* in the study area in the Northern Province and to attempt to model the findings, using the CLIMEX model, to predict possible further spread of *Boophilus microplus.*

CHAPTER 2. LITERATURE REVIEW

2. 1. The genus *Babesia*

2. 1. 1. Comparative morphology and strain differences.

- ***Babesia bigemina*** is a large *Babesia*, up to 5µm x 3 µm in size, occurring singly or in pairs in erythrocytes. Single forms are elongated or club-shaped; in pairs the angle between the merozoites is typically acute (Hoyte, 1976; Potgieter, 1977; Potgieter and Els, 1977; Levine, 1985).

- ***Babesia bovis*** is a small *Babesia*, up to 2.4µm x 1.5 µm in size, and one or two parasites are found in each erythrocyte (Neitz, 1941). Single *Babesia bovis* organisms are round, oval or irregular in shape, while paired forms are club shaped, sometimes with rounded ends. The angle between the paired organisms is often, but not always, obtuse (Hoyte, 1976; Potgieter, 1977; Levine, 1985).

- **Strain differences and antigenic variation in *Babesia* species.** Several researchers (Neitz, 1941; Riek, 1964; Callow, 1964; 1967; 1968; Johnston and Tammemagi, 1969; Curnow, 1973a; Doyle, 1977; Thompson *et al*., 1977; De Vos, 1978; Mahoney *et al*., 1979a; Smith *et al*., 1980; Callow *et al*., 1981) found evidence of immunological strain differences in both *Babesia bigemina* and *Babesia bovis.* Immunological differences between strains from different geographical locations have also been demonstrated with *Babesia bigemina* and *Babesia bovis* (Mahoney, 1974). Hall (1963) showed that when calves were challenged with a strain of *Babesia bigemina* other than that to which they had acquired passive immunity from their dams, the disease reaction was almost as severe as experienced by calves from fully susceptible mothers. Recovered cattle were also more resistant when challenged with the homologous strain when compared to heterologous strains (Callow, 1967; 1968; Johnston and Tammemagi, 1969; Smith *et al*., 1980).

The strains differ in virulence, and frequent cyclic transmission may cause this to increase (Callow, 1984). Some Australian strains of *Babesia bigemina* are relatively non-pathogenic (Johnston, 1968; Mahoney, 1974; James *et al*., 1985). Antigenic differences within a *Babesia bovis* strain are often found, and these revert to a common, strain-specific type after being transmitted through the tick vector *Boophilus microplus* (Curnow, 1973a). Cross-immunity is common between different strains in infected animals (Mahoney *et al*., 1979a; Mahoney *et al*., 1979b).

2. 1. 2. Diagnosis of redwater. Parasites are detected in appropriately stained peripheral blood smears by light microscopy, positive identification of the parasites under the microscope being the only way to confirm a presumptive diagnosis. *Babesia bovis* can also be detected in organ smears, particularly those made from brain tissue at necropsy (De Vos and Potgieter, 1994).

2. 1. 3. Geographical distribution in South Africa.

- ***Babesia bovis*** was probably reported for the first time in South Africa from the Cape Colony in 1905 (Potgieter and Els, 1977). In 1941 it was identified in Pretoria (Neitz, 1941) and was later recorded in coastal KwaZulu-Natal and Mpumalanga (De Vos, 1979; De Vos and Potgieter, 1983), and subsequently from the interior of KwaZulu-Natal (De Vos and Every, 1981). The known distribution of *Babesia bovis* is shown in Fig. 2. 1.

- ***Babesia bigemina*** is indigenous to southern Africa (Lawrence and Norval, 1979) and has been recorded in KwaZulu-Natal, Mpumalanga, Gauteng, the Northern Province and in large parts of the North West Province. It also occurs in parts of the Eastern Cape Province, the coastal parts of the Western Cape Province, parts of the Free State Province and in the north-eastern corner of the Northern Cape Province (De Vos, 1979). The known distribution of *Babesia bigemina* is shown in Fig. 2. 1

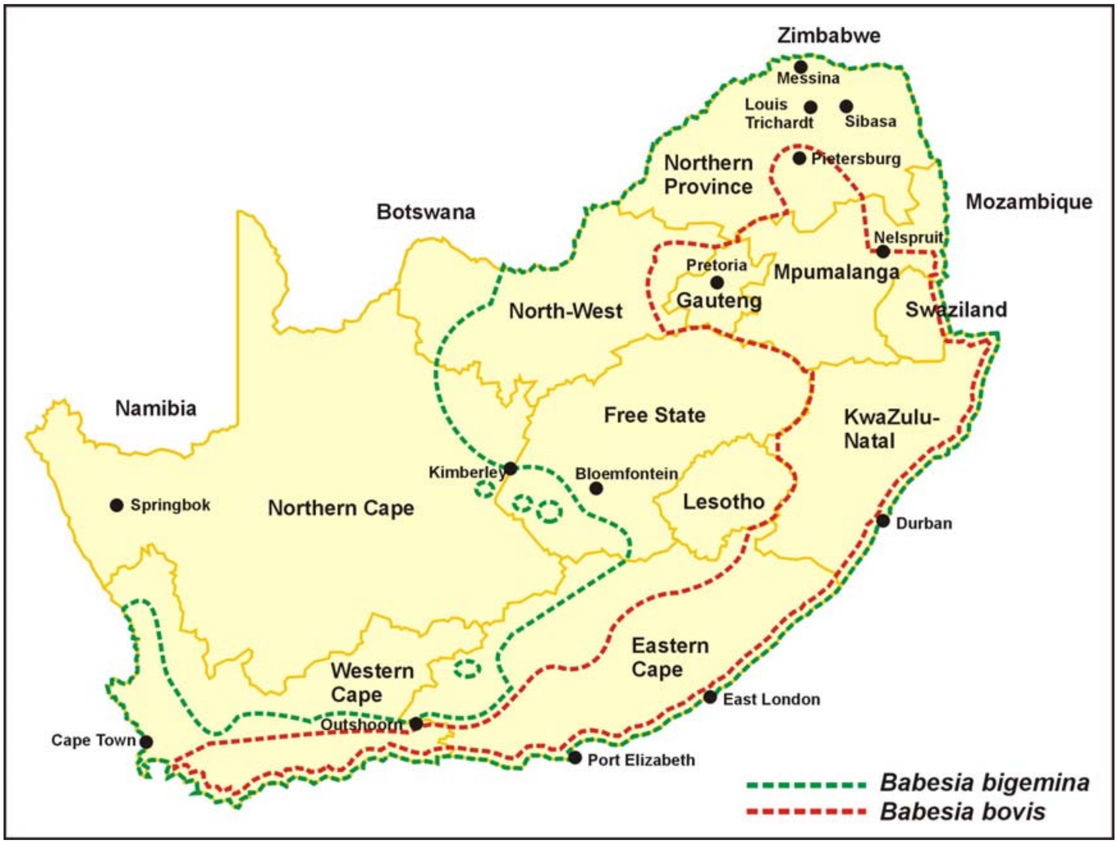


Fig. 2. 1. Map showing the geographical distribution of *Babesia bigemina* and *Babesia bovis* in South Africa (adapted from De Vos, 1979; De Vos and Every, 1981; De Vos and Potgieter, 1983; Jagger *et al.*, 1985; Wedderburn *et al.*, 1991).

2. 1. 4. Transmission by the vectors.

- ***Babesia bigemina*** is transmitted to cattle by the one-host ticks *Boophilus decoloratus* and *Boophilus microplus* (Riek, 1964; Potgieter, 1977; Potgieter and Els, 1977; Norval *et al*., 1983; Callow, 1984) and to a much lesser extent by the two-host tick *Rhipicephalus evertsi evertsi* (Büscher, 1988). The importance of *Rhipicephalus evertsi evertsi* as a vector of *Babesia bigemina* in the field is uncertain (Howell *et al*., 1978).

Adult female *Boophilus decoloratus* and *Boophilus microplus* ticks acquire the *Babesia bigemina* infection in the final stage of feeding; the infection is then passed transovarially to the larvae. The life cycle of each tick species is identical (Potgieter and Els, 1977). The larvae that hatch are infected but not infective; the sporozoites are transmitted to cattle by both nymphs and adults of the *Boophilus* species. The *Babesia bigemina* infection in *Boophilus* species is retained by transovarial transmission in the absence of reinfection from a susceptible host for at least two generations (Callow, 1965; Potgieter, 1977; Potgieter and Els, 1977; Gray and Potgieter, 1981; Dalgliesh and Stewart, 1983; Büscher, 1988).

Nymphs of *Rhipicephalus evertsi evertsi* have been thought to transmit *Babesia bigemina*, although with difficulty (Neitz, 1941; Büscher, 1988).

- ***Babesia bovis*** is transmitted by *Boophilus microplus*, the only known vector in southern Africa (Riek, 1966; Potgieter, 1977). The female tick is infected when engorging, and the infection is passed transovarially to the larvae. The larvae then transmit the sporozoites to cattle whilst feeding but clear themselves of the infection and neither nymphs nor adults transmit the infection (Potgieter, 1977; Potgieter and Els, 1979; Mahoney and Mirre, 1979; Gray and Potgieter, 1981; Dalgliesh and Stewart, 1983). The *Babesia bovis* infection is not transmitted to the next generation of ticks without going through its life cycle in the vertebrate host (Mahoney and Mirre, 1979).

2. 1. 5. Clinical signs of the diseases.

- **African redwater.** Clinical signs of bovine babesiosis caused by *Babesia bigemina* begin 7-21 days after the initial attachment of an infected tick (Callow and Dalgliesh, 1982). The first sign is usually a temperature exceeding 40°C, coupled with anorexia. The animal is depressed with haemoglobinuria as a consistent finding shortly after the onset of the disease. A clinically detectable anaemia soon develops and the animal may die if not treated. In more protracted cases there is marked icterus (Callow *et al.*, 1993; De Vos and Potgieter, 1994). *Babesia bigemina* infection is normally eliminated from the cattle within 6 months (Callow *et al*., 1974b).

- **Asiatic redwater.** The clinical signs of bovine babesiosis caused by *Babesia bovis* are similar to those of *Babesia bigemina* but the disease is more acute, has a shorter course with more severe signs and a higher mortality rate. Animals are weak and reluctant to move; they have an increased respiratory rate, fever and severe depression. At this stage haemoglobinuria is not usually present but diarrhoea is common and pregnant cattle may abort. Signs of central nervous system (CNS) involvement develop in some animals and manifest as nystagmus, circling, head pressing, aggression, convulsions and paralysis. The mortality rate is higher than that of African redwater and in nonfatal cases the recovery period is protracted (Smith *et al.* 1980; Norval *et al*., 1992b; Callow *et al.*, 1993; De Vos and Potgieter, 1994; Bock *et al.*, 1997).

Cattle which survive the acute disease become persistent carriers of the infections for periods of 1-4 years during which they show no clinical evidence of being infected (Mahoney, 1969; Callow, 1977). *Babesia bovis* infections can persist for 1-4 years and ticks feeding on the carriers can become infected (Johnston and Tammemagi, 1969; Mahoney *et al.*, 1973)

2. 1. 6. Pathogenesis of bovine babesiosis. The most important factor in bovine babesiosis is the invasion and breakdown of the erythrocytes by the parasites (McCosker, 1981). Haemolytic anaemia is a feature of both diseases, although in *Babesia bovis* infections acute cases may die before evidence of clinical anaemia develops (De Vos and Potgieter, 1994). The acute haemolytic phase lasts for about a week (Callow, 1977) and toxins and by-products of tissue necrosis can lead to serious clinical abnormalities (Callow, 1984).

2. 1. 7. Pathology.

- ***Babesia bigemina.*** There is rapid intravascular haemolysis and the serum haemoglobin levels are high, giving rise to haemoglobinuria. Kidney damage is usually evident and the kidneys are enlarged with degeneration of the convoluted tubules. The liver undergoes fatty degeneration and the gall bladder contains large amounts of bile. The spleen is enlarged. The carcass is anaemic with watery blood and pulmonary oedema a regular feature. Icterus is common in animals with the chronic form of the disease (Callow, 1977; Callow *et al.*, 1993; De Vos and Potgieter, 1994).

- ***Babesia bovis.*** Findings are similar to but more severe than with *Babesia bigemina* infection as proteolytic enzymes are released by the erythrocytes and coagulation is disturbed. There are also anoxic degenerative changes in the blood vessels of the brain, liver, kidneys and skeletal muscles. The cerebral cortex often shows a pink discoloration that is pathognomonic for the disease. Acute cases may die before any anaemia is noticed, although severe anaemia may develop in the more protracted cases. The post mortem examination shows intense congestion of most organs and icterus is seen in those cases which survive the initial disease (Smith *et al.*, 1980; Callow and Dalgliesh, 1982; Callow, 1984; Callow *et al.*, 1993; De Vos and Potgieter, 1994).

2. 1. 8. Immunity.

- **General immunity.** Previously unexposed adult cattle of all breeds develop severe disease on first infection with *Babesia* species (Trueman and Blight, 1978; De Vos and Potgieter, 1994; Bock *et al.*, 1997). Mortality from *Babesia bovis* in susceptible herds can be as high as 50 – 80 % (Callow, 1977; Norval *et al.*, 1992a; Bock *et al.*, 1997). *Babesia* *bovis* infection may persist in *Bos taurus* cattle for at least four years, and the immunity acquired by those cattle which eliminate the infection without treatment, may persist even longer. If the cattle are challenged regularly, this resistance will persist for the rest of their lives (Callow, 1967, 1968; Mahoney, 1969; Mahoney *et al*., 1973; Johnston *et al.*, 1978; Trueman and Blight, 1978; Mahoney *et al*., 1979b). The degree of acquired immunity to *Babesia bovis* is influenced by the degree of exposure to the parasite (Callow *et al*., 1974a). Immunity in calves that are naturally infected before the age of 5-7 months with both *Babesia bigemina* and *Babesia bovis* may also persist for at least four years (Mahoney *et al.*, 1973; Johnston *et al.*, 1978; Mahoney *et al.*, 1979b).

Mortality from *Babesia bigemina* can be as high as 5-10 % if susceptible cattle are brought into an endemic area (De Vos, 1979). Cattle clear themselves of *Babesia bigemina* infection within a period lasting from a few months to as long as two years, but can retain a sterile immunity (Callow, 1967; Mahoney, 1969; Löhr, 1972; Callow *et al.*, 1974b; Johnston *et al.*, 1978). An infection with *Babesia bigemina* can give some cross-protection against *Babesia bovis* but the reverse has not been demonstrated (Wright *et al.*, 1987).

- **Age-dependent immunity.** Age-dependent immunity allows the animal to become infected with the parasite without succumbing to disease (Riek, 1968). When a cow has been infected with *Babesia bovis* or

Babesia bigemina, she will produce antibodies to these parasites. New-born calves will absorb the antibodies secreted in the colostrum and be immune against infection against the homologous strain for 7-360 days, with a mean of 119 days (Hall, 1963; Hall *et al.*, 1968; Ross and Löhr, 1970; Callow, 1984). This immunity is sterile unless the calves become infected with the parasite (Hall, 1963; Ross and Löhr, 1970); the duration of such immunity depends on the amount of antibodies the calves have ingested (Riek, 1968). Should calves in this state of passive immunity become infected they generally do not manifest clinical signs but develop an acquired immunity (Latif *et al.*, 1979; Dallwitz *et al.*, 1986). Calves born to non-immune dams are susceptible to clinical disease (Hall, 1963), until an age-specific immunity takes over at 2-4 months and persists until the calf is 9 months of age (Riek, 1963; Trueman and Blight, 1978; Callow, 1984). This immunity is not dependent on the dam's immunological status (Callow, 1977; Riek, 1968; Mahoney and Ross, 1972; Mahoney, 1974; Mahoney *et al.*, 1979b).

- **Breed-dependent immunity.** *Bos indicus* cattle develop a relatively high degree of immunity after exposure to *Babesia bovis*, compared to that of *Bos taurus* cattle (Daly and Hall, 1955; Francis, 1966; Bigalke *et al.*, 1976; Johnston *et al.*, 1978; Johnston *et al.*, 1981; Callow, 1984; James *et al.*, 1985; Rechav and Kostrzewski, 1991; Bock *et al.*, 1997; 1999a). *Bos indicus* cattle have a lower level of *Babesia bovis* parasitaemia compared to *Bos taurus* cattle which are similarly infected (Johnston, 1967; Bock *et al*., 1997). In *Bos taurus* calves the passively acquired immunity to *Babesia bovis* does not prevent subclinical infection, while in *Bos indicus* calves such immunity may persist for several months and might in fact contribute to a potentially unstable situation by interfering with active immunization (Mahoney, 1974). *Bos indicus* cattle and their crosses rid themselves of patent parasitaemia sooner than *Bos taurus* breeds and after a while the parasite can no longer be detected in the blood (Johnston *et al.*, 1978). A similar difference has not been observed with *Babesia bigemina* (Johnston, 1967; Mahoney *et al*., 1973; Callow *et al*., 1974b). Under similar environmental conditions a *Bos indicus*-cross herd will have a lower rate of *Babesia bovis* transmission than a *Bos taurus* herd, and therefore needs a higher level of tick infestation to maintain endemic stability (Mahoney, 1979).A breed-dependent immunity to *Babesia bigemina* is less clear (Bock *et al.*, 1999b). Daly and Hall (1955) and Johnston (1967) found comparable immunological reactions to *Babesia bigemina* in different cattle breeds but De Vos (1979) and Bock *et al.* (1999b) found crossbred *Bos indicus* and *Bos indicus* steers significantly more resistant to *Babesia bigemina* than *Bos taurus* cattle. Resistance in *Bos indicus* varies according to the virulence of the parasite and, under similar conditions, *Bos taurus* cattle were found to be more susceptible to the clinical disease (Bock *et al*., 1999b). Jongejan *et al.* (1988) observed a low incidence of African redwater outbreaks in Zambia and concluded that this may have been due to the innate resistance of the local cattle breeds (Perry *et al.*, 1984).

2. 1. 9. Endemic stability to redwater. When redwater is first introduced into an area, cattle are highly susceptible to the disease, and a mortality rate of up to 40% has been reported in untreated Hereford cattle suffering from *Babesia bovis* infection (Callow, 1977; Trueman and Blight, 1978). De Vos (1979) reported mortality rates of 5 – 10 % in outbreaks of *Babesia bigemina* infections in South Africa. Endemic stability to both *Babesia bigemina* and *Babesia bovis* develops, but a stable situation for one *Babesia* species does not necessarily imply a stable situation for the other (De Vos, 1979). In Zimbabwe dipping was disrupted for a number of years and endemic stability to TBD developed rapidly after the initial heavy losses of nearly one million cattle. Subsequent losses were minimal (Norval, 1982; Norval *et al.*, 1983).

The maintenance of an endemically stable situation is dependent on a regular supply of ticks infected with *Babesia bigemina* and/or *Babesia bovis.* When the tick challenge is high a large number of infected ticks feed on the hosts, and there is a steady inoculation of the parasite (Callow, 1984). Young animals are protected by their age-specific immunity and are generally exposed to infection before the age of 9 months. Further repeated exposures to infected ticks ensure that a high level of antibodies is maintained in the animals and endemic stability develops. An infection rate in calves close to 100 % at the age of 9 months would indicate that endemic stability has been reached (Mahoney and Ross, 1972; Callow, 1977).

Norval *et al.* (1992b) defined endemic stability as "a climax relationship between host, vector and environment in which all co-exist with the virtual absence of clinical disease, while endemic instability means an incomplete relationship (between host, vector and environment) in which clinical disease occurs". In Zimbabwe Norval *et al*. (1983) described five different epidemiological situations for bovine babesiosis:

- Endemically stable situations (81 – 100 % positive sera)
- Situations approaching endemic stability (61 – 80 % positive sera)
- Endemically unstable situations (21 – 60 % positive sera)
- Minimal disease situations (1 – 20 % positive sera)
- Disease–free situations (0 % positive sera)

In an endemically stable region most of the calves would have seroconverted to *Babesia bovis* and/or *Babesia bigemina* by 9 months of age. Low infection rates in cattle over 9 months can lead to endemic instability and a risk of outbreaks of disease (Mahoney and Ross, 1972). Endemic stability can, however, be disrupted by intensive dipping or a change in climatic conditions (Bigalke *et al.*, 1976; De Vos, 1979; Norval, 1982; Callow, 1984). Cattle grazing for long periods on crop residue land, seasonal movement of herds in search of fresh grazing or abnormally cold winters may interrupt or delay transmission of TBD in the young animals and cause endemic instability (Dallwitz *et al*., 1986). *Boophilus* ticks rarely survive for longer than 6 months on pastures in the absence of hosts, and rotational grazing may prevent the young animals from reaching endemic stability to TBD (Bigalke *et al.*, 1976; Ardington, 1982).

2. 2. The tick vectors of bovine babesiosis in southern Africa.

Boophilus microplus has a worldwide distribution and is found in Asia, Australia, Central and South America, West Indies and parts of Africa. *Boophilus decoloratus* is found throughout Africa and is common in southern Africa (Brown *et al.*, 1990).

2. 2. 1. *Boophilus* species on domestic and wild animals in southern Africa.

- ***Boophilus decoloratus*** (African blue tick) is a one-host tick and one of the most common cattle ticks in South Africa. It has a wide host range but cattle are the main domestic animal hosts, while dogs, horses and donkeys also can be heavily infested (Theiler, 1962; Mason and Norval, 1980). Sheep and goats are also suitable hosts (Walker *et al.*, 1978).

  Wild animals can become infested with this tick, with Burchell's zebra (*Equus burchelli*), blue wildebeest (*Connochaetes taurinus*), black wildebeest (*Connochaetes gnou*), giraffe (*Giraffa camelopardalis*), African buffalo (*Syncerus caffer*), bushbuck (*Tragelaphus scriptus*), kudu (*Tragelaphus strepsiceros*), nyala (*Tragelaphus angasi*), eland (*Taurotragus oryx*), impala (*Aepyceros melampus*), grey rhebok (*Pelea capreolus*) and warthog (*Phacochoerus aethiopicus*) sometimes carrying quite heavy burdens (Horak, 1982; Horak *et al*., 1983; Horak *et al.,* 1984; Horak *et al*., 1986; Horak *et al*., 1988).

- ***Boophilus microplus*** (Asiatic cattle tick, pan tropical blue tick) is a one-host tick and cattle are the main hosts but it has also been found on other domestic animals such as sheep, goats, dogs and horses (Smith, 1983). Wild animals are rarely hosts of this tick (MacLeod and Mwanaumo, 1978; Walker, 1991; Boomker *et al*., 1983), but it has been collected off lion (*Panthera leo*), grey rhebok, sable antelope (*Hippotragus niger*), grey duiker (*Sylvicapra grimmia)* and African buffalo. There are indications that its potential range is similar to that of *Boophilus decoloratus* (Theiler, 1962; Horak *et al*., 1986; Walker, 1991).

2. 2. 2. Characteristics of the genus *Boophilus*. *Boophilus* species can be identified by the presence of an inornate scutum, a hexagonal *basis capitulum*, short mouth-parts, pale yellow legs, small eyes, absence of festoons, and the presence of anal plates in the male (Gothe, 1967a; Arthur and Londt, 1973; Howell *et al*., 1978; Walker *et al.*, 1978).

2. 2. 3. Comparative morphology. *Boophilus decoloratus* is difficult to distinguish from *Boophilus microplus* (Gothe, 1967a). Macroscopically, males and females of *Boophilus decoloratus* are somewhat lighter brown in colour than *Boophilus microplus* and the body of the *Boophilus decoloratus* female tends to be a little larger and more elongated than that of *Boophilus microplus.* The semi-engorged female *Boophilus microplus* is rounder with a slimmer "waist". Microscopically the principal features used for identification are the shape of the mouthparts of both males and females (Howell *et al*., 1978; Wedderburn *et al.,* 1991). *Boophilus decoloratus* has three rows of denticles on each side of the hypostome and a convex protuberance with setae on the medial aspect of the first palpal segments (Arthur and Londt, 1973). *Boophilus microplus* has four rows of denticles on each side of the hypostome and a concavity with no setae on the medial aspect of the first palpal segment.

The males of *Boophilus decoloratus* have long adanal plates, which reach beyond the posterior body margin and the long internal spurs are clearly visible outside the scutum. In *Boophilus microplus* the adanal plates do not reach beyond the posterior body margin and they have a short internal spur and an even shorter external spur (Gothe, 1967a; Arthur and Londt, 1973; Heyne, 1986).

2. 2. 4. Geographical distribution and seasonal incidence of *Boophilus decoloratus* and *Boophilus microplus* in southern Africa. The conditions for survival of both *Boophilus decoloratus* and *Boophilus microplus* are ideal over large areas of southern Africa (Theiler, 1962; McCosker, 1981). The two species occur together in many parts of the subcontinent, but because *Boophilus microplus* has more specific climatic requirements, *Boophilus decoloratus* has the wider distribution (De Vos, 1979). Temperature and precipitation (Theiler, 1962; Gothe, 1967b; De Vos, 1979) limit the spread of both ticks. The prevalence of both tick species seems to decline at higher altitudes in South Africa (Baker *et al.*, 1989), whilst *Boophilus decoloratus* is found in large numbers at higher altitude zones in Zimbabwe (Lawrence and Norval, 1979; Mason and Norval, 1980), Zambia (MacLeod and Mwanaumo, 1978), and Kenya (Gitau *et al.*, 1997).

2. 2. 4. 1. Distribution of *Boophilus decoloratus* in southern Africa. *Boophilus decoloratus* can survive in areas where there is a maximum of 90 days of frost spread over a period of 150 days a year (Gothe, 1967b). At lower temperatures there may be pockets of suitable climatic conditions where it can survive and develop (Theiler, 1949; Gothe, 1967b). The tick can survive in areas with an annual rainfall as low as 380 mm (Walker *et al*., 1978; De Vos, 1979), and it can tolerate even lower rainfall if the area is covered by bush rather than by open grassland (Theiler, 1949; Walker *et al*., 1978). Decreasing humidity seems to be the limiting factor in the tick's distribution (Theiler, 1949; Gothe 1967b).

- ***Boophilus decoloratus* in South Africa.** The climatic conditions in South Africa are not harsh enough to restrict the spread of *Boophilus decoloratus,* but cold conditions and low rainfall may limit its numbers and activity (Theiler, 1949; Gothe, 1967b; Rechav, 1982). In South Africa *Boophilus decoloratus* is widely distributed in the Northern Province, Gauteng, Mpumalanga and the eastern part of the North-West Province as well as KwaZulu-Natal. It also occurs in the northern and eastern part of the Free State Province, the northeastern and eastern parts of the Eastern Cape Province and in the southern coastal belt and winter rainfall areas of the Western Cape. It is absent from the desert shrub of Karoo veld, Namaqualand and the northwestern part of the Eastern Cape Province (Theiler, 1949; 1962; Baker and Ducasse, 1967; Londt *et al.*, 1979; Robertson, 1981; Rechav, 1982; Walker, 1991; Dreyer *et al.*, 1998a).

Boophilus decoloratus is present throughout the year in those areas where it normally occurs in South Africa (Baker and Ducasse, 1967; Robertson, 1981; Rechav, 1982). The main period of activity is from November to June, with peaks in July, September-October, December-January and March-April (Baker and Ducasse, 1967; Howell *et al.*, 1978; Londt *et al.*, 1979; Robertson, 1981; Rechav, 1982; Baker *et al.*, 1989; Spickett *et al*., 1989; Rechav and Kostrzewski, 1991; Tice, 1997). The evidence suggests that its life cycle probably has two to four generations *per annum* (Rechav, 1982; Rechav and Kostrzewski, 1991; Dreyer *et al.,* 1998a). Peaks were found in late autumn (March to May) and in winter (June to August) in the Free State Province, where a survey demonstrated that nearly 80 % of the annual *Boophilus* burden occurred during the cooler months of the year (Dreyer *et al*., 1998a)

- ***Boophilus decoloratus* in Zimbabwe.** The tick is present in most regions of the country but is more common in the higher rainfall eastern part of Zimbabwe (Theiler, 1962; Lawrence and Norval, 1979; Mason and Norval, 1980; Norval *et al.*, 1983). It is absent in those areas of the country which have been cleared of wild and domestic ungulates due to tsetse fly control, and small populations only are found in the dry south-western lowveld (Mason and Norval, 1980). *Boophilus decoloratus* is present in Zimbabwe throughout the year without exhibiting distinct seasonal peaks, and is thought to have an annual seasonal cycle of two to four generations (Jooste, 1966; Matson and Norval, 1977; Mason and Norval, 1980).

- ***Boophilus decoloratus* in Zambia.** The tick is widely distributed in the central, southern and western parts of Zambia (Theiler, 1962; MacLeod and Mwanaumo, 1978; Pegram *et al.*, 1986). It can survive in the hot and dry Luangwa Valley, where *Boophilus microplus* is absent, and in the cooler and wetter high-altitude areas in other districts (MacLeod and Mwanaumo, 1978). There are two periods of abundance in Zambia (March to July and September to December) and the tick probably completes two to five generations per year (MacLeod, 1970; Pegram *et al.*, 1986).

- ***Boophilus decoloratus* in Swaziland.** The tick is widespread in Swaziland (Theiler, 1949; Jagger *et al.*, 1985; Walker, 1991; Wedderburn *et al.*, 1991).

- ***Boophilus decoloratus* in Mozambique.** *Boophilus decoloratus* is probably present throughout the country (Theiler, 1962).

- ***Boophilus decoloratus* in Botswana.** The tick is mainly present along the eastern and south-eastern agricultural strip on the border with South Africa and Zimbabwe as well as in the Okavango Delta and in the north-eastern Chobe District (Theiler, 1962; Walker *et al.*, 1978; Walker, 1991).

- ***Boophilus decoloratus* in Namibia.** The tick occurs from the thornveld of Damaraland in the north to Windhoek in the south and it has also been collected in Ovamboland, Okavango and in the Caprivi Strip (Theiler, 1962; Biggs and Langenhoven, 1984; Walker, 1991).

2. 2. 4. 2. Distribution of *Boophilus microplus* in southern Africa. *Boophilus microplus* prefers warm and humid conditions and can survive in areas where there is a maximum of 60 days of frost, spread over a period of 150 days a year (Gothe, 1967b). The larvae are susceptible to cold and can only tolerate 0° C for 72 hours. Cold seems to be the limiting factor in the tick's distribution (Gothe, 1967b; De Vos, 1979). It is not known how tolerant adults are of cold. Temperatures must be at least 15-20° C for egg laying and larval hatching to occur, with a maximum upper threshold of 40 °C. The relative humidity must be at least 80 % for eggs to survive (Callow, 1984; Sutherst and Maywald, 1985), and the tick is absent in areas with annual rainfall of less than 500 mm (De Vos, 1979). The seasonal changes seem to be similar to those of *Boophilus decoloratus* (Arthur and Londt, 1973; De Vos, 1979; Baker *et al*., 1989).

- ***Boophilus microplus* in South Africa.** Cold conditions seem to have restricted the spread of *Boophilus microplus* in South Africa (Gothe, 1967a). The first recorded report of *Boophilus microplus* in South Africa was in 1908 when Howard stated that it was present in the southeastern districts of the then Cape Province (Theiler, 1962). In the survey by Theiler (1962), *Boophilus microplus* was found in the mild and humid coastal strip between Bredasdorp and Port Elizabeth where rainfall occurs all year round. More recent surveys have indicated that *Boophilus microplus* is present in northern Gauteng, parts of Mpumalanga, large areas of KwaZulu-Natal, parts of the Eastern Cape Province and along the southern Cape coast (Howell *et al.*, 1978; De Vos, 1979; Baker *et al.*, 1989; Walker, 1991). The main periods of seasonal activity are similar to *Boophilus decoloratus* (De Vos, 1979).

- ***Boophilus microplus* in Zimbabwe.** *Boophilus microplus* was probably introduced from Mozambique in the mid-1970s (Mason and Norval, 1980; Norval *et al.,* 1992a) when it was restricted to the eastern and northern part of the country. *Boophilus microplus* was later found close to the South African border, and there was serological evidence of *Babesia bovis* in the area (Mason and Norval, 1980; Norval *et al.*, 1983). The population dynamics are similar to those of *Boophilus decoloratus* (Mason and Norval, 1980; Norval *et al.*, 1983). After the drought of 1981-1984 *Boophilus microplus* was thought to have disappeared completely from Zimbabwe (Norval, unpublished data, cited by Norval *et al.*, 1992a). However, *Babesia bovis* antibodies were detected in the eastern and northern parts of Zimbabwe and *Boophilus microplus* was collected in the eastern and north-western part of the country (Katsande et al., 1996). *Boophilus microplus* is probably still present in these areas (Katsande *et al.*, 1999) and its presence in the south-eastern lowveld indicated that the tick could survive in most areas of Zimbabwe (Mason and Norval, 1980).

- ***Boophilus microplus* in Zambia**. *Boophilus microplus* was first recorded in northern Zambia (Theiler, 1962), whilst MacLeod and Mwanaumo (1978), Pegram *et al.* (1986) and Berkvens *et al*. (1998) reported that it was widely distributed in the eastern and northern sector of the country, where it had partially or totally displaced *Boophilus decoloratus.* The tick was found at intermediate altitudes in hot and dry areas where it would be expected to be absent in light of current knowledge of the tick's climatic requirements (Berkvens *et al*., 1998). Two patterns of seasonal abundance were present: in areas with low *Boophilus microplus* numbers peaks were recorded in April-May and in August. Four generations per year were found in areas with high tick numbers (Berkvens *et al*., 1998).

- ***Boophilus microplus* in Swaziland.** *Boophilus microplus* was first recorded in Swaziland during a survey in 1985 after a series of outbreaks of bovine babesiosis caused by *Babesia bovis* (Jagger *et al*., 1985). Its distribution in Swaziland is patchy, but it is present throughout the country (Wedderburn *et al*., 1991).

- ***Boophilus microplus* in Mozambique.** The tick is presumably present throughout Mozambique (Theiler, 1962), although definite reports on the distribution of the tick in this country are difficult to access.

- ***Boophilus microplus* in Namibia.** The tick has not been recorded in Namibia (Walker, 1991).

- ***Boophilus microplus* in Botswana.** The tick has not been recorded in Botswana (Walker *et al.*1978; Walker, 1991).

2. 2. 5. Interbreeding and competition between *Boophilus* species. Shortly after the introduction of *Boophilus microplus* into South Africa researchers stated that the tick (as *Boophilus fallax*, Howard, 1908; as *Boophilus annulatus*, Dönitz, 1910) was ousting *Boophilus decoloratus* from the latter's endemic areas (Theiler, 1962). Observations in the field of mixed infections of *Boophilus decoloratus* and *Boophilus microplus* revealed that, where the two species co-existed, there was a tendency for *Boophilus microplus* to displace *Boophilus decoloratus* partially or totally (MacLeod and Mwanaumo, 1978; Mason and Norval, 1980; Norval *et al*., 1983; Norval and Short, 1984; Norval and Sutherst, 1986; Sutherst, 1987b; Wedderburn *et al.*, 1991; Berkvens *et al*., 1998; Baker, 2001, personal communication).

This displacement is rapid and *Boophilus microplus* can completely displace *Boophilus decoloratus* in 4-10 generations, which would generally take 1-3 years to complete (Sutherst, 1987b). Several authors, who argued that it could be related to climatic factors, reproductive capability, interspecific competition on the host, adaptation to the environment and different resistance patterns to acaricides, have discussed the potential mechanisms for the displacement.

Boophilus decoloratus is more tolerant of low temperatures and dry conditions than *Boophilus microplus* (Theiler, 1949; Gothe, 1967b). Arthur and Londt (1973) described a shorter life cycle for *Boophilus microplus* than for *Boophilus decoloratus.* Spickett and Malan (1978) found that the two species were genetically incompatible as cross-matings produced sterile eggs. As *Boophilus* females mate once only (Londt, 1976), cross-matings would result in decreased *Boophilus* fertility. In areas where both species were present, their numbers were low, possibly because of this cross-mating tendency (Baker *et al.*, 1989).

The replacement of *Boophilus decoloratus* by *Boophilus microplus* seemed only to a small degree to be due to reproductive competition. Norval and Sutherst (1986) showed that the cross-matings were not random events, but that there was a tendency for assortative mating (i.e. each species will mate with their own species if this is possible) to occur. As a result there were fewer hybrid matings than would be expected if mating was random. Hilburn and Davey (1992) doubted these results and concluded that due to different development times of the two species, the number of assortative matings was probably higher.

The attatchment sites on the animal are similar for both species (Howell *et al.*, 1978). Norval and Short (1984) found that *Boophilus microplus* fed more successfully on cattle than did *Boophilus decoloratus* and more females of *Boophilus microplus* completed feeding and continued their developmental stages on cattle. The presence of *Boophilus microplus* on cattle appears to enhance their resistance to *Boophilus decoloratus*: a reduction in engorged weight resulted in *Boophilus decoloratus* females producing fewer eggs, which contributed to a decrease in their population. Interspecific competition on the host was the most likely explanation for the displacement of

Boophilus decoloratus by *Boophilus microplus* (Norval and Short, 1984). *Boophilus decoloratus* would probably not be present where *Boophilus microplus* is already well established (Norval *et al*., 1983).

Sutherst (1987b) showed in a computer model that the displacement was due to reproductive interference combined with faster population growth rates by *Boophilus microplus.* In warm, high rainfall areas this gave *Boophilus microplus* an advantage over *Boophilus decoloratus* of 3.5 in terms of population growth potential. In colder and drier areas with a resident wild ungulate population acting as a host reservoir for *Boophilus decoloratus*, the advantage was negligible and here *Boophilus decoloratus* would probably persist (Sutherst, 1987b).

Mason and Norval (1980) described a similar pattern of displacement in Zimbabwe, and suggested that the displacement of *Boophilus decoloratus* by *Boophilus microplus* may be due to changing environments, such as changes in weather patterns, or development of acaricide resistance by *Boophilus microplus.* In Zimbabwe displacement did take place in the absence of dipping as well as in areas where widly differing climate and weather conditions occurred.

In Zambia, MacLeod and Mwanaumo (1978) found that *Boophilus decoloratus* had been displaced by *Boophilus microplus* in large areas in the Central Province. Berkvens *et al*. (1998) found *Boophilus decoloratus* still present in areas with low stocking rates, indicating that less intense competition between the species favoured *Boophilus decoloratus.*

Where *Boophilus decoloratus* and *Boophilus microplus* occur together, their relative resistance to acaricides is uncertain. Baker *et al.* (1968) reported greater resistance of *Boophilus microplus* larvae to some acaricides, but from later trials it was concluded that *Boophilus microplus* was more susceptible to the most commonly used acarides than *Boophilus decoloratus* (Solomon *et al*., 1979; Baker *et al*., 1981).

2. 2. 6. Infection rates of *Babesia* species in *Boophilus* species. The infection rate can be defined as the proportion of tick larvae (*Babesia bovis*) or nymphs and adults (*Babesia bigemina*) harbouring the *Babesia.* The transmission of *Babesia* parasites to susceptible cattle is dependent on the proportion of ticks harbouring *Babesia* combined with the ability of these ticks to pass on the infection to cattle (Mahoney, 1974).

2. 2. 6. 1. Observed *Babesia bigemina* and *Babesia bovis* infections in ticks. Riek (1964; 1966) found infections of *Babesia bovis* and *Babesia bigemina* in *Boophilus microplus* of 90 % when the tick had fed on cattle with tick-transmitted infections. Johnston (1967) reported that *Babesia bigemina* infection in *Boophilus microplus* ranged from 2-10 % whilst *Babesia bovis* infections ranged from 0.06-0.47 %. Mahoney and Mirre (1971) recorded that 0.5-14 % of *Boophilus microplus* larvae contained *Babesia bovis* whilst 20-40 % contained *Babesia bigemina*. Mohammed (1976) found *Babesia bigemina* infections in *Boophilus decoloratus* that varied between 2 and 10 %. Gray and Potgieter (1981) reported close to 30 % of *Boophilus decoloratus* ticks infected with *Babesia bigemina*. By using Polymerase Chain Reaction (PCR), Smeenk *et al.* (2000) found that 5 % of *Boophilus decoloratus* were positive for *Babesia bigemina* while 60 % were positive for *Babesia bovis*. With *Boophilus microplus* the infection was even higher, with 69 % of the ticks being positive for *Babesia bovis*. Smeenk *et al*. (2000) were also able to demonstrate simultaneous infections with both *Babesia bigemina* and *Babesia bovis*. The *Babesia* DNA was extracted from the haemolymph. These findings, however, do not imply that the parasite can undergo further development in the salivary gland of the tick and thus become infective to cattle. There is strong experimental and epidemiological evidence that *Boophilus microplus* is the only vector of *Babesia bovis* in southern Africa (Potgieter, 1977; Potgieter and Els, 1979; Norval *et al*., 1983).

2. 6. 6. 2. Infectivity of ticks to cattle. Riek (1964) suggested that the majority of parasites ingested by a tick die, and that only a very small proportion undergoes further development. The infection rates of *Babesia* species in *Boophilus* ticks decrease as the ticks and the *Babesia* parasites go through developmental stages, with the result that the subsequent ability of ticks to transmit the infection is low. Studies by Mahoney and Mirre (1971) and Mahoney *et al*. (1981) showed that the infection of *Babesia bovis* in *Boophilus microplus* larvae was as low as 0.04-0.07 %. The infection of *Babesia bigemina* in *Boophilus microplus* larvae and nymphs was higher at 0.23 % (Mahoney and Mirre, 1971). Dallwitz *et al*. (1986) gave infection rates of these immature stages for *Babesia bovis* of 0.03 % and those for *Babesia bigemina* of 0.1-0.5 %. With such low prevalence of infection in the ticks the chance of animals becoming infected was also low (Callow, 1984). Nevertheless, only one infected tick is required to transmit *Babesia bigemina* or *Babesia bovis* to susceptible cattle (Mahoney and Mirre, 1971).

2. 2. 7. Environmental factors affecting the *Babesia* parasites in the tick. A number of different environmental factors, tick strains, methods of infection and parasite density in the vertebrate host can affect the development of the *Babesia* parasites in the tick (Riek, 1964; 1966; Mahoney *et al.*, 1981). The development of both *Babesia bigemina* and *Babesia bovis* in *Boophilus microplus* was slower at temperatures below 20° C (Riek, 1963; 1964; 1966), whilst higher temperatures stimulated the development (Dalgliesh and Stewart, 1979; 1982; Dalgliesh *et al.*, 1979; Ouhelli and Schein, 1988). Riek (1966; 1968) found that different strains of *Boophilus microplus* had different susceptibilities to infection with *Babesia bovis*, and heavy *Babesia* infections as well as virulent *Babesia bovis* strains could result in tick mortality (Riek, 1966, 1968; Dalgliesh *et al*., 1981; Callow, 1984). If the infection rate of *Babesia bigemina* and *Babesia bovis* in the host is too high, the engorged females will die (Riek, 1964; 1966).

2. 2. 8. Inoculation rate. This rate is a measure of the average daily probability that an animal in a herd will become infected with babesiosis (Mahoney and Ross, 1972; Mahoney, 1974). The inoculation rate (h) can be defined as the number of tick bites (m) received by the host per day, multiplied by the proportion of the vector population carrying infective forms of the organism (a) and the proportion of bites that successfully infect the host (b) (Mahoney, 1974). The formula to calculate the inoculation rate is:

$$h = mab$$

In this model the number of ticks biting each animal per day is important and the higher the inoculation rate, the higher the number of calves infected whilst being protected by age-specific resistance. In an endemically stable situation the inoculation rate ranged from 0,005 to 0,05, depending on the number of ticks in the field (Mahoney, 1979). A minimum number of *Boophilus* tick bites to maintain endemic stability in a herd of exotic cattle would be at least 20 bites per day. *Bos indicus* cattle, however, had a higher level of resistance against *Babesia bovis* than *Bos taurus* cattle and would need a minimum of 40 tick bites a day to maintain stability (Mahoney *et al.*, 1981). Smith (1983) used a computer model to calculate the number of *Boophilus* ticks necessary to maintain endemic stability and suggested 8-9 engorged ticks/day as the optimal number. Jongejan *et al.* (1988) calculated inoculation rates in calves in the range of 0.05-0.3 % for *Babesia bovis* and 0.3-0.6 % for *Babesia bigemina* and concluded that the situation seemed endemically unstable, but that there were no disease outbreaks. The age-specific prevalence rates, however, indicated endemic stability, (Jongejan *et al.*, 1988).

2. 2. 9. Breed resistance against *Boophilus* species. There are major differences between *Bos indicus* and *Bos taurus* breeds in their resistance to *Boophilus* spp. (Bonsma, 1981; Hewetson, 1981). Under similar environmental conditions *Bos indicus* cattle were infested with lower numbers of ticks (Johnston, 1967; Mahoney, 1979; Sutherst and Comins, 1979; Sutherst *et al*., 1979; Bonsma, 1981; Mahoney *et al.*, 1981; Kaiser *et al.* 1982; Rechav and Zeederberg, 1986; Rechav and Kostrzewski, 1991; Fourie *et al.*, 1996). The resistance in cattle is related to the thickness of the skin, the amount of subcutaneous muscles, the mobility of the cow's tail and the quality of the coat, all of which prevent the ticks from becoming attached and engorging fully (Francis, 1966; Bonsma, 1981). The numbers of ticks on *Bos indicus* and *Bos indicus/Bos taurus* crossbreeds were significantly less than on purebred *Bos taurus* cattle

(Francis, 1966; Utech *et al*., 1978; Bonsma, 1981; Sutherst and Utech, 1981; Norval *et al*., 1992b). Crosses with Zebu-type cattle seem to have particular resistance-building qualities (Sutherst and Utech, 1981; Spickett *et al*., 1989).

2. 2. 10. Other factors affecting host resistance against *Boophilus* species. An infection with *Babesia bovis* can cause immunosuppression against *Boophilus microplus*; and Callow and Stewart (1978) demonstrated that calves infected with *Babesia bovis* had a larger tick burden than uninfected calves. The *Babesia* parasite thus improves its chance of survival and transmission by increasing the number of its vectors, its prevalence being related to vector density (Callow and Stewart, 1978). Malnutrition reduces the resistance to ticks (O'Kelly and Seifert, 1969) and factors such as lactation, sex and age may affect resistance (Utech *et al.*, 1978; Sutherst and Utech, 1981). It appears that some animals have an innate resistance to ticks as they consistently carry fewer ticks than others do in the same group (Sutherst *et al*., 1979; Petney *et al.*, 1990; Latif *et al*., 1991; Dreyer *et al.*, 1998b). Resistance to a new tick species starts developing as soon as cattle are challenged and will increase after prolonged exposure. The resistance is proportional to the degree of tick challenge (Utech *et al*., 1978).

When calves become infested with *Boophilus microplus* it might take as long as 2 years before their level of resistance stabilizes. Calves normally carry lighter tick burdens than adult cattle as tick infestations have been found to be nearly three times higher on their dams. This suggests that young animals might be protected by some age-related resistance (Mahoney, 1979; Sutherst *et al*., 1979). Prolonged tick challenge later in life further promotes host resistance against ticks (Sutherst and Utech, 1981). Excessive grooming and close proximity between individuals can lead to a transfer of *Boophilus* spp. between animals (Sutherst *et al*., 1979) and this may be a mode of transfer when *Boophilus microplus* invades a new area (Mason and Norval, 1981).

2. 3. The role of models in tick control.

Computer models have been constructed to highlight the relationship between environmental factors and tick ecology and are used to predict the potential distribution of various tick species (Sutherst and Maywald 1985, 1986; Sutherst, 1987a; Sutherst *et al*., 1991; Sutherst *et al*., 1995; Sutherst, 1998). Models can be used to develop a holistic approach to a tick-parasite-host system and to decide on the best approach to combat disease in different parts of the world (Dallwitz *et al*., 1986).

2. 3. 1. CLIMEX. This computer-based system allows the prediction of the possible distribution and survival of a tick species, using known biological and climatical data (Sutherst and Maywald, 1985; Sutherst, 1987a). The model is used in an attempt to predict population growth during favourable and unfavourable seasons. An “Ecoclimatic Index” (EI) is derived which indicates the climatic favourability for the location of a tick species. The index runs from 0 to 100 and with a low EI, there is a greater likelihood of endemic instability. An EI of less than 20 indicates that the tick is not well suited to the environment, and the tick population will be low in that area. Transmission of TBD in these areas would be intermittent (Dallwitz *et al*., 1986; Sutherst and Maywald, 1986).
In areas where the EI is less than 5, ticks will not be able to heavily parasitize cattle. In these areas endemic stability may not be maintained, but it may be possible to eradicate

a tick species or vaccinate against TBD. A high EI normally identifies areas that may be permanently occupied by the tick (Sutherst and Maywald, 1985; Sutherst, 1987a).

The index is derived from a population growth index that shows the potential for an increase in the population, and four stress indices which describe the negative effects of extreme cold, dry, hot and wet conditions (Sutherst and Maywald, 1985). If the stress indices reach 100, they exclude persistence of the species in that environment (Sutherst, 1998). When dryness is the limiting factor, however, the presence of local swamps or irrigation may provide favourable habitats for the tick species even if the EI is low (Sutherst and Maywald, 1985).

2. 3. 2. Earlier predictions using the CLIMEX model. Sutherst and Maywald (1985) used the CLIMEX model to predict the possible spread of *Boophilus microplus* into new areas in South Africa. At that stage the tick had not been found in the Soutpansberg region, but there were indications that *Babesia bovis* was present (Sutherst and Maywald, 1985; Sutherst, 1987a; Gous, personal communication, 1999).

CHAPTER 3. MATERIALS AND METHODS

3. 1 Survey areas.

The survey area was in the Soutpansberg, Dzanani, Mutale, Thohoyandou and Vuvani Districts in the Northern Province of South Africa and the survey was carried out between May 1999 and December 2000. The region was chosen because of recent outbreaks of bovine babesiosis caused by *Babesia bovis* (Loock, personal communication, 1999). The area borders the Kruger National Park (KNP) to the east, Zimbabwe to the north, the Vivo-Dendron road (R 521) to the west and the Pietersburg-Giyani road (R 81) to the south. Sibasa and Louis Trichardt are the administrative centres of the veterinary services in this area. The communal farming areas were divided into different wards, which were each serviced by an animal health technician who would normally oversee the dipping. Five commercial farms and 30 communal dip tanks were included in the survey.

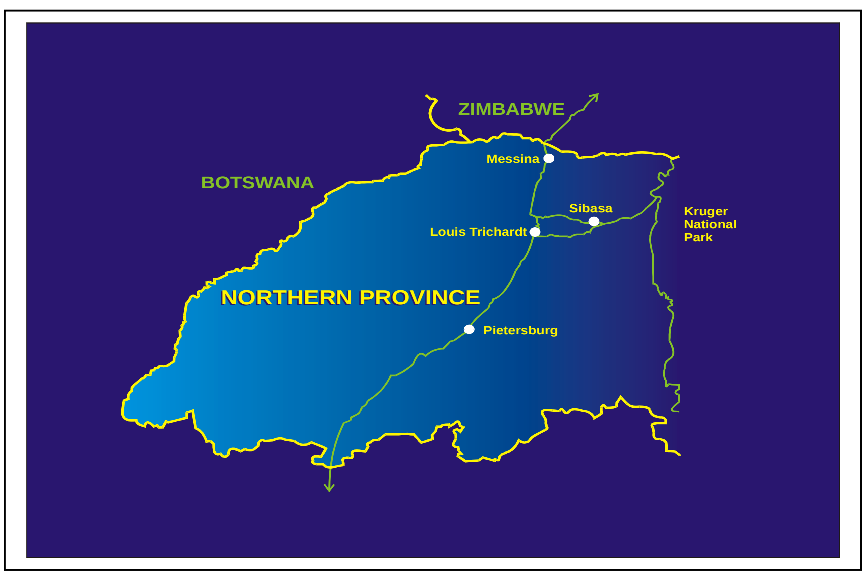


Fig. 3. 1. Map of the Northern Province of South Africa showing the survey area.

3. 1. 1. The communal farming areas. There were several dip tanks in each ward and each dip tank serviced the cattle in an area with a maximum radius of 20 km. The cattle belonged to several farmers and were maintained under traditional methods of animal husbandry. They were periodically taken on set days to the communal plunge-type dip tanks in order to control ectoparasites. The cattle dipped at the communal dip tanks situated in these areas were mostly indigenous cattle breeds or a mix of indigenous and exotic breeds. They were mainly kept for meat production, but were also milked for home consumption for most of the year. They usually grazed on unfenced communal land during the day and were not fed concentrates, but were allowed access to harvested maize fields when available. The pastures were moderately to heavily overgrazed and the animals were held in kraals at night with little or no housing facilities. No vaccination against *Babesia bigemina* or *Babesia bovis* was undertaken in the area. The cattle were not marked with ear-tags so individuals were not readily identifiable for repeated sampling.

Compulsory dipping was practised for many years (Bigalke *et al.*, 1976) and the cattle had been dipped regularly at weekly or fortnightly intervals. During 2000, however, the government subsidies for acaricides were discontinued and the farmers were expected to pay for the acaricide. Due to the poor economic conditions and serious flooding during February and March 2000, dipping had been discontinued in some areas and became irregular in others. Prior to 1996 Triatix (Amitraz + Ca-hydroxide, Intervet) was widely used, but all the dip tanks were using Grenade (Cyhalothrin, Intervet) at the time of the study.

3. 1. 1. 1. Location of the different communal dip tanks.

Thononda. Geographic co-ordinates: 22° 52' 52,2" S; 30° 15' 13,7" E. Nine hundred cattle were usually dipped at this dip tank.

Dzondo. Geographic co-ordinates: 23 ° 02' 39.0'' S; 30 ° 21' 56.2'' E. Three hundred and fifty cattle were usually dipped at this dip tank.

Guyuni. Geographic co-ordinates: 22° 48' 07.2'' S; 30° 31' 43.7'' E. Eight hundred cattle were usually dipped at this dip tank and 25% were young animals

Sendedza. Geographic co-ordinates: 22 ° 54' 17.6'' S; 30 ° 11' 04.2'' E. Nine hundred cattle were usually dipped at this dip tank.

Luvhanga. Geographic co-ordinates: 23 ° 01' 05.5'' S; 30 ° 31' 00.9'' E. Fourteen hundred cattle were usually dipped at this dip tank.

Muledzhi. Geographic co-ordinates: 22 ° 41' 23.7'' S; 30 ° 37' 07.8'' E. Six hundred cattle were usually dipped at this dip tank and 25 % were young animals.

Malavuwe. Geographic co-ordinates: 22 ° 52' 12.4'' S; 30 ° 37' 58.6'' E. Eight hundred cattle were usually dipped at this dip tank and 25 % were young animals.

Makwarani. Geographic co-ordinates: 22° 51’ 26.4 S; 0° 24’ 36.3’’ E. Six hundred cattle were usually dipped at this dip tank and 15% were young animals. The cattle were in poor condition and there had been excessive mortality in the calves due to internal parasites.

Lamvi. Geographic co-ordinates: 22° 39.2’ S; 30° 47.3’ E. Seven hundred cattle were usually dipped at this dip tank and 25 % were young animals.

Tshaulu. Geographic co-ordinates: 22° 48’ 24,5” S; 30° 45’ 04,4” E. Six hundred cattle were usually dipped at this dip tank and 20-25 % were young animals.

Davhana. Geographic co-ordinates: 23° 12.6’ S; 30° 28.6’ E. Fifteen hundred cattle were usually dipped at this dip tank.

Mahagala. Geographic co-ordinates: 22° 45’ 52,1” S; 30° 51’ 09,4” E. Six hundred cattle were dipped weekly at this dip tank and 25 % were young animals.

Matshena. Geographic co-ordinates: 22° 28’ 11.9’’ S; 30° 36’ 46.6’’ E. Nine hundred cattle were dipped fortnightly at this dip tank.

Phipidi. Geographic co-ordinates: 22° 56’ 41,9” S; 30° 24’ 16,0” E. Two hundred and thirty cattle were dipped every second week at this dip tank.

Shakadza. Geographic co-ordinates: 22° 36’ 48,6” S; 30° 32’ 59,7” E. Eight hundred cattle were dipped every second week at this dip tank.

Fesekraal 1. Geographic co-ordinates: 22° 24’ 04.4’’ S; 30° 35’ 04.4’’ E. Two hundred cattle were dipped every second week at this dip tank.

Matatani. Geographic co-ordinates: 22° 32’ 44,3” S; 30° 44’ 37,7” E. Nine hundred cattle were usually dipped at this dip tank. The dip tank was located in a dry area, but some of the cattle herds grazed on the southern slopes of the Soutpansberg Mountains where there were favourable conditions for tick survival.

Tshiendeulu. Geographic co-ordinates: 22° 49’ 20,3” S; 30° 11’ 06,7” E. Six hundred and fifty cattle were usually dipped at this dip tank.

Khakhu. Geographic co-ordinates: 22° 50’ 16,3” S; 30° 15’ 19,4” E. Twelve hundred cattle were usually dipped at this dip tank.

Murangoni. Geographic co-ordinates: 22° 54’ 23,4” S; 30° 23’10,8” E. Two hundred and seventy cattle were usually dipped at this dip tank.

Gondeni. Geographic co-ordinates: 22° 54’ 39,2” S; 30° 26’ 33,2” E. Six hundred cattle were usually dipped at this dip tank.

Savhani. Geographic co-ordinates: 22° 40’ 41,1” S; 30° 30’13” E. Twelve hundred cattle were dipped at this dip tank and they were dipped every second or third week.

Tshikotoni. Geographic co-ordinates: 22° 39' 34,3" S; 30° 26' 41,7" E. Eleven hundred cattle were dipped every second week and 15-20 % were young.

Mphephu. Geographic co-ordinates: 22 ° 52' 22.1'' S; 30 ° 06' 37.7'' E. A thousand cattle were usually dipped at this dip tank.

Keerweerder. Geographic co-ordinates: 22 ° 42' 34.2'' S; 30 ° 11' 01.0 '' E. Six hundred cattle were usually dipped at this dip tank and they were a mixture of Afrikander and Brahman breeds.

Masetoni. Geographic co-ordinates: 22 ° 42' 52.0'' S; 30 ° 53' 18.7'' E. Two hundred and eighty cattle were usually dipped at this dip tank.

Fripp. Geographic co-ordinates: 22 ° 48' 36.3'' S; 29 ° 57' 01.5'' E. Three hundred and seventy cattle were usually dipped at this dip tank.

Maunguwi. Geographic co-ordinates: 22° 49' 51.8'' S; 30° 03' 34.8'' E. Three hundred and eighty cattle were usually dipped at this dip tank.

Sambandou. Geographic co-ordinates: 22° 44.2' S; 30° 39.6' E. Nine hundred and fifty cattle were usually dipped at this dip tank.

Makonde Project. Geographic co-ordinates: 22° 46' 38.2'' S; 30° 32' 38.0'' E. One hundred and twenty cattle were usually dipped at this dip tank.

3. 1. 1. 2. Vegetation types and climatic conditions at the dip tanks. Two major veld types were found in the research area, namely Sour Lowveld Bushveld and Soutpansberg Arid Mountain Bushveld (Acocks, 1975; Low and Rebelo, 1996). A few of the dip tanks were situated in Mopani Bushveld and Mixed Lowveld Bushveld.

- **Sour Lowveld Bushveld.** This vegetation type was found mainly on the lower eastern slopes of the Soutpansberg Mountains at altitudes between 550 and 800 m. The annual rainfall in the area varied from 600 to 1000 mm, and temperatures varied from 2 ° C to 43 ° C with an average of 22 ° C. The soil types varied from deep sandy soils at the higher altitudes to more clay-like soils derived from granites and gneisses in the lower areas. Riverine forests and open tree savanna vegetation were common. Common trees and shrubs in the area included silver clusterleaf (*Terminalia sericea*), bushwillow (*Combretum collinum*), paperbark thorn (*Acacia sieberiana*), common hook-thorn (*Acacia caffra*), common wild fig (*Ficus thonningii*) and spineless monkey orange (*Strychnos madagascariensis*). In the shrub layer sickle bush (*Dichrostachys cinerea*), large sourplum (*Ximenia caffra*) and camel's foot (*Piliostigma thinningi*) were found. The common grasses consisted of yellow thatching grass (*Hyperthelia dissoluta*), common thatchgrass (*Hyparrhenia hirta*) and wiregrass (*Elionurus muticus*).

  The dip tanks which were located in this vegetation zone were Thononda, Dzondo, Guyuni, Sendedza, Luvhanga, Muledzhi, Malavuwe, Makwarani, Lamvi, Tshaulu, Mahagala, Phipidi, Tshiendeulu, Khakhu, Murangoni, Gondeni, Tshikotoni, Masetoni, Sambandou and Makonde Project.

- **Soutpansberg Arid Mountain Bushveld.** This vegetation type was found on the dry, hot, rocky northern slopes and summits of the Soutpansberg Mountains at altitudes between 300 and 2050 m. The annual rainfall in the area varied from 300 mm in the north to 500 mm on the plateau, and temperatures varied from 3 ° C to 44 ° C, with an average of 23 ° C. The soil was mainly acidic sandy, loamy and gravelly soil derived from sandstone, quartzite and shale. The main trees in this area were white seringa (*Kirkia acuminata*), red bushwillow (*Combretum apiculatum*), common hook-thorn, red seringa (*Burkea africana*), silver clusterleaf and Lebombo ironwood (*Androstachys johnsonii*). The shrubs included spineless monkey orange, velvet sweetberry (*Bridelia mollis*), redheart tree (*Hymenocardia ulmoides*) and shakama plum (*Hexalobus monopetalus*). Guinea grass (*Panicum maximum*), common fingergrass (*Digitaria eriantha*) and tassel three-awn (*Aristida congesta*) were common grasses in the area.

  The dip tanks that were located in this vegetation type included Matshena, Shakadza, Matatani, Savhani, Mphephu, Keerweerder, Fripp and Maunguwi.

- **Mopani Bushveld.** This vegetation type was located from the KNP to the Soutpansberg, to the north of the mountains and into Zimbabwe at altitudes between 300 and 700 m. The rainfall in the area varied from 250 to 550 mm per year, and the temperatures varied from 1.5 ° C to 42.5 ° C with an average of 22 ° C. The soil types varied from sandy and clay-like soils in the KNP to sandstone, shale and basalt north of the Soutpansberg and in the Limpopo Valley. Mopane (*Colophospermum mopane*), red bushwillow, knob thorn (*Acacia nigrescens*) and baobab (*Adansonia diditata*) were the most common trees. The shrub layer consisted of wild raisin bush (*Grewia* spp.), three-hook thorn (*Acacia senegal*), small sourplum (*Ximenia americana*) and other *Ximenia* spp. The common grasses consisted of common nine-awn grass (*Enneapogon cenchroides*), tassel three-awn, Kalahari sand quick (*Schmidtia pappophoroides*) and *Panicum* spp.

  Fesekraal was the only dip tank located in this vegetation type.

- **Mixed Lowveld Bushveld.** This vegetation type was found on flat and undulating landscapes at altitudes between 350 and 500 m. The annual rainfall in the area varied from 450 to 600 mm and the temperatures varied from – 4 ° C to 45 ° C, with an average of 22 ° C. The soil was sandy and clay-like in the higher parts, with a high sodium content in the lower parts. The vegetation was dense and bushy with open savanna in the low-lying areas and forest along the riverbanks. Common trees and bushes were red bushwillow, largefruit bushwillow (*Combretum zeyheri*) and silver clusterleaf. In the more low-lying places knobthorn, scented thorn (*Acacia nilotica*) and common falsethorn (*Albizia harveyi*) were found. The shrub layer consisted of hairy corkwood (*Commiphora africana*), wild grape (*Cissus cornifolia*) and sickle bush. The grass layer was poorly to moderately developed and among the grasses found were herringbone grass (*Pogonarthria squarrosa*), blueseed grass (*Tricholaena monachne*) and curlyleaf lovegrass (*Eragrostis rigidor*). Grasses such as Kalahari sand quick, spreading bristlegrass (*Aristida congesta*) and bushveld signalgrass (*Urochloa mosambicensis*) were also common.

Davhana was the only dip tank located in this vegetation type.

3. 1. 2. The commercial farming areas. The cattle on the commercial farms were mostly beef breeds which were bred for commercial sale, but also included some dairy breeds and stud animals. The breeds commonly found were Simmentaler, Friesian, Afrikander, Nguni, Jersey, Bonsmara and Brahman. On some of the farms the older cattle had been vaccinated against *Babesia bovis* and *Babesia bigemina* and these animals were not included in the serological testing. Some farmers had dip tanks and spray races on the premises, but hand spraying, hand-dressing and pour-ons were also used. Grazing on the farms was abundant and the cattle were in good condition.

3. 1. 2. 1. Detailed description of the five commercial farms.

- **Zwartrandjes Farm.** Geographic co-ordinates: 29 ° 52’ E; 23 ° 14’ S. This farm was owned by Mr. A. MacDonald and carried 160 Bonsmara cattle, 60 of which were younger than 12 months. The average annual rainfall on the farm was 400-500 mm but during 2000 the farm received 3 times the normal rainfall and serious flood damage occurred (MacDonald, personal communication, 2000). The cattle were checked daily for any signs of disease and clinical cases of redwater were treated with Berenil (Diminazene, Intervet). *Babesia bovis* had never been diagnosed on the farm. The tick burden on the farm was low and consisted mainly of *Rhipicephalus appendiculatus*, *Amblyomma hebraeum* and *Boophilus* species. The cattle were rotated between camps every two weeks. Certain camps, which were heavily shaded, appeared to carry more ticks than open camps (MacDonald, personal communication, 1999). The cattle were spray-dipped with Ektoban (Cymiazol + Cypermethrin, Bayer Animal Health) once a week in summer and every third week or more in winter.

  The herd was semi-closed and a certain number of new bulls were brought in every year. The 18-month-old weaners were usually only vaccinated against *Babesia bigemina*. Due to the unusually heavy rainfall in 2000, the farmer vaccinated all the calves under 6-7 months against both *Babesia bigemina* and *Babesia bovis* as a precautionary measure.

Consequently, serological testing on this farm was discontinued but tick collection was continued to determine whether *Boophilus microplus* was present.

- **Modderfontein Farm.** Geographic co-ordinates: 29° 53' E; 23° 29' S. This farm was owned by Mr. F. Oldreive and carried 570 cattle, most of which were Friesians, Bonsmara or Simbra, which is a mixture of Simmentaler and Brahman. The annual rainfall on the farm was 800-900 mm. Clinical cases of both African and Asiatic redwater had occurred on the farm and the incidence was highest in the age group 18-24 months. Fourteen clinical cases of Asiatic redwater were recorded in 1999 (Oldreive, personal communication, 2000). The tick burden was heavy and *Boophilus* were the most common tick species on the cattle.

  Five camps were grazed only by the heifers, five by the beef animals and seven by the dairy cattle. The heifer camps carried the heaviest tick burdens and the heifers were first allowed to graze in these camps at the age of 9-10 months. The cattle were hand-sprayed with Tikgard (Chlorfenvinphos + alphamethrin, Pfizer Animal Health) once a week in summer and once every three weeks in winter. Bayticol (Flumethrin, Bayer Animal Health) and Paracide (Alphamethrin, Pfizer Animal Health) were found to be ineffective, probably due to resistance (Oldreive, personal communication, 1999). The herds were semi-closed with only the bulls being brought in annually. The herds had never been vaccinated against bovine babesiosis.

- **Nooitgedacht Farm.** Geographic co-ordinates: 30 ° 04' E; 23 ° 08' S. The farm was owned by Mr. P. Ahrens and carried 250 Simmentaler stud cattle, of which 70 were younger than 12 months. Cattle were vaccinated against both *Babesia bovis* and *Babesia bigemina* when they were 2 years old. The management on the farm was good and the cattle were checked for disease and ticks on a regular basis. The cattle were kept nearly free of ticks. Although bovine babesiosis almost never occurred on the farm, 20 deaths due to *Babesia bovis* had recently been confirmed (Ahrens, personal communication, 1999).

  The cattle were plunge-dipped and after the outbreak of bovine babesiosis the owner chose to switch to Ektoban. The farm was later omitted from the survey due to low levels of ticks and low prevalence of antibodies to TBD. Tick control was strict and the cattle were dipped as soon as ticks appeared. The low prevalence of antibodies in the animals under 2 years was probably due to the vaccination of older cattle.

- **Mara Research Station.** Geographic co-ordinates: 29° 25' E; 23° 6' S. The herd consisted of 800 cattle which were mainly Bonsmara, Simmentaler, Afrikander, Nguni and Jersey crosses and 250 of them were younger than 12 months. The average annual rainfall was 450 mm but during the very wet year in 2000 double this amount was received. *Babesia bigemina* was present at Mara and had previously caused mortalities. The tick burden was low and consisted mainly of *Amblyomma* species, although during 2000 a heavier than normal *Boophilus* burden was recorded.

Two hundred camps were divided equally among the 15 herds and specific camps were allocated to each herd. The cattle were plunge-dipped with Ektoban when there were more than an average of 10 adult *Amblyomma hebraeum* per animal on a sample selected from each herd (Du Plessis *et al.*, 1992).

About 100 cattle were added to the station every year as replacements. These cattle were dipped on arrival, isolated for some weeks and then dipped again before they were introduced to the resident herd. In 2000, 60 cattle were tested for antibodies to *Babesia* species, but due to the structured camp system a random sample could not be taken. The serum samples were omitted from the survey in 2000, but tick collection was continued to determine whether *Boophilus microplus* was present.

- **Naboomkop Farm.** Geographic co-ordinates: 30° 20' E; 23° 09' S. This farm was owned by Mr. S. Wilson and carried 150 adult cattle and 50 young animals. The breeds were Brahman and Brahman x Bonsmara. The annual rainfall on the farm was 700 mm but in 2000 the farm received

1500 mm. Newly introduced cattle had recently succumbed to bovine babesiosis, and this was suspected to be an important cause of mortality among the young calves. Sick animals were not treated. The tick burden was heavy at times with abundant *Boophilus* species present but the burden was lighter than normal during the first half of 2000.

The farm was divided into several camps, and the camps that were most commonly used had the heaviest tick burdens (Wilson, personal communication, 1999). The cattle were hand-sprayed once a week with Pro-dip (Cypermethrin, Logos Agvet), Paracide and Bayticol. Triatix (Amitraz, Intervet) was not used as the farmer felt it was ineffective (Wilson, personal communication, 1999). The herd was open, and adult cows were added at irregular intervals. One or two bulls were brought in every year and 30 of the cows in the herd had been brought in as adults.

3. 1. 2. 2. Vegetation types and climatic conditions on the commercial farms. The commercial farms were located in three different vegetation type zones (Acocks, 1975; Low and Rebelo, 1996): Sour Lowveld Bushveld**,** Mixed Bushveld and Sweet Bushveld.

- **Sour Lowveld Bushveld.** The commercial farms located in this vegetation type were Nooitgedacht and Naboomkop.

- **Mixed Bushveld.** This vegetation type occurred in large parts of the Northern Province and varied from short, dense bushveld to open tree savanna. The area consisted of plains at an altitude of 700 to 1100 m. The rainfall varied from 350 to 650 mm per year, and temperatures varied from –8 ° C to 40 ° C with an average of 21 ° C. The soil was coarse, sandy and shallow, with underlying granite, quartzite, sandstone and shale. The open tree savanna consisted of silver clusterleaf, peeling plane (*Ochna pulchra*), wild raisin (*Grewia flava*) and red seringa (*Burkea africana*). On shallow soil red bushwillow, common hook-thorn (*Acacia caffra*), sicklebush and live-long (*Lannea discolor*) dominated the area. The most common grasses were fingergrass (*Digitaria eriantha*), Kalahari sand quick, broom grass (*Eragrostis pallens*) and purple spike cat's tail (*Perotis patens*).

  The commercial farms located in this vegetation type were Zwartrandjes and Modderfontein.

- **Sweet Bushveld.** This vegetation type was found in the dry and hot Limpopo River Valley and in the valleys of tributary rivers at altitudes between 800 and 950 m. The rainfall varied from 350 to 500 mm per year, and temperatures varied from –5 ° C to 40 ° C with an average of 21 ° C. The soil is deep and sandy, covering granite, quartzite and sandstone. The tree species most commonly found were silver clusterleaf, yellow pomegranate (*Rhigozum obovatum*), wild raisin, common corkwood

(*Commiphora pyracanthoides*) and shepherd's tree (*Boscia albitrunca*). Dense thickets of blue thorn (*Acacia erubescens*), black thorn (*Acacia mellifera*) and sicklebush are prominent features of this veld type. The grasses were dominated by sweetveld species such as Kalahari sand quick, broom grass and various *Aristida* species. Guinea grass, small panicum (*Panicum coloratum*) and blue buffalograss (*Cenchrus ciliaris*) were also common.

The commercial farm located in this vegetation type was Mara Research Station.

3. 2. Experimental design.

3. 2. 1. Sample selection. A 2-stage non-probability cluster sampling method was used to sample the cattle (Thrusfield, 1995). The farms/dip tanks in the Northern Province were the primary units and the individual animals at each dip tank/farm were the secondary sampling units. The primary units were selected by the State Veterinarian Dr. Pieter Loock and Veterinary Technologist Mr. T. Tshisamphiri. The sampling method was non-probability (convenience) sampling (Thrusfield, 1995), and farms/dip tanks were selected according to the following criteria:

- History of occurrence of *Babesia bigemina*/*Babesia bovis*.
- Number of cattle on the dip tank/farm.
- Geographical location.

- Usability of the crush.
- Farmers' willingness to participate in the study.

3. 2. 2. Sample population in the study area.

- **Dip tanks.** The total cattle population in Dzanani, Mutale, Thohoyandou and Vuvani districts was 103,252 heads, distributed among 142 dip tanks (1999 South African census). The number of cattle normally dipped at each tank varied from 200 to 1500. Based on an estimated herd prevalence of 60 %, a sample of 30 dip tanks would be sufficient to give 95 % confidence of being within 10 % of the true prevalence of *Babesia bovis* and *Babesia bigemina*. Cattle from 11 of the dip tank areas (for convenience these are referred to merely as "dip tanks") were selected in 1999 for inclusion in the study. Tick collection at two of these dip tanks was continued in 2000 to monitor any changes in the *Boophilus* population. None of the dip tanks were monitored for changes in serology in 2000. Nineteen new dip tanks were added to the survey in 2000. The 30 dip tanks that were sampled during 1999 and 2000 serviced 22,000 cattle.

- **Commercial farms.** There were 595 commercial farm units in the Soutpansberg district with a total cattle population of 128,200. The number of cattle on the commercial farms in the survey varied from

160 to 800 per farm. Five commercial farms were selected in 1999 for inclusion in the study and two of these farms were further monitored for changes in serology during 2000. Tick collection at four of the commercial farms was continued in 2000 to monitor any changes in the *Boophilus* population.

3. 2. 3. Blood collection. The prevalence of TBD in the survey areas was unknown, so 50 % prevalence was estimated with a desired confidence level of 95 %. (Thrusfield, 1995). The number of animals required for the serology test was calculated (Martin *et al*., 1987). The unit for analysis was the individual animal. Seropositive results were expressed as seroprevalence and defined as P = a/b where a was the number of positive animals and b the number of animals tested (Alvarez *et al*., 1996).

With the exception of one dip tank where only 41 cattle were bled, 60 cattle were bled at each dip tank and commercial farm. The animals were randomly selected according to the number of cattle at the dip tank/farm. The sample of animals was split into 4 to 14-month-old animals, and those older than 18 months. Where possible, a minimum of 30 animals in each age group was sampled.

Cattle were held in a crush prior to dipping and blood samples were taken. Blood samples were collected from the tail vein (*v. caudalis mediana*) into a 10 ml. Monoject* Vacutainer tube without anti-coagulant. The blood samples were carefully labelled, making sure that the age group was clearly indicated. Blood could not be collected from the same animals at subsequent collections as most animals on the communal lands were not marked in any way.

The blood samples were stored at room temperature for 4 hours to allow clotting, and were then centrifuged at 3000 rpm for 20 min. The sera were decanted into 4 ml cryotubes (Cryovial*) and stored at –10 C° at the Veterinary Laboratories at Sibasa/Louis Trichardt. The cryotubes were clearly marked with the year, date, dip tank/farm and age of the animal. They were later transferred on ice to the Onderstepoort Veterinary Institute (OVI), where the serum samples were analyzed for antibodies against *Babesia bigemina* and *Babesia bovis* using the Indirect Fluorescent Antibody test (IFAT) (Anon., 1984).

3. 2. 4. Tick collection. The sampling method chosen for tick collection was convenience sampling (Trushfield, 1995) to avoid injury to animals and the collectors. At each dip tank/commercial farm six young animals, aged 4-14 months, which carried a heavy *Boophilus* tick burden were sampled before dipping and care was taken to choose cattle from different owners.

Boophilus ticks were collected from young cattle in May, September, November and December 1999, in May, October and December 2000 and in February 2001. The ticks were collected early in the morning so that as many replete ticks as possible could be counted (Johnston, 1967). The calves were restrained on the ground with ropes and the *Boophilus* ticks were removed by hand. Templates on certain body areas were used when collecting the ticks and all adult ticks inside the templates were collected (Baker and Ducasse, 1967). Small templates (5 x 5 cm) were used on the neck, poll and dewlap and larger templates (10 x 10 cm) on the elbow region, knee and perineum. The ticks were preserved in 70 % ethanol and the containers were marked to indicate the name of the dip tank, a number (1 to 6) allocated to each animal and the sampling site on the animal.

In certain areas the tick burdens were very low and ticks from several animals were pooled. As many ticks as possible from these locations were collected into 70 % ethanol. The ticks were collected when the animals were held in crushes prior to dipping and stored in containers marked to indicate the sampling date of the dip tank. No templates were used for these collections.

In the ectoparasite laboratory in the Department of Veterinary Tropical Diseases at the Faculty of Veterinary Science at Onderstepoort, the *Boophilus* ticks were identified by the author as either *Boophilus decoloratus* or *Boophilus microplus* using a stereoscopic microscope. The ticks were distinguished as engorged females (e.f.), unengorged females (u.e.f), males (m.) and immatures (imm.) (Gothe, 1967a; Heyne, 1986).

3. 3. Serological procedures.

3. 3. 1. Detection of antibodies. The most widely used procedure for detection of antibodies to *Babesia* species is IFAT (Joyner *et al*., 1972; Anon., 1984; OIE, 1996), which is highly sensitive and specific (Anon., 1984; Todorovic and Long, 1976). In the routine testing done at the OVI titres over 1/80 are considered positive (Bessenger and Schoeman, 1983).

3. 4. CLIMEX mapping.

3. 4. 1. The CLIMEX maps. The CLIMEX maps and other support were provided by Dr. R. W. Sutherst, CSIRO Entomology, Long Pocket Laboratories, 120 Meiers Rd, Indooroopilly, Queensland, Australia 4068. The maps were created using ArcMap 8.1 Esri Inc. software.

3. 4. 2. Climatic information. Prof. Roland Schulze, Dep. of Agricultural Engineering, University of Natal, Pietermaritzburg, provided the climatic information. This information included daily maximum temperatures, daily minimum temperatures, rainfall and relative humidity collected at several sites in the Northern Province over the past 30 years. The data were processed using CLIMEX, and Ecoclimatic Indices for each dip tank/farm were then computed.

3. 5. Statistical analysis.

3. 5. 1. Computing of probabilities. Ms. Rina Owen and Mr. Solly Millard, University of Pretoria, used SAS software to compute the probabilities in the survey. The Chi-square test was used to decide if the differences between sampling years, age groups and farming models were statistically significant.

To compare the seroprevalence of *Babesia bovis* with that of *Babesia bigemina* the Wilcoxon Rank sum test for independent samples and BMDP software was used (Keller and Warrack, 2000).

CHAPTER 4. RESULTS

4. 1. Serological findings.

A total of 2201 blood samples were collected. With the exception of one dip tank where only 41 cattle were bled, 60 cattle were bled at each dip tank or farm. The sample was split into 4 to14 month-old animals, and animals older than 18 months. The results of the serological findings are summarized in Tables 4.1- 4.14.

4. 1. 1. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* collected from cattle at the communal dip tanks during 1999 and 2000.

Seroprevalence for *Babesia bovis* and *Babesia bigemina* are given in Tables 4.1-4. 6.

Table 4. 1. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in both age groups of cattle bled at dip tanks during 1999.

			Babesia bovis		*Babesia bigemina*	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Thononda	03.05.1999	60	38	**63.3%**	19	**31.7%**
Dzondo	04.05.1999	60	43	**71.7%**	35	**58.3%**
Guyuni	05.05.1999	60	40	**66.7%**	37	**61.7%**
Sendedza	06.05.1999	60	26	**43.3%**	38	**63.3%**
Luvhanga	07.05.1999	60	44	**73.3%**	39	**65.0%**
Muledzhi	10.05.1999	60	50	**83.3%**	43	**71.7%**
Malavuwe	11.05.1999	60	30	**50.0%**	37	**61.7%**
Makwarani	12.05.1999	60	42	**70.0%**	37	**61.7%**
Lamvi	13.05.1999	60	51	**85.0%**	30	**50.0%**
Tshaulu	14.05.1999	60	33	**55.0%**	42	**70.0%**
Davhana	21.05.1999	60	21	**35.0%**	13	**21.7%**
Total		**660**	**418**		**370**	
Mean				**63.3%**		**56.1%**

Table 4. 2. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle older than 18 months bled at dip tanks during 1999.

			Babesia bovis		*Babesia bigemina*	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Thononda	03.05.1999	30	20	**66.7%**	8	**26.7%**
Dzondo	04.05.1999	30	20	**66.7%**	23	**76.7%**
Guyuni	05.05.1999	30	22	**73.3%**	15	**50.0%**
Sendedza	06.05.1999	30	12	**40.0%**	16	**53.3%**
Luvhanga	07.05.1999	30	23	**76.7%**	20	**66.7%**
Muledzhi	10.05.1999	30	28	**93.3%**	21	**70.0%**
Malavuwe	11.05.1999	30	14	**46.7%**	12	**40.0%**
Makwarani	12.05.1999	30	27	**90.0%**	23	**76.7%**
Lamvi	13.05.1999	30	22	**73.3%**	14	**46.7%**
Tshaulu	14.05.1999	30	21	**70.0%**	19	**63.3%**
Davhana	21.05.1999	30	7	**23.3%**	1	**3.3%**
Total		**330**	**216**		**172**	
Mean				**65.5%**		**52.1%**

Table 4. 3. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle aged 4-14 months bled at dip tanks during 1999

			Babesia bovis		*Babesia bigemina*	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Thononda	03.05.1999	30	18	**60.0%**	11	**36.7%**
Dzondo	04.05.1999	30	23	**76.7%**	12	**40.0%**
Guyuni	05.05.1999	30	18	**60.0%**	22	**73.3%**
Sendedza	06.05.1999	30	14	**46.7%**	22	**73.3%**
Luvhanga	07.05.1999	30	21	**70.0%**	19	**63.3%**
Muledzhi	10.05.1999	30	22	**73.3%**	22	**73.3%**
Malavuwe	11.05.1999	30	16	**53.3%**	25	**83.3%**
Makwarani	12.05.1999	30	15	**50.0%**	14	**46.7%**
Lamvi	13.05.1999	30	29	**96.7%**	16	**53.3%**
Tshaulu	14.05.1999	30	12	**40.0%**	23	**76.7%**
Davhana	21.05.1999	30	14	**46.7%**	12	**40.0%**
Total		**330**	**202**		**198**	
Mean				**61.2%**		**60.0%**

Table 4. 4. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in both age groups of cattle bled at dip tanks during 2000.

			Babesia bovis		*Babesia bigemina*	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Mahagala	02.05.2000	60	53	**88.3%**	36	**60.0%**
Matshena	03.05.2000	60	8	**13.3%**	2	**3.3%**
Phiphidi	04.05.2000	41	29	**70.7%**	22	**53.7%**
Shakadza	05.05.2000	60	50	**83.3%**	42	**70.0%**
Fesekraal 1	08.05.2000	60	6	**10.0%**	8	**13.3%**
Matatani	12.05.2000	60	36	**60.0%**	15	**25.0%**
Tshiendeulu	17.05.2000	60	46	**76.7%**	30	**50.0%**
Khakhu	24.05.2000	60	57	**95.0%**	55	**91.7%**
Murangoni	25.05.2000	60	52	**86.7%**	46	**76.7%**
Gondeni	26.05.2000	60	47	**78.3%**	33	**55.0%**
Savhani	31.05.2000	60	48	**80.0%**	27	**45.0%**
Tshikotoni	01.06.2000	60	35	**58.3%**	12	**20.0%**
Mphephu	09.10.2000	60	32	**53.3%**	39	**65.0%**
Keerweerder	10.10.2000	60	0	**0.0%**	20	**33.3%**
Masetoni	23.10.2000	60	47	**78.3%**	38	**63.3%**
Fripp	25.10.2000	60	25	**41.7%**	39	**65.0%**
Maunguwi	13.12.2000	60	47	**78.3%**	37	**61.7%**
Sambandou	14.12.2000	60	49	**81.7%**	31	**51.7%**
Makonde Project	15.12.2000	60	33	**55.0%**	21	**35.0%**
Total		**1121**	**700**		**553**	
Mean				**62.4%**		**49.3%**

Table 4. 5. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle older than 18 months bled at dip tanks during 2000.

			Babesia bovis		*Babesia bigemina*	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Mahagala	02.05.2000	30	25	**83.3 %**	21	**70.0 %**
Matshena	03.05.2000	30	6	**20.0 %**	2	**6.7 %**
Phiphidi	04.05.2000	30	23	**76.7 %**	19	**63.3 %**
Shakadza	05.05.2000	30	27	**90.0 %**	24	**80.0 %**
Fesekraal 1	08.05.2000	30	5	**16.7 %**	7	**23.3 %**
Matatani	12.05.2000	30	27	**90.0 %**	11	**36.7 %**
Tshiendeulu	17.05.2000	30	25	**83.3 %**	14	**46.7 %**
Khakhu	24.05.2000	30	28	**93.3 %**	27	**90.0 %**
Murangoni	25.05.2000	30	27	**90.0 %**	23	**76.7 %**
Gondeni	26.05.2000	30	25	**83.3 %**	20	**66.7 %**
Savhani	31.05.2000	30	25	**83.3 %**	10	**33.3 %**
Tshikotoni	01.06.2000	30	22	**73.3 %**	9	**30.0 %**
Mphephu	09.10.2000	30	22	**73.3 %**	15	**50.0 %**
Keerweerder	10.10.2000	30	0	**0.0 %**	10	**33.3 %**
Masetoni	23.10.2000	30	25	**83.3 %**	18	**60.0 %**
Fripp	25.10.2000	30	16	**53.3 %**	20	**66.7 %**
Maunguwi	13.12.2000	30	24	**80.0 %**	20	**66.7 %**
Sambandou	14.12.2000	30	19	**63.3 %**	14	**46.7 %**
Makonde Project	15.12.2000	30	15	**50.0 %**	10	**33.3 %**
Total		**570**	**386**		**294**	
Mean				**67.7%**		**51.6%**

Table 4. 6. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle aged 4-14 months bled at dip tanks during 2000.

			Babesia bovis		*Babesia bigemina*	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Mahagala	02.05.2000	30	28	**93.3%**	15	**50.0%**
Matshena	03.05.2000	30	2	**6.7%**	0	**0.0%**
Phiphidi	04.05.2000	11	6	**54.5%**	3	**27.3%**
Shakadza	05.05.2000	30	23	**76.7%**	18	**60.0%**
Fesekraal 1	08.05.2000	30	1	**3.3%**	1	**3.3%**
Matatani	12.05.2000	30	9	**30.0%**	4	**13.3%**
Tshiendeulu	17.05.2000	30	21	**70.0%**	16	**53.3%**
Khakhu	24.05.2000	30	29	**96.7%**	28	**93.3%**
Murangoni	25.05.2000	30	25	**83.3%**	23	**76.7%**
Gondeni	26.05.2000	30	22	**73.3%**	13	**43.3%**
Savhani	31.05.2000	30	23	**76.7%**	17	**56.7%**
Tshikotoni	01.06.2000	30	13	**43.3%**	3	**10.0%**
Mphephu	09.10.2000	30	10	**33.3%**	24	**80.0%**
Keerweerder	10.10.2000	30	0	**0.0%**	10	**33.3%**
Masetoni	23.10.2000	30	22	**73.3%**	20	**66.7%**
Fripp	25.10.2000	30	9	**30.0%**	19	**63.3%**
Maunguwi	13.12.2000	30	23	**76.7 %**	17	**56.7 %**
Sambandou	14.12.2000	30	30	**100.0 %**	17	**56.7 %**
Makonde Project	15.12.2000	30	18	**60.0 %**	11	**36.7 %**
Total		**551**	**314**		**259**	
Mean				**57.0 %**	**Mean**	**47.0 %**

Seropositive reactors to *B. bovis* were found at 29 out of 30 (**97** %) dip tanks included in the study.

Seropositive reactors to *B. bigemina* were found at all (**100** %) of the dip tanks included in the study.

Year and age groups were compared with regard to the seroprevalences of *Babesia bovis* and *Babesia bigemina* at dip tanks during 1999 and 2000. The summaries are given in Tables 4. 7 and 4. 8.

Table 4. 7. Chi–square test of the differences in the seroprevalence of *Babesia bovis* and *Babesia bigemina* from cattle bled at the communal dip tanks during 1999 and 2000, compared by age. (p<0.05 is significant).

B. bovis	Young 1999 compared to Old 1999 p=0.2581 Not significant	Young 2000 compared to Old 2000 **p=0.0002** **Significant**
B. bigemina	Young 1999 compared to Old 1999 **p=0.0414** **Significant**	Young 2000 compared to Old 2000 p=0.1257 Not significant

Table 4. 8. Chi–square test of the differences in the seroprevalence of *Babesia bovis* and *Babesia bigemina* from cattle bled at the communal dip tanks during 1999 and 2000, compared by year. (p<0.05 is significant).

B. bovis	Young 1999 compared to Young 2000 p=0.2179 Not significant	Old 1999 compared to Old 2000 p=0.4866 Not significant	All 1999 compared to All 2000 p= 0.7078 Not significant
B. bigemina	Young 1999 compared to Young 2000 **p=0.0002** **Significant**	Old 1999 compared to Old 2000 p=0.8753 Not significant	All 1999 compared to All 2000 **p=0.0060** **Significant**

The highlighted cells show a downward trend.

4. 1. 1. 1. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* collected from cattle bled at dip tanks situated in the Sour Lowveld Bushveld veld type during 1999.

Seroprevalence for *Babesia bovis* and *Babesia bigemina* are given in Tables 4. 9-4. 14

Table 4. 9. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in both age groups of cattle bled at dip tanks situated in the Sour Lowveld Bushveld veld type during 1999.

			Babesia bovis		***Babesia bigemina***	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Thononda	03.05.1999	60	38	**63.3%**	19	**31.7%**
Dzondo	04.05.1999	60	43	**71.7%**	35	**58.3%**
Guyuni	05.05.1999	60	40	**66.7%**	37	**61.7%**
Sendedza	06.05.1999	60	26	**43.3%**	38	**63.3%**
Luvhanga	07.05.1999	60	44	**73.3%**	39	**65.0%**
Muledzhi	10.05.1999	60	50	**83.3%**	43	**71.7%**
Malavuwe	11.05.1999	60	30	**50.0%**	37	**61.7%**
Makwarani	12.05.1999	60	42	**70.0%**	37	**61.7%**
Lamvi	13.05.1999	60	51	**85.0%**	30	**50.0%**
Tshaulu	14.05.1999	60	33	**55.0%**	42	**70.0%**
Total		**600**	**397**		**357**	
Mean				**66.2%**		**59.5%**

Table 4. 10. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle older than 18 months bled at dip tanks situated in the Sour Lowveld Bushveld veld type during 1999.

			Babesia bovis		***Babesia bigemina***	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Thononda	03.05.1999	30	20	**66.7%**	8	**26.7%**
Dzondo	04.05.1999	30	20	**66.7%**	23	**76.7%**
Guyuni	05.05.1999	30	22	**73.3%**	15	**50.0%**
Sendedza	06.05.1999	30	12	**40.0%**	16	**53.3%**
Luvhanga	07.05.1999	30	23	**76.7%**	20	**66.7%**
Muledzhi	10.05.1999	30	28	**93.3%**	21	**70.0%**
Malavuwe	11.05.1999	30	14	**46.7%**	12	**40.0%**
Makwarani	12.05.1999	30	27	**90.0%**	23	**76.7%**
Lamvi	13.05.1999	30	22	**73.3%**	14	**46.7%**
Tshaulu	14.05.1999	30	21	**70.0%**	19	**63.3%**
Total		**300**	**209**		**171**	
Mean				**69.7%**		**57.0%**

Table 4. 11. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle aged 4-14 months bled at dip tanks situated in the Sour Lowveld Bushveld veld type during 1999.

Babesia bovis			***Babesia bovis***		***Babesia bigemina***	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Thononda	03.05.1999	30	18	**60.0%**	11	**36.7%**
Dzondo	04.05.1999	30	23	**76.7%**	12	**40.0%**
Guyuni	05.05.1999	30	18	**60.0%**	22	**73.3%**
Sendedza	06.05.1999	30	14	**46.7%**	22	**73.3%**
Luvhanga	07.05.1999	30	21	**70.0%**	19	**63.3%**
Muledzhi	10.05.1999	30	22	**73.3%**	22	**73.3%**
Malavuwe	11.05.1999	30	16	**53.3%**	25	**83.3%**
Makwarani	12.05.1999	30	15	**50.0%**	14	**46.7%**
Lamvi	13.05.1999	30	29	**96.7%**	16	**53.3%**
Tshaulu	14.05.1999	30	12	**40.0%**	23	**76.7%**
Total		**300**	**188**		**186**	
Mean				**62.7%**		**62.0%**

Table 4. 12. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in both age groups of cattle bled at dip tanks situated in the Sour Lowveld Bushveld veld type during 2000.

			Babesia bovis		*Babesia bigemina*	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Mahagala	02.05.2000	60	53	**88.3%**	36	**60.0%**
Phiphidi	04.05.2000	41	29	**70.7%**	22	**53.7%**
Tshiendeulu	17.05.2000	60	46	**76.7%**	30	**50.0%**
Khakhu	24.05.2000	60	57	**95.0%**	55	**91.7%**
Murangoni	25.05.2000	60	52	**86.7%**	46	**76.7%**
Gondeni	26.05.2000	60	47	**78.3%**	33	**55.0%**
Tshikotoni	01.06.2000	60	35	**58.3%**	12	**20.0%**
Masetoni	23.10.2000	60	47	**78.3%**	38	**63.3%**
Sambandou	14.12.2000	60	49	**81.7%**	31	**51.7%**
Makonde Project	15.12.2000	60	33	**55.0%**	21	**35.0%**
Total		**581**	**448**		**324**	
Mean				**77.1%**		**55.7%**

Table 4. 13. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle older than 18 months bled at dip tanks situated in the Sour Lowveld Bushveld veld type during 2000.

			Babesia bovis		*Babesia bigemina*	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Mahagala	02.05.2000	30	25	**83.3 %**	21	**70.0 %**
Phiphidi	04.05.2000	30	23	**76.7 %**	19	**63.3 %**
Tshiendeulu	17.05.2000	30	25	**83.3 %**	14	**46.7 %**
Khakhu	24.05.2000	30	28	**93.3 %**	27	**90.0 %**
Murangoni	25.05.2000	30	27	**90.0 %**	23	**76.7 %**
Gondeni	26.05.2000	30	25	**83.3 %**	20	**66.7 %**
Tshikotoni	01.06.2000	30	22	**73.3 %**	9	**30.0 %**
Masetoni	23.10.2000	30	25	**83.3 %**	18	**60.0 %**
Sambandou	14.12.2000	30	19	**63.3 %**	14	**46.7 %**
Makonde Project	15.12.2000	30	15	**50.0 %**	10	**33.3 %**
Total		**300**	**234**		**175**	
	Mean			**78 %**		**58.3 %**

Table 4. 14. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle aged 4-14 months bled at dip tanks situated in the Sour Lowveld Bushveld veld type during 2000.

Babesia bovis			***Babesia bovis***		***Babesia bigemina***	
Dip tank	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Mahagala	02.05.2000	30	28	**93.3%**	15	**50.0%**
Phiphidi	04.05.2000	11	6	**54.5%**	3	**27.3%**
Tshiendeulu	17.05.2000	30	21	**70.0%**	16	**53.3%**
Khakhu	24.05.2000	30	29	**96.7%**	28	**93.3%**
Murangoni	25.05.2000	30	25	**83.3%**	23	**76.7%**
Gondeni	26.05.2000	30	22	**73.3%**	13	**43.3%**
Tshikotoni	01.06.2000	30	13	**43.3%**	3	**10.0%**
Masetoni	23.10.2000	30	22	**73.3%**	20	**66.7%**
Sambandou	14.12.2000	30	30	**100.0 %**	17	**56.7 %**
Makonde Project	15.12.2000	30	18	**60.0 %**	11	**36.7 %**
Total		**281**	**214**		**149**	
Mean				**76.2 %**		**53.0 %**

Seropositive reactors to both *B. bovis* and *B. bigemina* were found at all (**100** %) of the dip tanks situated in Sour Lowveld Bushveld during 1999 and 2000.

Year and age groups were compared with regard to the seroprevalences of *Babesia bovis* and *Babesia bigemina* at dip tanks situated in the Sour Lowveld Bushveld during 1999 and 2000. The summaries are given in Tables 4. 15 and 4. 16.

Table 4. 15. Chi–square test of the differences in the seroprevalence of *Babesia bovis* and *Babesia bigemina* from cattle bled at the communal dip tanks situated in the Sour Lowveld Bushveld veld type during 1999 and 2000, compared by age. (p<0.05 is significant).

B. bovis	Young 1999 compared to Old 1999 p=0.0700 Not significant	Young 2000 compared to Old 2000 p=0.5971 Not significant
B. bigemina	Young 1999 compared to Old 1999 p=0.2122 Not significant	Young 2000 compared to Old 2000 p=0.1979 Not significant

Table 4. 16. Chi–square test of the differences in the seroprevalence of *Babesia bovis* and *Babesia bigemina* from cattle bled at the communal dip tanks situated in the Sour Lowveld Bushveld veld type during 1999 and 2000, compared by year. (p<0.05 is significant).

B. bovis	Young 1999 compared to Young 2000 **p=0.0004** **Significant**	Old 1999 compared to Old 2000 **p=0.0202** **Significant**	All 1999 compared to All 2000 **p=0.0001** **Significant**
B. bigemina	Young 1999 compared to young 2000 **p=0.0287** **Significant**	Old 1999 compared to Old 2000 p=0.7410 Not significant	All 1999 compared to All 2000 p=0.1941 Not significant

The highlighted cells show a downward trend.

4. 1. 2. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* collected from cattle on the commercial farms during 1999 and 2000.

Seroprevalence for *Babesia bovis* and *Babesia bigemina* are given in Tables 4. 17-4. 22.

Table 4. 17. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in both age groups of cattle bled on the commercial farms during 1999.

			Babesia bovis		*Babesia bigemina*	
Farm	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Zwartrandjes	13.09.1999	60	9	**15.0%**	39	**65.0%**
Modderfontein	14.09.1999	60	36	**60.0%**	39	**65.0%**
Nooitgedacht	15.09.1999	60	3	**5.0%**	3	**5.0%**
Mara Res. St.	16.09.1999	60	3	**5.0%**	32	**53.3%**
Naboomkop	17.09.1999	60	6	**10.0%**	32	**53.3%**
Total		**300**	**57**		**145**	
Mean				**19.0%**		**48.3%**

Table 4. 18. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle older than 18 months bled on commercial farms during 1999.

			Babesia bovis		*Babesia bigemina*	
Farm	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Zwartrandjes	13.09.1999	30	2	**6.7%**	18	**60.0%**
Modderfontein	14.09.1999	30	22	**73.3%**	20	**66.7%**
Nooitgedacht	15.09.1999	30	2	**6.7%**	2	**6.7%**
Mara Res. St.	16.09.1999	30	2	**6.7%**	18	**60.0%**
Naboomkop	17.09.1999	42	5	**11.9%**	26	**61.9%**
Total		**162**	**33**		**84**	
Mean				**20.4%**		**51.9 %**

Table 4. 19. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle aged 4-14 months bled on commercial farms during 1999.

			Babesia bovis		***Babesia bigemina***	
Farm	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Zwartrandjes	13.09.1999	30	7	**23.3%**	21	**70.0%**
Modderfontein	14.09.1999	30	14	**46.7%**	19	**63.3%**
Nooitgedacht	15.09.1999	30	1	**3.3%**	1	**3.3%**
Mara Res. St.	16.09.1999	30	1	**3.3%**	14	**46.7%**
Naboomkop	17.09.1999	18	1	**5.6%**	6	**33.3%**
Total		**138**	**24**		**61**	
Mean				**17.4%**		**44.2%**

Seropositive reactors to both *B. bovis* and *B. bigemina* were found on all (**100** %) of the commercial farms included in the study.

Table 4. 20. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in both age groups of cattle bled on commercial farms during 2000.

			Babesia bovis		***Babesia bigemina***	
Farm	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Modderfontein	09.05.2000	60	40	**66.7%**	18	**30.0%**
Naboomkop	16.05.2000	60	29	**48.3%**	39	**65.0%**
Total		**120**	**69**		**57**	
Mean				**57.5%**		**47.5%**

Table 4. 21. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle older than 18 months bled on commercial farms during 2000.

			Babesia bovis		*Babesia bigemina*	
Farm	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Modderfontein	09.05.2000	30	20	**66.7%**	6	**20.0%**
Naboomkop	16.05.2000	30	10	**33.3%**	20	**66.7%**
Total		**60**	**30**		**26**	
Mean				**50.0%**		**43.3%**

Table 4. 22. Seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle aged 4-14 months bled on commercial farms during 2000.

			Babesia bovis		*Babesia bigemina*	
Farm	**Collection date**	**No. tested**	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Modderfontein	09.05.2000	30	20	**66.7%**	12	**40.0%**
Naboomkop	16.05.2000	30	19	**63.3%**	19	**63.3%**
Total		**60**	**39**		**31**	
Mean				**65.0%**		**51.7%**

Year and age groups were compared with regard to the seroprevalences of *Babesia bovis* and *Babesia bigemina* on commercial farms during 1999 and 2000. The summaries are given in Table 4. 23 and 4. 24.

Table 4. 23. Chi–square test of the differences in the seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* from cattle bled at commercial farms during 1999 and 2000, compared by age. (p<0.05 is significant).

B. bovis	Young 1999 compared to Old 1999 p=0.5121 Not significant	Young 2000 compared to Old 2000 p=0.0965 Not significant
B. bigemina	Young 1999 compared to Old 1999 p=0.1864 Not significant	Young 2000 compared to Old 2000 p=0.3607 Not significant

Table 4. 24. Chi–square test of the differences in the seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* from cattle bled at commercial farms during 1999 and 2000, compared by year. (p<0.05 is significant).

B. bovis	Young 1999 compared to Young 2000 **p=0.0001** **Significant**	Old 1999 compared to Old 2000 **p=0.0001** **Significant**	All 1999 compared to All 2000 **p=0.0001** **Significant**
B. bigemina	Young 1999 compared to Young 2000 p=0.3332 Not significant	Old 1999 compared to Old 2000 p=0.2596 Not significant	All 1999 compared to All 2000 p=0.8773 Not significant

The highlighted cells show a downward trend from 1999 to 2000.

4. 1. 3. A comparison of the seroprevalences of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle bled at communal dip tanks and commercial farms during 1999 and 2000.

The significance values are given in Tables 4. 25 and 4. 26

Table 4. 25. Chi-square test of the differences in the seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in cattle bled at the communal dip tanks and the commercial farms during 1999 and 2000. (p<0.05 is significant).

B. bovis	Dip tanks 1999 compared to Commercial farms 1999 **p=0.0001 Significant**	Dip tanks 2000 compared to Commercial farms 2000 p=0.2890 Not significant
B. bigemina	Dip tanks 1999 compared to Commercial farms 1999 **p=0.0261 Significant**	Dip tanks 2000 compared to Commercial farms 2000 p=0.7030 Not significant

Table 4. 26. Chi-square test of the differences in the seroprevalence of antibodies to *Babesia bovis* and *Babesia bigemina* in all cattle bled at the communal dip tanks and the commercial farms during 1999 and 2000 compared by year. (p<0.05 is significant).

B. bovis	Young 1999 compared to Young 2000 **p=0.0020 significant**	Old 1999 compared to Old 2000 **p=0.0001 significant**	All 1999 compared to All 2000 **p=0.0001 significant**
B. bigemina	Young 1999 compared to Young 2000 **p=0.0103 Significant**	Old 1999 compared to Old 2000 p=0.6804 Not significant	All 1999 compared to All 2000 **p=0.0366 Significant**

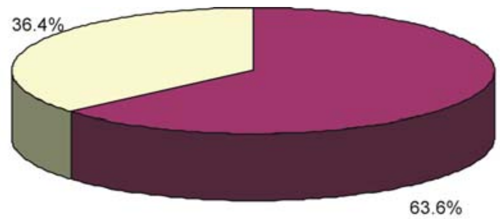
The highlighted cells show a downward trend.

Differences in *B. bovis* seroprevalence from 1999 to 2000 when all cattle at the communal dip tanks and commercial farms were compared. The seroprevalence to *B. bovis* in the all the young cattle in the survey was significantly higher (p=0.0020) in 2000 than in 1999 (Table 4. 26). The seroprevalence in all the old cattle in the survey was significantly higher (p=0.0001) in 2000 than in 1999. The seroprevalence to *B. bovis* in all cattle in the survey was significantly higher (p=0.0001) in 2000 than in1999.

Differences in *B. bigemina* seroprevalence from 1999 to 2000 when all cattle at the communal dip tanks and commercial farms were compared. The seroprevalence to *B. bigemina* in the all the young cattle in the survey was significantly lower (p=0.0103) in 2000 than in 1999 (Table 4. 26). The seroprevalence in all the older cattle in the survey was lower in 2000 than in 1999, but the difference was not significant (p=0.6804). The seroprevalence to *B. bigemina* in all cattle in the survey was significantly lower (p=0.0366) in 2000 than in 1999.

4. 2. Endemic stability to *Babesia bovis* and *Babesia bigemina.*

4. 2. 1. Endemic stability to *Babesia bovis* and *Babesia bigemina* in cattle at the communal dip tanks during 1999 and 2000. The results are shown in Fig. 4. 1-4. 4.

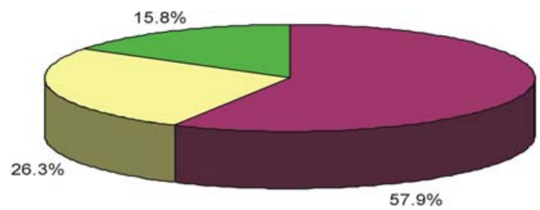


***Babesia bovis*:** 7 dip tanks with more than 61 % seropositive cattle: Endemic stability or a situation approaching endemic stability **63.6 %**

4 dip tanks with 21 – 60 % seropositive cattle: Endemically unstable situation **36.4 %**

0 dip tanks with 0 – 20 % seropositive cattle: Minimal disease or disease-free situation **0.0 %**

Fig. 4. 1. Endemic stability to *Babesia bovis* recorded at the 11 dip tanks in the survey during 1999.

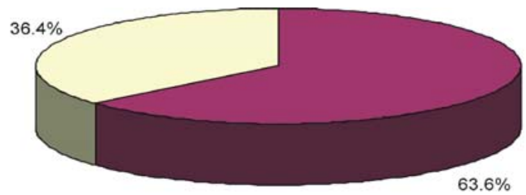


***Babesia bovis*:** 11 dip tanks with more than 61 % seropositive cattle: Endemic stability or a situation approaching endemic stability **57.9 %**

5 dip tanks with 21 – 60 % seropositive cattle: Endemically unstable situation **26.3 %**

3 dip tanks with 0 – 20 % seropositive cattle: Minimal disease or disease-free situation **15.8 %**

Fig. 4. 2. Endemic stability to *Babesia bovis* recorded at the 19 dip tanks in the survey during 2000.

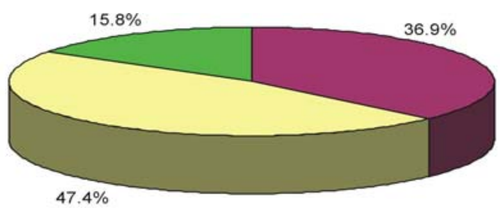


***Babesia bigemina*:** 7 dip tanks with more than 61% seropositive cattle:
Endemic stability or a situation approaching endemic stability **63. 6 %**

4 dip tanks with 21 – 60 % seropositive cattle:
Endemically unstable situation **36. 4 %**

0 dip tanks with 0 – 20 % seropositive cattle:
Minimal disease or disease-free situation **0. 0 %**

Fig. 4. 3. Endemic stability to *Babesia bigemina* recorded at the 11 dip tanks in the survey during 1999.



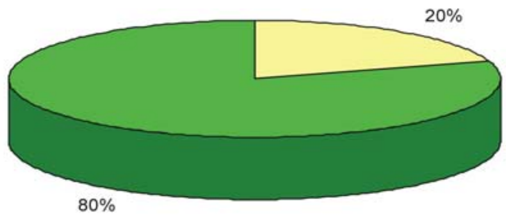
***Babesia bigemina*:** 7 dip tanks with more than 61% seropositive cattle:
Endemic stability or a situation approaching endemic stability **36.9 %**

9 dip tanks with 21 – 60 % seropositive cattle:
Endemically unstable situation **47.4 %**

3 dip tanks with 0 – 20 % seropositive cattle:
Minimal disease or disease-free situation **15.8 %**

Fig. 4. 4. Endemic stability to *Babesia bigemina* recorded at the 19 dip tanks in the survey during 2000.

4. 2. 2. Endemic stability to *Babesia bovis* and *Babesia bigemina* recorded on the commercial farms during 1999 and 2000. The results are shown in Fig. 4. 5-4. 8

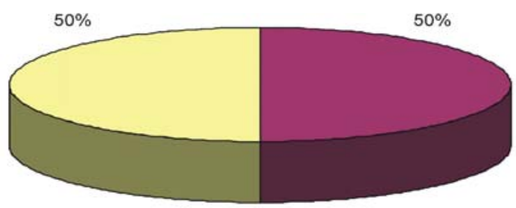


***Babesia bovis*:** 0 commercial farms with more than 61% seropositive cattle: Endemic stability or a situation approaching endemic stability **0.0 %**

1 commercial farm with 21 – 60 % seropositive cattle: Endemically unstable situation **20.0 %**

4 commercial farms with 0 – 20 % seropositive cattle: Minimal disease or disease-free situation **80.0 %**

Fig. 4. 5. Endemic stability to *Babesia bovis* recorded on the 5 commercial farms in the survey during 1999.

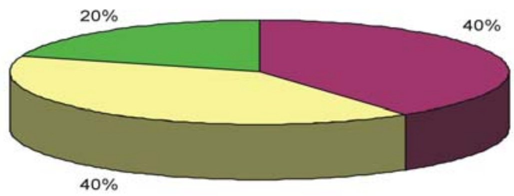


***Babesia bovis*:** 1 commercial farm with more than 61% seropositive cattle: Endemic stability or a situation approaching endemic stability **50.0 %**

1 commercial farm with 21 – 60 % seropositive cattle: Endemically unstable situation **50.0 %**

0 commercial farms with 0 – 20 % seropositive cattle: Minimal disease or disease-free situation **0.0 %**

Fig. 4. 6. Endemic stability to *Babesia bovis* recorded on the 2 commercial farms in the survey during 2000.

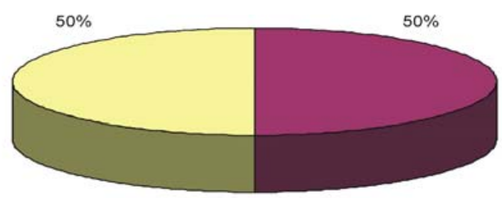


***Babesia bigemina*:** 2 commercial farms with more than 61% seropositive cattle: Endemic stability or a situation approaching endemic stability **40.0 %**

2 commercial farms with 21 – 60 % seropositive cattle: Endemically unstable situation **40.0 %**

1 commercial farm with 0 – 20 % seropositive cattle: Minimal disease or disease-free situation **20.0 %**

Fig. 4. 7. Endemic stability to *Babesia bigemina* recorded on the 5 commercial farms in the survey during 1999.



***Babesia bigemina*:** 1 commercial farm with more than 61% seropositive cattle: Endemic stability or a situation approaching endemic stability **50.0 %**

1 commercial farms with 21 – 60 % seropositive cattle: Endemically unstable situation **50.0 %**

0 commercial farm with 0 – 20 % seropositive cattle: Minimal disease or disease-free situation **0.0 %**

Fig. 4. 8. Endemic stability to *Babesia bigemina* recorded on the 2 commercial farms in the survey during 2000.

4. 3. Tick collection results from the survey area during 1999 and 2000.

The results of the tick collections are summarized in Tables 4.27-4. 30. A total of 25,042 *Boophilus* ticks were collected in the study area from 29 dip tanks and 5 commercial farms. Of these 1,530 (**6.1** %) were *Boophilus decoloratus* and 23,512 (**93.9** %) *Boophilus microplus*.

Thirteen percent of the *Boophilus microplus* ticks and **9.2** % of the *Boophilus decoloratus* ticks were males.

4. 3. 1. Tick collection results from the communal dip tanks during 1999 and 2000.

The results of the tick collections are given in Tables 4. 27 and 4.28.

Table 4. 27. *Boophilus* ticks collected from cattle at dip tanks during 1999.

Dip tank	May 1999		November 1999		December 1999		Total	
	B. dec.	***B. micro.***	***B. dec.***	***B. micro.***	***B. dec.***	***B. micro.***	***B. dec.***	***B. micro.***
Thononda	73	25			0	554	**73**	**579**
Dzondo	0	534					**0**	**534**
Guyuni	0	448					**0**	**448**
Sendedza	133	6			37	404	**170**	**410**
Luvhanga	4	517					**4**	**517**
Muledzhi	0	29	0	596			**0**	**625**
Malavuwe	0	570					**0**	**570**
Makwarani	0	524					**0**	**524**
Lamvi	0	286	0	668			**0**	**954**
Tshaulu	0	496					**0**	**496**
Davhana			0	259			**0**	**259**
Total	**210**	**3435**	**0**	**1523**	**37**	**958**	**247**	**5916**

B. dec. = *Boophilus decoloratus* *B. micro*. = *Boophilus microplus*

In 1999 *Boophilus microplus* was found together with *Boophilus decoloratus* at 3 of the 11 dip tanks, *Boophilus microplus* only was found at 8 dip tanks and at none of the dip tanks was *Boophilus decoloratus* found on its own. Of the 6,163 *Boophilus* ticks collected at the dip tanks in 1999, 247 (**4** %) were *Boophilus decoloratus* whilst 5,916 (**96** %) were *Boophilus microplus.*

Table 4. 28. *Boophilus* ticks collected from cattle at dip tanks during 2000.

Dip tank name	May/ June 2000		October 2000		December 2000		Total	
	B. dec.	*B. micro.*	*B. dec.*	*B. micro.*	*B. dec.*	*B. micro.*	*B. dec.*	*B. micro.*
Thononda	0	1691					**0**	**1691**
Mahagala	0	901					**0**	**901**
Matshena	0	0					**0**	**0**
Phiphidi	0	1224					**0**	**1224**
Shakadza	0	474					**0**	**474**
Fesekraal 1	0	14					**0**	**14**
Matatani	0	1256					**0**	**1256**
Tshiendeulu	39	941					**39**	**941**
Khakhu	0	1413					**0**	**1413**
Murangoni	0	1491					**0**	**1491**
Gondeni	0	905					**0**	**905**
Savhani	0	916					**0**	**916**
Tshikotoni	0	543					**0**	**543**
Mphephu			4	383			**4**	**383**
Keerweerder			35	1			**35**	**1**
Masetoni			0	251			**0**	**251**
Fripp			0	20			**0**	**20**
Maunguwi					0	1562	**0**	**1562**
Sambandou					0	714	**0**	**714**
Makonde Project					0	306	**0**	**306**
Total	**39**	**11,769**	**39**	**655**	**0**	**2582**	**78**	**15,006**

B. dec. = *Boophilus decoloratus* *B. micro.* = *Boophilus microplus*

In 2000 *Boophilus microplus* was found together with *Boophilus decoloratus* at 3 of the 20 dip tanks, *B. microplus* only was found at 16 of the dip tanks, and at 1 of the dip tanks no *Boophilus* ticks were found. *Boophilus decoloratus* was never found on its own at any of the dip tanks. Of the 15,084 *Boophilus* ticks collected at the dip tanks in 2000, only 78 (**0.52** %) were *Boophilus decoloratus* and 15,006 (**99.48** %) were *Boophilus microplus*.

4. 3. 2. Tick collection results from the commercial farms during 1999 and 2000.

The results of the tick collections are given in Tables 4. 29 and 4. 30.

Table 4. 29. *Boophilus* ticks collected from cattle on commercial farms during 1999.

	September 1999		**November 1999**		**December 1999**		**Total**	
	B. dec.	***B. micro.***	***B. dec.***	***B. micro.***	***B. dec.***	***B. micro.***	***B. dec***	***B. micro.***
Nooitgedacht	0	17					**0**	**17**
Zwartrandjes	63	0	45	0	265	0	**373**	**0**
Modderfontein	16	189	36	50			**52**	**239**
Mara Res. St.	1	0					**1**	**0**
Naboomkop	9	45	16	42			**25**	**87**
Total	**89**	**251**	**97**	**92**	**265**	**0**	**451**	**343**

B. dec. = *Boophilus decoloratus* *B. micro.* = *Boophilus microplus*

In 1999 *Boophilus microplus* was found together with *Boophilus decoloratus* on 2 of the 5 farms, *Boophilus microplus* only was found on 1 farm and *Boophilus decoloratus* was found on its own on 2 farms. A total of 794 *Boophilus* ticks were collected from the commercial farms during 1999 and of these were 451 (**56.8** %) *Boophilus decoloratus* and 343 (**43.2** %) were *Boophilus microplus*.

Table 4. 30. *Boophilus* ticks collected from cattle on commercial farms during 2000 and 2001.

Farm name	May/June 2000		October 2000		December 2000		February 2001		Total	
	B. dec.	*B. micro.*	*B. dec.*	*B. micro.*	*B. dec.*	*B. micro.*	*B. dec.*	*B. micro.*	*B. dec.*	*B. micro.*
Zwartrandjes	123	0	96	0					**219**	**0**
Modderfontein	158	715			5	129			**163**	**844**
Mara Res. St.	44	0					328	0	**372**	**0**
Naboomkop	0	1403							**0**	**1403**
Total	**325**	**2118**	**96**	**0**	**5**	**129**	**328**	**0**	**754**	**2247**

B. dec. = *Boophilus decoloratus* *B. micro.* = *Boophilus microplus*

In 2000 *Boophilus microplus* was found together with *Boophilus decoloratus* on 1 of the 4 farms, *Boophilus microplus* only was found on 1 farm whilst *Boophilus decoloratus* was found on its own on 2 farms. Of the 3,001 *Boophilus* ticks collected from the commercial farms in 2000, 754 were *Boophilus decoloratus* (**25.1** %) and 2,247 (**74.9** %) were *Boophilus microplus.*

4. 4. Displacement of *Boophilus decoloratus* by *Boophilus microplus* in the survey area.

4. 4. 1. Tick collection results recorded during the displacement process.

The results of the tick collections are given in Table 4. 31.

Table 4. 31. Tick collections obtained by repeatedly sampling farms and/or dip tanks where *Boophilus decoloratus* and *Boophilus microplus* co-existed during the survey.

Dip tank/farm	May 1999		Sept. 1999		Nov. 1999		Dec. 1999		May/June 2000		Dec. 2000	
	B. dec.	*B. micro.*	*B. dec.*	*B. micro.*	*B. dec.*	*B. micro.*	*B. dec.*	*B. micro.*	*B. dec.*	*B. micro.*	*B. dec.*	*B. micro.*
Thononda	73	25					0	554	0	1691		
Sendedza	133	6					37	404				
Modderfontein			16	189	36	50			158	715	5	129
Naboomkop			9	45	16	42			0	1430		

B. dec. = *Boophilus decoloratus* *B. micro.* = *Boophilus microplus*

4. 4. 2. Ecoclimatic Indices for *Boophilus microplus* and *Boophilus decoloratus* recorded at the communal dip tanks and commercial farms.

Table 4. 32. Ecoclimatic Indices for *Boophilus microplus* and *Boophilus decoloratus.*

	Ecoclimatic Index	
Dip tank/farm	***B. microplus***	***B.decoloratus***
Fesekraal	0	1
Matshena	0	1
Matatani	0	4
Shakadza	8	22
Lamvi	8	22
Tshikotoni	6	19
Savhani	9	20
Masetoni	4	17
Guyuni	7	20
Sambandou	4	16
Malavuwe	2	16
Mahagala	7	22
Maunguwi	19	27
Makonde	5	18
Tshaulu	6	22
Tshiendeulu	15	21
Khakhu	12	18
Makwarani	16	21
Mphephu	10	21
Thononda	16	21
Sendedza	19	25
Murangoni	18	22
Gondeni	23	29
Phiphidi	20	26
Muledzhi	23	29
Fripp	5	19
Keerweerder	0	3
Luvhanga	4	16
Dzondo	20	27
Mara	0	4
Nooitgedacht	9	20
Naboomkop	5	17
Davhana	2	6
Zwartrandjes	3	13
Modderfontein	4	14

4. 5. Comparison of the *Boophilus* tick numbers and the serology results obtained during 1999 and 2000.

The serology results from communal dip tanks and commercial farms were compared with the *Boophilus* tick species found at the time of the bleeding. The results are given in Tables 4. 33-4. 37.

Table 4. 33. Mean seroprevalence of *Babesia bovis* and *Babesia bigemina* from those dip tanks/farms where only *Boophilus decoloratus* was recorded in 1999.

		Babesia bovis		***Babesia bigemina***	
	No. tested	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Zwartrandjes	60	9	**15.0%**	39	**65.0%**
Mara Res.St.	60	3	**5.0%**	32	**53.3%**
Total	**120**	**12**		**71**	
		Mean	**10.0%**		**59.2%**

Table 4. 34. Mean seroprevalence of *Babesia bovis* and *Babesia bigemina* from those dip tank/farms where *Boophilus decoloratus* and *Boophilus microplus* co-existed in 1999.

		Babesia bovis		***Babesia bigemina***	
	No. tested	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Thononda	60	38	**63.3%**	19	**31.7%**
Sendedza	60	26	**43.3%**	38	**63.3%**
Luvhanga	60	44	**73.3%**	39	**65%**
Modderfontein	60	36	**60%**	39	**65%**
Naboomkop	60	6	**10%**	32	**53.3%**
Total	**300**	**150**		**167**	
		Mean	**50.0%**		**55.7%**

Table 4. 35. Mean seroprevalence of *Babesia bovis* and *Babesia bigemina* from those dip tank/farms where only *Boophilus microplus* was recorded in 1999.

		Babesia bovis		*Babesia bigemina*	
	No. tested	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Dzondo	60	43	**71.7%**	35	**58.3%**
Guyuni	60	40	**66.7%**	37	**61.7 %**
Muledzhi	60	50	**83.3%**	43	**71.7 %**
Malavuwe	60	30	**50.0%**	37	**61.7 %**
Makwarani	60	42	**70.0%**	37	**61.7 %**
Lamvi	60	51	**85.0%**	30	**50.0 %**
Tshaulu	60	33	**55.0%**	42	**70.0 %**
Nooitgedacht	60	3	**5.0%**	3	**5.0 %**
Total	**480**	**292**		**264**	
		Mean	**60.8%**		**55.0%**

Table 4. 36. Mean seroprevalence of *Babesia bovis* and *Babesia bigemina* from those dip tanks/farms where *Boophilus decoloratus* and *Boophilus microplus* co-existed in 2000.

		Babesia bovis		*Babesia bigemina*	
	No. tested	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Tshiendeulu	60	46	**76.7%**	30	**50.0%**
Mphephu	60	32	**53.3%**	39	**65.0%**
Keerweerder	60	0	**0.0%**	20	**33.3%**
Modderfontein	60	40	**66.7%**	18	**30.0%**
Total	**240**	**118**		**107**	
		Mean	**49.2%**		**44.6%**

Table 4. 37. Mean seroprevalence of *Babesia bovis* and *Babesia bigemina* from those dip tanks/farms where only *Boophilus microplus* was recorded in 2000.

		Babesia bovis		*Babesia bigemina*	
	No. tested	**No. pos.**	**% pos.**	**No. pos.**	**% pos.**
Mahagala	60	53	**88.3%**	36	**60.0%**
Phiphidi	41	29	**70.7%**	22	**53.7%**
Shakadza	60	50	**83.3%**	42	**70.0%**
Fesekraal	60	6	**10.0%**	8	**13.3%**
Matatani	60	36	**60.0%**	15	**25.0%**
Khakhu	60	57	**95.0%**	55	**91.7%**
Murangoni	60	52	**86.7%**	46	**76.7%**
Gondeni	60	47	**78.3%**	33	**55.0%**
Savhani	60	48	**80.0%**	27	**45.0%**
Tshikotoni	60	35	**58.3%**	12	**20.0%**
Masetoni	60	47	**78.3%**	38	**63.3%**
Fripp	60	25	**41.7%**	39	**65.0%**
Maunguwi	60	47	**78.3%**	37	**61.7%**
Sambandou	60	49	**81.7%**	31	**51.7%**
Makonde Proj.	60	33	**55.0%**	21	**35.0%**
Naboomkop	60	29	**48.3%**	39	**65.0%**
Total	**941**	**643**		**501**	
		Mean	**68.3%**		**53.2%**

Table 4. 38. Summary of seroprevalence of *Babesia bigemina* and *Babesia bovis* related to vector occurrence at all dip tanks/farms sampled during 1999.

	Dip tanks/farms where *B. decoloratus* and *B. microplus* were recorded	**Dip tanks/farms where only *B. microplus* was recorded**	**Dip tanks/farms where only *B. decoloratus* was recorded**
B. bovis	50.0 %	60.8 %	10.0 %
B. bigemina	55.7 %	55.0 %	59.2

Table 4. 39. Summary of seroprevalence of *Babesia bigemina* and *Babesia bovis* related to vector occurrence at all dip tanks/farms sampled during 2000.

	Dip tanks/farms where *B.decoloratus* and *B.microplus* were recorded	**Dip tanks/farms where only *B. microplus* was recorded**
B. bovis	49.2%	68.3%
B. bigemina	44.6%	53.2%

CHAPTER 5. DISCUSSION

5. 1. Serological findings.

Serological tests are useful tools to assess the degree of exposure of a herd to tick-borne diseases, and to act as sensitive markers for the presence of certain tick vectors. In the present survey 2201 blood samples from cattle at 30 communal dip tanks and 5 commercial farms were screened for antibodies to *Babesia bovis* and *Babesia bigemina*, using the IFAT. The results are detailed in Tables 4. 1–4. 26.

5. 1. 1. Interpretation of the IFAT. The IFA test can also detect colostral antibodies, and the mean extinction point of colostral antibodies to *Babesia bigemina* is 119 days (Ross and Löhr, 1970). Antibodies against *Babesia bigemina* or *Babesia bovis* in calves older than four months are considered to be a sign of *Babesia* infection (Mahoney *et al*., 1981). Antibodies are detected by IFAT 2-4 weeks after infection (Anon., 1984) and are detectable for several years (Mahoney *et al*., 1973; Mahoney *et al*., 1979a; Mahoney *et al*., 1981). Chronically infected animals may have low titres and the sera could be classified as negative (Anon., 1984). Drug sterilization of *Babesia bigemina* and *Babesia bovis* in infected animals is followed by a rapid loss of reactivity in the test, without loss of immunity (Callow *et al*., 1974a; 1974b; Callow *et al*., 1993). The interpretation of serological reactions in older cattle therefore requires some caution, and should be guided by careful characterization of the test employed, e.g. a negative test in a previously infected animal may not necessarily indicate the loss of infection and immunity (Callow *et al*., 1974b; Mahoney, 1974; Anon., 1984). Todorovic and Long (1976) found positive IFAT reactions in subclinical infections, but the prevalence and incidence might be under-estimated in serological surveys of babesiosis (Anon, 1984).

5. 1. 2. Serological cross-reactions. Although the IFAT is widely used for detection of antibodies to *Babesia bovis* and *Babesia bigemina*, serological cross-reactions may occur. Shortly after *Babesia bigemina* infection, false positive reactions to *Babesia bovis* can occur, and these may last for several weeks (Smith *et al*., 1980; Bessenger and Schoeman, 1983). A positive *Babesia bovis* test can give positive titres to *Babesia bigemina* (Smith *et al*., 1980; Bessenger and Schoeman, 1983). *Babesia occultans* and several other *Babesia* species can give cross-reactions during acute disease or shortly afterwards (Bessenger and Schoeman, 1983; Papadopoulos *et al*., 1996; De Waal, personal communication, 2000). High titres are sometimes obtained against the homologous antigen while lower titres are obtained against the heterologous antigen (Smith *et al*., 1980). This might be due to mixed infections or to cross-reactions, and the use of IFAT is not always satisfactory for diagnosing infections in areas where both *Babesia bovis* and *Babesia bigemina* are present.

In the field mixed infections are common and cross-reactions tend to increase when the cattle are infected with both *Babesia* species (Papadopoulos *et al.*, 1996). However, Tjornehoj *et al.* (1996) found no evidence of cross-reaction between the two species at a dilution of 1/90. Zwart and Brocklesby (1979) and Smith *et al.* (1980) concluded that the test is sufficiently specific to determine the prevalence of *Babesia bovis* and *Babesia bigemina* where the species coexist. The IFAT is regarded as a reliable test for studying *Babesia bovis* (Johnston *et al*., 1973; OIE, 1996), but might be lacking in sensitivity and specificity for *Babesia bigemina* (Callow, 1979). Low titres of *Babesia bovis* antibodies in areas where the parasite does not occur can be explained by these cross-reactions (Bessenger and Schoeman, 1983). Todorovic and Long (1976) reported only negative samples in cattle from known *Babesia*-free areas and concluded that the IFAT was reliable when indicating absence of infection.

5. 1. 3. Serological findings from cattle sampled at communal dip tanks during 1999 and 2000.

- ***Babesia bovis.*** *Babesia bovis* was widespread in the cattle population in the survey area, with positive reactors in **97** % of the herds and an overall seroprevalence of **63** % over the two years. There was a non-significant (p=0.4866) increase in seroprevalence in the older cattle from 1999 to 2000. The younger cattle showed a non-significant decrease in *Babesia bovis* from 1999 to 2000 (p=0.2179). For all the cattle in the survey there was a non-significant decrease in seroprevalence from 1999 to 2000 (p=0.7078). Many of the herds in the drier areas were endemically unstable to *Babesia bovis*, and clinical cases of Asiatic redwater were common (Loock, 2000, personal communication, Fig. 1. 1). The lower transmission rate in the younger animals in 2000 may have contributed to this instability.

Virtually all (10/11) of the dip tanks sampled in 1999 were located in the Sour Lowveld Bushveld (Low and Rebelo, 1996), but only half (10/20) of those sampled in 2000 were located in this veld type. When selecting dip tanks for sampling in 2000, an effort was made to include dip tanks in the drier parts of the survey area in order to determine the limit of *Babesia bovis* occurrence. This may have skewed the serology results in 2000, and to eliminate this confounding factor, dip tanks with similar rainfall, minimum and maximum temperatures and vegetation were compared in 1999 and 2000.

There was a significant (p=0.0001) increase in the seroprevalence for *Babesia bovis* from 1999 to 2000 in cattle inhabiting Sour Lowveld Bushveld. When the seroprevalence of *Babesia bovis* in cattle in Sour Lowveld Bushveld was compared with that of cattle at all the dip tanks, the overall slight decrease seen in the dip tank herds from 1999 to 2000 seemed to be due to the selection of dip tanks, rather than to a real decrease in transmission of the blood parasite.

Boophilus microplus has a patchy distribution in South Africa and serological surveys have indicated that *Babesia bovis* is unevenly distributed in the country. Few of these surveys have been conducted in communal farming areas. Tice *et al.* (1998) tested young cattle from four communally grazed areas in North West Province and Mpumalanga, and found that the prevalence of *Babesia bovis* varied greatly from year to year and from area to area. None of the four areas were endemically stable for *Babesia bovis*, yet no outbreaks of clinical disease were recorded (Tice *et al*., 1998). Dreyer *et al*. (1998c) reported that the seroprevalence of *Babesia bovis* was close to 20 % in the Free State Province, and concluded that these findings were due to factors other than tick transmission. *Boophilus microplus* had never been reported in the Free State and there had been no outbreaks of disease due to *Babesia bovis*.

Extensive surveys have been made of the distribution of *Babesia bovis* in communal farming areas in other southern African countries (Norval *et al*., 1983; Bryant and

Norval, 1985; Jagger *et al*., 1985; Jongejan *et al.*, 1988; Katsande *et al*., 1999; Smeenk *et al*., 2000; Backx, personal communication, 2001). In some areas in Zimbabwe *Babesia bovis* was present at 50 to 100 % of the dip tanks. Most herds were endemically unstable but the trend seemed to favour greater stability in the areas where the blood parasite was common (Norval *et al*., 1983; Bryant and Norval, 1985; Katsande *et al*., 1999; Smeenk *et al*., 2000).

Babesia bovis was present throughout Mozambique and the herds appeared to be endemically stable (Backx, personal communication, 2001). Surveys indicated that *Babesia bovis* was common in cattle in many of the areas sampled in Swaziland (Jagger *et al*., 1985) and Zambia (Jongejan *et al.*, 1988); 50-80 % of the herds contained seropositive cattle, but they were mostly in an unstable or minimal-disease situation.

In the present survey *Babesia bovis* was more widespread and the seroprevalence was higher in the communal farming areas than was reported by many of the previous studies. The situation resembled that found in Zimbabwe, where *Babesia bovis* was common among the cattle at the dip tanks where *Boophilus microplus* was present and where there was minimal tick control.

- ***Babesia bigemina*.** The present study indicated that *Babesia bigemina* was widespread in the communal farming area with seropositive reactors in all of the herds surveyed. The reduction in the *Babesia bigemina* seroprevalence from 1999 (**56.1** %**)** to 2000 (**49.3** %) was significant (p=0.006); this was attributed to a decrease in the prevalence in the young animals (p=0.0002). The seroprevalence of *Babesia bigemina* was lower than in many earlier studies, and clinical outbreaks of bovine babesiosis due to *Babesia bigemina* were reported in the study area (Loock, 2000, personal communication, Fig. 1. 1).

Boophilus decoloratus is widespread in South Africa (Walker, 1991) and previous serological surveys undertaken in communal farming areas in South Africa have shown that *Babesia bigemina* infections were more common than *Babesia bovis* infections. Tice *et al.* (1998) found positive reactors in all of the communal areas tested, and even though the prevalence was low, there were no reports of clinical disease. Dreyer *et al*. (1998c) recorded 60-70 % seroprevalence of *Babesia bigemina* in two communal grazing areas in the Free State Province, without any clinical cases being reported.

In surveys in other southern African countries it was found that *Babesia bigemina* was widespread in the communal grazing areas. In Zimbabwe *Babesia bigemina* was present in nearly all of the herds sampled, and the herds were only endemically unstable if they were dipped too frequently (Norval *et al*., 1983; Bryant and Norval, 1985; Katsande *et al*., 1999; Smeenk *et al*., 2000). Similar results were reported from Swaziland (Jagger *et al*., 1985) and Zambia (Jongejan *et al*., 1988), where few clinical cases were reported.

There was an overall non-significant (p=0.1941) decrease in seroprevalence of *Babesia bigemina* from 1999 to 2000 at dip tanks located in Sour Lowveld Bushveld. The decrease in the young cattle was significant (p=0.0287), however, which confirms the finding of an overall lower transmission rate in the young animals at all dip tanks during 2000.

5. 1. 4. Serological findings from cattle sampled on the commercial farms during 1999 and 2000. The sample size for the commercial farms was small. In 1999 five farms were included in the survey, but three of them were not re-sampled in 2000. Low seroprevalence of *Babesia bovis* and *Babesia bigemina* was found on these farms in 1999. Two of the farmers vaccinated their cattle in 2000 and the third farm, the Mara Research Station, was not included in 2000 due to a sampling error. The blood sampling at Mara was done on cattle from one herd only and this herd had grazed in the few camps that were specifically allotted to the herd. It was felt that the sample was not representative for all the cattle at the Research Station. The serology results from the commercial farms in 2000 thus came from only two units.

- ***Babesia bovis.*** In the present study *Babesia bovis* was widespread in the commercial farming areas, and the prevalence was higher than reported in most of the earlier studies. Seropositive cattle were found on all of the farms during 1999 and 2000, and the seroprevalence increased significantly (p=0.0001) from **19** % in 1999 to **57.5**% in 2000. This could be attributed to the high infection rates in both old and young animals. The seroprevalence of *Babesia bovis* on the commercial farms in 2000 was similar to the seroprevalences reported by de Vos (1979) on farms where poor tick control was practised.

In earlier surveys in southern Africa it was found that *Babesia* infections in cattle kept on commercial farms were less widespread and at a lower seroprevalence than in cattle kept in communal grazing areas (De Vos, 1979; De Vos and Every, 1981; Gray and de Vos, 1981; Norval *et al*., 1983; De Vos and Potgieter, 1983; Smeenk *et al*., 2000). De Vos (1979) reported great variation in the seroprevalence of *Babesia bovis* between farms in South Africa. The variation was dependent on the presence of the tick vector and the efficiency of the dipping programme. In areas favourable for *Boophilus microplus*, almost all of the commercial herds where poor tick control was practised had seropositive reactors to *Babesia bovis*, and these herds were endemically stable (De Vos, 1979). On other farms where the tick control was better, the seroprevalence of *Babesia bovis* was only 10-30 % and the herds were endemically unstable. Other surveys in South Africa (De Vos and Every, 1981; Gray and de Vos, 1981; De Vos and Potgieter, 1983) reported that *Babesia bovis* was present on less than 40 % of the farms and the seroprevalence varied between 2-60 %.

Studies conducted in Zimbabwe (Norval *et al.*, 1983; Smeenk *et al*., 2000) revealed that *Babesia bovis* was present on 20-75 % of the commercial farms, with a seroprevalence of less than 20 % in most herds. Most of the commercial farms in these studies were endemically unstable for *Babesia bovis*.

- ***Babesia bigemina.*** In the present survey all of the commercial farms had seropositive reactors to *Babesia bigemina* and the parasite appeared to be widespread. The seroprevalence of *Babesia bigemina* decreased slightly from **48** % in 1999 to **47.5** % in 2000. These results were in agreement with those of earlier studies in South Africa (De Vos, 1979; Gray and de Vos, 1981; de Vos and Potgieter, 1983).

Large areas of South Africa are favourable for *Boophilus decoloratus*, and *Babesia bigemina* has been found over large parts of the country, with seropositive cattle reported from 80-100 % of the commercial farms (De Vos, 1979; Gray and de Vos, 1981; de Vos and Potgieter, 1983). On farms with good tick control the seroprevalence in the cattle was low, and these herds were endemically unstable. On farms where less efficient tick control was practised, nearly 100 % of the cattle were seropositive, and these herds were endemically stable to *Babesia bigemina*.

In Zimbabwe *Babesia bigemina* was present on only 40-70 % of the commercial farms sampled, and the seroprevalence was low (Norval *et al*., 1983; Smeenk *et al*., 2000). Most of the farmers practised intensive tick control with the result that their herds were in an endemically unstable situation.

Both *Babesia bovis* and *Babesia bigemina* were widespread in the commercial herds sampled in this region of South Africa, but the sample size (n = 5) was too small to extrapolate the results to other areas. Antibodies to both *Babesia bovis* and *Babesia bigemina* were found in cattle on all of the commercial farms during 1999 and 2000. The prevalence of *Babesia bovis* in the present survey was higher than in most of the earlier studies in southern Africa (De Vos, 1979; Norval *et al.*, 1983; Gray and de Vos, 1981). The prevalence of *Babesia bigemina* in herds on the commercial farms was comparable to earlier results from farms with medium tick control (Norval *et al.*, 1983; Gray and de Vos, 1981).

The transmission of *Babesia bovis* and *Babesia bigemina* occurred readily on both commercial farms sampled in 2000. The percentage of cattle seropositive to *Babesia bovis* increased from **19** % in 1999 to **57.5** % in 2000 (p=0.0001). The increase could be attributed to the high transmission rate in both old and young animals. The seroprevalence of *Babesia bigemina* remained constant. The high transmission rate of *Babesia bovis* on the commercial farms coincided with an increase in *Boophilus microplus* numbers during the same period.

The unusually high rainfall in 2000 appeared to have had more influence on tick control and transmission of TBD in the commercial farm herds when compared with the communally grazed cattle. The commercial farms experienced a substantial increase in *Boophilus microplus* numbers during the survey period, and there were losses due to redwater amongst cattle in the 18-24-month-old group, as well as amongst cattle introduced from other areas. One can speculate that the frequent rain prevented the commercial farmers from carrying out their normal tick control programme, thus enhancing tick survival. During the flooding the commercial farmers had a much wider choice of grazing areas available for their cattle than the communal farmers, and they could graze their cattle on higher ground less affected by the rising water. In these areas the *Boophilus microplus* larvae would have been able to survive and multiply.

5. 1. 5. Statistical significance of the serological results from the communal dip tanks and the commercial farms. In 1999 the seroprevalence of both *Babesia bigemina* and *Babesia bovis* was significantly higher (p=0.0261 and p=0.0001, respectively) in the communally grazed cattle when compared with that of the cattle on the commercial farms. In 2000, however, the seroprevalences of the communally grazed herds and the commercial farm herds showed no significant difference either for *Babesia bovis* (p=0.2890) or for *Babesia bigemina* (p=0.7030) at the end of the season. The details are found in Table 4. 25.

When all the animals sampled are considered, there was an overall significant increase (p=0.0001) in seroprevalence of *Babesia bovis* from 1999 to 2000. The overall seroprevalence for *Babesia bigemina* decreased significantly (p=0.0366) from 1999 to 2000. The non-significant (p=0.6804) decline in the older cattle was offset by a significant decline in the younger animals (p=0.0130). The details are found in Tables 4- 26.

The higher seroprevalence of *Babesia bovis* compared to *Babesia bigemina* found at the communal dip tanks was difficult to explain. One would expect to find a higher transmission rate and seroprevalence for *Babesia bigemina* when compared to those of *Babesia bovis*, due to the higher infection rate of *Babesia bigemina* in the *Boophilus* ticks (Mahoney, 1969; De Vos, 1979; De Vos and Potgieter, 1983). Several studies from southern Africa (Norval *et al*., 1983; Bryant and Norval, 1985; Smeenk *et al*., 2000; Spickett, personal communication, 2001) have reported a higher seroprevalence of *Babesia bovis* compared to *Babesia bigemina* where both parasites co-exist. Tjornehoj *et al*. (1996) monitored an outbreak of Asiatic redwater in Malawi where, within a period of three months, **75** % of previously negative animals had seroconverted to *Babesia bovis*, compared to only **36** % to *Babesia bigemina*.

In the present study the prevalence of *Babesia bovis* in the communally grazed areas in 1999 was non-significantly higher (p=0.3278) than that of *Babesia bigemina* (**63.3** % compared to **56.1** %) but in 2000 the difference (**62.4** % compared to **49.3** %) was significant (p=0.0141).

A similar pattern was found on the commercial farms. In 1999 the seroprevalence for *Babesia bovis* was low (**19** %), probably due to low *Boophilus microplus* numbers. In 2000, when *Boophilus microplus* became more numerous, the seroprevalence of *Babesia bovis* increased and eventually exceeded that of *Babesia bigemina.*

The overall seroprevalence of *Babesia bovis* in all animals in 1999 was not significantly (p=0.6291) different from that of *Babesia bigemina*. This may have been as a result of the low seroprevalence of *Babesia bovis* on the commercial farms due to low *Boophilus microplus* numbers in 1999. In 2000 there was an influx of *Boophilus microplus* onto these farms, the transmission of both *Babesia* species was good and the seroprevalence of *Babesia bovis* for all animals in the survey was significantly (p=0.0142) higher than that of *Babesia bigemina.*

5. 2. Endemic stability to *Babesia bovis* and *Babesia bigemina.*

The concept of endemic stability has been reviewed in Chapter 2. Briefly, Mahoney and Ross (1972) and Norval *et al.* (1983) developed different models for bovine babesiosis which were based on the serological results from a group of young cattle aged up to 9 months. In the present study the serological data from the whole herd was used as a basis for any conclusions on endemic stability. The inclusion of the older animals in this study may have overestimated the number of endemically stable herds, but would give a more realistic view of the real risk of TBD outbreaks.

5. 2. 1. Endemic stability to *Babesia bovis* and *Babesia bigemina* found in the cattle sampled at the dip tanks during 1999 and 2000.

The results are shown in Fig. 4. 1-4. 4.

- ***Babesia bovis***. The percentage of communally grazed herds in the study area which were endemically stable or were approaching stability to *Babesia bovis* was close to **60** % and did not change much from 1999 to 2000. Positive cattle to *Babesia bovis* were found at **100** % of the dip tanks during 1999 and at **97** % of the dip tanks in 2000. More herds appeared to be in a minimal disease or disease-free situation in 2000 compared to 1999, but many of these dip tanks were located in areas that were marginal for the survival of *Boophilus* ticks.

- ***Babesia bigemina***. The percentage of communally grazed herds in this study, which had reached endemic stability or were approaching endemic stability to *Babesia bigemina* was high (**60** %) in 1999, but declined to less than **40** % in 2000. The reasons for the loss of endemic stability to *Babesia bigemina* appeared to be due to a lower infection rate in the younger animals in 2000, as well as unfavourable conditions for *Boophilus* tick survival.

Most of the farmers in the communal farming areas had previously had access to a free government programme where dipping was carried out weekly or fortnightly. From 2000 the farmers had to pay for these services, and dipping became irregular. During February and March 2000 dipping was impossible in much of the study area, as dip tanks were flooded or were inaccessible due to the exceptionally high rainfall. Under these circumstances one would have expected TBD to become a serious problem, but the exact level of clinical disease was unknown. Few clinical cases of TBD were reported to the State Veterinarian in 2000, and all these cases came from the Thohoyandou district, which had the easiest access to the laboratory at Sibasa. Communication in the survey area was severely hampered for the rest of the year as farmers were unable to notify the veterinarians if problems were encountered, so the real extent of losses due to redwater was unknown.

When dip tanks located in Sour Lowveld Bushveld were compared for endemic stability to *Babesia bovis*, there was a slight increase in herds that were endemically stable or were approaching endemic stability (**70** % in 1999 to **80** % in 2000). The percentage of herds that were endemically stable or were approaching endemic stability to *Babesia bigemina* decreased from **70** % in 1999 to **30** % in 2000. The trend towards less endemic stability to *Babesia bigemina*, which was recorded in the total sample in 2000, was real and was not affected by location of the dip tank.

Norval *et al*. (1983) reported that the loss of endemic stability to *Babesia bigemina* was more common in areas where *Boophilus microplus* was well established, and they suggested that *Babesia bigemina* may be transferred less efficiently by *Boophilus microplus* than by *Boophilus decoloratus*. The influx of *Boophilus microplus* into the survey area was followed by a decrease in *Boophilus decoloratus* and this may have affected the transmission of *Babesia bovis* and *Babesia bigemina*. In the present study fewer communally grazed herds had reached endemic stability for *Babesia bigemina* when compared with *Babesia bovis*, and this may have occurred because of the dominance of the *Boophilus microplus* population.

5. 2. 2. Endemic stability to *Babesia bovis* and *Babesia bigemina* found in the cattle sampled on the commercial farms during 1999 and 2000.

The results are shown in Fig. 4. 5-4. 8.

- ***Babesia bovis***. In 1999, **80** % (4/5) of the commercial farm herds were in a minimal disease or disease-free situation. In 2000, 1 of the 2 herds in the survey was approaching endemic stability and the other was endemically unstable. One of the main reasons for the shift in endemic stability status was the increase in the *Boophilus microplus* population on the commercial farms, and this was followed by increased transmission of *Babesia bovis*. Both commercial herds, which were bled twice during the survey period, moved towards greater endemic stability to *Babesia bovis*.

- ***Babesia bigemina.*** In 1999, 2 of 5 of the herds were endemically stable or approaching endemic stability to *Babesia bigemina*, 2 herds were endemically unstable and 1 herd was disease-free. In 2000, 1 of 2 herds was approaching endemic stability whilst 1 herd was unstable. None of the herds were in a minimal disease situation in 2000.

Although the number of commercial farms included in the survey was small, some trends could be seen. Transmission of *Babesia bovis* increased significantly in 2000, while transmission of *Babesia bigemina* remained constant during the survey period. Both factors were probably due, directly or indirectly, to the increase in *Boophilus microplus* numbers.

5. 2. 3. Endemic stability to *Babesia bovis* and *Babesia bigemina* at the dip tanks compared with endemic stability on the commercial farms.

- ***Babesia bovis.*** In 1999, over **60** % of the communally grazed herds had reached endemic stability to *Babesia bovis*, compared with none of the commercial farm herds. None of the communal herds was disease-free or in a minimal disease situation, but **80** % of the commercial farm herds were in this group in 1999. During 2000, the communally grazed herds in the survey area shifted slightly towards instability to *Babesia bovis* but the two commercial farms moved towards greater stability. The difference between the two farming systems had almost disappeared.

- ***Babesia bigemina.*** In 1999, over **60** % of the communally grazed herds had reached endemic stability to *Babesia bigemina*, compared with **40** % of the commercial farm herds. In 1999, none of the communally grazed herds were disease-free or in a minimal disease situation, but **20** % of the commercial farm herds were in this group. During 2000, the communally grazed herds shifted towards instability with less than **40** % approaching stability and **60** % being endemically unstable or in a minimal disease situation. The commercial farm herds, however, moved towards endemic stability. The change with *Babesia bigemina* on the commercial farms was not as dramatic as with *Babesia Boris*, and little difference could be detected between the two farming systems. The two commercial herds in this survey had reached the same level of endemic stability as the communally grazed herds.

5. 2. 4. Correlation between the endemic stability to *Babesia bovis* and *Babesia bigemina* at dip tanks/farms and the presence of specific *Boophilus* species. In 1999, **60** % of the dip tank/farm herds where only *Boophilus microplus* was recorded had reached endemic stability or were approaching endemic stability for both *Babesia bovis* and *Babesia bigemina*. In 2000, **60** % of these dip tank/farm herds had reached endemic stability or were approaching stability for *Babesia bovis*, whilst only **45** % had reached endemic stability or were approaching stability for *Babesia bigemina.*

Although the tick collection method was not standardised, it appeared that there were more ticks present on the cattle in 2000 than in 1999. The number of *Boophilus* ticks that were collected at the dip tanks/farms doubled from 1999 to 2000 and virtually all were *Boophilus microplus*.

In the present study there endemic stability to *Babesia bigemina* appeared to decline when only *Boophilus microplus* was collected at the dip tanks/farms. These findings support those of Norval *et al.* (1983), who reported less endemic stability for *Babesia* *bigemina* when *Boophilus microplus* was well established. They suggested that *Babesia bigemina* was transmitted less efficiently by *Boophilus microplus* than by *Boophilus decoloratus.*

5. 3. Tick collection in the survey area.

5. 3. 1. General discussion of the tick results. The climate in the survey area was well suited for both *Boophilus* species, but whilst *Boophilus decoloratus* was endemic in the region (Theiler, 1949; Walker 1991), *Boophilus microplus* had never been recorded there (Theiler, 1962; De Vos, 1979; Baker *et al*., 1989; Walker, 1991).

At the start of the survey there was a lack of information on the distribution of the two *Boophilus* species in Venda and Soutpansberg, and selected criteria (Chapter 3. 2. 1) were used to decide which communally grazed areas and commercial farms should be included in the survey.

It would appear that the spread of *Boophilus microplus* in the study area was rapid (Loock, 1999, personal communication). The first cattle losses due to *Babesia bovis* were reported as early as 1984, and outbreaks of Asiatic redwater have since escalated (Loock, 1999, personal communication). The losses were especially heavy in the communally farmed areas where the exact mortality details were often not known due to under-reporting by the farmers, but in certain areas the communal farmers lost nearly half their cattle (Loock, 1999, personal communication; Tshisamphiri, 2000, personal communication). Initially the cattle in the study area were fully susceptible to *Babesia bovis*, and the disease outbreaks appeared to be similar to those reported from Zimbabwe, where the small-scale farmers also suffered heavy losses after the breakdown of dipping and the concomitant influx of *Boophilus microplus* (Norval, 1982; Norval *et al*., 1992a).

Some of the commercial farmers had also lost cattle due to *Babesia bovis*, probably due to acaricide resistance or a breakdown in dipping routines, and some of them had then vaccinated their cattle to reduce further losses (Ahrens, 1999, personal communication).

In 1999 *Boophilus microplus* was already dominant in the communally farmed area. Low numbers of *Boophilus decoloratus* were collected even at dip tanks far north and west in the study area during 2000.

5. 3. 2. Reasons for the variation in tick numbers in the study area. There were large variations in the number of ticks found on both communally grazed and commercial cattle, and the following reasons may explain these variations:

- Some dip tanks/farms maintained a strict dipping schedule throughout the study period.

- Some dip tanks/farms were located in areas that were too dry for large populations of *Boophilus* ticks to survive.

- At some of the dip tanks/farms the cattle had been dipped a few days prior to sampling, and in these cases an effort was made to go back to these locations for a second tick collection at a later stage.

- At some of the dip tanks/farms where both *Boophilus decoloratus* and *Boophilus microplus* were present, the tick numbers may have been low due to reproductive interference (Spickett and Malan, 1978; Norval and Sutherst, 1986).

The number of ticks collected from cattle at each dip tank/farm also varied according to the number of collectors involved, the number of days that had elapsed since the last dipping, as well as the location of the dip tank/farm. As many ticks as possible were collected from each dip tank/farm and an attempt was made to collect at least 400 ticks at each location. If few ticks were collected at the first sampling, the area was re-sampled in order to get more information on which *Boophilus* species were present. At certain dip tanks/farms small numbers of ticks were consistently collected, due either to harsh climatic conditions or to strict dipping regimens, and only one sampling was possible at many of these dip tanks. The inconsistency of the sampling procedure must therefore be taken into consideration before a possible association between the relative abundance of *Boophilus* ticks and the seroprevalence of *Babesia bovis* and *Babesia bigemina* can be concluded.

During the first part of the survey an attempt was made to collect *Boophilus* ticks from selected body sites on 6 animals at each dip tank/farm. This practice was soon discontinued, however, as there were few ticks on the cattle in certain regions of the survey area. As one of the main objectives of the study was to assess the proportion of *Boophilus microplus* in relation to *Boophilus decoloratus*, it was felt that sampling specific predilection sites of attachment could be over-ridden.

5. 3. 3. Tick collection from cattle at the dip tanks. *Boophilus* species were the most common ticks on cattle at the dip tanks and were common on communally grazed cattle. This finding is in line with other surveys on communally grazed land where intensive dipping programmes had been practised for a number of years (Norval, 1978, 1979). High stocking rates and limited pasture made ideal conditions for *Boohpilus* larval survival and host finding (Norval, 1978; 1979; Solorio-Rivera *et al.*, 1999), and high *Boophilus* tick numbers were observed on the cattle at most of the dip tanks during this survey. Overgrazing on communally grazed land removes the ground cover, and the larvae and nymphs of 2- and 3-host tick species are not able to develop. These overgrazed pastures are, however, ideal for the survival of *Boophilus* larvae (Rechav, 1982). *Boophilus* ticks increase substantially under these circumstances (Norval, 1978; 1979) and when intensive dipping is discontinued, these ticks are the first to appear in large numbers. Dipping had been carried out intermittently at most of the dip tanks in Venda and as a consequence the *Boophilus* numbers were high. After a period of low tick numbers in February - April 2000 due to flooding, the numbers soon increased again (Tshisamphiri, 2000, personal communication) when the combined effects of government dipping policy and the ruined dip tanks became effective. At nearly 80 % of the dip tanks (n = 30) at least 400 *Boophilus* ticks were collected during both 1999 and 2000.

During 1999 *Boophilus microplus* was already the dominant *Boophilus* species in the area and was collected from cattle at all (**100** %) of the communal dip tanks, whilst *Boophilus decoloratus* was found at only **30** % of the dip tanks. At these dip tanks *Boophilus decoloratus* comprised **23** % of the total *Boophilus* count. When selecting the dip tanks for sampling in 2000, an effort was made to go as far north and west in the survey area as possible in order to find areas where *Boophilus microplus* had not yet spread. Despite this, *Boophilus microplus* was found at **95** % of the dip tanks and *Boophilus decoloratus* at only **16** % of the dip tanks. At these dip tanks *Boophilus decoloratus* comprised **14.7** % of the total *Boophilus* count.

Of the 6163 *Boophilus* ticks collected at the dip tanks during 1999, **4** % were *Boophilus decoloratus*. During 2000 only **1** % of the 15,084 *Boophilus* ticks collected were *Boophilus decoloratus*.

This survey showed that both *Boophilus* species were widespread in the communal grazing areas covered, and that *Boophilus microplus* was the dominant *Boophilus* species at all of the communal dip tanks sampled. Norval *et al*. (1983) reported finding *Boophilus decoloratus* from all climatic zones in Zimbabwe, and this tick was collected from **58** % of the communal grazing areas whilst *Boophilus microplus* was collected from **26** % of these areas.

5. 3. 4. Tick collections from cattle on the commercial farms. In general the cattle on commercial farms in the survey area carried fewer *Boophilus* ticks than cattle on communally grazed land. Pasture management and lower stocking rates on the commercial farms prevented overgrazing, and 2- and 3-host tick species were more common on these farms than *Boophilus* species (Norval, 1978, 1979). Rotation of pastures was a common practice on the commercial farms, but this practice was not feasible on the communally grazed land. Pasture spelling can reduce tick numbers drastically (Johnston *et al.*, 1981; Bigalke *et al.*, 1976).

Tick numbers were also low as a result of the harsh climatic conditions (Mara Research Station) or frequent dipping (Nooitgedacht Farm and Zwartrandjes Farm). The two most westerly farms sampled were the ones where only *Boophilus decoloratus* was found. At Modderfontein Farm and Naboomkop Farm both *Boophilus* species were found during 1999 and the tick numbers may initially have been low due to frequent dipping and reproductive interference (Spickett and Malan, 1978; Sutherst, 1987a). This changed in 2000 when *Boophilus microplus* became the dominant tick on these farms and *Boophilus decoloratus* almost disappeared. On both farms the total *Boophilus* numbers collected increased substantially as *Boophilus microplus* displaced *Boophilus decoloratus* partially or totally. This change on the commercial farms was reflected in the displacement observed in the communally grazed areas.

Most of the commercial farms had strict dipping routines that were applied to new cattle on arrival, whilst introductions into communal herds occurred without such precautions. The spread of *Boophilus microplus* into new areas is associated with cattle movements as this is a one-host tick that seldom feeds on hosts other than cattle (Sutherst and Comins, 1979). One can speculate that the spread of *Boophilus microplus* to commercial farms may have occurred after a breakdown of fences, which resulted in communally grazed cattle straying onto commercial farms. Occasional flooding may also have contributed to this spread (Callow *et al.*, 1976a).

A total of 3795 *Boophilus* ticks were collected from the commercial farms during the survey. During 1999 *Boophilus decoloratus* was the dominant *Boophilus* tick on the commercial farms and was collected from cattle at 4 (**80** %) of the 5 farms, whilst *Boophilus microplus* was found on 3 (**60** %) of the farms. Of the 794 *Boophilus* ticks collected during 1999, **57** % were *Boophilus decoloratus* and **43** % were *Boophilus microplus*. During 2000 *Boophilus microplus* became the dominant *Boophilus* species on the four commercial farms surveyed: **75** % of the 3001 *Boophilus* ticks collected were *Boophilus microplus* and only **25** % were *Boophilus decoloratus*.

Norval *et al*. (1983) reported *Boophilus decoloratus* from **9** % of the commercial farms sampled in Zimbabwe, while *Boophilus microplus* was collected from only **3** % of these localities. These figures are much lower than the findings in this survey and possibly reflect a change in attitude to dipping amongst the commercial farmers in the study area. In 1999 only one of the commercial farmers attempted to keep cattle tick-free, whereas the others all tolerated a certain number of ticks on the cattle.

5. 3. 5. *Boophilus* tick collections from the communal dip tanks compared with those from the commercial farms. *Boophilus microplus* was found in larger numbers at the communal dip tanks than on the commercial farms. On average, more than four times as many *Boophilus* ticks were collected from each dip tank compared with each commercial farm in 1999. During 2000, more *Boophilus* were collected from the commercial farms than during 1999, but more than half of this total came from only one farm, namely Naboomkop. On this farm few ticks were collected during 1999 when both *Boophilus* species were present, but large numbers of ticks were found on the farm in 2000 when only *Boophilus microplus* was present. Few ticks were found on the other commercial farms. During 2000, the average number of ticks collected at the communal dip tanks was almost double that from the commercial farms.

Boophilus decoloratus was never found on its own at any of the dip tanks in the communally grazed areas, but was present on **40** % of the commercial farms in 1999 and **50** % in 2000. *Boophilus microplus* was found on its own on **20** % of the commercial farms in 1999 compared with **73** % of the dip tanks. In 2000 *Boophilus microplus* was found on its own on **25** % of the commercial farms, compared with **84** % of the dip tanks.

5. 4. Displacement of *Boophilus decoloratus* by *Boophilus microplus* in the survey area during 1999/2000.

5. 4. 1. Introduction. The distribution of any tick species is never static and often changes in response to a range of factors, which include the movement of animals, changes in local dipping programmes, different resistance to acaricides and drought. Historically, tick species have been introduced onto new continents (Curnow, 1973b; Payne and Osorio, 1990), and once established their spread is often rapid (Lawrence and Norval, 1979; Barré *et al*., 1987; Walker, 1987; Peter *et al*., 1998). In some cases the newly introduced tick species has been able to compete successfully with or even displace a closely related species (MacLeod and Mwanaumo, 1978; Rechav *et al*., 1982; Norval, 1983; Sutherst, 1987a; Berkvens *et al*., 1998.)

In the early 1900s, the first research reports suggested that *Boophilus microplus* was displacing *Boophilus decoloratus* in the south-eastern Cape (as *Boophilus fallax*, Howard, 1908 or as *Boophilus annulatus*, Dönitz, 1910. Cited by Theiler, 1962). Other reports from southern Africa later confirmed this displacement (Spickett and Malan, 1978; MacLeod and Mwanaumo, 1978; Mason and Norval, 1980; Baker *et al*., 1981;

Norval *et al*., 1983; Norval and Short, 1984; Norval and Sutherst, 1986; Sutherst, 1987b; Berkvens *et al*., 1998). The actual process of displacement of *Boophilus decoloratus* by *Boophilus microplus* has never been documented in South Africa. One of the objectives of the present study was aimed at assessing the relative numbers of *Boophilus microplus* in relation to *Boophilus decoloratus* in the study area and monitoring the possible displacement of *Boophilus decoloratus* by *Boophilus microplus* in the field.

5. 4. 2. Tick findings in the survey area showing displacement. The *Boophilus decoloratus* numbers in the survey area declined from **4** % of the total *Boophilus* count in 1999 to **1** % in 2000. Where changes in the tick population at the dip tank/farm could be followed through several samplings, there was a clear tendency for *Boophilus microplus* to displace *Boophilus decoloratus.*

The two tick species were found to co-exist at 6 communal dip tanks (Thononda, Sendedza, Luvhanga, Tshiendeulu, Mphephu and Keerweerder) and 2 commercial farms (Naboomkop and Modderfontein). It was only possible to repeat samplings at a few of these dip tanks, but on the commercial farms repeated samplings were made.

During 1999 and 2000 *Boophilus decoloratus* co-existed with *Boophilus microplus* at **23** % of the dip tanks or farms (n = 35). During the study *Boophilus decoloratus* appeared to have been completely displaced by *Boophilus microplus* at Thononda dip tank and Naboomkop Farm and partially displaced at Sendedza dip tank and Modderfontein Farm (Table 4. 31). At Luvhanga, Tshiendeulu and Mphephu dip tanks the numbers of *Boophilus decoloratus* were low and at Keerweerder dip tank only one

Boophilus microplus tick was found out of 36 *Boophilus* ticks collected and there were no positive reactors to *Babesia bovis*.

- **Thononda dip tank.** In May 1999, **75** % of the *Boophilus* ticks collected were *Boophilus decoloratus* and **25** % were *Boophilus microplus*. In December the same year, all (**100 %**) were *Boophilus microplus.* It appeared that *Boophilus decoloratus* was no longer present at this dip tank and the finding was confirmed in May 2000 when all *Boophilus* ticks were *Boophilus microplus.*

- **Sendedza dip tank.** In May 1999, **95.7** % of the *Boophilus* ticks collected were *Boophilus decoloratus* and only **4.3** % were *Boophilus microplus.* In December 1999, **8.4** % of the ticks were *Boophilus decoloratus* and **91.6** % were *Boophilus microplus*. There was a tendency at this dip tank towards total displacement of *Boophilus decoloratus* by *Boophilus microplus.* The dip tank was destroyed in the flooding in February 2000 and no more ticks could be collected from this site.

- **Modderfontein Farm.** At the first tick collection from this farm (September 1999) **7.8** % of the *Boophilus* ticks collected were *Boophilus decoloratus* and **92.2** % were *Boophilus microplus*. In November 1999, **41.9** % were *Boophilus decoloratus* and **58.1** % *Boophilus microplus.* In May 2000, **18.1** % were *Boophilus decoloratus* and **81.9** % *Boophilus microplus.* In December 2000, only **3.7** % of the ticks were *Boophilus* *decoloratus* whilst **96.3** % were *Boophilus microplus.* There was a strong tendency towards displacement of *Boophilus decoloratus* by *Boophilus microplus* on this farm.

- **Naboomkop Farm.** At the first collection from the farm (September 1999) **16.7** % of the *Boophilus* ticks collected were *Boophilus decoloratus* and **83.3** % were *Boophilus microplus*. In November 1999, **27.6** % of the ticks were *Boophilus decoloratus* and **72.4** % were *Boophilus microplus*. In May 2000, all (**100** %) of the ticks were *Boophilus microplus.* It appeared that *Boophilus decoloratus* was no longer present on the farm and the displacement of *Boophilus decoloratus* by *Boophilus microplus* seemed complete.

The findings in this survey strongly support other surveys where rapid spread of *Boophilus microplus* and the displacement of *Boophilus decoloratus* have been reported (MacLeod and Mwanaumo, 1978; Mason and Norval, 1980; Baker *et al.*, 1981; Norval *et al.*, 1983; Berkvens *et al.*, 1998). The hypothesis that *Boophilus decoloratus* can be displaced by *Boophilus microplus* in 1-3 years also seems to have been confirmed (Sutherst, 1987a).

5. 4. 3. The use of the CLIMEX Ecoclimatic Index (EI) and the CLIMEX maps.
CLIMEX is a computer-software programme designed to model the effects of climate on the distribution and relative abundance of plants and animals. CLIMEX uses climatic data together with biological data and known geographic distribution data of species. The model can be used to predict the spread and potential for development of an organism into a new geographical area (Sutherst and Maywald, 1985).

The results are found in Table 4. 32 and the maps are shown in Fig. 5. 1-5. 4.

The communal dip tanks and commercial farms in this survey were scattered over a large area in Venda and Soutpansberg. All the different locations had different possibilities for the survival and development of *Boophilus* ticks and consequently for the development of endemic stability to *Babesia bovis* and *Babesia bigemina* in the herds.

The higher the EI for a location, the larger the potential for survival of the tick and therefor for an increase in the population. If the value is lower than about 20 on a scale of 1-100, then this is an indication that the environment is moderately favourable for the tick, but the tick species will still be able to survive and multiply. With an EI of less than 5, the tick numbers on the cattle would be too low to achieve endemic stability to either *Babesia* species by tick transmission alone (Sutherst and Maywald, 1985; 1986; Sutherst, 1987b).

The EIs in the survey area and the matching maps of the possible distribution of *Boophilus decoloratus* and *Boophilus microplus* were computed using 30 years of climatic data (by courtesy of Prof. R. Schulze, University of Natal).

At some of the dip tanks/farms in the survey area the rainfall was marginal for the survival of either *Boophilus* species. There were marked differences in the EIs for *Boophilus decoloratus* and *Boophilus microplus* at all locations in the survey, with *Boophilus decoloratus* being the more drought-resistant tick. Some of the dip tanks/farms had EIs for *Boophilus decoloratus* of less than 5, and few *Boophilus* ticks were found in these areas. The EI for *Boophilus decoloratus* was always higher than for *Boophilus microplus*; even so, *Boophilus decoloratus* was collected at only 8 of the 35 dip tanks/farms in the survey. This skewed result clearly shows the displacement of *Boophilus decoloratus* by *Boophilus microplus.*

The mean annual rainfall in the survey area over the past 6 years was almost double that of the 5 previous years (Data from The South African Weather Bureau). With double the average rainfall it was possible to show the expected distribution of the two species during a prolonged wet spell or as a result of a permanent change in the weather pattern (Fig. 2 and 4).

During a wet cycle conditions for growth and development of *Boophilus microplus* were favourable even in areas with normally low EIs, as small amounts of rainfall in the dry months would considerably reduce the dry stress and make the area more suitable for *Boophilus.* At Messina the EI for *Boophilus decoloratus* is 4 and the EI for *Boophilus microplus* is 0 in years with average rainfall. With 25 mm of rain or irrigation per week during the dry months the EIs can change to 39 and 24, respectively (Sutherst, personal communication, 2001). The unusually heavy rainfall in the study area over the past few years may explain why *Boophilus microplus* had spread so successfully into the drier part of this region.

In Fig. 5. 2 it is clear that the areas suitable for *Boophilus microplus* have vastly expanded. Large numbers of *Boophilus microplus* were found at dip tanks situated outside the area where this species would normally be expected to occur. Some of the communal herds grazed in areas located on the southern slopes of the Soutpansberg Mountains whilest the dip tank was located on the dry northern slopes (Tshisamphiri, personal communication, 2001). In other instances a non-perennial river, a vlei or a waterhole may constitute a perfect microclimate for the tick (Sutherst and Maywald, 1985).

The climate in many parts of Venda is suitable for *Boophilus microplus* and the tick will probably continue to spread and may become permanently established in many of the different ecological areas adjacent to the dip tanks. To the west of Louis Trichardt as well and in the Mopani veld north of the Soutpansberg Mountains *Boophilus decoloratus* will probably persist, as the climatic conditions are much drier with an EI for *Boophilus microplus* close to 0 in years of average or lower rainfall.

Cattle normally develop some resistance to *Boophilus microplus* after a few months of exposure (Wagland, 1975; 1978; Sutherst *et al*., 1979; Sutherst and Utech, 1981). As a consequence *Boophilus microplus* loses some advantage over *Boophilus decoloratus* in herds which have recently been invaded. As a result of the large fluctuations in annual rainfall, coupled with the small difference in host adaptations of each species, a seesaw effect can develop whereby each species sequentially displaces the other. This has been observed in Zimbabwe and in Swaziland (Norval *et al*., 1992b; Wedderburn *et al*., 1991). These areas can become highly unstable for both *Babesia bovis* and *Babesia bigemina*, as *Boophilus* numbers never increase enough to maintain the endemic stability to either parasite.

A pattern of introduction, disappearance and re-introduction of *Boophilus microplus* can be expected in some of the regions where the tick was found in substantial numbers during this survey. Earlier reports have shown that this can happen in South Africa (Bigalke *et al*., 1976). A similar occurrence has been described in Zimbabwe where

Rhipicephalus appendiculatus was introduced at the start of a wet climatic cycle and disappeared at the end of a dry cycle (Norval and Perry, 1990). In other regions *Boophilus microplus* will be able to establish itself permanently, especially in those areas where the EI is close to or over 20.

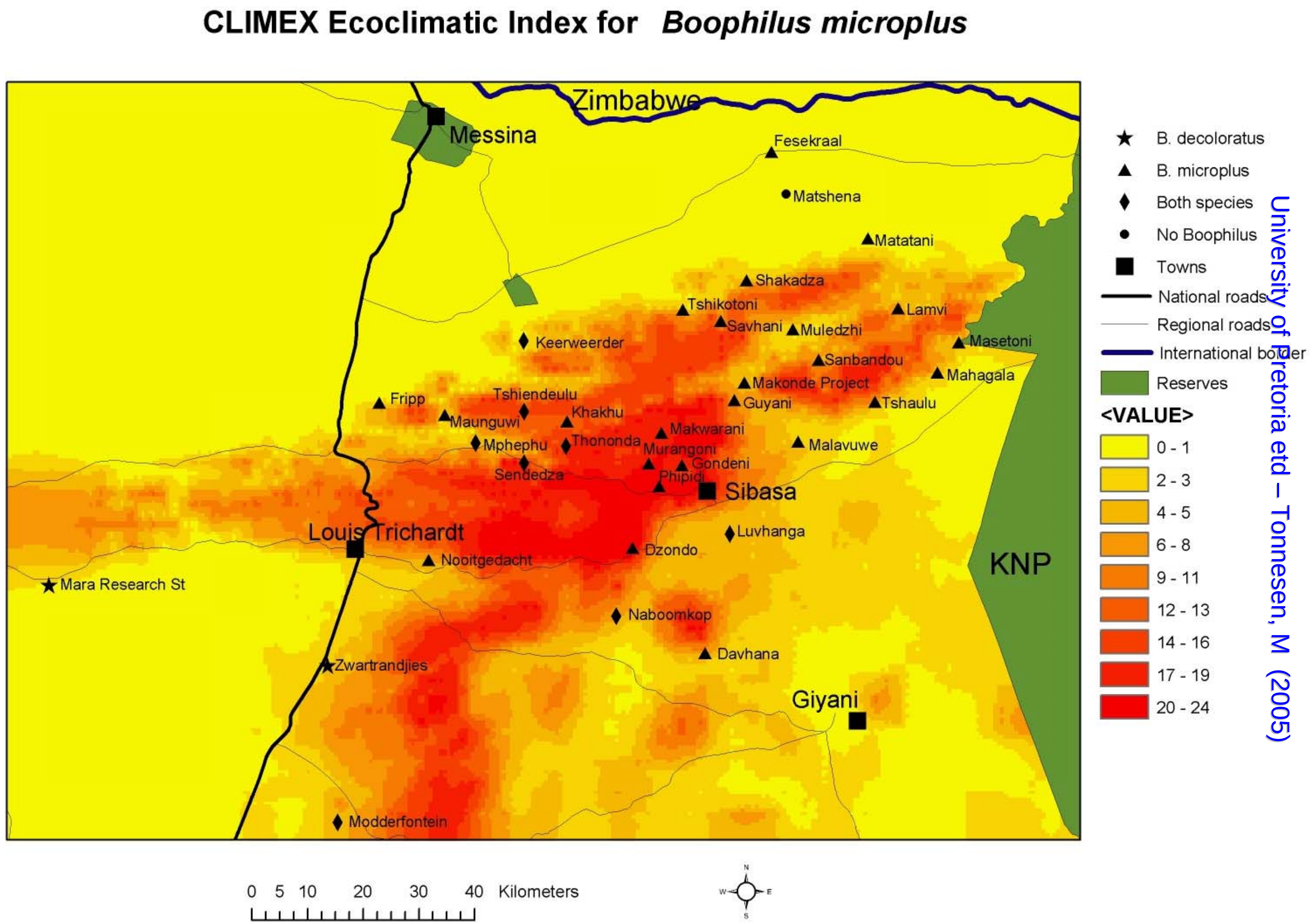


Fig. 5. 1. CLIMEX Ecoclimatic Index map illustrating the predicted distribution of *Boophilus microplus* in years with average rainfall.

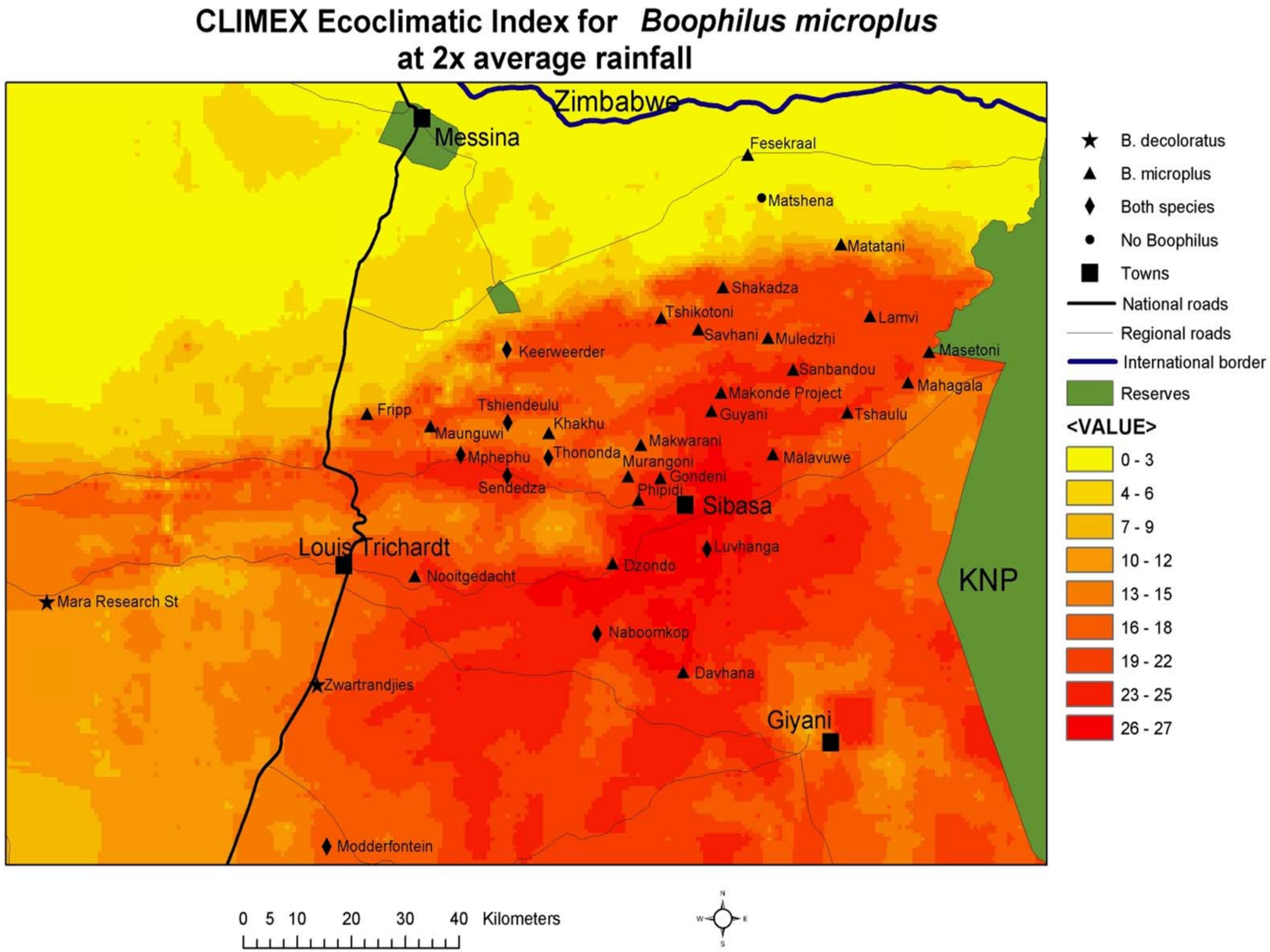


Fig. 5. 2. CLIMEX Ecoclimatic Index map illustrating the predicted distribution of *Boophilus microplus* in years with double average rainfall.

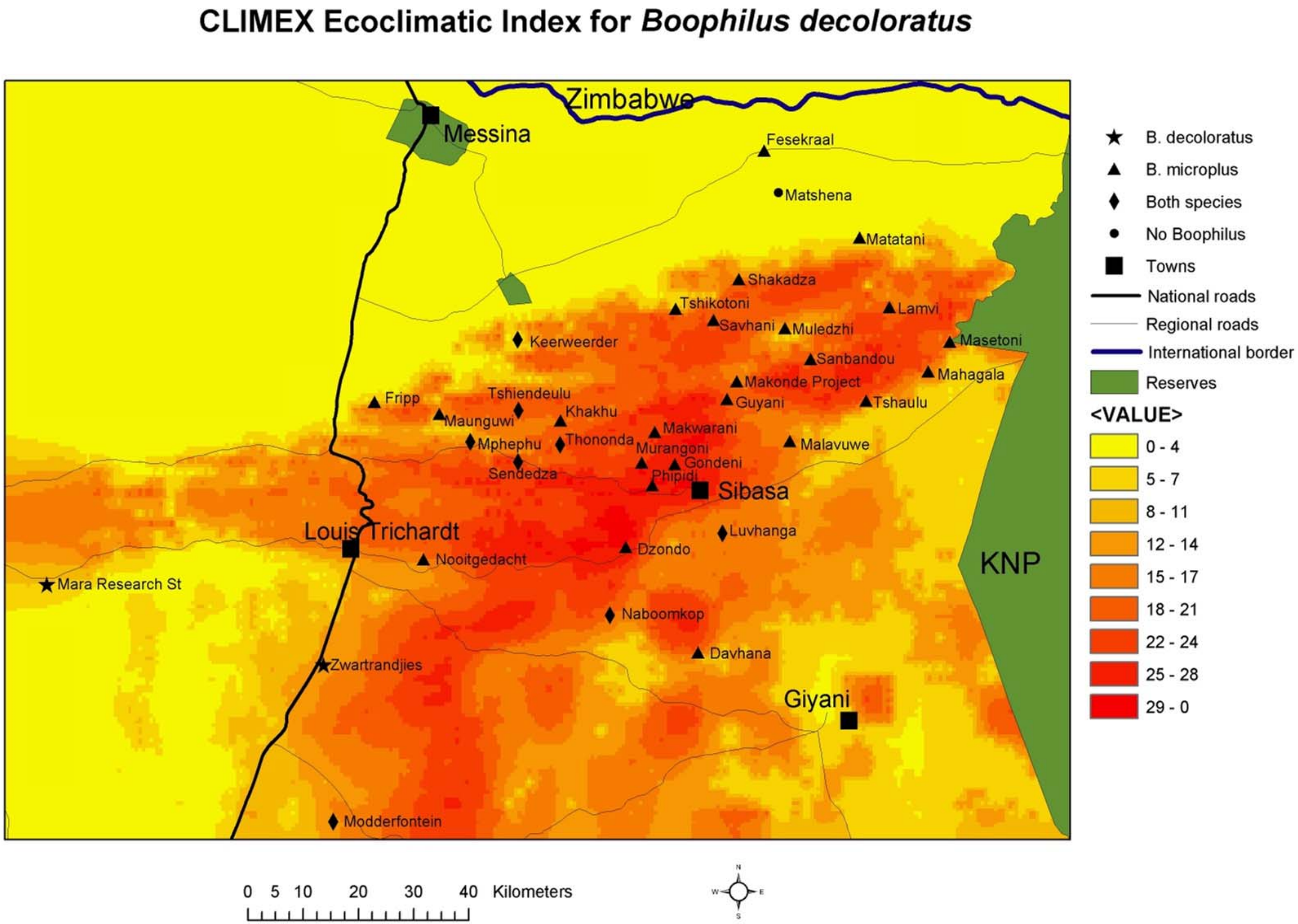


Fig. 5. 3. CLIMEX Ecoclimatic Index map illustrated the predicted distribution of *Boophilus decoloratus* in years with average rainfall.

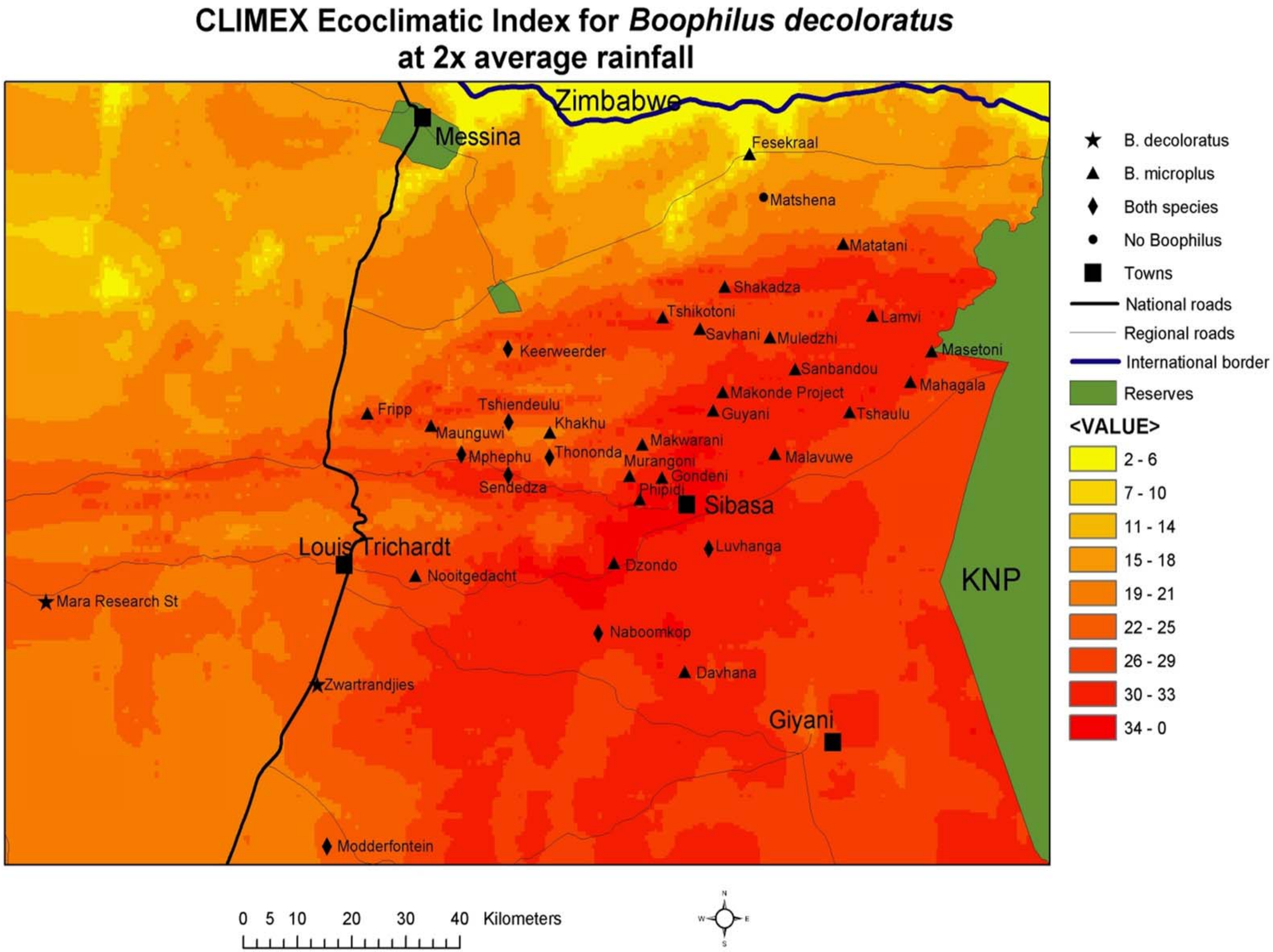


Fig. 5. 4. CLIMEX Ecoclimatic Index map illustrating the predicted distribution of *Boophilus decoloratus* in years with double average rainfall.

5. 5. Possible association between the *Babesia* seroprevalence and the presence of *Boophilus* tick species collected at the communal dip tanks and the commercial farms during 1999 and 2000.

5. 5. 1. Introduction. During the present survey the seroprevalences of *Babesia bovis* and *Babesia bigemina* in cattle at the communal dip tanks/commercial farms were compared with the presence or absence of the specific *Boophilus* tick species. The seroprevalences in the herds were also compared with the numbers of ticks collected off the cattle at the time of the bleeding. By monitoring the change in *Babesia* seroprevalence, a possible association between seropositive cattle and the ongoing displacement of *Boophilus decoloratus* by *Boophilus microplus* was followed. The findings in this survey were compared with those of Norval *et al*. (1983).

5. 5. 2. Possible association between the *Boophilus* tick species on the cattle and the mean *Babesia bovis* seroprevalence.

The results were presented in Tables 4. 23-4. 27

During 1999, the mean seroprevalence of *Babesia bovis* was **10** % at the dip tanks/farms where *Boophilus decoloratus* was the only *Boophilus* tick recorded. At the dip tanks/farms where both *Boophilus* species were present, the mean seroprevalence of *Babesia bovis* was **50** %, compared with **60.8** % in herds where only *Boophilus microplus* occurred. The latter difference was significant (p=0.0001).

In 2000, where both *Boophilus* species were present at the dip tanks/farms, the mean seroprevalence for *Babesia bovis* was **49.2** %, compared with **68.3** % where only *Boophilus microplus* was present. The difference was significant (p=0.0001).

This increase in the mean seroprevalence of *Babesia bovis* was to be expected, as high tick numbers at those dip tanks/farms where only *Boophilus microplus* was present would have ensured effective transmission of *Babesia bovis*. This was supported by the fact that many of the herds at these dip tanks/farms had reached or were approaching endemic stability. The strong correlation between the presence of *Boophilus microplus* and *Babesia bovis* confirms that *Boophilus microplus* is the main and probably the only vector of *Babesia bovis* in South Africa (Potgieter, 1977; Norval *et al*., 1983).

When *Boophilus microplus* is introduced, the prevalence of *Babesia bovis* in a herd would be expected to rise. The high *Babesia bovis* prevalence found in most herds in the survey was probably associated with a longstanding infection with *Babesia bovis*, and the veterinary records indicated that *Babesia bovis* had been in the area for the past 15 years. The fact that *Boophilus decoloratus* was still present and had not been totally replaced indicated that the infection at the dip tank/farm was recent and a low seroprevalence of *Babesia bovis* can be expected.

5. 5. 2. 1. Factors affecting the *Babesia bovis* seroprevalence during the survey. Small numbers of positive reactors to *Babesia bovis* were detected in the absence of *Boophilus microplus* at some of the dip tanks/farms. As *Boophilus microplus* is the only known vector of *Babesia bovis* in this part of Africa (Potgieter, 1977; Norval *et al*., 1983), other possible explanations for these positive findings needed to be considered. These include:

- Seropositive cattle may have been brought in from the outside as a cultural exchange (lobola) or as replacement cattle (Norval *et al.,* 1983). In the northern part of Venda cattle may have been smuggled across the border from Zimbabwe (Tshisamphiri, personal communication, 2000).

- *Boophilus microplus* may have been present at the dip tanks/farms in small numbers without being detected. (Norval *et al.*, 1983). This would give a low transmission rate for *Babesia bovis* and a minimal-disease situation would develop. Dreyer *et al.*, (1998c) recorded seropositive reactors to *Babesia bovis* in 20 % of the cattle without actually finding *Boophilus microplus* in a sample of 230,000 *Boophilus* ticks. It is possible that small numbers of *Boophilus microplus* may have been overlooked at some of the dip tanks/farms, as the tick was not expected to be present in that area (Dreyer *et al*., 1998c). In the present study, the numbers of ticks collected at the dip tanks/commercial farms where only *Boophilus decoloratus* was found were very low, so the possibility of *Boophilus microplus* being present here could not be excluded.

- The IFAT may have given false positive reactions. Shortly after infection with *Babesia bigemina* false positive reactions to *Babesia bovis* are seen (Smith *et al*., 1980; Bessenger and Schoeman, 1983), and the low mean seroprevalence of *Babesia bovis* in areas where the parasite does not normally occur may be explained by these cross-reactions (Bessenger and Schoeman, 1983; Papadopoulos *et al*., 1996). Various authors, however, have concluded that the test is sufficiently specific to determine the prevalence of *Babesia bovis* and *Babesia bigemina* where the species coexist (Zwart and Brocklesby, 1979; Smith *et al*., 1980; Tjornehoj *et al*., 1996).

5. 5. 3. Possible association between the *Boophilus* tick species on cattle and the mean *Babesia bigemina* seroprevalence.

The results were presented in Tables 4. 23-4. 27

During 1999, the seroprevalence of *Babesia bigemina* was **59.2** % when only *Boophilus decoloratus* was recorded at the dip tank/farms. Seroprevalence was **55.7** % where both *Boophilus* species were present and **55.0** % where only *Boophilus microplus* was collected; these differences were not significant (p=0.7125).

During 2000 the seroprevalence to *Babesia bigemina* was **44.6** % where both *Boophilus* species were present and **53.2** % where only *Boophilus microplus* was found; the difference was significant (p=0.0166).

The composition of the *Boophilus* population at the communal dip tanks/farm herds where both *Boophilus* species were present changed from 1999 to 2000. Few *Boophilus* ticks were found on the cattle at the 5 dip tanks/farms included in this group in 1999 and **23** % of these were *Boophilus decoloratus.* In 2000 there were only 4 dip tanks/farms in the group where both *Boophilus* species were found, and although the mean number of collected *Boophilus* ticks more than doubled, only **10.4** % were *Boophilus decoloratus.*

Only small numbers of *Boophilus decoloratus* were present in the total tick sample during 2000. The low transmission rate in the young animals that year, together with the few *Boophilus decoloratus* ticks collected, may explain why the transmission of *Babesia bigemina* in the group with both *Boophilus* species present was initially low (**44.6** %) when compared to the transmission in the similar group during 1999 (**55.7** %). The average *Boophilus* numbers collected from cattle at each dip tank/farm increased in 2000, and it is possible that the relatively high seroprevalence of *Babesia bigemina* at dip tanks/farms where *Boophilus microplus* was dominant was a result of the general increase in *Boophilus* numbers. In 2000 the seroprevalence of *Babesia bigemina* in the group of dip tanks/farms with both *Boophilus* species present (**44.6** %) changed to 53.2 % where only *Boophilus microplus* present. The seroprevalence of *Babesia bigemina* at dip tanks/farms with only *Boophilus microplus* present in 2000 was still lower than in the similar group (**55** %) the previous year.

The increasing *Boophilus microplus* numbers at the dip tanks/farms and the low transmission rate of *Babesia bigemina* may be the underlying reason for the low seroprevalence of *Babesia bigemina* in 2000. The number of dip tanks/farms where both *Boophilus* species were found was too small to draw any conclusions.

No significant association was found between the presence of *Boophilus decoloratus* and the seroprevalence of *Babesia bigemina*. During 1999 the seroprevalence of *Babesia bigemina* decreased as *Boophilus decoloratus* was displaced by *Boophilus microplus*, but during 2000 the seroprevalence of *Babesia bigemina* increased.

Because of the loss of endemic stability in the areas where *Boophilus microplus* was dominant, Norval *et al*. (1983) suggested that *Babesia bigemina* was transmitted less efficiently by *Boophilus microplus* than by *Boophilus decoloratus*. It was not possible to confirm this hypothesis in this project. There was, however, a sharp decline in endemic stability to *Babesia bigemina* in areas dominated by *Boophilus microplus* in the present study. More research is needed on the transmission of *Babesia bigemina* by the *Boophilus* species in Africa.

5. 5. 4. Possible association between the seroprevalence of *Babesia bovis* and *Babesia bigemina* and the presence of *Boophilus* ticks at the dip tanks/farms. At dip tanks/farms where both *Boophilus* species were present, there was no significant difference in the mean seroprevalence for *Babesia bigemina* and *Babesia bovis* either in 1999 or in 2000. During both these years the sample size was small, 5 dip tanks/farms in 1999 and 4 dip tanks/farms in 2000.

There was a non-significant (p=0.5781) difference in the seroprevalences of *Babesia bigemina* and *Babesia bovis* collected at the 8 dip tanks/farms with only *Boophilus microplus* present in 1999, with the seroprevalence of *Babesia bovis* being higher than that of *Babesia bigemina*. This trend was confirmed in 2000 with a significant difference (p=0.0112) between the seroprevalences of the two *Babesia* species at the dip tanks/farms where only *Boophilus microplus* was present. When all 24 dip tanks/farms with only *Boophilus microplus* present during both 1999 and 2000 were compared, the seroprevalence of *Babesia bovis* was significantly higher than that of *Babesia bigemina* (p=0.0065). When all 22 dip tanks where only *Boophilus microplus* was present during both 1999 and 2000 were compared, the difference in seroprevalence of *Babesia bovis* and *Babesia bigemina* was even more significant (p=0.0028). The results are in Tables 4. 24-4. 4.27

5. 5. 5. Possible association between the relative abundance of *Boophilus* ticks at the dip tanks/farms compared with the seroprevalence to *Babesia bovis* and *Babesia bigemina*. In areas which were marginal for tick survival, tick numbers were low and this resulted in a low transmission rate for both *Babesia* species. In order for 100 % transmission to occur during the first nine months of their lives exotic cattle need to be bitten by at least 20 and indigenous breeds by at least 40 ticks a day, respectively (Mahoney, 1979).

When the dip tanks/farms with similar *Boophilus* tick numbers were grouped according to high or low mean tick counts, the low tick counts generally coincided with the low seroprevalence for both *Babesia bovis* and *Babesia bigemina*. If the mean seroprevalence of *Babesia bigemina* and *Babesia bovis* in cattle at 12 of the dip tanks with the lowest tick counts at the time of bleeding were compared, the seroprevalence of *Babesia bigemina* (**43.6** %) was non-significantly (p=0.0912) higher than for *Babesia bovis* (**29.2** %). This was to be expected as the dip tanks/farms with lowest tick numbers contained a large proportion of locations where *Boophilus decoloratus* was still present.

As tick numbers increased, there was a general increase in seroprevalence for both *Babesia bigemina* and *Babesia bovis*. When the mean seroprevalences of *Babesia bigemina* and *Babesia bovis* in cattle at 12 dip tanks with the highest tick numbers at the time of bleeding were compared, then the seroprevalence of *Babesia bigemina* (**55.5** %)

was significantly (p=0.0060) lower than that of *Babesia Bovis* (**75.9** %). The increase in tick numbers was apparently due to an influx of *Boophilus microplus.*

According to Australian surveys *Boophilus microplus* transmits *Babesia bigemina* more effectively than *Babesia bovis* (Callow and Hoyte, 1961; Callow, 1964; 1967; Mahoney, 1969; Mahoney and Mirre, 1971; Mahoney *et al*., 1973; Callow *et al*., 1976a; Mahoney *et al*., 1981; Büscher, 1988; Bock *et al.*, 1999a; 1999b). In the present survey the transmission of *Babesia bigemina* appeared to decline, as *Boophilus microplus* became more numerous. The findings in **5. 1. 3**, **5. 5. 4** and **5. 5. 5** may be due to the fact that *Boophilus microplus* transmits *Babesia bigemina* less effectively than it transmits *Babesia bovis* (Norval *et al*., 1983). More research is needed on how *Boophilus microplus* transmits *Babesia* species in Africa.

5. 5. 6. Changes in seroprevalence of *Babesia bovis* and *Babesia bigemina* in the cattle at single dip tanks/farms where displacement of *Boophilus decoloratus* by *Boophilus microplus* was monitored. The number of dip tank/farms where both *Boophilus* species occurred was low (n=8).

- At Modderfontein Farm both tick species were found at the first sampling in May 1999 and *Boophilus decoloratus* was still present in small numbers in December 2000. The seroprevalence on this farm reflected the change in the tick population; in 1999, **60** % of the herd was positive to *Babesia bovis* and **65** % was positive to *Babesia bigemina*. By 2000, **66.7** % of the herd were positive to *Babesia bovis* whilst only **30** % were now positive to *Babesia bigemina*.

- Naboomkop Farm initially had low seroprevalence to *Babesia bovis* (**10** %) and higher seroprevalence of *Babesia bigemina* (**53.3** %), and there were few *Boophilus* ticks present. In 2000, *Boophilus microplus* had displaced *Boophilus decoloratus* and the seroprevalence to *Babesia bovis* had increased to **48.3** % and the seroprevalence to *Babesia bigemina* had increased to **65.0** %. The latter increase may have been due to a sharp increase in the *Boophilus* numbers.

- At Tshiendeulu dip tank only **4** % of the *Boophilus* ticks were *Boophilus decoloratus* and the serology results reflect the changes in an area where *Boophilus microplus* was dominant: **76.7** % of the herd was positive for *Babesia bovis* and **50** % was positive for *Babesia bigemina*.

- At Sendedza dip tank the serology reflected the pattern that one might expect in an area where *Boophilus microplus* was increasing, with a prevalence of *Babesia bovis* (**43.3** %) being lower than that of *Babesia bigemina* (**63.3** %). In May 1999 *Boophilus decoloratus* was the most common tick at this dip tank; in December 1999 the displacement of *Boophilus decoloratus* by *Boophilus microplus* was almost complete.

- At Luvhanga dip tank there were few *Boophilus decoloratus* remaining and the seroprevalence to *Babesia bovis* was higher (**73.3** %) than that of *Babesia bigemina* (**65** %).

- At Thononda dip tank both *Boophilus* species were collected on the first sampling but the tick numbers were low and only 98 *Boophilus* ticks were collected. The seroprevalence to *Babesia bovis* was **63.3** % and the seroprevalence to *Babesia bigemina* was **31.7** %. During subsequent sampling, when high numbers of ticks were collected, *Boophilus microplus* appeared to have displaced *Boophilus decoloratus.*

- At Mphephu dip tank *Boophilus microplus* had almost replaced *Boophilus decoloratus,* but the serology still showed a pattern with higher seroprevalence of *Babesia bigemina* (**65.0** %) and lower seroprevalence of *Babesia bovis* (**53.3** %).

- At Keerweerder dip tank few ticks were collected and only one was *Boophilus microplus.* The serology indicated low transmission of TBD and there were no reactors to *Babesia bovis*. At this dip tank *Boophilus microplus* may have been introduced recently.

5. 5. 7. The ability of the different *Boophilus* ticks to transmit *Babesia* species. Most of the research on *Boophilus microplus* and the ability of the tick to transmit the *Babesia* species has been done in Australia or with Australian strains (Callow and Hoyte, 1961; Callow, 1964; 1967; Mahoney, 1969; Mahoney and Mirre, 1971; Mahoney *et al*., 1973; Callow *et al*., 1976a; Mahoney *et al.*, 1981; Büscher, 1988; Bock *et al.*, 1999a; 1999b). When Spickett and Malan (1978) crossed Australian and South African strains of *Boophilus microplus,* they reported hybrid sterility in the next generation. This crossing may indicate that in order to adapt to the new environment, the South African strain of *Boophilus microplus* has become genetically different from the Asian strain originally imported into the two continents at the end of the 19th century. In addition, one can speculate that the Australian research on the transmission of *Babesia bigemina* by *Boophilus microplus* does not necessarily apply to the disease transmission by the African strain of *Boophilus microplus.* This complex problem merits further research.

6. 1. 1. Conclusions on the displacement of *Boophilus decoloratus* by *Boophilus microplus*.

When *Boophilus microplus* first invaded the southern part of Africa in the late 1890s, it seemed to have been contained along the coastal areas of the Eastern Cape and KwaZulu-Natal and the spread into the hinterland was slow. Over the years there were occasional reports of finding *Boophilus microplus* in new places, but the tick did not invade all areas which were climatically favourable to it. One can speculate that *Boophilus microplus* needed time to adapt to the conditions on the new continent. Spickett and Malan (1978) crossed Australian and South African *Boophilus microplus* and reported hybrid sterility between the two strains. De la Fuente *et al*. (2000) used DNA techniques to show that the genetic variation between the *Boophilus microplus* strains from South America and Australia was greater than between some strains of *Boophilus microplus* and *Boophilus annulatus*.

The above findings may indicate that the strains of *Boophilus microplus* from isolated gene pools had changed considerably from the original tick imported from Asia more than a century previously. One can speculate that these changes resulted in *Boophilus microplus* gradually becoming better adjusted to the local climate and vegetation. This adjustment may then have resulted in the rapid spread and concurrent displacement of *Boophilus decoloratus* seen in parts of southern Africa since 1970 (Mason and Norval, 1980; Baker *et al*., 1981; Norval *et al*., 1983; Berkvens *et al*., 1998).

Boophilus species are one-host ticks and they only spread through the movement of cattle. Over the past 30 years there has been a paradigm shift in attitudes towards tick burdens on cattle (Mahoney and Ross, 1972; Callow, 1977; Norval, 1982; Norval *et al*., 1983). Undipped cattle are regularly moved over large distances. This paradigm shift in the philosophy of dipping may have facilitated the spread of *Boophilus microplus* into new areas at a time when the tick had become truly adapted to southern Africa.

A possible development of different levels of acaricide resistance in the two *Boophilus* species may explain why *Boophilus microplus* seems to displace *Boophilus decoloratus* in the field. Acaricide resistance develops quickly in one-host ticks (Norval *et al*., 1992b), but resistance patterns of the two species collected from the same farms are not often available (Baker *et al*., 1981). There is at present no reason to believe that one of the two ticks is more resistant to widely used acaricides than the other. The displacement process seems to be due to factors other than acaricide resistance.

Until recently, researchers have suggested that the spread of *Boophilus microplus* into new areas in Africa was contained or delayed by the creation of a zone of sterile hybrids where *Boophilus decoloratus* and *Boophilus microplus* overlap. The spread of *Boophilus microplus* was supposed to be slowed down and this zone would prevent *Boophilus microplus* from occupying all the climatologically favourable areas on the continent (Sutherst and Maywald, 1985; Sutherst, 1987a; Norval *et al*., 1992a; De Vos and Potgieter, 1994). Such sterile hybrids do occur when *Boophilus microplus* and *Boophilus annulatus* interbreed (Hilburn *et al*., 1991; Hilburn and Davey, 1992).

Interspecific matings between *Boophilus microplus* and *Boophilus decoloratus* in the laboratory produced sterile eggs (Spickett and Malan, 1978). When *Boophilus decoloratus* and *Boophilus microplus* were placed on cattle in equal numbers, the two species showed a mating preference for their own kind (assortative mating) and only 10 % of the egg mass was sterile, presumably from interspecific mating (Norval and Sutherst, 1986). There was no evidence that any hybrid offspring had been produced (Norval and Sutherst, 1986).

It would appear that a viable *Boophilus decoloratus*/*Boophilus microplus* hybrid similar to the sterile hybrids described between *Boophilus microplus* and *Boophilus annulatus* is unlikely. One may speculate that the shorter life cycle of *Boophilus microplus*, combined with a possible larger number of the *Boophilus microplus* males available for mating (Mason and Norval, 1980; Hilburn *et al*., 1991; Hilburn and Davey, 1992), may give *Boophilus microplus* a reproductive advantage over *Boophilus decoloratus*. The amount of assortative mating between the two species reported by Norval and Sutherst (1986) is uncertain (Hilburn *et al.*, 1991). There seems to be a zone of reproductive interference where *Boophilus decoloratus* and *Boophilus microplus* overlap, but the experimental evidence of this zone is too small to draw any conclusion on the extent to which it contributes to contain or prevent the spread of *Boophilus microplus.* The present study suggested that reproductive interference was ineffective in preventing *Boophilus microplus* from spreading when the climatic conditions were favourable. The displacement in these areas appeared to be rapid and total.

If the EI (Sutherst and Maywald, 1985) for *Boophilus microplus* is low, *Boophilus decoloratus* may co-exist for some time with *Boophilus microplus* during the displacement process, or it may be found on its own. When the EI for *Boophilus microplus* is high, *Boophilus decoloratus* appears to disappear quickly when *Boophilus microplus* is introduced into an area. The present distribution of *Boophilus microplus* in South Africa is unknown, so mapping the presence of this tick should be a high priority.

In Africa there are large geographical areas at risk to colonization by *Boophilus microplus* (Sutherst *et al.*, 1995), should the tick continue to spread from its present sites in the southern and eastern parts of the continent. Tanzania has already reported on the spread of *Boophilus microplus* (Lynen, 2001, personal communication), and it is known that large areas in neighbouring Kenya are suitable for tick survival (Sutherst and Maywald, 1986). The cattle in East Africa have had little exposure to *Babesia bovis*, and if *Boophilus microplus* becomes established, the cost to the cattle industry could be substantial. The present study illustrates that there is a strong possibility that *Boophilus microplus* can easily spread into new areas of Africa.

6. 1. 2. The association between the Ecoclimatic Indices, tick numbers and *Babesia* serology recorded at each dip tank/farm.

By comparing the tick collections, *Babesia* serology and the EI at each locality, the following deductions were possible.

- **Dip tanks/farms with an EI between 0-4 for *Boophilus microplus* and an EI between 1-10 for *Boophilus decoloratus*.** The cattle on these dip tanks/farms carried few *Boophilus* ticks and the seroprevalence for both *Babesia bovis* and *Babesia bigemina* was low. In these areas EI was higher for *Boophilus decoloratus* than for *Boophilus microplus*, and the presence of *Boophilus microplus* was patchy whilst *Boophilus decoloratus* was more common. The dip tanks/farms in this group included Fesekraal, Matshena, Davhana, Keerweerder, Mara, and Zwartrandjes. The cattle on these dip tanks/farms should be vaccinated or dipped intensively when the first *Boophilus* ticks appear on the cattle.

- **Dip tanks/farms with an EI between 5-10 for *Boophilus microplus*.** *Boophilus* tick numbers were higher at these locations and *Boophilus microplus* was common on the cattle. Seroprevalence of *Babesia bovis* was generally higher than for *Babesia bigemina*, and at some dip tanks/farms the herds were approaching endemic stability to *Babesia bovis*. In case of a prolonged drought the seesaw effect would be felt in these locations. The herds at these dip tanks/farms may reach endemic stability simply by not dipping too often and by making sure that there are always ticks on the calves and the young cattle. The farmers should be informed about possible changes of the *Boophilus* species after a prolonged dry spell.

- **Dip tanks/farms with an EI between 11-23 for *Boophilus microplus*.** The cattle at these dip tanks/farms carried large numbers of *Boophilus microplus* ticks and would require periodic dipping to avoid tick worry. Some of the herds at these dip tanks/farms may reach endemic stability to TBD by simply not being dipped too often. The high EI makes it unlikely that the cattle will ever be free of *Boophilus microplus*.

Boophilus decoloratus was still present on the cattle at some of these dip tank/farms, but it either disappeared or the numbers were greatly reduced during the survey.

6. 1. 3. Recommendations.

Communal dip tanks with low tick numbers. Endemic stability to TBD would probably not be attained at communal dip tanks where very low tick numbers were recorded. There will never be enough ticks at these dip tanks to infect all the young cattle and make the herd endemically stable. The majority of the cattle in these herds will be susceptible to TBD. If they become infected as adults, after ticks multiply under favourable conditions, outbreaks of disease may occur. Farmers at these dip tanks should vaccinate their cattle or at least be prepared to dip when *Boophilus* ticks appear. In our survey this group included Matshena, Fesekraal 1 and Keerweerder. Communal farmers in the drier areas in the northern and western part of Venda must also be prepared for the possible incursion of *Boophilus microplus* during wet cycles.

Communal dip tanks with large tick numbers. At these dip tanks there should be sufficient ticks on the cattle to achieve endemic stability to *Babesia bovis*. The young cattle would need 20-40 *Boophilus* tick bites daily to infect them with babesiosis. Care should be taken not to dip cattle under nine months of age too frequently, as a well-designed dipping schedule would enhance their natural immunity to *Babesia*. At most of these dip tanks *Boophilus microplus* was common and the cattle herds at some of these dip tanks had reached or were close to endemic stability to *Babesia bovis*.

In most of the communal areas where *Boophilus microplus* was absent but where *Boophilus decoloratus* was common, endemic stability to *Babesia bigemina* was the norm. If *Boophilus microplus* invades these areas, the farmers will probably experience outbreaks of Asiatic redwater and intensive dipping will be necessary to control *Boophilus microplus*. This change in dipping routines may decrease the number of cattle that were seropositive to *Babesia bigemina* and this would disturb the endemic stability for this disease.

Some of the dip tanks had already reached endemic stability to *Babesia bovis*. This was sometimes not the case with *Babesia bigemina*, however, and breakdown of dipping could result in clinical disease from *Babesia bigemina* until endemic stability eventually would be reached.

It would appear that endemic stability to bovine babesiosis could easily be lost by one or two seasons of poor transmission of both *Babesia* species. Climatic changes or any changes in the dipping schedules could lead to lower transmission of either *Babesia* species, endemic instability and outbreaks of clinical disease. Some of the dip tanks were located in areas with few *Boophilus* ticks, so the young cattle would not get enough tick bites to achieve endemic stability. These herds should be vaccinated against TBD. Most of the communal herds in this survey should attempt to reach endemic stability to TBD, however, and this could be achieved through minimal dipping and other integrated control strategies. Vaccination against TBD would be too costly for most communal farmers (Norval, 1982), as the cattle would probably have to be dipped anyway to prevent severe tick worry.

Commercial farms. With good management it should be possible for the commercial farmers to achieve a minimal disease situation for *Babesia bovis* and *Babesia bigemina* in their herds. There is a strong correlation between host density and the risk of getting infected with *Babesia* species (Solorio-Rivera *et al*., 1999) and the host density is lower on the commercial farms than in the communally grazed areas. Regular dipping, pasture spelling and vaccination may be economically feasible for the commercial farmers.