# INTERACTIONS BETWEEN TICKS AND DOGS IN THE GREATER BLOEMFONTEIN

by

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# CHAPTER 1

## GENERAL INTRODUCTION

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## BACKGROUND

Since the beginning of mankind, animals have played a major role in human relationships (Bergler 1988). Animals have always been an essential part of our history, culture, and existence (Rowan 1988). Domestic animals have been a source of help and support to man in hunting, protection, stimulation and delight (Bergler 1988). There are large differences between different peoples and different cultures and thus also in the type of animals kept, their numbers and the attitudes towards them. Man's self image, socially (cultural, religious and economic) is reflected in the significance he attaches to his animals (Bergler 1988).

Dogs play an important role in the everyday life of man. They perform numerous tasks like protecting property and cattle, hunting and even serve as draught animals in some cultures (Bergler 1988). They also perform a deeper role by fulfilling psychological needs in being a playmate and companion for both children and adults alike. Pets, but especially dogs, are even used extensively by clinical psychologists as aids to therapy (Bergler 1988). As companions, dogs present a great deal. They offer a safe outlet for human needs for contact with another warm being and may also satisfy the needs for intimacy (Rowan 1988). Hence, any form of suffering that the dog experiences affects the owner as well. In this regard parasites are among the most notorius irritants for both pets and their owners.

Two of the tick species that commonly infest dogs in South Africa (*Rhipicephalus sanguineus* and *Haemaphysalis leachi*) are also important vectors of disease to man and dog. Both of these ticks are known vectors of spotted fever group rickettsioses (Kelly and Mason 1991), Q fever (Howell, Walker and Nevill 1983) and *R. sanguineus* can also transmit *Ehrlichia chaffeensis* the causative agent of human ehrlichiosis (Dumler and Bakken 1995). As far as dogs are concerned *R. sanguineus* are considered the main vector of *Ehrlichia canis* (Van Heerden 1992) and *H. leachi* the main vector of *Babesia canis* (Horak 1995). Except for the transmission of pathogens some ticks can also affect the physical condition of dogs. This is because they are capable of causing a variety of afflictions such as pain and irritation due to the bite, inflammation and secondary bacterial infections at the feeding site, tick toxicosis and tick bite allergic reactions (Sonenshine 1991).

According to Horak (1995) the species composition of ticks collected from dogs is a reflection of the environments in which the dogs are kept. Dogs confined to kennels, houses or small gardens are those likely to be infected with *R. sanguineus*, whereas dogs from large suburban properties, peri-urban small holdings and from farms are frequently infested with *H. leachi*. A three-host ixodid tick may spend more than 80% of its life off the host (Norval 1977). During this period the detached engorged ticks need a suitable microhabitat for oviposition and incubation or moulting. The off-host microhabitat choice of detached ticks must be such that it increases the probability that the following instar will find a suitable host (Belozerov 1982). According to Chilton and Bull (1993) selection of an off-host refuge site may involve a compromise between two alternative behaviours, namely, avoidance of desiccation or predation and that directed towards host detection.

The tick infestation patterns of dogs in the Free State are unknown and in-depth studies on the biology of the main tick species (*R. sanguineus* and *H. leachi*), which infest dogs in South Africa, is also sadly lacking. In view of the close relationship between man and dog and the fact that the aforementioned ticks and others can transmit diseases to man and dogs, a study on dogs in the greater Bloemfontein area was initiated with the following broad objectives:

- To determine the diversity and seasonal dynamics of the most dominant tick species infecting dogs in selected localities in the greater Bloemfontein area.
- To investigate aspects (oviposition, longevity and moulting) of the biology of *R. sanguineus* and *H. leachi* under both laboratory and natural conditions.
- To investigate the attitudes of dog owners with regard to tick infestations of their dogs.
- To investigate the general physical condition of the dogs in the selected study localities.
- To investigate the seroprevalence of tick transmitted diseases in the selected study localities

# STUDY AREA

The present study was conducted from February 1995 to October 1996 in the Free State Province of South Africa. Initially, sampling took place in only three localities, namely, East End, Brandwag and Batho which are all suburbs of Bloemfontein (29°07′;26°12′). These localities differ from each other in various aspects. Perhaps the most important would be the differences in culture, animal husbandry, development and socio-economic standing. From November 1995, three new localities were introduced, namely, Heidedal (a suburb of Bloemfontein), Botshabelo (29°14′; 26°42′) and Thaba Nchu (29°12′; 26°50′) located 55 and 65km, respectively, east of Bloemfontein. All of these localities together form what is commonly known as the Bloemfontein-Botshabelo-Thaba Nchu region (BBT region; Fig. 1.1).

During the "Apartheid era" the government's policies were designed to prevent an influx of "non-whites" into the so-called "white" areas. In fact, strict control measures imposed racial segregation (Krige 1995). The immediate relevance that this fact has on the present study only becomes clear on close scrutiny. As a direct result of the racial segregation of the past, the BBT region can be regarded as being one of "Apartheid's" creations (Krige 1996). This has led to a "clustering" of groups of people of common colour, economic status, and very importantly, similar culture. Both Botshabelo and Thaba Nchu are classified as resource poor environments. The above-mentioned localities were chosen to represent an adequate spectrum of different socio-economic standings, geographic distribution and demographic profiles of the greater Bloemfontein area.

*DESCRIPTION OF SAMPLING LOCALITIES*

The word locality refers to a sampling site or group of sites Three areas were used in the present study, namely, Bloemfontein, Botshabelo and Thaba Nchu. In order to achieve structured sampling these areas were further divided on the basis of inherent differences. Bloemfontein was sub-divided into Brandwag, Universitas, Willows, East End (SPCA), Heidedal and Batho. Botshabelo is sub-divided into 19 zones, with the oldest zones in the centre and the youngest on the outskirts. These zones represent different sections or blocks within Botshabelo, a few of which were used as sampling divisions Thaba Nchu was sub-divided into Motlatla, Mokwena and Seloshesha. It should be noted that each sub-division consisted of several individually owned properties. All the sites were not necessarily used in every part of the present study, e.g. Batho was only used for determining diversity, prevalence, relative density and seasonal dynamics. These sampling sites or localities differed from each other in terms of socio-economic levels, infrastructure (or levels of development) and cultural background. These differences, in turn, influenced the way in which animals are treated and hence the welfare of those animals. The distinctions that were made were based largely on facts like municipal boundaries and geographical position, but also partly on personal perceptions and therefore contain a degree of subjectivity

When considering the degree of development (or infrastructure) factors like tarred roads, veterinarian clinics, shopping centres, number of open plots (yeld) and availability of transport were considered In this regard Bloemfontein was regarded as being the most developed, followed by Thaba Nchu and then Botshabelo. Botshabelo is grossly overcrowded (Lye and Murray 1980) and 60% of the occupants are unemployed

(Moniez and Monson 1994). The area surrounding Thaba Nchu (the peri-urban areas in which sampling was done) is similar to Botshabelo.

In Botshabelo and Thaba Nchu there is evidence of poor town planning and housing consists mainly of shacks, huts and very few brick homes. There are no water-borne sewage facilities and people rely on street taps for drinking water. The roads are very poor and the telephone service is totally inadequate (Moniez and Monson 1994). To put it mildly, both the Botshabelo and Thaba Nchu areas represent the other end of the spectrum when it comes to development.

Another important fact is that they also represent a completely different culture, which formed an important facet of the present study. Hunting forms part of their lifestyles and the use of spears, bow and arrow, traps, pitfalls and dogs are not uncommon (De Wet 1986). Cattle, which also form an important part of the culture, are allowed to roam freely because there are no fenced-in grazing camps. They also share a common grazing area. Theft of cattle is common and therefore a member of the family accompanies cattle at all times during grazing periods (Jeppe 1980). This and the fact that dogs are used for hunting is important with regard to the present study because the dogs usually accompany the person looking after the cattle and thus spend a lot of time in the open veld.

This was taken a step further by looking at the degree of urbanisation. Bloemfontein, which is urban, is classified as a city because it meets the set criteria. Botshabelo and Thaba Nchu are not cities but are classified as urban settlements (Krige 1988). For the purpose of the present study, the whole of Bloemfontein and Seloshesha were regarded as being "urban: formal". Botshabelo, Mokwena and Motlatla were regarded as being "urban: informal" Both these categories are urban because they fall within the municipal boundaries but the reason for the distinction was that the "urban: informal" has a rural element. The "urban: informal" areas present an overlap between the rural and urban way of life. Thus it can be said that, although urban, the activities practised in these localities are rural-like because of the presence of cattle and the practices associated with it. The differences in the daily activities of the "urban: formal" and the "urban: informal" areas were the reasons for distinguishing between them. These differences can impact on the attitudes towards dogs because the reasons for keeping a dog could be different.

When considering the socio-economic levels. factors like the type of home (cost thereof) and type (and number) of vehicles available was looked at. The political history of South Africa resulted in a group distinction based on socio-economic as well as cultural differences. It was accepted that there were exceptions but that generally the previously white localities were regarded as being on a higher socio-economic level (the affluent localities). In this regard, Brandwag, Willows and Universitas were the highest, followed by Heidedal, then Thaba Nchu, Batho and lastly Botshabelo. When considering the cultural differences, Brandwag, Willows and Universitas were grouped together. Heidedal, Botshabelo and Thaba Nchu were each regarded as a group on their own. To prevent repetition Batho. although very different to the Brandwag grouping and very similar to Botshabelo, was not included in this part of the study. By looking at Botshabelo and the criteria used for this part of the present study, the category Batho represented was covered by that of Botshabelo.

East End is a single locality situated very near to Heidedal. Initially it was used as a tick source and in terms of socio-economic differences and cultural differences, it was difficult to group it with the other localities. However, it should be noted that the SPCA is surrounded by veld and that there are other resident animals on site besides dogs These include pigs, horses and stray cats.

The locality selection described in Chapter 2 was used for the sole purpose of covering a large enough area to be representative of the greater Bloemfontein. In Chapter 6 it is explained how cultural and socio-economic differences were considered when choosing the localities. In Chapter 7 different levels of urbanisation, as described above were used as a distinguishing factor in choosing the localities. For the chapters that dealt with laboratory investigations, most ticks were collected from East End (SPCA).

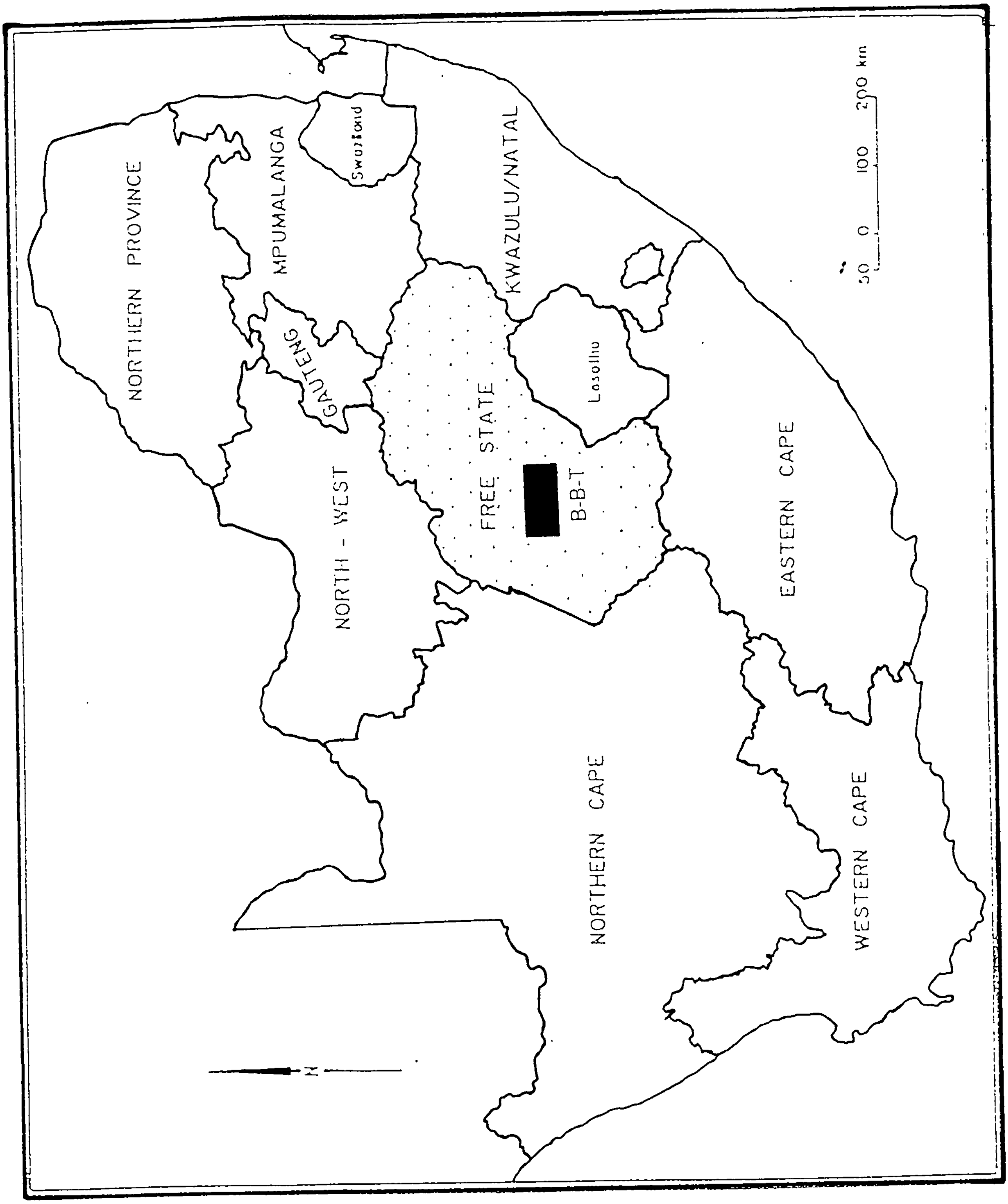


Figure 1.1 Map of South Africa (adapted from Krige 1995). The Free State Province is indicated by [dotted box] and the Bloemfontein-Botshabelo-Thaba Nchu Region is indicated by [black box].

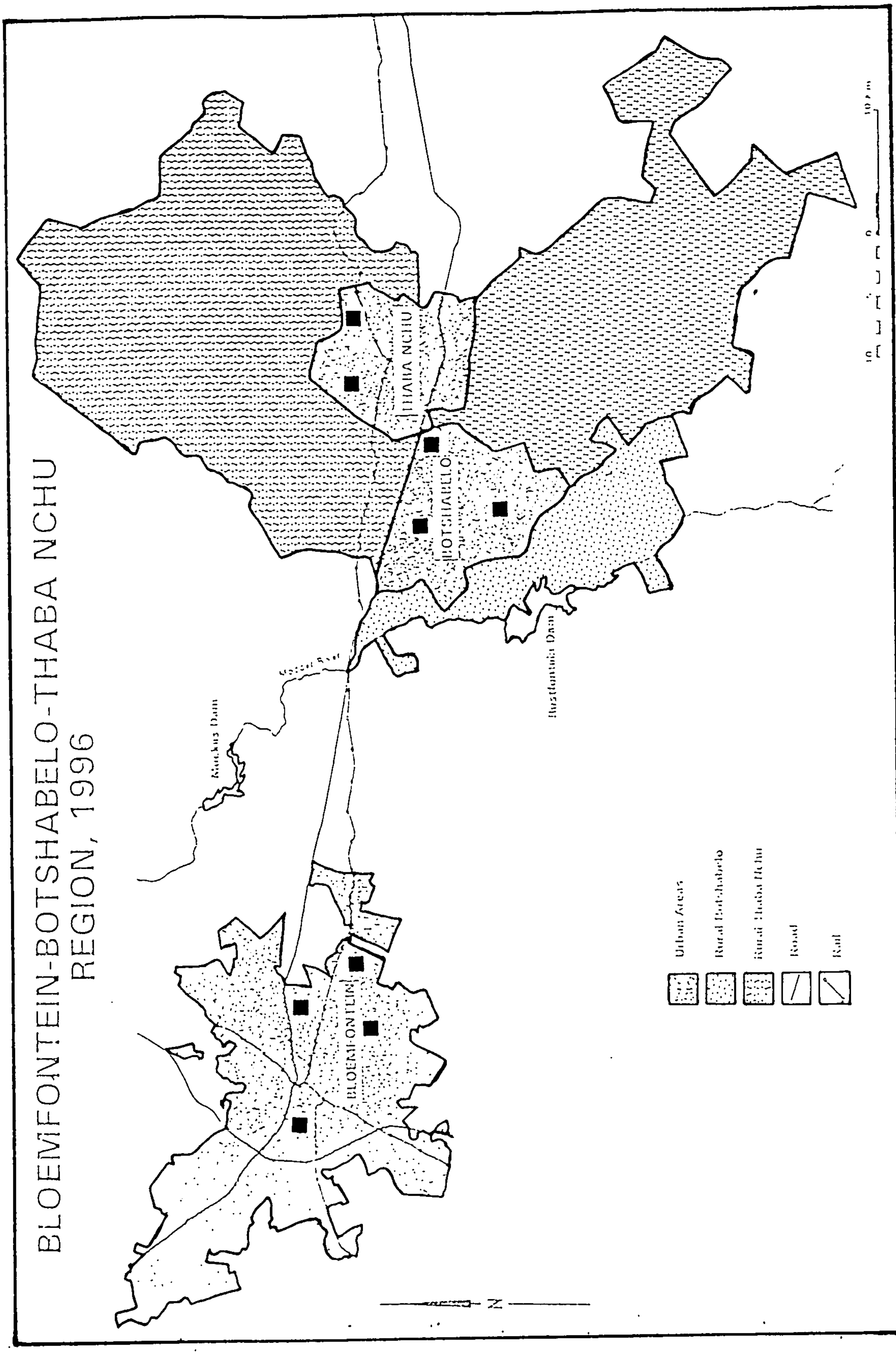


Figure 1.2 The Bloemfontein-Botshabelo-Thaba Nchu Region (adapted from Krige 1995). ■ indicates the sampling sites or localities.

# CLIMATE

The climate of the study area, to a large extent, depends on wind patterns, which bring rain from the east (i.e the Indian Ocean) during the summer months. The Drakensberg Mountains cause these winds to rise abruptly and hence to disgorge before they enter the plateau on the other side. What is left of the moisture will precipitate in progressively smaller amounts as they move to the west (Lye and Murray 1980).

*ATMOSPHERIC TEMPERATURES*

Summer temperatures in the central Free State Region are moderate to warm and winters are cold with frost occurring from April to the end of September (Mostert, Roberts, Heslinga and Coetzee 1971). The mean maximum and minimum atmospheric temperatures, measured at the Bloemfontein Airport meteorological station, are shown in Fig. 1.3a.

*RAINFALL*

The central Free State is a summer rainfall area and thunderstorms are common for 65.6 days of the year (De Wet 1986; Mostert 1958). Rainfall decreases from east to west across the Free State region and it is unpredictable, varying in incidence from year to year. Over 80% of the annual rains occur between the months of October and April (de Waal 1990; Mostert 1958). The average annual rainfall for the central Free State is from 650mm in the east to 450mm in the west (Mostert, Roberts, Heslinga and Coetzee 1971). The mean monthly rainfall (1930-1995) is shown in Fig. 1.3(b). In general the highest rainfall is recorded during February and the lowest during June. The study area is also characterised by the occurrence of periodic droughts (Mostert 1958). This and the fact that the temperature fluctuations during winter are extreme are of great biological importance (Mostert 1958).

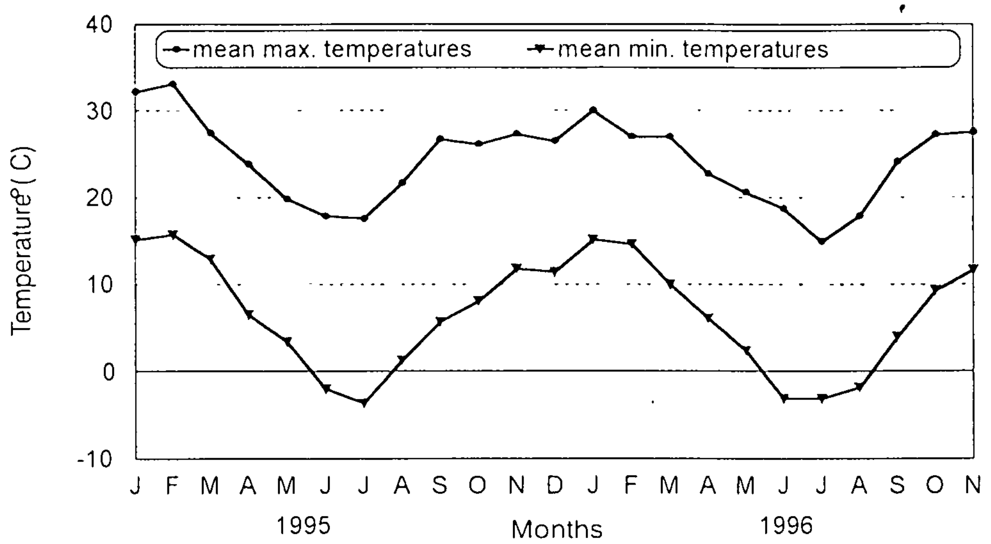


**Figure 1.3(a).** The mean monthly minimum ( —▾—) and maximum ( —•—) atmospheric temperatures in the Bloemfontein area for the period from January 1995 to November 1996.

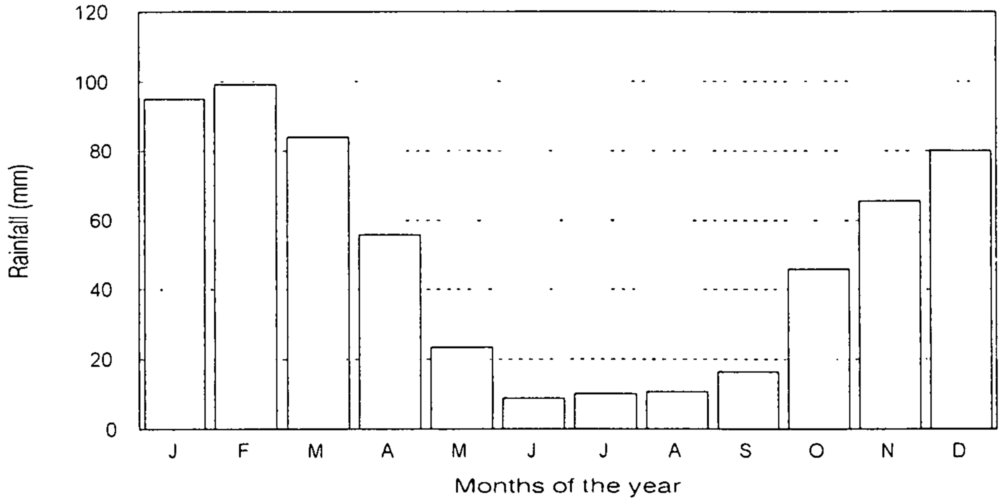


**Figure 1.3(b).** Bar graph showing the mean monthly rainfall for the Bloemfontein area (1930 to 1995).

# TOPOGRAPHY AND ALTITUDE

The larger part of the central Free State is rather flat and lies between 1200m and 1500m above sea level, the eastern area being higher above sea level (1200 – 1800m) Botshabelo and Thaba Nchu are situated at this higher, eastern altitude The whole area is drained by the Modder River and its tributaries (Mostert 1958).

# VEGETATION

The veld-type in the Bloemfontein area is classified as Dry *Cymbopogon-Themeda* and Botshabelo and Thaba Nchu as transitional *Cymbopogon-Themeda* veld (Acocks 1988). Large bushes and trees are not characteristic of this area (De Wet 1986; Mostert, Roberts, Heslinga and Coetzee 1971; Mostert 1958).

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# CHAPTER 2

## TICK DIVERSITY, SEASONALITY AND SITES OF ATTACHMENT

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## INTRODUCTION

Surveys on the tick infestations of dogs have been conducted in various places in South Africa. These studies have shown that dogs in South Africa are parasitised by at least 15 different tick species (Horak 1982; Theiler 1962; Howell, Walker and Nevill 1983). Three species, however, commonly infest dogs in South Africa, namely, *Rhipicephalus sanguineus*, *Rhipicephalus simus* and *Haemaphysalis leachi* (Horak, Guillarmod, Moolman and De Vos 1987). Very little is known about the ticks infesting dogs in the Free State Province of South Africa and nothing is known about the ticks that infest dogs in the greater Bloemfontein area. This type of information is important in terms of zoonosis and disease transmission. Likewise, data on the seasonal occurrence of ticks are important in terms of planning control strategies.

For many years man has been trying to control tick infestations, with limited success due to the tick's versatility, resilience and resourcefulness (Fourie and Kok 1992). Previous attempts focussed on chemical control (Matthewson 1984) but because of the development of resistance to chemicals a new approach has received attention, namely, integrated pest management (Fourie and Kok 1992). Knowledge of the tick's bionomics is essential for this to be effective in that it serves to highlight any parts in the life cycle of the tick that may be susceptible to control strategies (Fourie and Kok 1992). Determining the attachment sites of these tick species has, for example, enormous implications on control especially with regard to the use of "spot on" acaracidal treatment. This is further complicated by specific factors which influence the attachment sites which a particular ticks species prefers (Fourie and Van Zyl 1991).

Very little is known about the seasonal prevalence of *R. sanguineus* in the southern hemisphere (Horak 1982) and in South Africa it has been determined in only a few areas such as the Gauteng region. The seasonal prevalence of *H. leachi* is unknown (Horak 1982).

The objectives of the present study were:

- To investigate the diversity, prevalence and relative density of the ticks infesting dogs in the greater Bloemfontein area.
- To examine the seasonal dynamics of ticks in this area.
- To examine the differences in the sites of attachment of the different tick species.

# MATERIALS AND METHODS

*STUDY LOCALITIES*

During February 1995 sampling was conducted in three localities, namely, the SPCA (East End), Brandwag and Batho. In November 1995, the project was expanded to include additional sampling areas. These areas were Heidedal, Botshabelo and Thaba Nchu. Sampling in these areas was terminated in October 1996. These areas are described in Chapter 1.

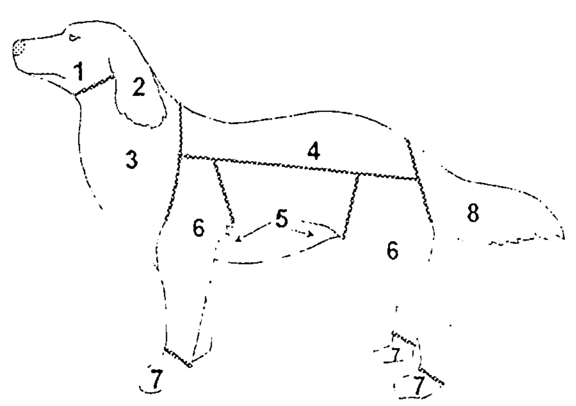
*TICK DIVERSITY, PREVALENCE, RELATIVE DENSITY AND SEASONAL DYNAMICS*

Within each locality three long (hair length > 4cm) and three short haired dogs were selected (different breeds and sex). For the sake of consistency the same dogs were used each month, which was easily done because the dogs used, belonged to individuals at fixed addresses, except for East End (at the SPCA). Care was taken to arrange with the owners not to treat their dogs for the duration of the present study so as not to affect the results. In East End (at the SPCA) the sampling was done in the same set of kennels. At the SPCA new arrivals were dipped only on arrival and not sampled until ticks started attaching again. Dogs are brought to the SPCA from all over the greater Bloemfontein area and are often heavily infested with ticks. This gives an inaccurate indication of the tick burden at the SPCA and is the reason for dipping the dogs on arrival.

During each sampling occasion all ticks were removed from the dogs by means of thorough palpation of the dog's entire pelage. Ticks were removed according to the recommendations made by Needham (1985). According to Needham (1985) a tick should be grasped as close as possible to the skin of the host with a blunt, curved forceps. The tick should subsequently be pulled straight upward so as not to break off the mouths parts, which are often crucial in the identification of ticks. This method was followed in the present study, the only exception being that a blunt straight forceps was used which was found to be more effective when angled close to the skin of the host. The ticks that were removed were placed in labelled vials and fixed in 70% ethanol. The labels included the date, locality, size of dog, dog/kennel number and any other relevant information. In the laboratory the ticks were identified to species level with the aid of a stereo-microscope and quantified. The data obtained from this was used to determine the diversity, prevalence, relative density and seasonal dynamics of the most prominent species. The prevalence was determined by dividing the number of dogs infected with a particular tick species by the number of dogs sampled. This was expressed as a percentage (Kassai 1999). The relative density was determined by dividing the total number of individuals of a particular tick species by the total number of individual dogs sampled (Kassai 1999).

*ATTACHMENT SITES AND HAIR LENGTH*

At the SPCA (East End), a distinction was made between dogs with long and short hair and also between the different body regions on which the ticks attached. Twelve dogs, six of which had long and six, which had short hair, were examined fortnightly. The 12 dogs included the dogs that were used to determine the seasonal occurrence of the ticks. Sampling took place from April 1995 to December 1995 and thus included summer and winter months. The body of the dog was divided into eight different regions, namely; head, ears, neck, back, abdomen, legs, toes, and tail (Fig. 2.1). Collection of ticks was done as described above. The ticks collected from each of the eight body regions were placed in separately labelled bottles for later identification and quantification. Although all tick developmental stages found were sampled, only the adults were used in the final analysis.




1. Head
2. Ears
3. Neck
4. Back
5. Abdomen
6. Legs
7. Feet
8. Tail

**Figure 2.1.** Body regions on which ticks were collected.

In the beginning of 1996 the "sites of attachment" survey was terminated because the number of dogs sampled were deemed adequate. Sampling at the SPCA (East End) was, however, continued in an attempt to reinforce the results obtained in 1995.

# RESULTS

*TICK DIVERSITY, PREVALENCE AND RELATIVE DENSITY*

A total of 21 636 adult ticks representing nine different species were collected during the present study. Only two species, however, dominated in all the localities, namely, *Rhipicephalus sanguineus* and to a lesser extent *Haemaphysalis leachi*. The prevalence of *R. sanguineus* was much higher than that of *H. leachi*. A total of 73.5% and 22.4% of all the dogs sampled were infested by *R. sanguineus* and *H. leachi*, respectively (Table 2.1). This difference is reinforced by the relative density difference. The relative density of *R. sanguineus* and *H. leachi* was 27.4 and 5.8, respectively. The number of dogs infested by other ticks constituted less than 2%. The seven other species found constituted less than 1.2% of the total sample and included *Boophilus decoloratus*, *Hyalomma truncatum*, *Rhipicephalus evertsi evertsi*, *Ixodes rubicundus*, *Rhipicephalus* sp., *Rhipicephalus gertrudae* and *Rhipicephalus follis* (Table 2.1, Fig. 2.2). Of the "less than 1.2%" group, *Boophilus decoloratus* and *Rhipicephalus gertrudae* were the dominant species. Except for the above mentioned adult ticks *Amblyomma marmoreum* and *Otobius megnini* nymphs were occasionally collected from dogs.

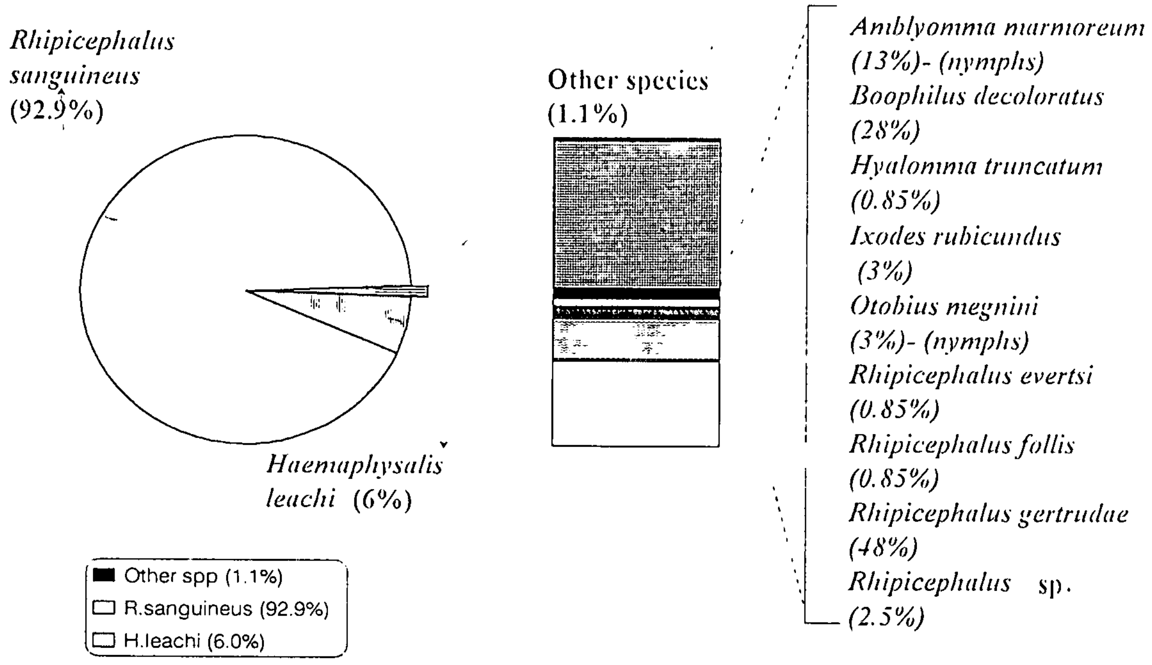



**Figure 2.2.** Pie diagram showing the species diversity of the ticks collected from dogs in the greater Bloemfontein area.

**Table 2.1.** Adult ticks collected from dogs from various localities in the greater Bloemfontein area

| *Tick species* | *East End* | | *Brandwag* | | *Batho* | | *Botshabelo* | | *Thaba Nchu* | | *Heidedal* | | *Total* | *Relative density* | *% dogs infested (Prevalence)* |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | *F* | *M* | *F* | *M* | *F* | *M* | *F* | *M* | *F* | *M* | *F* | *M* | | | |
| *Boophilus decoloratus* | 2 | 1 | 0 | 0 | 0 | 1 | 21 | 10 | 23 | 7 | 0 | 0 | 65 | 4.6 | 1.4 |
| *Haemaphysalis leachi* | 239 | 318 | 12 | 2 | 6 | 0 | 105 | 236 | 126 | 179 | 17 | 50 | 1290 | 5.8 | 22.4 |
| *Hyalomma truncatum* | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 2 | 2 | 0.1 |
| *Ixodes rubicundus* | 0 | 0 | 0 | 0 | 0 | 7 | 0 | 0 | 0 | 0 | 0 | 0 | 7 | 7 | 0.1 |
| *Rhipicephalus evertsi* | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 2 | 2 | 0.1 |
| *Rhipicephalus follis* | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 1 | 0 | 0 | 0 | 0 | 2 | 1 | 0.2 |
| *Rhipicephalus gertrudae* | 0 | 0 | 0 | 0 | 0 | 0 | 52 | 60 | 0 | 0 | 0 | 0 | 112 | 9.3 | 1.2 |
| *Rhipicephalus sp.* | 0 | 0 | 0 | 0 | 1 | 5 | 0 | 0 | 0 | 0 | 0 | 0 | 6 | 2 | 0.3 |
| *Rhipicephalus sanguineus* | 1222 | 2243 | 123 | 219 | 1090 | 1260 | 1805 | 2842 | 3186 | 5012 | 491 | 657 | 20150 | 27.4 | 73.5 |
| TOTAL | 1467 | 2562 | 135 | 221 | 1097 | 1273 | 1984 | 3149 | 3335 | 5198 | 508 | 707 | 21636 | | |

The occurrence of ticks within the different study localities is summarised in Table 2.2, which indicates which species were found and more importantly, where they were found *R. sanguineus* and *H. leachi* were found in all the localities and *B. decoloratus* and *R. gertrudae* were found in Batho, the SPCA (East End), Botshabelo and Thaba Nchu. It should be noted that *B. decoloratus* and *H. truncatum* were found on dogs at the SPCA (East End) and furthermore, *B. decoloratus* was also found on dogs in Batho, Botshabelo and Thaba Nchu.

**Table 2.2.** Adult tick species found at different sampling localities

| Localities vs Species | SPCA (East End) | Brandwag | Heidedal | Batho | Botshabelo | Thaba Nchu |
|---|---|---|---|---|---|---|
| *Boophilus decoloratus* | + | - | - | + | + | + |
| *Haemaphysalis leachi* | + | + | + | + | + | + |
| *Hyalomma truncatum* | + | - | - | - | - | - |
| *Ixodes rubicundus* | - | - | - | + | - | + |
| *Rhipicephalus evertsi* | + | - | - | - | - | - |
| *Rhipicephalus follis* | - | - | - | - | + | - |
| *Rhipicephalus gertrudae* | - | - | - | - | + | + |
| *Rhipicephalus sp.* | - | - | - | + | - | - |
| *Rhipicephalus sanguineus* | + | + | + | + | + | + |

(+) = species present; (-) = species absent

The average number of ticks per dog found at each locality for the duration of the present study is presented graphically in Fig. 2.3. *H. leachi* was present in all six localities but the numbers were very low varying from a mean of 0.07 in Batho to 2.21 in Botshabelo. Most *R. sanguineus* ticks ($\bar{x}$ = 48.82) were collected from dogs in Thaba Nchu followed by Botshabelo ($\bar{x}$ = 30.18). The least number ($\bar{x}$ = 5.59) was collected from dogs in Brandwag. At all the localities less *H. leachi* compared to *R. sanguineus* ticks were collected.

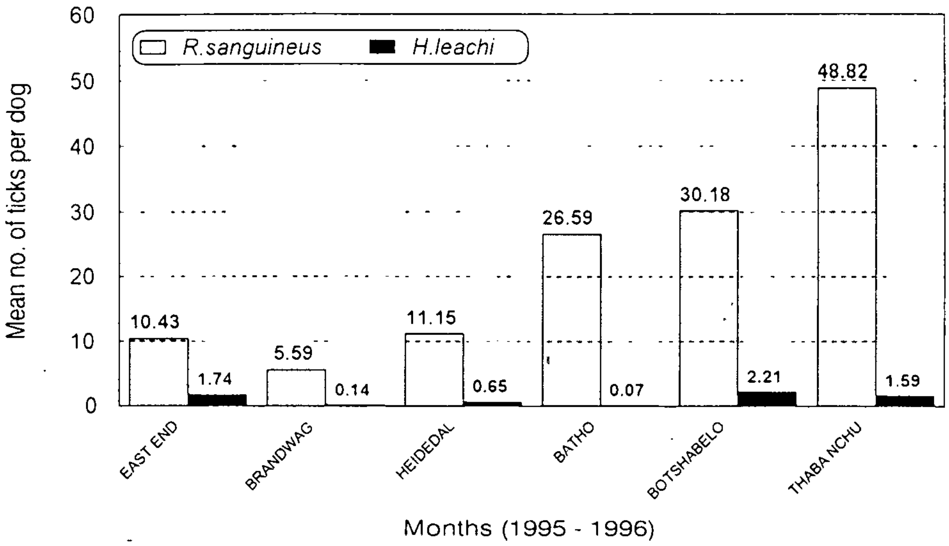



**Figure 2.3.** The mean number of *Rhipicephalus sanguineus* and *Haemaphysalis leachi* ticks collected per dog in the different localities.

*SEASONAL DYNAMICS*

Except for *R. sanguineus* and *H. leachi* the other tick species' numbers were too small to determine seasonal dynamics. The seasonal dynamics of these ticks in the different study localities are presented graphically in Figs 2.4a-f. In Brandwag (Fig. 2.4a) *R. sanguineus* displayed a peak during March 1995. During the winter months few or no ticks were sampled and the mean tick burdens on the dogs only started to increase during December 1996. No *H. leachi* ticks were collected from dogs except from December – February 1996 and then in low numbers.

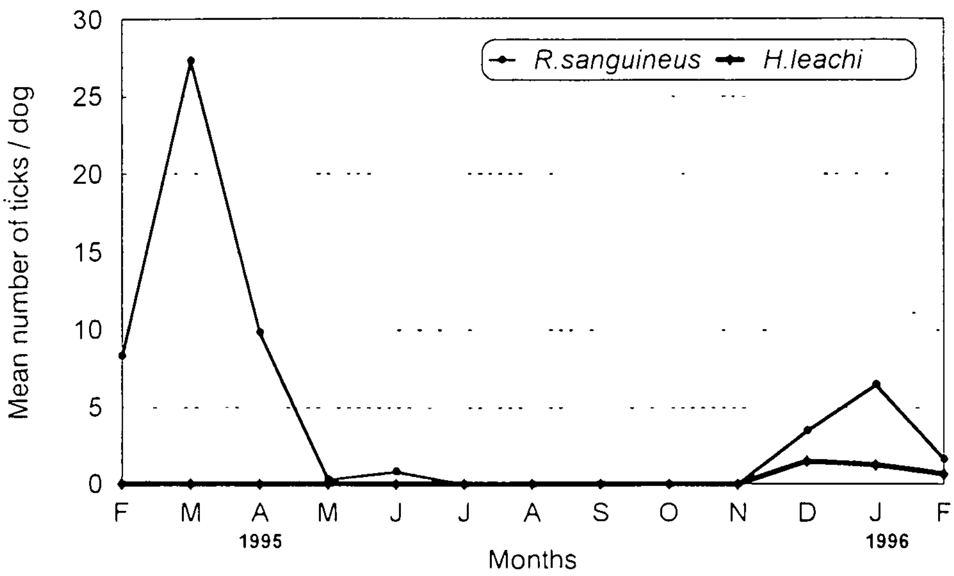



**Figure 2.4(a).** The mean monthly number of *Rhipicephalus sanguineus* and *Haemaphysalis leachi* ticks collected from dogs in Brandwag.

At the SPCA (East End) (Fig. 2.4b) *R. sanguineus* displayed a peak in March 1995 after which it steadily dropped in numbers with the onset of the winter months. Mean tick burdens again increased from July and peaked in August 1995. During 1996 the mean tick burdens were variable but the highest *R. sanguineus* tick burden was recorded during January 1996. For the duration of the study period the number of ticks collected was variable with no further distinct peaks. The number of *H. leachi* ticks that were collected remained very low for the duration of the present study with slight increases in January, July and October 1996.

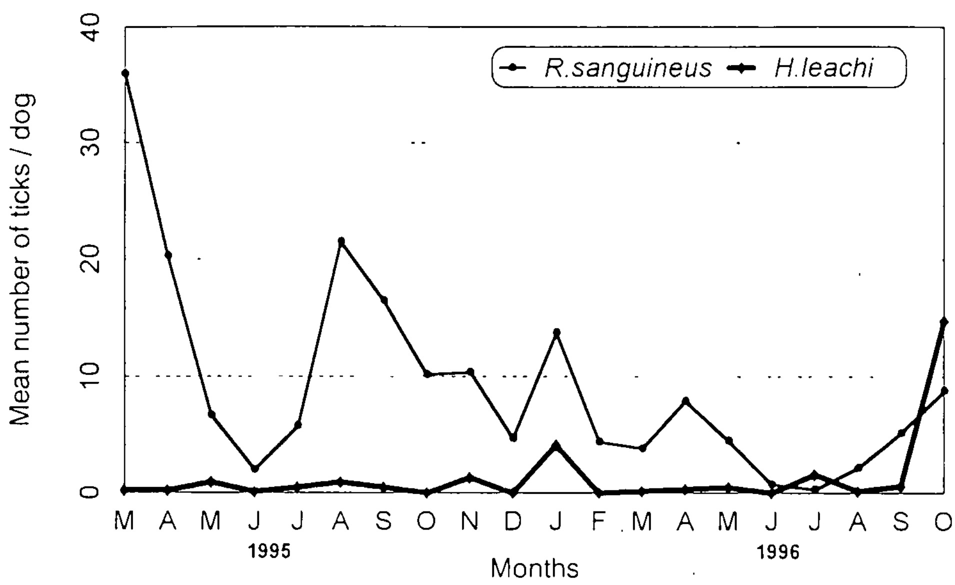



**Figure 2.4(b).** The mean monthly number of *Rhipicephalus sanguineus* and *Haemaphysalis leachi* ticks collected from dogs at the SPCA in East End.

In Heidedal (Fig 2.4c), *R. sanguineus* displayed peaks in January and March 1996. During the cold winter months the numbers were low but increased again from August to October. *H. leachi* ticks were collected only during January-February 1995 and August-October 1996.

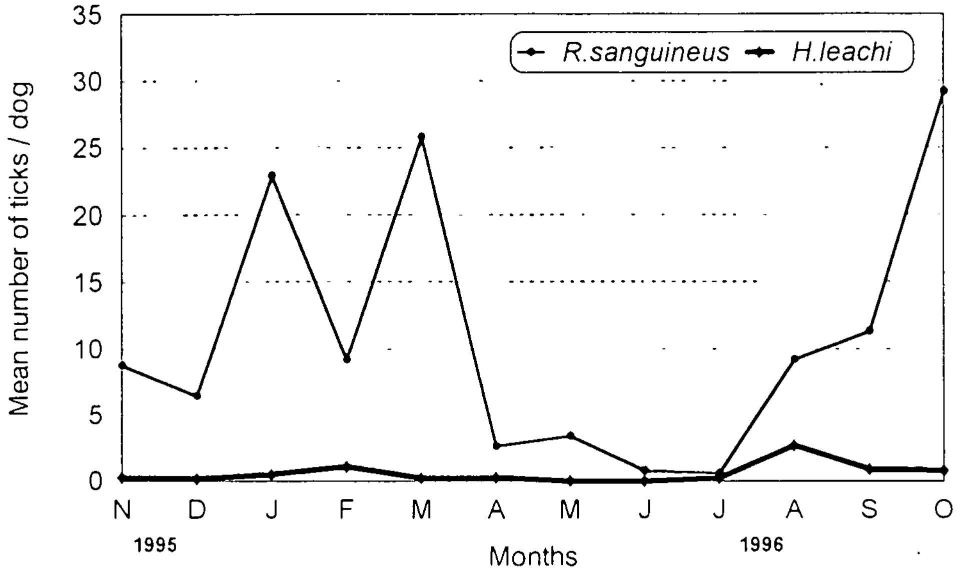



**Figure 2.4(c).** The mean monthly number of *Rhipicephalus sanguineus* and *Haemaphysalis leachi* ticks collected from dogs in Heidedal.

In Batho (Fig. 2.4d) *R. sanguineus* numbers decreased from February 1995 to reach a low during June. Tick numbers then increased again and reached a distinct peak during January 1996. No *H. leachi* ticks were collected except during August 1995.

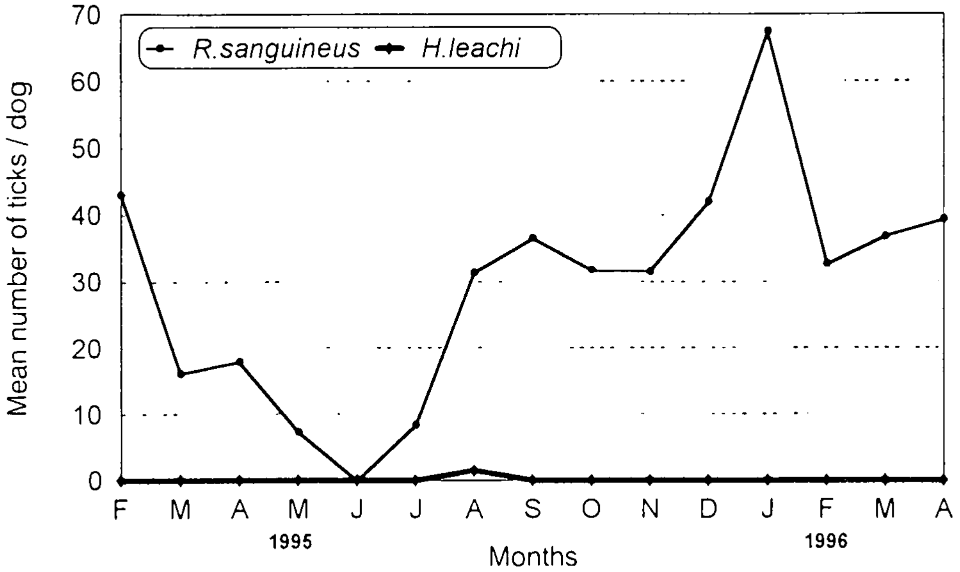



**Figure 2.4(d).** The mean monthly number of *Rhipicephalus sanguineus* and *Haemaphysalis leachi* ticks collected from dogs in Batho.

In Botshabelo (Fig. 2.4e) *R. sanguineus* displayed a distinct peak during January 1995 after which mean tick burdens declined steadily. *R. sanguineus* tick burdens started to increase from September again. No *H. leachi* ticks were collected except during February 1995 and October 1996.

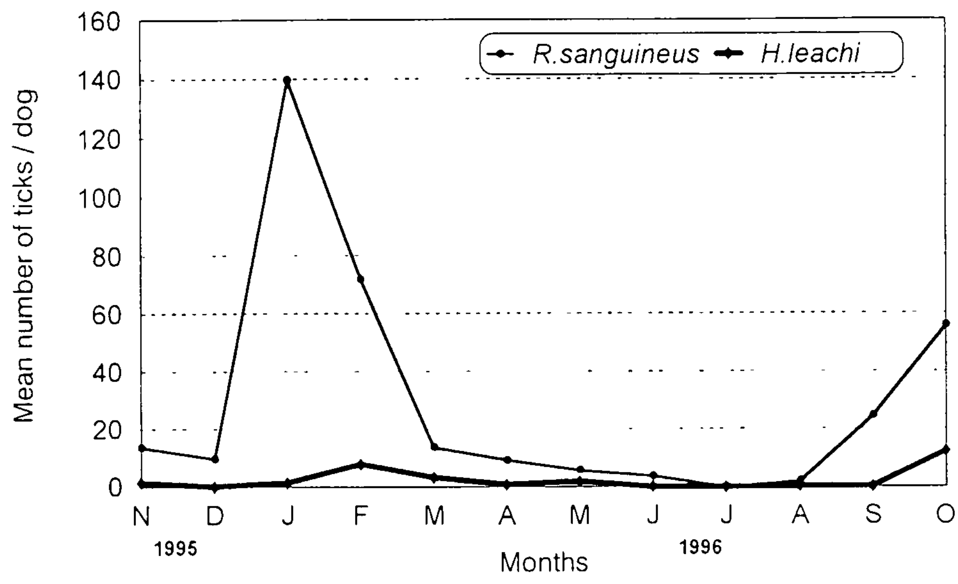



**Figure 2.4(e).** The mean monthly number of *Rhipicephalus sanguineus* and *Haemaphysalis leachi* ticks collected from dogs in Botshabelo.

In Thaba Nchu (Fig. 2.4f) *R. sanguineus* was most abundant during summer and autumn. The highest mean tick burden was recorded during January 1996. Few *R. sanguineus* ticks were collected during June – August 1996. Except during November-December 1995 and October 1996 no *H. leachi* ticks were collected.

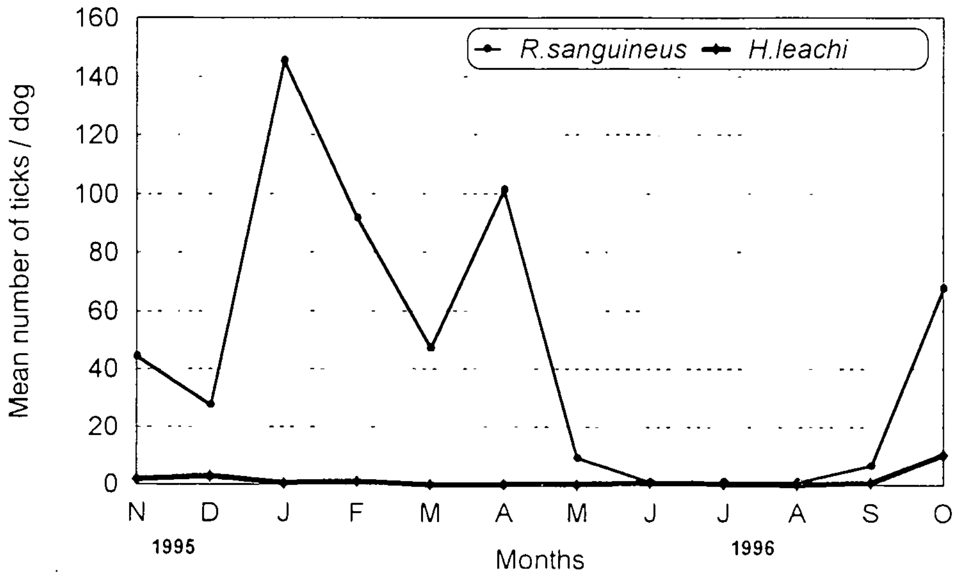



**Figure 2.4(f).** The mean monthly number of *Rhipicephalus sanguineus* and *Haemaphysalis leachi* ticks collected from dogs in Thaba Nchu.

The mean number of ticks for all the localities (data pooled) for the duration of the present study is graphically presented in Fig. 2.5. Mean minimum and maximum temperatures, obtained from the weather bureau in Bloemfontein, are superimposed on the same graph. Dogs were infested with ticks throughout the year. The highest burdens of *R. sanguineus* ticks were, however, recorded during the warm months of January and February (Fig. 2.5). *H. leachi* ticks were most abundant during early summer (October to November).

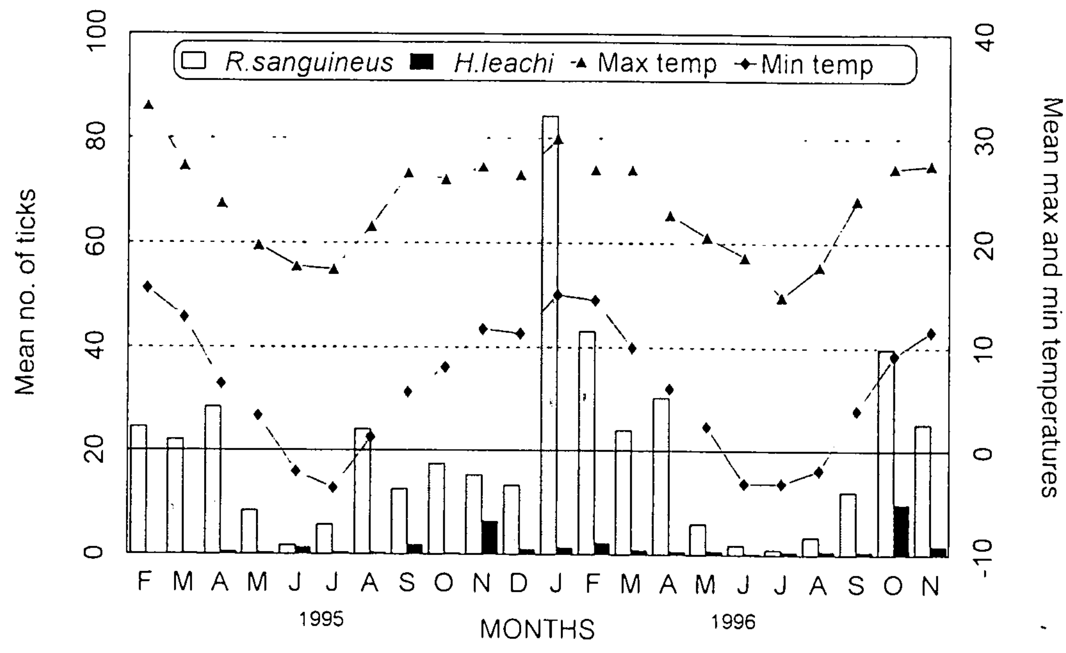



**Figure 2.5.** Seasonal occurrence of *Rhipicephalus sanguineus* and *Haemaphysalis leachi* ticks (all study areas pooled) in relation to temperature for the period from February 1995 to November 1996.

ATTACHMENT SITES AND HAIR LENGTH

This was investigated only at the SPCA (East End). Most *R. sanguineus* ticks attached on the back (29%), on the ears (19%) or on the neck (19%) of the dogs (Fig. 2.6). A significant percentage (12%) of the ticks attached on the paws of the dogs. Very few *R. sanguineus* ticks were found on the tail region (1%). In the case of *H. leachi* most of the ticks attached on the back (34%), on the neck (31%) or on the legs (22%) of the dogs. No *H. leachi* ticks were found attached to the ears, on the paws or tails of the dogs.

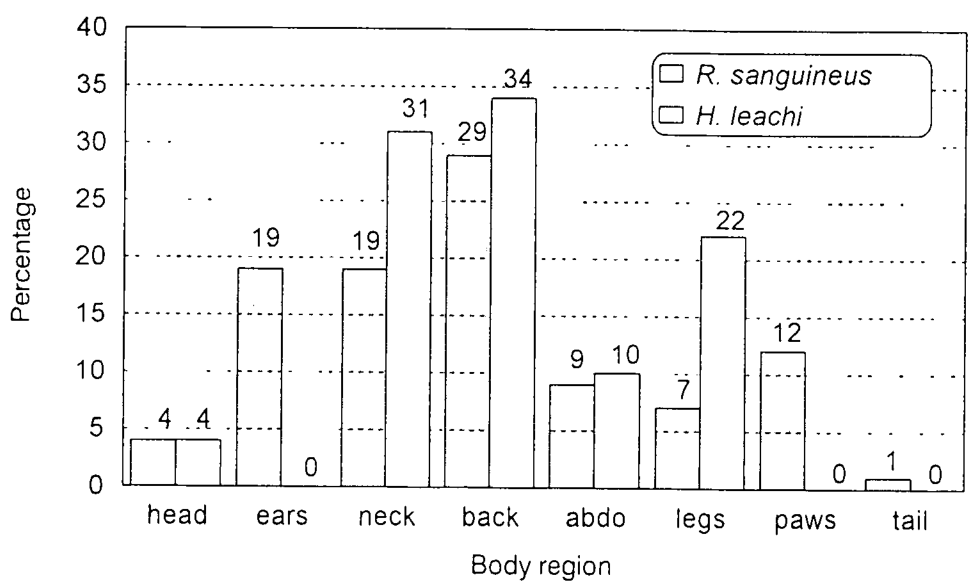



**Figure 2.6.** Percentages of *Rhipicephalus sanguineus* and *Haemaphysalis leachi* ticks attached to different regions examined.

Figure 2.7 gives a graphical presentation of the distribution of *R. sanguineus* on the body regions of dogs with long and short hair, respectively. In general the attachment of *R. sanguineus* ticks to dogs with long and short hair was fairly similar. The only conspicuous difference being that in long haired dogs 37% of the ticks attached to the back compared to the 22% on short haired dogs. Conversely, on short haired dogs 13 % of the ticks attached to the abdomen compared to the 4% on long haired dogs.

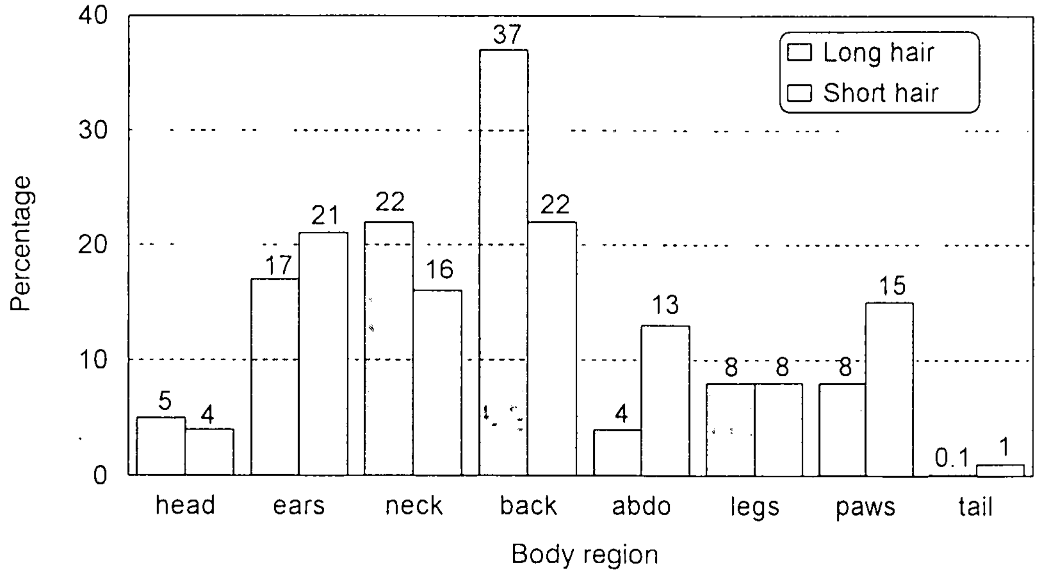



**Figure 2.7.** Percentages of *Rhipicephalus sanguineus* attached to different regions examined on long and short-haired dogs.

# DISCUSSION

*TICK DIVERSITY, PREVALENCE AND RELATIVE DENSITY*

In the present study nine different adult tick species were found to infest dogs. Results from the present study indicated that the most prevalent species infesting dogs (and the species with the greatest relative density) in the central Free State region is *R. sanguineus*, followed by *H. leachi*, to a lesser extent. This is important when considering control measures. According to the results of other surveys conducted in different regions of South Africa a total of 16 different tick species, belonging to six different genera, have been found to infest dogs. In the Grahamstown region, Horak *et al.* (1987) found that 14 different tick species infested the dogs. The most prevalent species in terms of relative density was *H. leachi* followed by *Rhipicephalus simus*. Horak (1995) also conducted a study at Onderstepoort and sampled ticks from dogs examined at the out-patients clinic and the results indicated that the most prevalent species was *H. leachi*, followed by *R. sanguineus*. At Moboloka in the North West Province, Bryson, Höhn, Horak and Kirkpatrick (1995) conducted a study, which indicated that six different tick species infest dogs in that area. The most prevalent species was *R. sanguineus* followed by *H. leachi*, to a lesser extent.

The main natural hosts of the species (other than *R. sanguineus* and *H. leachi*) sampled in the present study are not the domestic dog but rather wild and domestic ruminants (Howell, Walker and Nevill 1983). They can thus be viewed as being incidental infestations. The following is a brief discussion on the tick species infesting dogs in the present study.

*Amblyomma marmoreum*

This species is endemic to Africa and is known as the tortoise tick. It is a three-host tick (Rechav and Fielden 1995) and the adults are specific parasites of reptiles like snakes and tortoises (Walker 1991). Immature stages have been found to feed on cattle, sheep, goats, various carnivores and dogs. This was shown to be true in the present study as only nymphs were found on the dogs that were infested by this tick. It has a wide distribution in South Africa (Horak, MacIvor, Petney and De Vos 1987; Rechav and Fielden 1995; Theiler and Salisbury 1959; Walker 1991).

*Boophilus decoloratus*

*B. decoloratus*, better known as the blue tick, is a one-host tick which primarily infests cattle (Norval 1977; Walker 1991). A one-host life cycle minimises the time this tick has to spend in harsh microclimatic conditions and eliminates the need for intermediate hosts (Norval 1982). Its distribution is limited to areas where the rainfall is not less than 375mm per annum (Theiler 1969). It produces three generations per year and poses a serious threat to cattle (Dreyer, Fourie and Kok 1998). In Batho, Botshabelo and Thaba Nchu dogs may wander through areas frequented by cattle and as such becomes subject to infestation. *B. decoloratus* ticks were also sampled from dogs at the SPCA (East End) which was unsuspected. A plausible explanation for this observation is that many of the dogs at the SPCA come from resource poor environments such as Batho. Although dogs are normally dipped at the SPCA it is possible that the dip was ineffective or that some of the dogs may not have been dipped at all, thus accounting for the presence of *B. decoloratus*.

*Haemaphysalis leachi*

One of the most commonly found ticks which parasitises the domestic dog is *H. leachi*, which is also found on cattle and wild carnivores (Hoogstraal 1956). *H. leachi* is a three-host tick and its immatures infest rodents by preference (Hoogstraal 1956; Howell *et al.* 1978; Keirans 1992). Its distribution is limited to areas where the rainfall is not lower than 508mm per annum (Walker 1991). It is responsible for the transmission of *Babesia canis*, which causes babesiosis in dogs (Walker, Mehlitz and Jones 1978). The absence of rodents in a study locality can result in very low numbers, as was the case in the present study. *H leachi* is a common and widely distributed species of tick and is probably found in most regions in which a suitable host occurs (Walker 1970). Its distribution in many parts of South Africa is still unknown. There is little information about this tick in the OFS and according to Horak et al. (1987), there is still a great need for further research on their morphology and taxonomy. *H. leachi* ticks prefer the higher rainfall areas (Theiler 1962) and cannot survive in desert or semi-desert regions, except where the micro-climate is favourable (Walker 1970; Howell *et al.* 1983).

Few *H. leachi* ticks were found during the present study. Most of the *H. leachi* ticks were sampled at the SPCA (East End). The kennels are situated next to an open veld and therefore it is possible that rodents, attracted to the dog food in the kennels, may have allowed *H. leachi* nymphs to drop off. They then moult and the emerging adults are then able to infest the dogs in the kennels. This explains the presence of *H. leachi* the SPCA (East End). The fact that the dogs in these areas wander into the veld means that they are more likely to become infested with *H. leachi*. Horak *et al.* (1987) mentioned other factors which could serve as possibilities for their scarcity, for example the males being small and difficult to collect, or that the removal of the engorging females results in the absence of a source of attraction for the males. Horak *et al.* (1987), however, also state that they believe that the presence of a satisfactory rodent-carnivore relationship is probably more important than the other factors possibly involved.

### *Hyalomma truncatum*

Adults of this tick feed on a variety of domesticated and wild ungulates. They also sometimes infest dogs (Horak and MacIvor 1987). The immatures prefer the Cape hare (*Lepus capensis*) and the scrub hare (*Lepus saxatilis*) or even rodents (Theiler 1962; Horak and MacIvor 1987). Its distribution in South Africa is vast and almost throughout the country (Walker 1991) and extends throughout the continent (Linthicum, Logan, Kondig, Gordon and Bailey 1991). The tick is known to transmit Crimean-Congo haemorrhagic fever (Linthicum, Logan, Kondig, Gordon and Bailey 1991) and some strains cause sweating sickness in cattle(Howell *et al.* 1983). In dogs, large necrotic lesions develop at feeding sites. Only two *H. truncatum* ticks were sampled during the present study. Dogs occurring on farms and small holdings, however, are frequently infested by *Hyalomma* spp ticks (Fourie, personal communication).

### *Ixodes rubicundus*

This tick is known as the Karoo paralysis tick and its hosts include sheep, goats and cattle (Fourie and Horak 1991; Walker 1991). They also parasitise wild animals such as the caracal, mountain reedbuck and the eland (Horak, Moolman and Fourie 1987; Walker 1991) and dogs in *I. rubicundus* regions often pick up the ticks. It is a three-host tick and the rock elephant shrew (*Elephantulus myurus*) is the main host for the immatures (Fourie, Horak and Van den Heever 1992). The tick can cause paralysis in a variety of domestic and wild ungulates (Spickett and Heyne 1988). *I. rubicundus* is confined to hilly or mountainous terrain in localities which have a distinctive Karoo vegetation. Only seven *I. rubicundus* ticks were collected in one locality (Batho). In Batho several koppies occur which may explain the occurrence of this tick on the dogs.

*Othious megnini*

This tick is also known as the Spinose ear tick and was originally introduced into South Africa from America (Theiler and Salisbury 1958). The larvae and nymphs of this tick normally parasitise the ears of domestic animals like cattle, sheep, goats, horses, cats, pigs, rabbits and man (Jagannath and Lokesh 1989). Its distribution is limited by high rainfall and hence it will not be found in high rainfall areas (Theiler and Salisbury 1958). The occurrence of nymphs in the ear can lead to the perforation of the ear drum and nervous disorders in some animals (Jagannath and Lokesh 1989).

*Rhipicephalus evertsi evertsi*

This tick is also known as the red-legged tick and has a variety of hosts, which include cattle, sheep, goats and horses (Walker *et al.* 1978; Walker 1991). All of the parasitic stages frequently feed on the same host. Its distribution is limited to regions with a minimum rainfall of 250mm per annum but is found throughout South Africa (Walker 1991). It is also responsible for diseases such as spring lamb paralysis (Howell *et al.* 1983), East Coast Fever (*Theileria parva*) and Redwater (*Babesia bigemina*) , amongst others (Theiler 1950). Only two *Rhipicephalus evertsi evertsi* ticks were sampled from dogs in the present study. As such it can be assumed that dogs are very infrequently infested by these ticks.

***Rhipicephalus follis and Rhipicephalus gertrudae***

Both these species were previously classified as being *R. capensis*. They are almost exclusively parasites of large herbivores but *R. follis* has been found on antelopes and *R. gertrudae* has been found on sheep, goats, horses and donkeys (Walker 1991). The distribution of these two species is wide in South Africa and includes the Free State. *R. follis* is thought to be found exclusively in South Africa (Walker 1991). Only two *R. follis* but 112 *R. gertrudae* were collected during the present study, both species only in Botshabelo. In a previous study (Dreyer, Fourie and Kok 1998) conducted in Botshabelo both species were recorded on cattle.

***Rhipicephalus sp.***

Previously, this tick was sampled in the south-western Free State and was referred to as *R. punctatus* (Horak and Fourie 1992). Recently, however, studies have shown that it is a new species which belongs to *Rhipicephalus pravus* group (Dreyer, Fourie and Kok 1998). With a certain infestation density, it is capable of causing paralysis in sheep and goats. Hence, it is commonly known as the brown paralysis tick (Fourie and Kok 1992). The most important host for its immatures is the rock elephant shrew (*Elephantulus myurus*), which is found in areas with rocky outcrops and hills (Fourie, Horak and Van den Heever 1992). Only six of these ticks were sampled from dogs in the present study and only in

Batho. The koppies that occur in Batho may explain the occurrence of this tick on the dogs.

***Rhipicephalus sanguineus***

*R. sanguineus* has perhaps the widest distribution of all ticks that infest dogs (Hoogstraal 1956; Bechara, Szabó, Mukai and Rosa 1993). It is a three-host tick and its hosts include mice, rabbits, rats, shrews and hedgehogs. It's preferred host is, however, the dog (Hoogstraal 1956; Walker 1970; Koch 1982). Its distribution is throughout South Africa but is rare in the drier areas (Walker 1991). The exact distribution of *R. sanguineus* in South Africa has, however, never been determined. Apparently these ticks are absent in the drier regions of the country (Howell, Walker and Nevill 1983) but this could be due to the absence of man-made habitations and hence dogs (Heath 1974). The absence of these ticks in the desert regions also correlates with the absence of diseases in the same regions (Keirans 1992). It is responsible for the transmission of *Ehrlichia canis* which causes ehrlichiosis in dogs (Sainz, Tesouro, Rodriguez, Mayoral and Mazzucchelli 1995; Pusterla, Pusterla, Deplazes, Wolfensberger, Müller, Hörauf, Reusch and Lutz 1998). It is certainly regarded as being the main vector of the "spotted fever group rickettsiae" (Mumcuoglu, Burgan, Ioffe-Uspensky and Manor 1993).

Except for *R. sanguineus* and *H. leachi*, the other ticks which infested dogs in the present study represent less than 2% of the total number of ticks sampled. These species can thus be viewed as being incidental infestations.

There are two phenomena that influence the individual interactions between ticks and hosts (Sonenshine 1975). The first is tick-dependant, which includes host predilection and opportunism. The second is host-dependant and this includes factors such as activity patterns, habitat utilisation, social behaviour, home range, and body size (Barnard 1989). In the present study habitat utilisation would be an important factor. This, however, does not mean that any tick, found in the same area as dogs, will automatically infest dogs. Specific appetence responses may also be a limiting factor (Barnard 1989). The infestation of dogs by ticks that have other natural hosts depends largely on sympatry. This is particularly true in places such as Botshabelo and Thaba Nchu. In these areas, people farm on a small scale and the cattle are often kept in their backyards. This means that the dogs and the cattle share the same areas and presents spatial sympatry for dogs (found on these properties) and ticks species which normally parasitise cattle. The dogs also move around with their owners, or wander around on their own, and are thus exposed to various habitats and hence tick species.

Batho, Botshabelo and Thaba Nchu differed from localities such as Brandwag in that the dogs in Brandwag were restricted to a particular property. This lessens the possibility of dogs coming into contact with tick species other than those found on the property. If proper tick control is practised certain tick species can also be eradicated from the property.

*SEASONAL DYNAMICS*

Despite the fact that much has been written about *R. sanguineus* (Heath 1974), it has been pointed out by Hoogstraal (1956) that not much is known about its biology and ecology. Additional information has been published (Heath 1974) but not very recently. *R. sanguineus* inhabits both wet and arid, tropical and sub-tropical regions but has a predilection for warm habitats. *R. sanguineus* ticks prefer the warmer summer months in the Northern Hemisphere (Dipeolu 1975) but according to Horak (1982) their seasonal preference has not been determined in the Southern Hemisphere. In the Northern Hemisphere it has been shown that *R. sanguineus* favours the summer months from March-September. This tick does, however, occur throughout the year with numbers dwindling during the colder months (Amin and Madbouly 1973; Dipeolu 1975; Mumcuoglu *et al.* 1993; Inokuma, Tamura and Onishi 1996). The results of the present study show that *R. sanguineus* ticks were dominant and were also most abundant during the summer months with low numbers during the winter months. The numbers of *R. sanguineus* ticks were particularly high in Botshabelo during January, February and April and in Thaba Nchu during January and February. This can most probably be attributed to the lack of tick control in these localities (see Chapter 6). The dog owners from the more affluent localities engage in attempts to control ticks on a regular basis, which results in the tick burdens being lower.

*R. sanguineus* ticks are able to survive at fairly high temperatures (Heath 1981; Mumcuoglu *et al.* 1993) of up to 44°C (Heath 1974). Low temperatures explain the lack of activity during the cold winter months (Arthur 1962 after Heath 1974). Furthermore, Stella (1938) found that although *R. sanguineus* was found on dogs throughout the year, the adult ticks were fewer during winter. Those that were found were always in a lethargic or inert (very slow moving) state. This can be substantiated by the findings of Mumcuoglu, Burgan, Ioffe-Uspensky and Manor (1993) who also found that higher numbers of *R. sanguineus* are found during the summer months and fewer during the winter months.

Although the numbers of ticks during winter are very low in all localities, the fact that the numbers never reach zero implies that this species does not undergo diapause. Stella (1938) also points out that, although in a state of lethargy, adult ticks are often found between the hairs of dogs. So, it could be said that the life cycle of the ticks is put "on hold" during the cold winter months but not in the form of diapause. According to Heath (1974) any environmental temperature lower than 7°C is probably an unsuitable habitat for *R. sanguineus* (Heath 1974). This, however, is contrary to the findings of the present study where, during July 1995, a mean minimum temperature of -3.6°C was recorded and yet ticks were still attached to the dogs Although, it should be noted that ticks that are attached to the dog are exposed to the dog's body temperature and will therefore not be affected in the same manner as free roaming ticks. In Pretoria North, a study conducted by Horak (1982) also showed that *R. sanguineus* is most abundant during the summer months, from October to April. Furthermore, despite the low number of adult ticks during the winter months, the ticks were active throughout the year.

During the present study it was not possible to determine the exact seasonal occurrence of *H. leachi* due to the fact that relatively few ticks were collected from dogs. The results do, however, suggest that these ticks are most abundant during spring and early summer. As was the case with *R. sanguineus*, these ticks can attach to dogs during both winter and summer. On a property in one of the more affluent localities a dog was found infested with fairly heavy burdens of larvae, nymphs and adults indicating that immatures of these ticks may become adapted to feed on dogs.

A previous study conducted on smallholdings near Grahamstown showed that *H. leachi* was most abundant during the winter months (Horak *et al.* 1987). In a previous study conducted in Nigeria by Dipeolu (1975), it was found that *H. leachi* showed no definitive pattern of seasonal dynamics but it should be noted that the climate is also more sub-tropical. Factors such as rainfall and temperature (Walker 1991), amongst others, influence the seasonal relative density and distribution of tick species. The seasonal relative density of *H. leachi* is still unknown in large parts of South Africa (Horak *et al.* 1987).

*ATTACHMENT SITES AND HAIR LENGTH*

In the present study, *R. sanguineus* was found mostly on the ears, on the back and the neck with very few ticks in the tail region. Mumcuoglu *et al.* (1993), however, reported that *R. sanguineus* is found mostly on the ears, head and abdomen. Walker (1970) mentions that adults are often most numerous on the ears but may be found on any part of the body. Howell *et al.*(1983), on the other hand, states that adults are usually found on the ears, neck and between the toes, but with heavy infestations the ticks are found attached to various other body regions. These facts and the results of the present study suggest that there are other factors involved which have an influence on the tick's site of attachment. Mumcuoglu *et al.* (1993) speculated that the reason for the head being a predilection site is because ticks often attach to dogs when the dog is moving forward and that grooming is not possible on the head. The fact that very few ticks were found on the tail region in the present study is possibly due to the fact that dogs can effectively groom the tail and genital parts. Generally the results of the present study, except for the differences in percentage of ticks attaching to the back and abdomen, seems to be fairly similar to those reported in the literature.

In the present study few *H. leachi* ticks were collected. The results, however, seem to indicate that these ticks mainly attached on the neck and back while according to Howell *et al.*(1983) they feed mainly on the head and shoulders of dogs but can be found on other parts of the body in the case of heavy infestations.

It should be noted that this information could be of significant importance with regard to control. There are several factors that influence the attachment sites of ixodid ticks to their host such as density of the ticks (Andrews and Petney 1981), interspecific interaction between ticks (Andrews, Petney and Bull 1982), habitat (Balashov 1972; Wilkinson 1972) and the time of the season (Evans 1952). These influencing factors must be taken into account when sampling ticks so as to ensure reliable results. Furthermore, they are of particular importance when it comes to control. Knowing the correct preferred attachment sites of a particular tick species makes the application of acaricides such as "spot-on" formulations more effective (Fourie and Van Zyl 1991). Hence, fundamental knowledge on the predilection attachment sites of certain tick species and the factors which influence them are important to optimise chemical control.

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# CHAPTER 3

## DEVELOPMENTAL BIOLOGY - EGGS

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## INTRODUCTION

Research on the egg-stage of ticks has generally been neglected. It is the only immobile stage in the tick's life cycle and the numbers laid in relation to the percentage that hatches determine the success of the tick's following generation (Dipeolu, Amoo and Akinboade 1991). Mating stimulates females to engorge rapidly for a period of about 48 hours. The period after they have engorged fully is later followed by detachment and drop off (Toutoungi, Gern and Aeschlimann 1995). After engorgement and detachment the female ixodid tick, requiring no further feeding, finds a suitable place or microclimate and then assumes a sedentary habit until oviposition commences. The period from the day of detachment to the day egg laying begins is known as the pre-oviposition period (Toutoungi *et al.* 1995). This is followed by the next event in the tick's life cycle namely, oviposition (Sweatman and Koussa 1968).

In studies conducted by Sweatman it was shown that longevity and oviposition of engorged female ticks are affected by physical and biological factors (Sweatman 1967; Dipeolu 1984). The non-parasitic egg stage is considered to be the most susceptible stage of the tick's life cycle (Sweatman 1967; Theiler 1969). This stage also gives an indication of the tick's microclimatic range of tolerance (Sweatman 1967) and may be the stage that limits a tick's geographical distribution (Theiler 1969; McEnroe 1977). Conversely, the tick's temperature and relative humidity preferences reflect the climate within its geographical range (Heath 1981). One of the influencing factors is the relative humidity in the environment. The saturation deficit, which is a more comprehensive means of looking at relative humidity, provides a measure of the drying capacity of the immediate surrounding atmosphere at a given temperature (Teel 1984).

Although *R. sanguineus* occurs very widely, research conducted on the tick elsewhere cannot be directly extrapolated to the local situation. In South Africa the tick is exposed to different sets of selection pressures that may have resulted in specific adaptations. Possible differences in the biology of *R. sanguineus* that occurs in South Africa and elsewhere can only be discovered through comparative research. Irrespective of the very wide geographic distribution of *R. sanguineus*, the biology of this tick is still not understood properly (Heath 1979; Bechara, Szabó. Ferreira and Garcia 1995) and almost no research on the reproduction of this tick has been done in South Africa. This 's also true for *Haemaphysalis leachi*.

The objectives of the present study were to investigate the effects of temperature and relative humidity on oviposition and incubation and to determine and compare the fecundity and conversion efficiency index values for *R. sanguineus* and *H. leachi*.

# MATERIALS AND METHODS

*(a) PRE-OVIPOSITION AND INCUBATION PERIOD*

Fully engorged ticks were collected, weighed and placed at different temperature and relative humidity regimes (Fig. 3.1). The number of engorged females used in the present study, for each temperature and relative humidity regime, ranged between three and four for *R. sanguineus* and four and eight for *H. leachi*. A total of 51 *R. sanguineus* and 82 *H. leachi* ticks were used. Females were examined every second day to determine the onset of oviposition. After the commencement of egg-laying, observations were made on a daily basis to determine the onset of hatching.

*(b) DAILY EGG PRODUCTION*

Fully engorged *R. sanguineus* and *H. leachi* females were collected from dogs at the SPCA (East End) in Bloemfontein, during March 1995. Each female was weighed on an electronic scale (Sartorius: Type R200D; accurate to 0.001 mg) and placed separately in glass vials (10ml), the tops of which were closed with plastic gauze to allow free airflow. Five ticks of each species were used. The vials were placed inside one-litre plastic containers filled with a 100ml $KH_2PO_4$ saturated salt solution (Winston and Bates 1960) that provided a 90±2% RH. These bottles, covered with black plastic, were placed in temperature controlled incubation cabinets at 25±1°C. All the engorged females were observed daily to determine the onset of oviposition. At the onset of oviposition the daily number of eggs produced was determined. This was done by counting the individual eggs laid (at the same time each day) and this was done daily until no more eggs were deposited. The data were used to determine the egg-laying pattern for each species.

### (c) *FECUNDITY AND CONVERSION EFFICIENCY INDEX (CEI) VALUES AND THE INFLUENCE OF TEMPERATURE AND RELATIVE HUMIDITY.*

In the second part of the present study ticks were collected in the same manner as mentioned above but this time engorged females were exposed to different temperature and relative humidity regimes (Fig. 3.1). This was done to investigate the effect of temperature and relative humidity on egg production. The number of engorged females used in the present study, for each temperature and relative humidity regime, ranged between two and four for *R. sanguineus* and between two and eight for *H. leachi*. A total of 46 *R. sanguineus* and 70 *H. leachi* ticks were used. The sample sizes were dependent on the availability of the engorged ticks at the time. The mass of the eggs laid by these ticks was determined after the termination of egg laying. The mass of the eggs laid was then converted to the number of eggs laid, according to Koch (1983). This was done because of time constraints and because counting the number of eggs laid is time consuming. To do this, the masses of 50 and 100 eggs, respectively, taken daily (with each day representing a different stage of the oviposition period), were determined prior to the commencement of the present study. The mean mass per egg for *R. sanguineus* and *H. leachi* was calculated as $3.96 \times 10^{-5}$ mg and $4.02 \times 10^{-5}$ mg, respectively. The reason why eggs from different stages were used is because the egg mass changes during the course of oviposition. It was shown by Dipeolu *et al.* (1991) that eggs laid towards the end of the oviposition period were heavier. Furthermore, when a few eggs were laid the eggs were heavier, compared to when numerous eggs were laid. In order, therefore, to determine the mean mass of the eggs, samples were taken during the different stages of the oviposition period. This process of weighing the eggs and converting it to the number of eggs laid per engorged female was continued until the female ticks ceased to lay any more eggs. These results were used to calculate the fecundity for the different temperature and relative humidity regimes, for each species. The fecundity can be expressed as the mean number of eggs produced per mg female mass and was determined at each temperature and RH combination (Van der Lingen, Fourie, Kok and van Zyl 1999). The mass of the eggs laid by each engorged female was used to determine the Conversion Efficiency Index (CEI) which is the percentage body mass converted into egg mass. The CEI was calculated according to the following formula (Drummond & Whetstone 1970):

$$CEI = \left(\frac{m_1}{m_2}\right) * 100$$

where CEI = Conversion Efficiency Index (%)

$m_1$ = mass of egg cluster

$m_2$ = mass of engorged female

***(c) STATISTICAL ANALYSIS***

The following analyses were performed for both *R. sanguineus* and *H. leachi*:

(i) The relationship between the mass of the eggs laid and the mass of the engorged female.

(ii) The relationship between the reciprocal of the pre-oviposition time and the temperature and the Critical Developmental Temperature (CDT).

(iii) The relationship between the reciprocal of the incubation time and the temperature and the Critical Developmental Temperature (CDT).

These analyses were used to determine the critical developmental temperatures (CDT) for each of the above-mentioned cases. This gives an indication of the temperature below which development will take place very slowly (or not at all) in that particular part of the tick's life cycle.

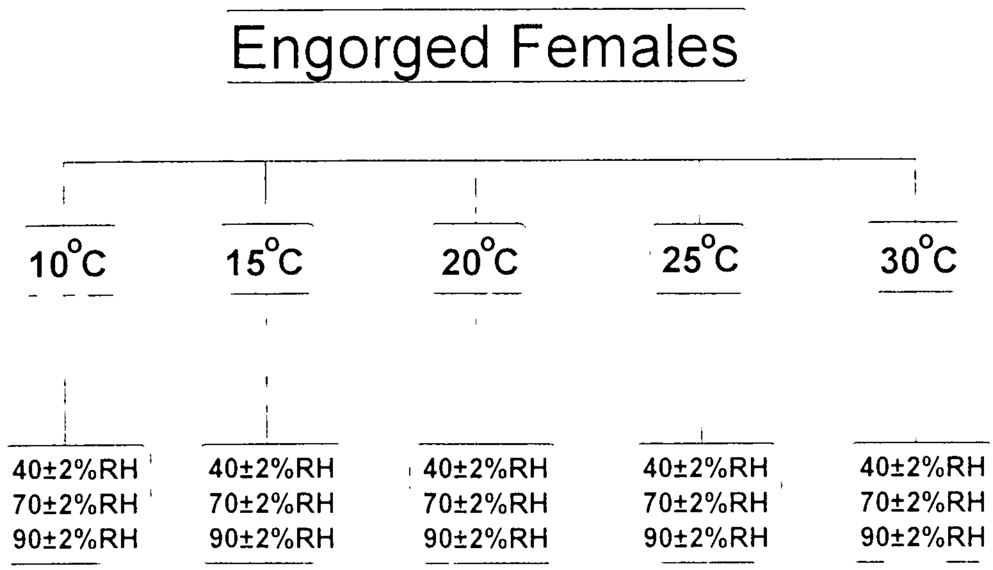


**Figure 3.1.** Different combinations of temperature and relative humidities used in the observations on oviposition and incubation periods of *R. sanguineus* and *H. leachi*.

# RESULTS

*(a) PRE-OVIPOSITION AND INCUBATION PERIOD*

At 10°C no eggs were laid. At any given relative humidity, the mean pre-oviposition time for both species decreased, as the temperature increased. The mean minimum and maximum pre-oviposition periods for *R. sanguineus* ticks were 4.8 (25°C, 90%RH) and 21.0 (15°C, 40%RH) days, respectively. The mean minimum and maximum pre-oviposition periods for *H. leachi* were 4.3 (30°C, 90%RH) and 12.1 (15°C, 40%RH) days, respectively (Tables 3.1 a & b).

The mean minimum and maximum incubation periods for *R. sanguineus* were 19.0 (30°C, 90%RH) and 72.0 (15°C, 90%RH) days respectively. The incubation period decreased with an increase in temperature. Relative humidity did not appear to influence the incubation period. The mean minimum and maximum incubation periods for *H. leachi* were 15.5 (30°C, 90%RH) and 66.7 (15°C, 40%RH) days respectively. The incubation period decreased with an increase in temperature. The relative humidity did not appear to have an effect on the incubation period. Eggs kept at 40%RH and temperatures of 20°C and 30°C did not hatch. At 25°C only one of the eight egg batches hatched (Table 3.1b).

**Table 3.1(a).** Summary of the pre-oviposition and incubation period of *R. sanguineus* eggs at different regimes of temperature and relative humidity.

| CONDITIONS | | PRE-OVIPOSITION PERIOD (DAYS) | | | INCUBATION PERIOD (DAYS) | | | n | Successful Hatch |
|---|---|---|---|---|---|---|---|---|---|
| TEMP (°C) | RH (%) | $\bar{x}$ | Min. | Max. | $\bar{x}$ | Min. | Max. | | |
| 10 | 40 | 0 | 0 | 0 | 0 | 0 | 0 | 3 | 0 |
| 10 | 70 | 0 | 0 | 0 | 0 | 0 | 0 | 3 | 0 |
| 10 | 90 | 0 | 0 | 0 | 0 | 0 | 0 | 3 | 0 |
| 15 | 40 | 21.0 | 21 | 21 | 0 | 0 | 0 | 3 | 0 |
| 15 | 70 | 17.0 | 14 | 20 | 71.7 | 70 | 73 | 3 | 3 |
| 15 | 90 | 18.0 | 17 | 20 | 72.0 | 70 | 73 | 3 | 3 |
| 20 | 40 | 13.3 | 8 | 23 | 42.7 | 35 | 46 | 3 | 3 |
| 20 | 70 | 18.0 | 8 | 23 | 37.0 | 32 | 47 | 3 | 3 |
| 20 | 90 | 8.7 | 8 | 9 | 46.3 | 46 | 47 | 3 | 3 |
| 25 | 40 | 5.0 | 5 | 5 | 22.0 | 19 | 25 | 4 | 4 |
| 25 | 70 | 5.0 | 5 | 5 | 22.0 | 19 | 25 | 4 | 4 |
| 25 | 90 | 4.8 | 4 | 5 | 22.3 | 22 | 23 | 4 | 4 |
| 30 | 40 | 5.3 | 5 | 6 | 20.5 | 18 | 23 | 4 | 4 |
| 30 | 70 | 5.0 | 5 | 5 | 19.3 | 17 | 22 | 4 | 4 |
| 30 | 90 | 5.0 | 5 | 5 | 19.0 | 19 | 19 | 4 | 4 |

**Table 3.1(b).** Summary of the pre-oviposition and incubation period of *H. leachi* eggs at different regimes of temperature and relative humidity.

| CONDITIONS | | PRE-OVIPOSITION PERIOD (DAYS) | | | INCUBATION PERIOD (DAYS) | | | n | Successful Hatch |
|---|---|---|---|---|---|---|---|---|---|
| TEMP(°C) | RH (%) | $\bar{x}$ | Min. | Max. | $\bar{x}$ | Min. | Max. | | |
| 10 | 40 | 0 | 0 | 0 | 0 | 0 | 0 | 4 | 0 |
| 10 | 70 | 0 | 0 | 0 | 0 | 0 | 0 | 4 | 0 |
| 10 | 90 | 0 | 0 | 0 | 0 | 0 | 0 | 4 | 0 |
| 15 | 40 | 12.1 | 11 | 13 | 66.7 | 64 | 68 | 7 | 3 |
| 15 | 70 | 11.1 | 10 | 14 | 50.0 | 45 | 53 | 7 | 7 |
| 15 | 90 | 11.3 | 11 | 12 | 39.7 | 32 | 46 | 8 | 7 |
| 20 | 40 | 10.0 | 8 | 13 | 0 | 0 | 0 | 4 | 0 |
| 20 | 70 | 8.3 | 8 | 9 | 34.0 | 34 | 34 | 4 | 3 |
| 20 | 90 | 9.0 | 9 | 9 | 32.0 | 32 | 33 | 4 | 4 |
| 25 | 40 | 5.4 | 5 | 6 | 24.0 | 24 | 24 | 8 | 1 |
| 25 | 70 | 7.4 | 5 | 8 | 23.8 | 21 | 26 | 8 | 8 |
| 25 | 90 | 7.0 | 5 | 7 | 24.8 | 24 | 25 | 8 | 8 |
| 30 | 40 | 4.8 | 4 | 5 | 0 | 0 | 0 | 4 | 0 |
| 30 | 70 | 4.8 | 4 | 5 | 18.5 | 17 | 21 | 4 | 4 |
| 30 | 90 | 4.3 | 4 | 5 | 15.5 | 15 | 16 | 4 | 4 |

The relationship between the reciprocal of the pre-oviposition period and temperature, for *R. sanguineus* was given by the following equation:

$y = a + bx + cx^2$, where

$a = -0.2096$

$b = 0.02316$

$c = -0.0002985$

$y =$ 1/ pre-oviposition period (days)

$x =$ temperature in °C

$R^2 = 93.35\%$

The relationship between the reciprocal of the pre-oviposition period and temperature for *H. leachi* was given by the following equation:

$y = a + bx$, where

$a = -0.08824$

$b = 0.01017$

$y =$ 1/ pre-oviposition period (days)

$x =$ temperature in °C

$R^2 = 91.38\%$

Figs. 3.2 & 3.3 graphically present the above-mentioned relationships. The $R^2$ values for *R. sanguineus* and *H. leachi* were 93.35% and 91.38%, respectively. The developmental zero (intersection of the regression line with the x-axis) or Critical Developmental Temperature (CDT) for *R. sanguineus* and *H. leachi* was calculated as 10.5°C and 8.7°C, respectively.

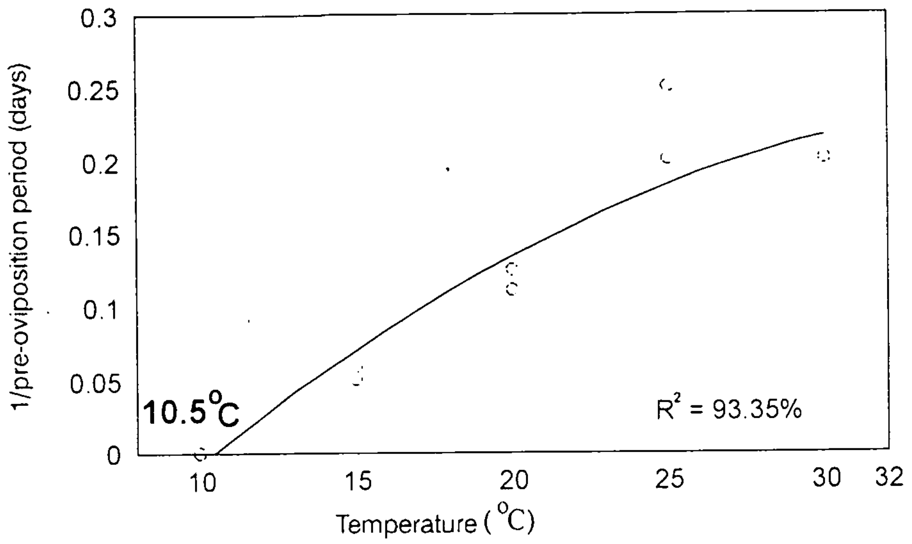


**Figure 3.2.** Graphical representation of the relationship between the inverse of the pre-oviposition period (days) and the temperature (°C) for *R. sanguineus*. The critical developmental temperature is 10.5°C.

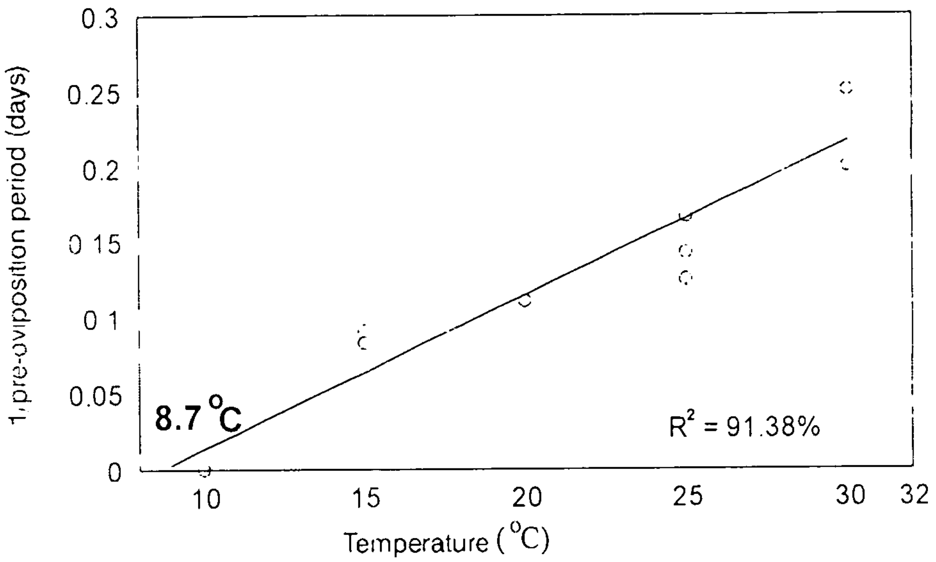


**Figure 3.3.** Graphical representation of the relationship between the inverse of the pre-oviposition period (days) and the temperature (°C) for *H. leachi*. The critical developmental temperature is 8.7°C.

The relationship between the reciprocal of the incubation period and temperature for *R. sanguineus* was given by the following relationship:

$y = a + bx$, where

$a = -0.02610$

$b = 0.002582$

$y =$ 1/incubation period

$x =$ temperature in °C

$R^2 = 98.24\%$

The relationship between the reciprocal of the incubation period and the temperature for *H. leachi* was given by the following relationship:

$y = a + bx + cx^2$, where

$a = -0.02874$

$b = 0.003563$

$c = -3.899 \times 10^{-5}$

$y =$ 1/incubation period

$x =$ temperature in °C

$R^2 = 86.45\%$

Figs. 3.4 & 3.5 graphically present the above-mentioned relationships. The $R^2$ values for *R. sanguineus* and *H. leachi* were 98.24% and 86.45%, respectively. The developmental zero (intersection of the regression line with the x-axis) or Critical Developmental Temperature (CDT) for *R. sanguineus* and *H. leachi* was calculated as 10.1°C *and* 8.9°C, respectively.

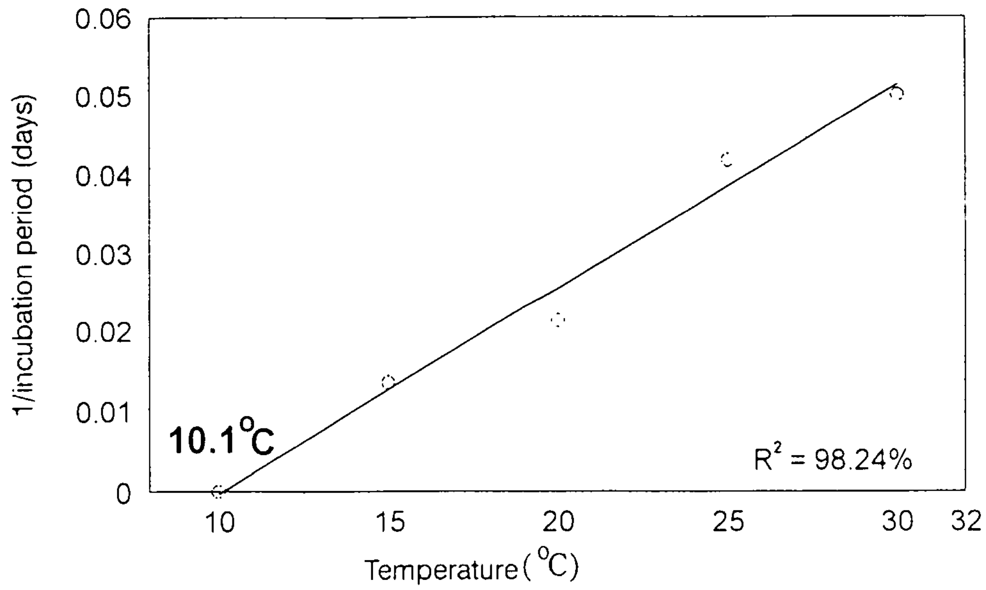


**Figure 3.4.** Graphical representation of the relationship between the inverse of the incubation period (days) and the temperature (°C) for *R. sanguineus*. The critical developmental temperature is 10.1°C.

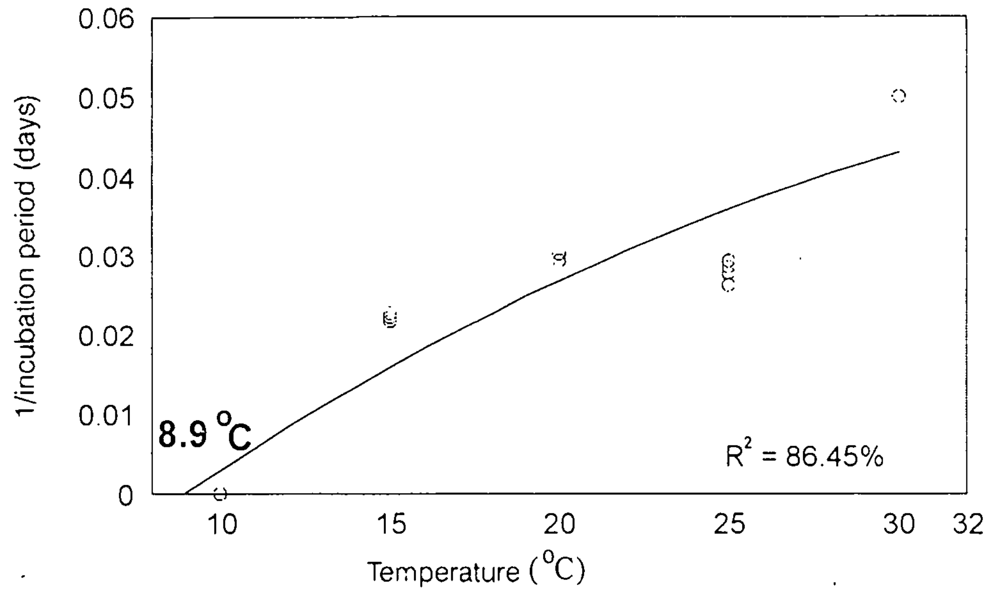


**Figure 3.5:** Graphical representation of the relationship between the inverse of the incubation period (days) and the temperature (°C) for *H. leachi*. The critical developmental temperature is 8.9°C.

*(b) DAILY EGG PRODUCTION*

The number of eggs laid per day (25°C; 90%RH) by *R. sanguineus* and *H. leachi* is graphically presented in Fig. 3.6. In the case of *R. sanguineus* ticks, egg laying started between four and six days after detachment. The egg laying reached a peak after eight days, after which daily egg production started declining immediately. The females stopped laying eggs after 21 days, which is not clearly shown on the graph due to the increment being too small. The first day and the last four days are characterised by the laying of very few eggs. In the case of *H. leachi* ticks, egg laying also started between four and five days after detachment. Egg laying reached a peak after six days, which was maintained until day eight, after which an immediate decline in mean daily egg production, until day 15, was observed. The females also stopped laying eggs after about 21 days.

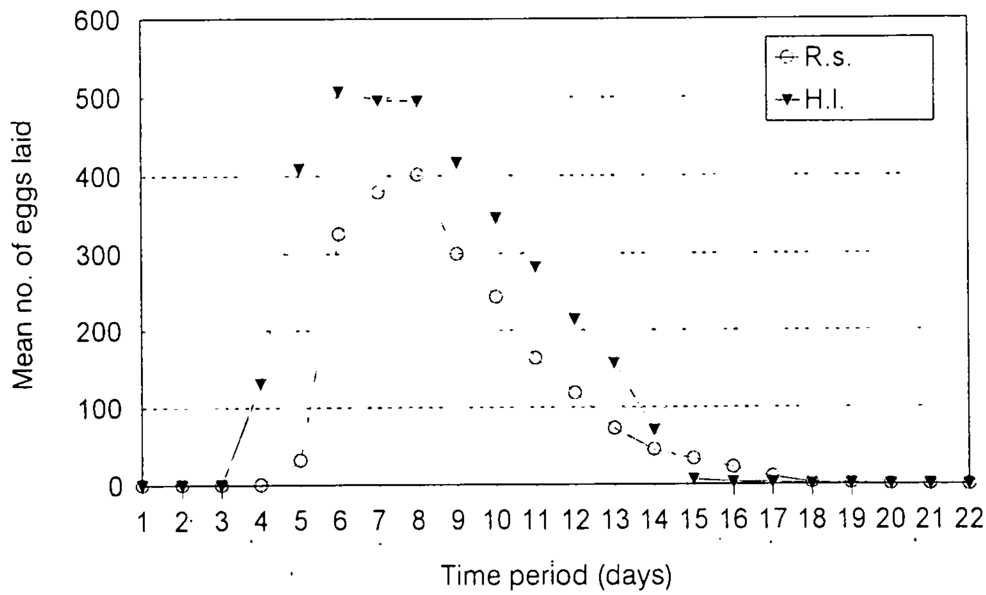


**Figure 3.6.** Average number of eggs laid per day by *R. sanguineus* and *H. leachi* at 25±1 °C and 90±2 % RH.

### *(c) FECUNDITY AND CONVERSION EFFICIENCY INDEX (CEI) VALUES AND THE INFLUENCE OF TEMPERATURE AND RELATIVE HUMIDITY.*

A summary of the number of eggs laid and the fecundity of the ticks at different combinations of temperature and relative humidity are given in Tables 3.2 a & b. For both *R. sanguineus* and *H. leachi* ticks, no eggs were laid at 10°C. The fecundity (mean number of eggs laid per mean female engorgement mass (mg)) of *R. sanguineus* ranged from a minimum of 10.3 eggs per mg body mass (15°C, 40%RH) to a maximum of 17.2 eggs per mg body mass (25°C, 90%RH). The fecundity of *H. leachi* ranged from a minimum of 7.7 eggs per mg body mass (15°C, 40%RH) to a maximum of 16.3 eggs per mg body mass (15°C, 90%RH). In general, for both species, the fecundity at a specific temperature increased with an increase in relative humidity (Tables 3.2 a & b). During the present study the maximum number (3232) of eggs laid by *R. sanguineus* was recorded at a temperature of 30°C and 90±2 %RH. In the case of *H. leachi*, maximum number (4801) of eggs laid was recorded at a temperature of 15°C and 70±2 %RH.

**Table 3.2(a).** Summary of number of eggs laid and fecundity of *R. sanguineus* females exposed to different temperatures and relative humidities.

| CONDITION | | FEMALE | NUMBER OF EGGS | | | | n |
|---|---|---|---|---|---|---|---|
| TEMPERATURE (°C) | RH (%) | $\bar{x}$ Mass (g) | $\bar{x}$ | Min. | Max. | Fecundity | |
| 10 | 40 | 0.120 | 0 | 0 | 0 | 0 | 3 |
| 10 | 70 | 0.146 | 0 | 0 | 0 | 0 | 3 |
| 10 | 90 | 0.166 | 0 | 0 | 0 | 0 | 3 |
| 15 | 40 | 0.144 | 1490 | 884 | 2096 | 10.3 | 2 |
| 15 | 70 | 0.141 | 1970 | 1717 | 2147 | 13.97 | 3 |
| 15 | 90 | 0.116 | 1414 | 960 | 1591 | 12.2 | 3 |
| 20 | 40 | 0.142 | 1768 | 1061 | 2424 | 12.5 | 3 |
| 20 | 70 | 0.135 | 1970 | 1490 | 2601 | 14.6 | 3 |
| 20 | 90 | 0.116 | 1742 | 1313 | 2096 | 15.0 | 3 |
| 25 | 40 | 0.179 | 2702 | 2652 | 2727 | 15.1 | 2 |
| 25 | 70 | 0.083 | 1288 | 783 | 1818 | 15.5 | 4 |
| 25 | 90 | 0.132 | 2276 | 1970 | 2748 | 17.2 | 3 |
| 30 | 40 | 0.088 | 1086 | 404 | 2475 | 12.3 | 4 |
| 30 | 70 | 0.114 | 1717 | 833 | 2525 | 15.1 | 3 |
| 30 | 90 | 0.095 | 1566 | 732 | 3232 | 16.5 | 4 |

**Table 3.2(b).** Summary of number of eggs laid and fecundity of *H. leachi* females exposed to different temperatures and relative humidities.

| CONDITION | | FEMALE | NUMBER OF EGGS | | | | n |
|---|---|---|---|---|---|---|---|
| TEMPERATURE (°C) | RH (%) | $\bar{x}$ Mass (g) | $\bar{x}$ | Min. | Max. | Fecundity | |
| 10 | 40 | 0.178 | 0 | 0 | 0 | 0 | 3 |
| 10 | 70 | 0.213 | 0 | 0 | 0 | 0 | 3 |
| 10 | 90 | 0.192 | 0 | 0 | 0 | 0 | 3 |
| 15 | 40 | 0.229 | 1766 | 821 | 3557 | 7.7 | 7 |
| 15 | 70 | 0.254 | 3682 | 2786 | 4801 | 14.5 | 7 |
| 15 | 90 | 0.198 | 3234 | 2736 | 3781 | 16.3 | 7 |
| 20 | 40 | 0.181 | 1915 | 1202 | 2910 | 10.6 | 4 |
| 20 | 70 | 0.241 | 3781 | 2786 | 4552 | 15.7 | 3 |
| 20 | 90 | 0.163 | 2438 | 1418 | 4279 | 14.95 | 4 |
| 25 | 40 | 0.244 | 1965 | 1716 | 2189 | 8.1 | 2 |
| 25 | 70 | 0.250 | 3234 | 2562 | 4204 | 12.94 | 8 |
| 25 | 90 | 0.242 | 3707 | 3110 | 4702 | 15.3 | 8 |
| 30 | 40 | 0.176 | 1791 | 1194 | 2637 | 10.2 | 3 |
| 30 | 70 | 0.186 | 2711 | 1592 | 4204 | 14.6 | 4 |
| 30 | 90 | 0.227 | 3682 | 3134 | 4204 | 16.2 | 4 |

The CEI values for *R. sanguineus* exposed to different temperatures and relative humidities are summarised in Table 3.3a. The lowest value (41.0%) was recorded at 15°C and 40±2 %RH and the highest value (68.1%) at 25°C and 90±2 %RH (Table 3.3a). In general, the lowest CEI values were recorded at the low (40%) relative humidity.

**Table 3.3(a).** Conversion Efficiency Index (%) values for *R. sanguineus* females exposed to different temperatures and relative humidities.

| CONDITIONS | | FEMALE | | | EGG MASS (g) | | | CEI (%) | n |
|---|---|---|---|---|---|---|---|---|---|
| TEMP. (°C) | RH (%) | $\bar{x}$ | MIN. | MAX. | $\bar{x}$ | MIN. | MAX. | | |
| 10 | 40 | 0.120 | 0.127 | 0.117 | 0 | 0 | 0 | 0 | 3 |
| 10 | 70 | 0.146 | 0.080 | 0.180 | 0 | 0 | 0 | 0 | 3 |
| 10 | 90 | 0.166 | 0.160 | 0.170 | 0 | 0 | 0 | 0 | 3 |
| 15 | 40 | 0.144 | 0.111 | 0.177 | 0.059 | 0.035 | 0.083 | 41.0 | 2 |
| 15 | 70 | 0.141 | 0.137 | 0.146 | 0.078 | 0.068 | 0.085 | 55.3 | 3 |
| 15 | 90 | 0.116 | 0.104 | 0.123 | 0.056 | 0.038 | 0.063 | 48.3 | 3 |
| 20 | 40 | 0.142 | 0.094 | 0.194 | 0.070 | 0.042 | 0.096 | 49.3 | 3 |
| 20 | 70 | 0.135 | 0.168 | 0.119 | 0.078 | 0.059 | 0.103 | 57.8 | 3 |
| 20 | 90 | 0.116 | 0.088 | 0.139 | 0.069 | 0.052 | 0.083 | 59.5 | 3 |
| 25 | 40 | 0.179 | 0.174 | 0.184 | 0.107 | 0.105 | 0.108 | 59.8 | 2 |
| 25 | 70 | 0.083 | 0.060 | 0.110 | 0.051 | 0.031 | 0.072 | 61.4 | 4 |
| 25 | 90 | 0.132 | 0.118 | 0.0142 | 0.09 | 0.078 | 0.094 | 68.1 | 3 |
| 30 | 40 | 0.088 | 0.041 | 0.166 | 0.043 | 0.016 | 0.098 | 48.9 | 4 |
| 30 | 70 | 0.114 | 0.080 | 0.154 | 0.068 | 0.033 | 0.100 | 59.7 | 3 |
| 30 | 90 | 0.095 | 0.055 | 0.179 | 0.062 | 0.029 | 0.128 | 65.3 | 4 |

The CEI values for *H. leachi* exposed to different temperatures and relative humidities are summarised in Table 3.3b. The lowest value (31.0%) was recorded at 15°C and 40±2 %RH and the highest value (65.7%) at 15°C and 90±2 %RH (Table 3.3b). In general, as in the case of *R. sanguineus*, the lowest CEI values were recorded at the low (40%) relative humidity.

**Table 3.3(b).** Conversion Efficiency Index (%) values for *H. leachi* females exposed to different temperatures and relative humidities.

| CONDITIONS | | FEMALE | | | EGG MASS (g) | | | CEI (%) | n |
|---|---|---|---|---|---|---|---|---|---|
| TEMP. (°C) | RH (%) | $\bar{x}$ | MIN. | MAX | $\bar{x}$ | MIN. | MAX. | | |
| 10 | 40 | 0.178 | 0.145 | 0.205 | 0 | 0 | 0 | 0 | 3 |
| 10 | 70 | 0.213 | 0.181 | 0.262 | 0 | 0 | 0 | 0 | 3 |
| 10 | 90 | 0.192 | 0.147 | 0.259 | 0 | 0 | 0 | 0 | 3 |
| 15 | 40 | 0.229 | 0.192 | 0.237 | 0.071 | 0.033 | 0.143 | 31.0 | 7 |
| 15 | 70 | 0.254 | 0.209 | 0.326 | 0.148 | 0.112 | 0.193 | 58.3 | 7 |
| 15 | 90 | 0.198 | 0.173 | 0.242 | 0.130 | 0.110 | 0.152 | 65.7 | 7 |
| 20 | 40 | 0.181 | 0.139 | 0.228 | 0.077 | 0.0483 | 0.117 | 42.5 | 4 |
| 20 | 70 | 0.241 | 0.185 | 0.280 | 0.152 | 0.112 | 0.183 | 63.0 | 3 |
| 20 | 90 | 0.163 | 0.119 | 0.194 | 0.098 | 0.057 | 0.172 | 60.1 | 4 |
| 25 | 40 | 0.244 | 0.214 | 0.273 | 0.079 | 0.069 | 0.088 | 32.4 | 2 |
| 25 | 70 | 0.250 | 0.183 | 0.337 | 0.130 | 0.103 | 0.169 | 52.0 | 8 |
| 25 | 90 | 0.242 | 0.210 | 0.290 | 0.149 | 0.125 | 0.189 | 61.6 | 8 |
| 30 | 40 | 0.176 | 0.144 | 0.235 | 0.072 | 0.048 | 0.106 | 40.9 | 3 |
| 30 | 70 | 0.186 | 0.122 | 0.267 | 0.109 | 0.064 | 0.169 | 58.6 | 4 |
| 30 | 90 | 0.227 | 0.210 | 0.250 | 0.148 | 0.126 | 0.169 | 65.2 | 4 |

The relationship between the mass of the eggs laid and the mass of the female, at 30°C and 90%RH, respectively, was given by the following linear regression relationships:

$y = a + bx$, where (for *R. sanguineus*)

$a = 1.05462$

$b = 25157.3$

$y =$ mass of eggs in g

$x =$ mass of engorged female in g

$R^2 = 99.95\%$

and (for *H. leachi*)

$a = 13.3528$

$b = 24574.3$

$y =$ mass of eggs in g

$x =$ mass of engorged female in g

$R^2 = 94.35\%$

These relationships are represented graphically by Figs. 3.7 and 3.8, and showed a highly significant positive correlation between the engorgement mass of the female and the mass of the eggs laid. The $R^2$ values for *R. sanguineus* and *H. leachi* were 99.95% and 94.35% respectively. The greater the engorgement mass, the greater the mass of the eggs laid.

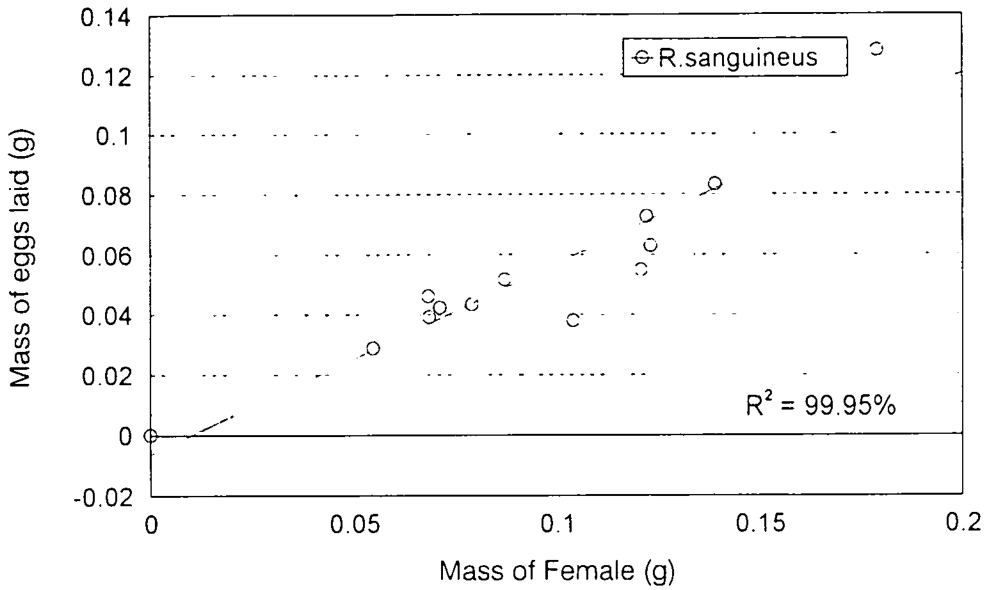


**Figure 3.7.** Linear regression to show the relationship between the mass of the eggs laid and the mass of the *R. sanguineus* females.

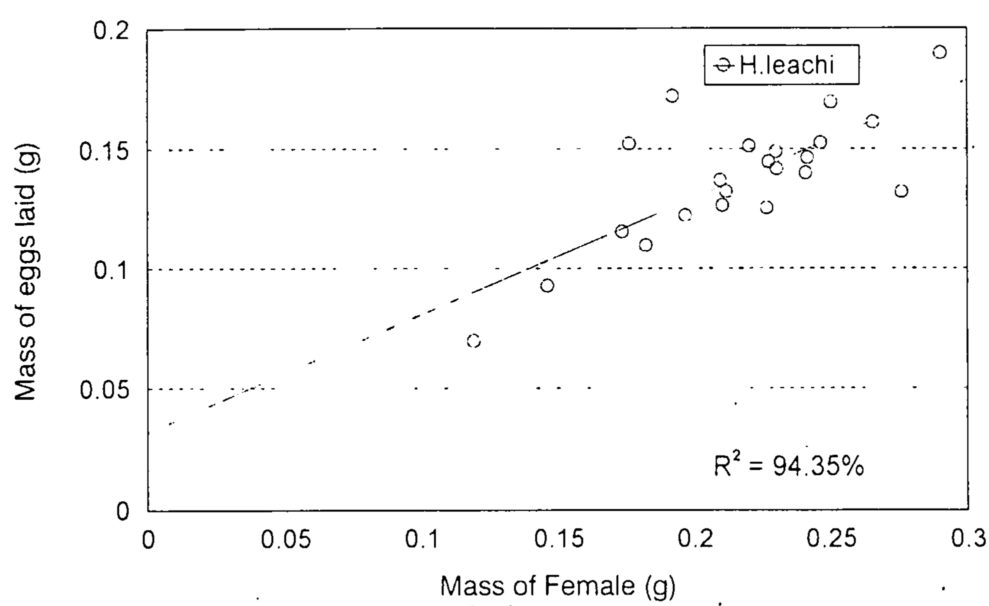


**Figure 3.8.** Linear regression to show the relationship between the mass of the eggs laid and the mass of the *H. leachi* females.

# DISCUSSION

### *(a) PRE-OVIPOSITION AND INCUBATION PERIOD*

Longevity and oviposition of engorged female ticks are affected by various factors, both physical and biological. Physical factors include temperature and relative humidity (Sweatman 1967; Branagan 1973; Dipeolu 1983), the effects of which have been demonstrated in the present study. The findings of the present study indicate that *R. sanguineus* and *H. leachi* are able to lay eggs between the temperature range of above 10°C and 30°C. The maximum temperature used in the present study was 30°C, however, these ticks can lay eggs at higher temperatures. Sweatman and Koussa (1968) experimented with *R. sanguineus* and found that the temperature zone of tolerance was between 10 and 40°C and that *R. sanguineus* did not oviposit at 10°C but did survive the longest at this temperature. Heath (1979) demonstrated that *R. sanguineus* is capable of laying eggs at temperatures as high as 38°C.

Exposure to lower temperatures resulted in a longer pre-oviposition period for both species and relative humidity had no significant effect. This indicates a correlation between temperature and pre-oviposition period and is supported by the findings of Sweatman (1967). According to Sweatman (1967) pre-oviposition periods for *R. sanguineus* increased dramatically below 20°C. An increase of temperature increases the metabolic rate and shortens the pre-oviposition and oviposition periods (Dipeolu, 1983), hence the shorter pre-oviposition periods at higher temperatures. In ixodid ticks, there is generally a decrease in the pre-oviposition period with an increase in temperature (Fujimoto 1992; Guglielmone 1992). This was shown to be the case for *Boophilus decoloratus, Boophilus geigyi* (Dipeolu 1983), *I. rubicundus* (Van Der

Lingen, Fourie, Kok and Van Zyl 1999) and *Dermacentor andersoni* (Arthur 1962) amongst others, and is also true for *R. sanguineus* and *H. leachi*.

The critical developmental temperature (CDT) was determined for both *R. sanguineus* and *H. leachi*. The CDT reflects the lowest temperature at which these species will lay eggs. Temperature is regarded as being the determining factor in this stage of a tick's life cycle (MacLeod 1935; Fujimoto 1992). This and the fact that there is a correlation between temperature and the pre-oviposition period suggests that in an attempt to produce eggs for experimental purposes it is advisable to place fully engorged females at moderately higher temperatures (25°C) and at higher humidities (70%), even though relative humidity did not appear to have any effect. This will ensure rapid oviposition after which lower temperatures such as 10°C can be used for the storage of those eggs. This will ensure retardation in the development of the eggs, which can be kept for later use. During the present study *H. leachi* did not lay eggs at 10°C, which is above the critical developmental temperature recorded during the present study. It should be noted that the incubators, in which the engorged females were kept, displayed a degree of variation of up to 2°C that may have affected the results. Secondly, if smaller temperature divisions had been used, the results may have reflected a CDT closer to 10°C.

It has also been suggested that the eggs, having been laid, are most susceptible to dehydration, which may be the one factor that limits a tick's geographical distribution (Theiler 1969; Rechav and von Maltzahn 1977; Koch 1983). Hair, Sauer and Durham (1975) have also suggested that the normal distribution of adult ticks is correlated to their ability to resist dehydration. Eggs, unlike the other developmental stages, cannot replenish lost water even in atmospheres with a relative humidity of 95% (Sauer and Hair 1971; Rechav and von Maltzahn 1977). The interaction between temperature and relative humidity is significant for percent hatch (Teel 1984) and eggs subjected to low relative humidities (and high temperatures) experience weight loss, which in turn affects the percentage eggs that successfully hatch (Teel 1984). Conversely, if the temperature is too low and the required CDT has not been met, hatching will not occur. The CDT for the incubation period for *R. sanguineus* and *H. leachi* was determined in the present study and reflects the lowest temperatures at which the eggs of these species will hatch. This suggests that ticks are found only in areas where the tick's CDT requirement is met and the dehydration level can be tolerated, hence the different geographic distribution areas for different tick species

The present study has shown that exposure of eggs to lower temperatures results in a prolonged incubation period for both species. The shortest incubation period was recorded at the highest temperature used. *R. sanguineus* showed almost complete independence of relative humidity. This, however, was not necessarily true for *H. leachi*. This indicates a correlation between the temperature and the incubation period and is supported by the findings of Heath (1979) who reported that the incubation periods of *R. sanguineus* take longer at lower temperatures. Nuttall (1915) also reported incubation periods that extend to 75 days at temperatures as low as 12°C. Heath (1979) further suggested that rapid development results in a shortened development period for a generation and in so doing, increases the survival rate in harsh environments.

*(b) DAILY EGG PRODUCTION*

The findings of the present study depicted a common pattern for both *R. sanguineus* and *H. leachi*. The eggs were laid, slowly at first, and then a peak was reached within a week of the onset of oviposition. Egg production subsequently rapidly slowed down over several days until oviposition stopped completely. At lower temperatures (<25°C), however, engorged females may take longer to reach this peak (Sweatman and Koussa 1968). *H. leachi* produced more eggs on a daily basis compared to *R. sanguineus*. Results gained in the present study for *R. sanguineus* were similar to what Sweatman (1967) recorded. A similar pattern has been illustrated for many other ticks species such as *Hyalomma anatolicum anatolicum* (Snow and Arthur 1966), *Amblyomma nitens* (Drummond, Whetstone, Ernst and Gladney 1969), *Boophilus decoloratus* (Dipeolu 1983) and *Amblyomma variegatum* (Arthur 1962), amongst others. Although the daily egg production patterns of *R. sanguineus* and *H. leachi* are fairly similar the number of eggs produced daily is different. The fact that *H. leachi* laid more eggs than *R. sanguineus* is probably due to the fact that female *H. leachi* ticks have a larger average engorgement mass, and thus lay more eggs. The larger number of eggs produced can perhaps be seen as an adaptation to more adverse environmental conditions (Van Der Lingen *et al.* 1999) by increasing the probability of eggs hatching successfully. The same would apply to the rapid developmental rates of eggs (Heath 1979).

*(c) FECUNDITY AND CONVERSION EFFICIENCY INDEX (CEI) VALUES AND THE INFLUENCE OF TEMPERATURE AND RELATIVE HUMIDITY.*

The results of the present study showed a linear correlation between the mass of the eggs (and the number of eggs deposited) and the weight of the engorged female, for both *R. sanguineus* and *H. leachi*. Sweatman and Koussa (1968) and Nager (1968) found the same for *R. sanguineus*. The correlation between the number of eggs laid and the weight of the engorged female (for other species) was also found by other authors to be highly significant (Drummond and Whetstone 1970; Honzakova, Olejnicek, Cerný, Daniel and Dusbabek 1975; Dipeolu, Amoo and Akinboade 1991; Toutoungi, Gern and Aeschlimann 1995). The number of eggs laid depends on the weight of the engorged female (Honzakova, Olejnicek, Cerný, Daniel and Dusbabek 1975). There is, however, a weight below which engorged ixodid females generally do not lay eggs (Nagar 1968). Despite the significance of weight Sweatman and Koussa (1968) found, however, that the period taken to lay the eggs is only affected by the temperature (and not the weight). Weight, as an influencing factor, was not specifically tested in the present study. *R. sanguineus*, *Dermacentor variabilis* (Nagar 1968) and the *Hyalomma* species (Arthur 1962) are examples of species for which this is true. Semi-engorged ticks also lay eggs (Honzakova, Olejnicek, Cerný, Daniel and Dusbabek 1975) but the CEI and the potential production of viable eggs increases only with an increased engorgement weight (Toutoungi *et al* 1995). The entire engorged mass is not used for egg production. This implies that the remainder of the engorgement weight is used for other metabolic processes (Bassal and Hefnawy 1972; Dipeolu 1983).

While both species produced a large number of eggs, the fact that *H. leachi* generally laid more than *R. sanguineus* (i.e. per set of conditions), had not significantly affected the fecundity. The lowest fecundity values were recorded at lower temperatures. There, however, appeared to be no conspicuous relationship between fecundity and the temperature or relative humidity. Similar results have been obtained for other species such as *Hyalomma truncatum* (12.6 eggs/mg body mass) (Linthicum, Logan, Kondig, Gordon and Bailey 1991), *Ixodes hexagonus* (5.2 eggs/mg body mass) (Toutoungi, Gern and Aeschlimann 1995), *I. ovatus* (12.4 eggs/mg body mass) (Fujimoto 1989), *I. ricinus* (7.7 eggs/mg body mass) (Honzákova, Olejnicek, Cerný, Daniel and Dusbábek 1975) and *I. rubicundus* (17.7 eggs/mg body mass) (Van Der Lingen, Fourie, Kok and Van Zyl 1999). The laying of large numbers of eggs can possibly be seen as an adaptation to harsh environments, which increases the surviving numbers.

The results of the present study showed that *R. sanguineus* and *H. leachi* converted large percentages of their engorgement body mass into eggs. There is a trend for the CEI in the case of *R. sanguineus* and *H. leachi* to increase with an increase in relative humidity. These measurements may, however, be artefactual, since at higher temperatures moisture loss from eggs would have been considerably greater, resulting in lower values. The CEI for *R. sanguineus* and *H. leachi* found were not unlike those for other ixodid species exposed to similar temperatures (20-35°C) and relative humidities (>80%). For example, the CEI value for *Amblyomma hebraem* is 65% (Norval 1977), *Boophilus annulatus* is 53.3% (Davey 1993), *Hyalomma dromedarii* is 74% (Hagras and Khalil 1988) and *Ixodes hexagonus* is 40.3% (Toutoungi *et al.* 1995). Both *R. sanguineus* and *H. leachi* appear to be adapted for the environments in which they occur. By producing more eggs through efficient body mass conversion both species have increased the chances of survival of the next tick generation.

Research on the egg stage has been generally neglected despite the fact that information about it is of great importance. It is the only immobile stage in the life cycle of the tick and the numbers laid, in conjunction with the percentage hatch, is crucial for the propagation of the tick's generation (Dipeolu, Amoo and Akinboade 1991). The present study has provided some basic information regarding the biology of egg laying of both *R. sanguineus* and *H. leachi*. The fact that these ticks are both relatively widely distributed and have disease spreading abilities makes their control of significant importance. Basic information on the biology of these tick species, such as generated and investigated in the present study, which can aid in the formulation of integrated tick control strategies, is of paramount importance.

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# CHAPTER 4

## DEVELOPMENTAL BIOLOGY - LARVAE

# CHAPTER 4

# DEVELOPMENTAL BIOLOGY - LARVAE

## INTRODUCTION

For a tick to survive, it has to be able to conserve water. This is particularly important in humidity conditions that are ever fluctuating. Each of the four stages in the life cycle of a tick has its own moisture requirements (Heath 1981). This, and the fact that temperature affects ticks, makes it crucial that a tick is able to find a host as quickly as possible in order for the next generation to be successful (Amoo and Dipeolu 1985). Finding a host is made easier when the tick drops off in an area where the host is most likely to be found. This requires synchronisation between the tick's detachment and the activity of the host, which increases the odds that the tick will find a host (Yeruham, Hadani, Galker, Rosen and Gunders 1995). When the adult tick detaches, it finds itself in a microclimate. Although the adult is capable of surviving harsh conditions, its eggs and larvae (which hatch form those eggs) may not survive those conditions. In order to understand more about a tick's microclimatic requirements it is important to learn more about the water balance of the various stages, such as the unfed, newly hatched larvae, and newly moulted nymphs and adults (Knülle 1966).

Laboratory investigations provide useful information regarding the ecological requirements of a tick species (Heath 1981). A tick's survival under variable field conditions requires a suitable temperature and a high humidity (Hair and Howell 1970; Sauer and Hair 1971), and much information on the requirements can be obtained in the laboratory. Although *Rhipicephalus sanguineus* has a very wide geographic distribution, very little work has been done on the biology of the larvae in comparison to some other species. Even less has been done in South Africa. This is also true for *Haemaphysalis leachi* and very little literature is available on its biology.

*AIMS AND OBJECTIVES*

The objectives of the present study were to determine:

(a) the extent to which temperature and relative humidity influence the survival of unfed (flat) larvae;

(b) the extent to which the temperature and relative humidity influence the pre-moult period and moulting success of engorged larvae;

(c) the extent to which varying periods of desiccation influence the moulting success of engorged larvae;

(d) whether or not a circadian drop-off rhythm exists for fully engorged larvae which detach from their host.

# MATERIALS AND METHODS

*(a) SURVIVAL OF FLAT LARVAE*

Engorged females of *R. sanguineus* and *H. leachi* were obtained in the same manner as described in Chapter 3. Eggs were kept at 25°C and 90±2%RH until they hatched. The newly hatched larvae were then kept at the same conditions for a period of 12 days to allow the cuticle and mouth parts to harden. Exposure at a favourable humidity gives the cuticle time to undergo age related changes that diminish permeability (Heath 1981). The larvae were then transferred to glass vials (10ml), 20 larvae in each, which were covered with gauze to allow free airflow. The glass vials were placed inside one-litre plastic containers and covered with black plastic. These one-litre bottles were then allocated to each of the different temperature and relative humidity regimes (Fig. 4.1). The larvae in these containers were then observed every second day, at the same time each day, until all the larvae had died. The time taken for all 20 larvae to die was recorded.

*(b) PRE-MOULT PERIOD AND MOULTING SUCCESS*

Flat *R. sanguineus* and *H. leachi* larvae were placed on Beagle dogs and field mice (*Rhabdomys pumilio*), respectively, to feed to repletion. Fully engorged larvae were collected in a container placed below the different cages in which the dogs and mice were kept (described in detail in section d, on circadian rhythms). The fully engorged larvae that were collected were placed in 10ml glass vials (10 larvae per vial) and covered with plastic gauze to allow free airflow. One vial was then kept in the dark placed at each of different temperature and relative humidity regimes as illustrated in Fig. 4.1. Observations were made every second day, at the same time each day, to determine the moulting success at the different temperature and humidity regimes. Larvae were only regarded as being dead when they became discoloured (black), and if they did not display any locomotor activity in response to the heating of the vial by hand or by breathing on the tick. To remove all uncertainty, regarding the larvae thought to be dead, observations were continued for another two weeks (Koch and Tuck 1986).

*(c) MOULTING SUCCESS OF LARVAE EXPOSED TO VARYING RELATIVE HUMIDITIES*

Flat larvae of both species were fed on dogs as previously described. The fully engorged larvae were divided into four groups, namely, A, B, C and D. Each group consisted of five glass vials (10ml) with 20 engorged larvae in each. Both groups A and B were exposed to the same conditions of temperature and relative humidity but in a different order. The ticks in group A were first placed in a desiccator (25°C and 0%RH) and then transferred to a higher humidity (25°C and 90±2%RH). The period of exposure to the low humidity varied from one to five days for each sub-group. The ticks in group B were first placed in a container with a 90±2%RH and then in the desiccator. The period of exposure to the high humidity varied between one to five days for each sub group. Observations were made every second day to determine if any engorged larvae had moulted and also to determine the moulting success. Larvae belonging to group C were exposed to only 0%RH and group D larvae only to 90±2%RH.

*(d) DETERMINATION OF POSSIBLE DROP-OFF RHYTHMS OF LARVAE*

Fully engorged *R. sanguineus* and *H. leachi* females were collected as described previously. They were allowed to lay eggs at 25°C (90±2%RH). These eggs were allowed to hatch under the same conditions. Two week old larvae were exposed to

20°C and 70±2%RH at a photoperiod of 14L:10D for one week and subsequently fed on Beagle dogs using the same photoperiod. Under the supervision of a veterinarian, the dogs were anaesthetised with Rompun® (Bayer; Active ingredient: Xylazine 20mg/ml) and the larvae then placed on the dog and allowed to disperse on their own. The reason for the use of the drug was to prevent the dogs from scratching off any of the larvae during the initial period of attachment. A total of 200 larvae were placed on each dog and two dogs were used per tick species. The number of engorged larvae, which detached, were counted and taken as a percentage of the number of ticks retrieved. The cages in which the dogs were kept had a specially designed base made from wooden strips, which were positioned so as to allow the engorged larvae to fall through into the tray below. The engorged larvae were prevented from escaping by a water-filled channel on the border of the tray. The ticks, which had fallen through into the tray below, were collected every two hours in the case of the dogs. This was done by washing them into a bucket below and through a sieve to remove and count the engorged larvae that had been retrieved. This process was repeated three times for each species and the data for each species was pooled. The same process was followed, using mice instead of dogs, for both species but the engorged larvae were collected every hour. This was repeated three times for each species and proved to be unsuccessful for *R. sanguineus*. In the case of *H. leachi*, which was successful, the data were pooled.

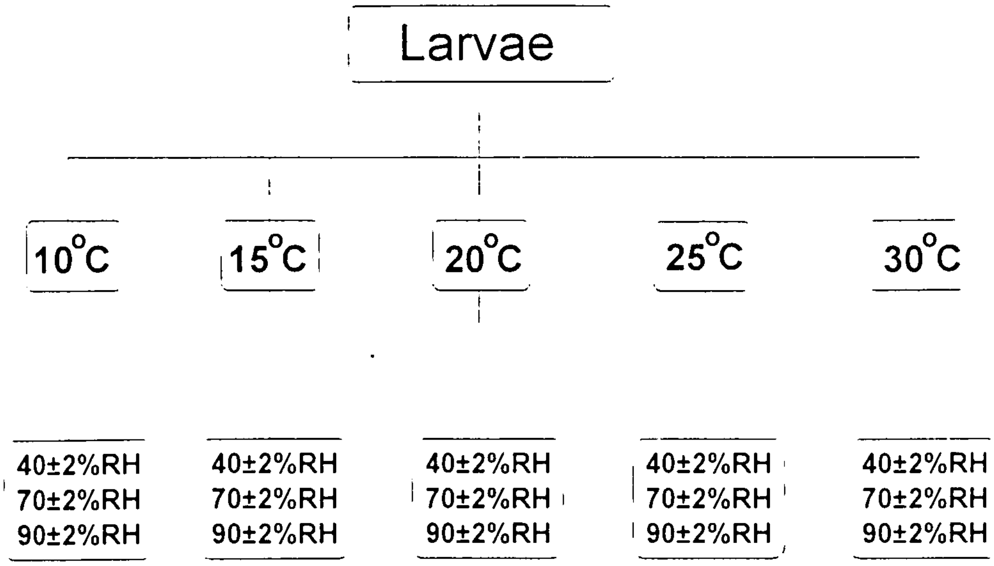


**Figure 4.1.** Different combinations of temperature and relative humidities to which flat and engorged *Rhipicephalus sanguineus* and *Haemaphysalis leachi* larvae were exposed.

# RESULTS

*(a) SURVIVAL OF FLAT LARVAE*

The mean survival times of *R. sanguineus* flat larvae exposed to different temperature and relative humidity regimes are summarised in Table 4.1(a) and presented graphically in Fig 4.2. The mean survival time increased with an increase in relative humidity and decreased with an increase in temperature. The longest mean survival time (133.5 days) was recorded at 15°C and 90±2%RH. The shortest mean survival time (11 days) was recorded at 30°C and 40±2%RH. At 10°C and 90±2%RH the survival rate decreased unexpectedly.

**Table 4.1(a).** Summary of the mean survival time (days) of flat *R. sanguineus* larvae exposed to different temperatures and relative humidities.

| TEMP. (°C) | RH (±2%) | Min. no. of Days | Max. no. of Days | Mean. no. of Days | n |
|---|---|---|---|---|---|
| 10 | 90 | 13 | 45 | 29.0 | 20 |
| 10 | 70 | 85 | 160 | 122.5 | 20 |
| 10 | 40 | 20 | 52 | 36.0 | 20 |
| 15 | 90 | 96 | 171 | 133.5 | 20 |
| 15 | 70 | 48 | 84 | 66.0 | 20 |
| 15 | 40 | 8 | 32 | 20.0 | 20 |
| 20 | 90 | 61 | 116 | 88.5 | 20 |
| 20 | 70 | 33 | 66 | 49.5 | 20 |
| 20 | 40 | 7 | 25 | 16.0 | 20 |
| 25 | 90 | 54 | 81 | 67.5 | 20 |
| 25 | 70 | 17 | 35 | 26.0 | 20 |
| 25 | 40 | 3 | 20 | 11.5 | 20 |
| 30 | 90 | 48 | 76 | 62.0 | 20 |
| 30 | 70 | 17 | 34 | 25.5 | 20 |
| 30 | 40 | 3 | 19 | 11.0 | 20 |

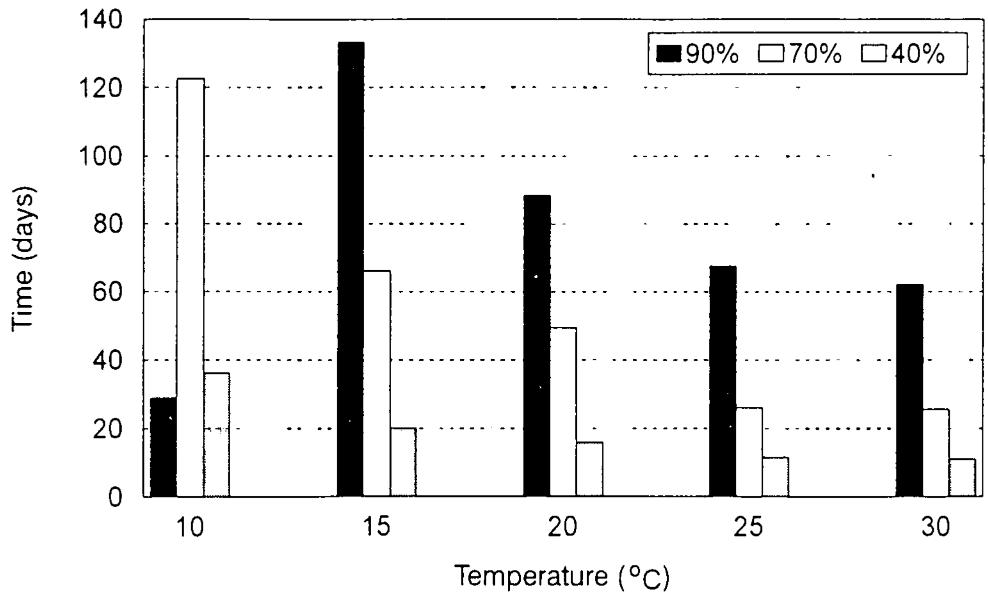


**Figure 4.2.** Mean survival time (days) of *R. sanguineus* larvae exposed to different temperatures and relative humidities.

The mean survival times for *H. leachi* flat larvae exposed to different temperature and relative humidity regimes are summarised in Table 4.1(b) and presented graphically in Fig 4.3. The survival time generally decreased with an increase in temperature and increased with an increase in relative humidity. The longest mean survival time (188 days) was recorded at 15°C and 90±2%RH. The shortest survival time (41 days) was recorded at 25°C; 40±2%RH and 30°C; 40±2%RH. This was an indication that a low relative humidity influenced the survival of *H. leachi* negatively.

**Table 4.1(b).** Summary of the survival time (days) of flat *H. leachi* larvae exposed to different temperatures and relative humidities.

| TEMP. (°C) | RH (±2%) | Min. no. of Days | Max. no. of Days | Mean. no. of Days | n |
|---|---|---|---|---|---|
| 10 | 90 | 50 | 254 | 152.0 | 20 |
| 10 | 70 | 50 | 207 | 128.5 | 20 |
| 10 | 40 | 40 | 56 | 48.0 | 20 |
| 15 | 90 | 152 | 224 | 188.0 | 20 |
| 15 | 70 | 110 | 165 | 137.5 | 20 |
| 15 | 40 | 36 | 50 | 43.0 | 20 |
| 20 | 90 | 81 | 150 | 115.5 | 20 |
| 20 | 70 | 80 | 106 | 93.0 | 20 |
| 20 | 40 | 36 | 51 | 43.5 | 20 |
| 25 | 90 | 102 | 134 | 118.0 | 20 |
| 25 | 70 | 36 | 50 | 43.0 | 20 |
| 25 | 40 | 34 | 48 | 41.0 | 20 |
| 30 | 90 | 122 | 164 | 143.0 | 20 |
| 30 | 70 | 56 | 82 | 69.0 | 20 |
| 30 | 40 | 34 | 48 | 41.0 | 20 |

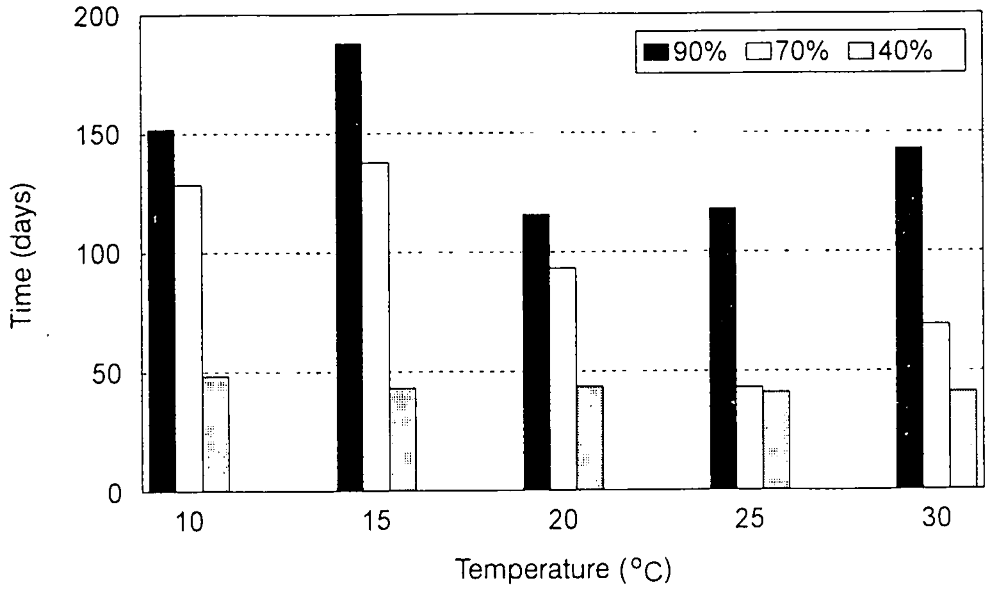


**Figure 4.3.** Mean survival time (days) of *H. leachi* larvae exposed to different temperatures and relative humidities.

*(b) PRE-MOULT PERIOD AND MOULTING SUCCESS*

Results on the pre-moult period and moulting success of engorged *R. sanguineus* and *H. leachi* larvae are summarised in Table 4.2. In the case of *R. sanguineus*, none of the larvae moulted at 10°C. At 15°C, larvae moulted within 33-38 days; at 20°C within 17-19 days; at 25°C within 9-14 days and at 30°C within 8-11 days. Moulting success ranged from 60% at 15°C (70±2%RH) and 25°C (40±2%RH) to 100% at 25°C (90±2%RH) and 15°C (70±2%RH).

In the case of *H. leachi*, none of the larvae moulted at 40±2%RH, at all temperatures. At 10°C larvae moulted within 149-192 days; at 15°C within 29-50 days; at 20°C within 16-25; at 25°C within 12-17 days and at 30°C within 10-18 days. With the exception of zero at 40±2%RH the moulting success ranged from 10% at 10°C (70±2%RH) to 100% at 15°C (90±2%RH) and 30°C (90±2%RH).

**Table 4.2.** Summary of the pre-moult period and moulting success of engorged *R. sanguineus* and *H. leachi* larvae which were exposed to varying temperatures and relative humidities.

| TEMP. (°C) | RH (±2%) | PRE-MOULT PERIOD (DAYS) | | MOULTING SUCCESS (%) | n |
|---|---|---|---|---|---|
| | | MIN. | MAX. | | |
| ***R. sanguineus*** | | | | | |
| 10 | 90 | 0 | 0 | 0 | 10 |
| 10 | 70 | 0 | 0 | 0 | 10 |
| 10 | 40 | 0 | 0 | 0 | 10 |
| 15 | 90 | 35 | 38 | 60 | 10 |
| 15 | 70 | 35 | 36 | 100 | 10 |
| 15 | 40 | 33 | 34 | 70 | 10 |
| 20 | 90 | 17 | 19 | 80 | 10 |
| 20 | 70 | 17 | 19 | 80 | 10 |
| 20 | 40 | 17 | 19 | 90 | 10 |
| 25 | 90 | 9 | 14 | 100 | 10 |
| 25 | 70 | 9 | 14 | 90 | 10 |
| 25 | 40 | 9 | 11 | 60 | 10 |
| 30 | 90 | 8 | 11 | 90 | 10 |
| 30 | 70 | 8 | 11 | 80 | 10 |
| 30 | 40 | 8 | 11 | 70 | 10 |
| ***H. leachi*** | | | | | |
| 10 | 90 | 149 | 176 | 20 | 10 |
| 10 | 70 | 192 | 192 | 10 | 10 |
| 10 | 40 | 0 | 0 | 0 | 10 |
| 15 | 90 | 29 | 30 | 100 | 10 |
| 15 | 70 | 34 | 50 | 90 | 10 |
| 15 | 40 | 0 | 0 | 0 | 10 |
| 20 | 90 | 16 | 23 | 80 | 10 |
| 20 | 70 | 19 | 25 | 90 | 10 |
| 20 | 40 | 0 | 0 | 0 | 10 |
| 25 | 90 | 12 | 16 | 90 | 10 |
| 25 | 70 | 14 | 17 | 60 | 10 |
| 25 | 40 | 0 | 0 | 0 | 10 |
| 30 | 90 | 10 | 18 | 100 | 10 |
| 30 | 70 | 10 | 18 | 80 | 10 |
| 30 | 40 | 0 | 0 | 0 | 10 |

The relationship between the reciprocal of the pre-moult period and the temperature, for *R. sanguineus* (Fig. 4.4), was given by the following relationship:

$y = a + bx$

where $a = -0.0679684$

$b = 0.0063925$

$y =$ 1/moulting time (days)

$x =$ temperature in °C

$R^2 = 97.74\%$

The relationship between the reciprocal of the pre-moult period and the temperature, for *H. leachi* (Fig. 4.5), was given by the following relationship:

$y = a + bx + cx^2$

where $a = -0.09216$

$b = 0.01093$

$c = -0.0001804$

$y =$ 1/ moulting time (days)

$x =$ temperature in °C

$R^2 = 92.95\%$

The regression equations for the two tick species are graphically presented in Figs. 4.4 & 4.5. The Critical Development Temperature was 10.6°C and 10.1°C for *R. sanguineus* and *H. leachi*, respectively.

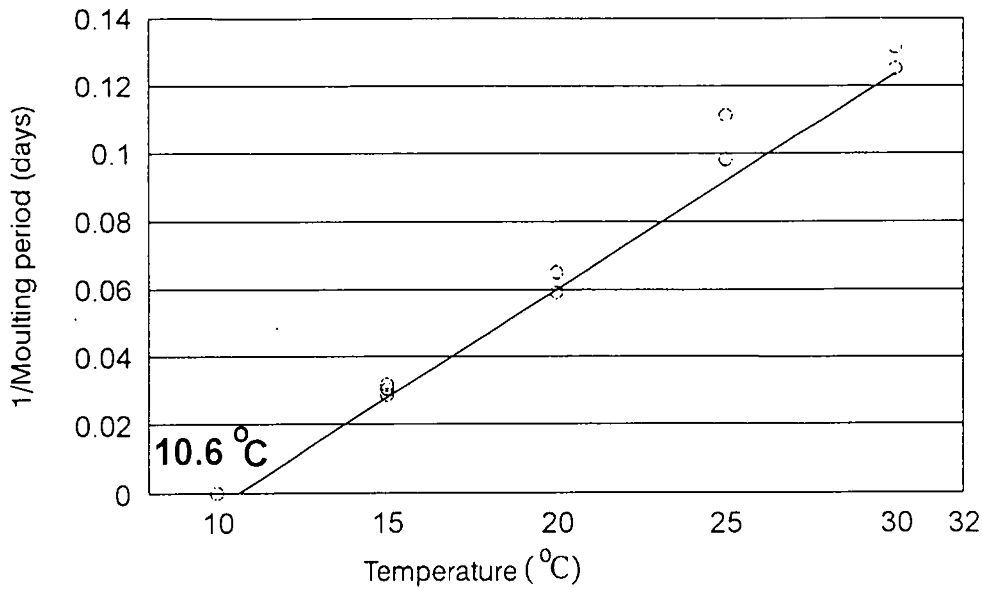


**Figure 4.4.** Graphical representation of the relationship between the reciprocal of the pre-moult period (days) and the temperature (°C) for *R. sanguineus* larvae at 90±2%RH. The critical temperature is 10.6 °C.

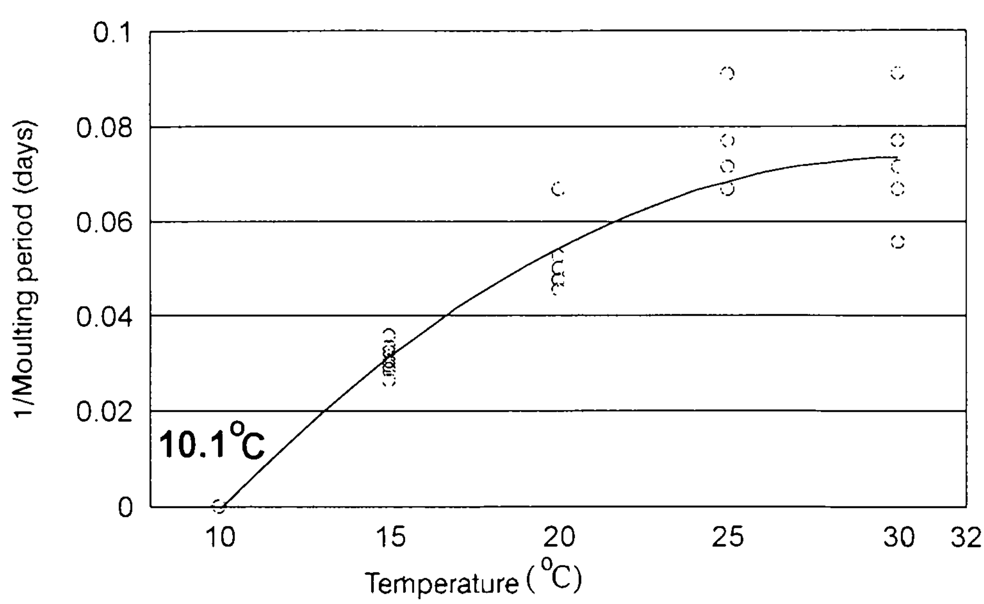


**Figure 4.5.** Graphical representation of the relationship between the reciprocal of the pre-moult period (days) and the temperature (°C) for *H. leachi* larvae at 90±2%RH. The critical temperature is 10.1 °C.

*(c) MOULTING SUCCESS OF LARVAE EXPOSED TO VARYING RELATIVE HUMIDITIES*

The group of *R. sanguineus* larvae exposed constantly to either a low (0%, group C) or high (90%, group D) relative humidity had a moulting success of 5 and 80%, respectively (Table 4.3a). The minimum and maximum time it took for the larvae to moult for the above two groups was 14 and 11-14 days, respectively. Moulting success for both groups A and B varied with values ranging between 10-75% and 15-45%, respectively. The mean pre-moult periods for groups A and B varied between 13-13.5 and 11-13 days, respectively (Table 4.3a).

**Table 4.3(a).** Summary of the moulting success of *R. sanguineus* larvae which were exposed to a constant temperature of 25°C and varying relative humidities (namely, High: 90%; Low: 0%), in total darkness.

| GROUP* | Exposure period (days) | X (days) | MIN. (days) | MAX. (days) | Moulting success (%) | n |
|---|---|---|---|---|---|---|
| A | 0%RH | | | | | |
| | 1 | 13.0 | 11 | 15 | 68 | 19 |
| | 2 | 13.5 | 11 | 16 | 75 | 20 |
| | 3 | 13.5 | 11 | 16 | 68 | 19 |
| | 4 | 13.0 | 11 | 15 | 53 | 19 |
| | 5 | 13.0 | 11 | 15 | 10 | 20 |
| B | 90±2%RH | | | | | |
| | 1 | 11.0 | 11 | 11 | 15 | 20 |
| | 2 | 11.5 | 11 | 12 | 25 | 20 |
| | 3 | 12.0 | 11 | 13 | 25 | 20 |
| | 4 | 13.0 | 11 | 15 | 45 | 20 |
| | 5 | 13.0 | 11 | 15 | 45 | 20 |
| | | | | | | |
| C | 0%RH | 14 | 14 | 14 | 5 | 20 |
| D | 90±2%RH | 12.5 | 11 | 14 | 80 | 20 |

* Group A: 0%RH for 1-5 days and 90%RH until moult
B: 90%RH for 1-5 days and 0%RH until moult
C: constant 0%RH
D: constant 90%RH

The group of *H. leachi* larvae exposed constantly to either a low (0%, group C) or high (90%, group D) relative humidity had a moulting success of 0 and 95%, respectively (Table 4.3b). The minimum and maximum time it took for the larvae to moult for group B was 11-20 days. Moulting success for group A was zero and varied for group B with values ranging between zero to 10%. The pre-moult period for group B was 12 days. The results indicate that relative humidity has a significant effect on the moulting success of *H. leachi*.

**Table 4.3(b).** Summary of the moulting success of *H. leachi* larvae that were exposed to a constant temperature of 25°C and varying relative humidities (namely, High: 90%; Low: 0%), in total darkness.

| GROUP* | Exposure period (days) | X (days) | MIN. (days) | MAX. (days) | Moulting success (%) | n |
|---|---|---|---|---|---|---|
| A | 0%RH | | | | | |
| | 1 | 0 | 0 | 0 | 0 | 20 |
| | 2 | 0 | 0 | 0 | 0 | 20 |
| | 3 | 0 | 0 | 0 | 0 | 20 |
| | 4 | 0 | 0 | 0 | 0 | 20 |
| | 5 | 0 | 0 | 0 | 0 | 20 |
| B | 90±2%RH | | | | | |
| | 1 | 0 | 0 | 0 | 0 | 20 |
| | 2 | 0 | 0 | 0 | 0 | 20 |
| | 3 | 0 | 12 | 12 | 5 | 20 |
| | 4 | 0 | 0 | 0 | 0 | 20 |
| | 5 | 14 | 14 | 14 | 10 | 20 |
| | | | | | | |
| C | 0%RH | 0 | 0 | 0 | 0 | 20 |
| D | 90±2%RH | 13 | 11 | 20 | 95 | 20 |

* Group A: 0%RH for 1-5 days and 90%RH until moult
B: 90%RH for 1-5 days and 0%RH until moult
C: constant 0%RH
D: constant 90%RH

*(d) DETERMINATION OF POSSIBLE DROP-OFF RHYTHMS OF LARVAE*

The larvae of both *R. sanguineus* and *H. leachi* only started detaching three days after being placed on their host. Most of the larvae detached during the daytime (Fig. 4.6). The highest drop-off percentage (19.7%) was recorded at 3pm. In the case of *H. leachi* larvae fed on dogs the larvae detached during both the photo and scoto-phases. No specific pattern was discernible (Fig. 4.7). With regard to the *H. leachi* fed on mice, most of the ticks detached during the photo-phase with peak detachment periods recorded between 6:00-8:00am and 2:00-6:00pm (Fig. 4.8). The highest drop-off percentages were 5.3% (at 18:00pm), 10% (at 6:00am) and 16.3% (at 16:00pm), respectively.

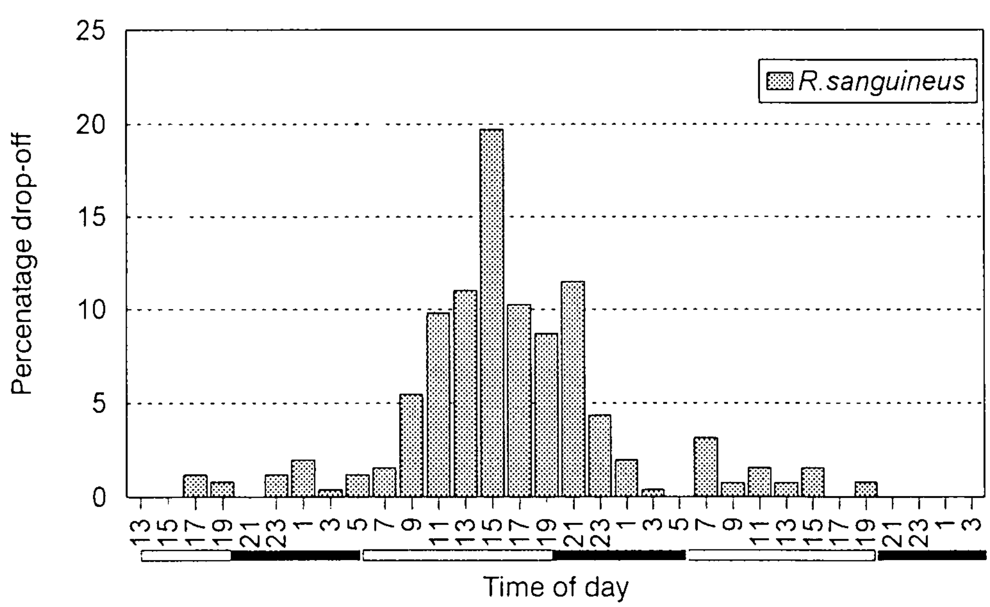


**Figure 4.6.** Histogram showing the time specific detachment pattern of *R. sanguineus* larvae, fed on dogs. (The horizontal bar indicates the light and dark phases viz. 14L:10D)

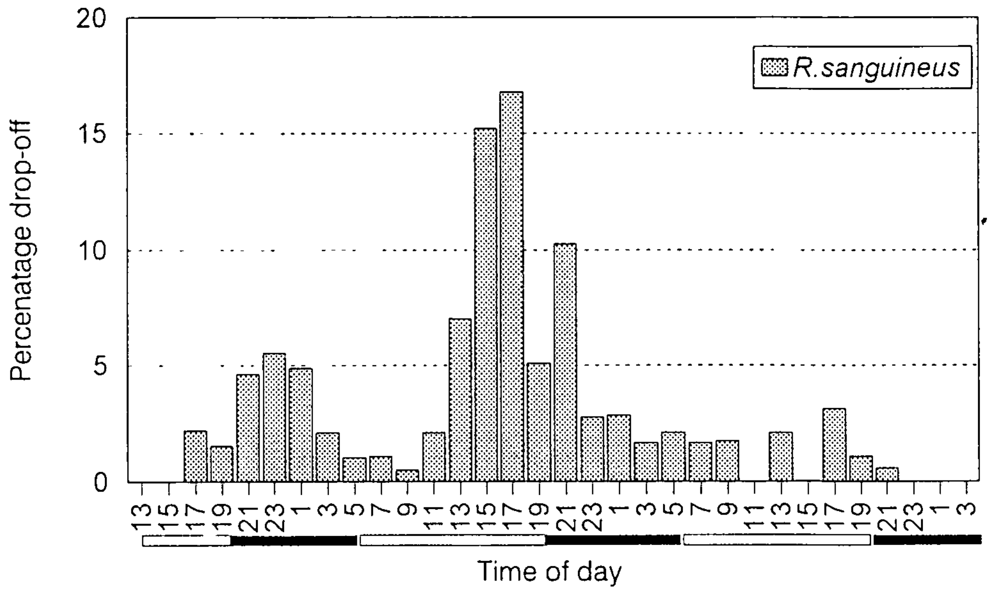


**Figure 4.7.** Histogram showing the time specific detachment pattern of *H. leachi* larvae, fed on dogs. (The horizontal bar indicates the light and dark phases viz. 14L:10D)

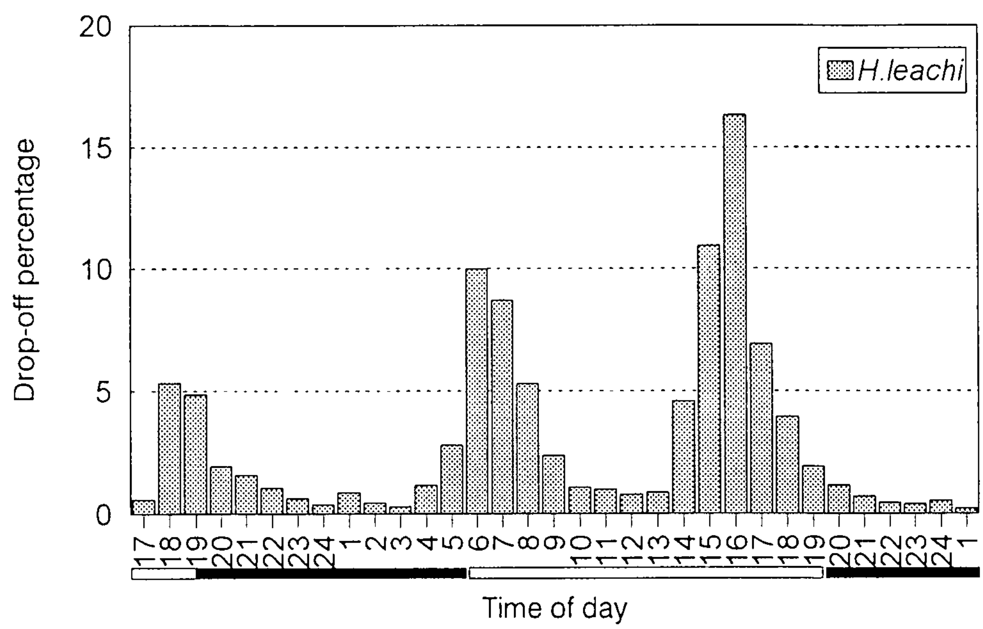


**Figure 4.8.** Histogram showing the time specific detachment pattern of *H. leachi* larvae, fed on mice. (The horizontal bar indicates the light and dark phases viz. 14L:10D)

# DISCUSSION

### *(a) SURVIVAL OF FLAT LARVAE*

In general the trend for both *R. sanguineus* and *H. leachi* was an increase in survival time with a decrease in temperature and an increase in relative humidity. The only exception to this was *R. sanguineus*, which displayed a decrease in survival time at 10°C (90±2%RH). This is possibly an indication that this tick is not adapted to survive in conditions where the condensation is very high. The maximum survival times for *R. sanguineus* and *H. leachi* larvae were recorded at lower temperatures. The shortest survival times for *R. sanguineus* and *H. leachi* larvae were recorded at higher temperatures but in combination with low relative humidities. According to Amoo and Dipeolu (1985), extremely low humidities cause the death of larvae, which had not yet found a host, but the survival increases with an increase in relative humidity or when they are allowed to feed. This trend has been found to be true for other species also, although each species appears to have a different threshold, below which its longevity rapidly declines. Zahler and Gothe (1995) showed that *Dermacentor reticulatus* displays similar results to *R. sanguineus* under similar conditions. At a temperature of 27°C and relative humidity ranging between 15-50%RH, *D. reticulatus* larvae survived for only one day. Hitchcock (1955) showed that *Boophilus microplus* can survive for up to 240 days at 22°C and 90%RH but only for up to 12 days when exposed to 70%RH. The longevity of *Amblyomma americanum* larvae also drops to a few days when exposed to 69%RH (Lancaster and McMillan 1955). Arthur (1951) showed that *Ixodes hexagonus* survived for up to 15 months at humidities close to saturation but died rapidly under dry conditions. The present study has shown that at all temperatures and relative humidities, the longevity of *H. leachi* larvae, in general, was longer compared to that of *R. sanguineus*. This may be indicative of a higher resistance to desiccation for *H. leachi*. Generally both these species appear to be adapted to harsher conditions.

It is a well-established fact that ticks can starve for considerable lengths of time. This ability to survive in the absence of a host depends mainly on humidity levels. Above certain humidities, ticks can survive for long periods (months, even years). If the humidity is below a certain level then ticks die in a relatively short period (Knülle 1966). The effect of temperature on flat larvae is far greater than on eggs, and primarily determines their survival time (Zahler and Gothe 1995) but there are other contributing factors. These factors include examples such as the ability of non-engorged larvae to absorb atmospheric water or to imbibe water from a host's tissues (Yoder and Spielman 1992). Larvae, which are not on a host, survive best at high relative humidities and only tolerate low humidities for very short periods of time (Zahler and Gothe 1995).

The surface area to volume ratio of larvae is greater than that of adults. This implies that larvae may be more susceptible compared to nymphs and adults (Dipeolu 1984), and the fact that larvae feed so briefly (Needham and Teel 1986, 1991), make the availability of water a critical limiting factor in a tick's distribution. Dipeolu (1984) studied the effect of different conditions in the field on the life cycles of several different species. This included all stages of the life cycle and compared the effect of the dry season and the rainy season. The results of his study have shown that the maximum life cycle was shortened during the dry season. This indicated that temperature and rainfall have an effect on hatching patterns, the various developmental patterns and the degree of survival of those forms. The differences in the life cycles recorded were largely the result of variation during the egg and larval stage of the life cycle. This was shown to be true for *Amblyomma variegatum*, *Boophilus decoloratus*, *Hyalomma rufipes* and *Hyalomma truncatum*. These results indicate that the earlier stages of the life cycle are more susceptible to environmental conditions and can have a negative effect on the life cycle as a whole if the eggs, and thus larvae too, are exposed to unfavourable conditions. Koch and Tuck (1986) compared *R. sanguineus* adults and nymphs and clearly showed that adults are far less susceptible to dehydration. When comparing these results to those obtained by others like Gothe and Hamel (1973) then it can be said that larvae are certainly the most susceptible to dehydration. This can indeed be explained by the difference in the surface area to volume ratio of the different developmental forms and would have certain implications in terms of mortality. If large numbers of eggs are laid to produce large numbers of larvae and if the mortality rate is high as a result of the larvae being more susceptible, then the large numbers of larvae can be seen as an adaptation to harsher conditions.

***(b) PRE-MOULT PERIOD AND MOULTING SUCCESS***

The general trend for both *R. sanguineus* and *H. leachi* was a decrease in the duration of the pre-moult period with an increase in temperature. This is a general phenomenon for many tick species (Heath 1981; Koch and Tuck 1986). Within a particular temperature, relative humidity did not appear to have any effect on the pre-moult period of *R. sanguineus* but it did have a definite effect on *H. leachi*. No moulting takes place at 40%RH. The critical developmental temperature (CDT) for both species was higher than 10°C, which explains the fact that none of the *R. sanguineus* ticks moulted at 10°C.

The tolerance that *R. sanguineus* displays towards desiccating conditions is a major factor contributing to its success in the more arid environments (Koch and Tuck 1986). The weight loss of *R. sanguineus* larvae increases with temperature and they can lose up to 43.1% by the time of moulting (Heath 1981). In the present study the maximum temperature used was 30°C, compared to the temperatures exceeding 40°C used by Heath (1981), and weight loss during moulting was not taken into account. It is therefore not possible to compare the results of the present study with the specific findings of Koch and Tuck (1986) and Heath (1981). It can, however, be said that *R. sanguineus* displayed a higher moulting success at 30°C (90±2%RH) than at 15 °C (90±2%RH) (Table 4.2). It should be noted that, although a higher humidity is generally more favourable in terms of reduced water loss, at 10°C and even 15°C (to a lesser extent) it proved to be unfavourable for *R. sanguineus*. This was due to the excess moisture build-up resulting from the low rate of evaporation under these conditions. For *H. leachi*, the opposite was true. These ticks displayed a higher moulting success rate at lower temperatures and high humidities. This makes sense because these ticks tend to burrow into the soil where the moisture content is most probably much higher than in the cracks in which *R. sanguineus* prefers to settle (Fourie, Kok and Visagie-unpublished data).

Koch and Tuck (1986) found that a high percentage (84%) of *R. sanguineus* larvae moult at 15%RH and 27°C. In fact, with temperatures and relative humidities ranging from 20-35°C and 35-95%RH, respectively, *R. sanguineus* displayed a moulting success of between 83-100%. With temperatures below 27°C, the larval pre-moult period was between 18.9 and 20.1 days but was as low as 6.2 days at temperatures higher than 27°C. The present study showed that with temperatures below 25°C, the premoult period increased. The maximum temperature used in the present study was 30°C therefore it is possible that the premoult period will be shorter if higher temperatures were used. Engorged larvae are able to moult with an 86% success rate under desiccating conditions of 35°C and relative humidity of 35% as well as at all other temperature-humidity combinations (Koch and Tuck 1986). Koch and Tuck (1986) also found that engorged larvae do not moult at 10°C, which is similar to the observations made in the present study. According to Koch and Tuck (1986) relative humidity does not affect moulting except that larvae take a little longer to moult at 35°C and low humidities. Moulting success of *R. sanguineus* larvae is very high at temperatures from 20 to 35°C and relative humidities of 15 to 95% (Koch and Tuck 1986). The present study compared favourably to the findings of Koch and Tuck (1986).

Heath (1974) found that the theoretical development zero (or CDT) for *R. sanguineus* larvae is near 18°C but Koch and Tuck (1986) found it to be 10.8°C, which would appear to support the CDT found in the present study. The point is a low temperature slows down the development process of all the stages of the life cycle of the tick. For *H. leachi*, the CDT recorded was slightly higher than the lowest temperature used (10°C). However, a low percentage of the larvae moulted at 10°C and 90%RH. This could be explained by the fact that the incubation cabinets do display some variation even if set at a particular temperature. When breeding with ticks the ideal temperature and relative humidity that should be used is 25°C and 90%RH, respectively, as this represents a low risk of desiccation and a reasonably fast developmental rate. However, if long-term storage of ticks is required then the slower developmental rate resulting from lower temperatures (10°C) and the increased survival rate of a high relative humidity (90%) should be used.

The inability of the engorged stages of a tick's life cycle to "control" their water balance is of great significance to a tick's survival (Lees 1946). In this regard, *R. sanguineus* and *H. leachi* larvae show a preference for distinct temperatures and relative humidities when exposed to a wide range of conditions. Essentially, the difference between unfed ticks and engorged ticks is that the unfed ticks are able to move around in search of a more favourable environment whereas the engorged tick has a very limited time frame to do so (Heath 1981). This impacts on the survival rate of engorged larvae and reinforces the need for engorged larvae to detach in a favourable environment.

*(c) MOULTING SUCCESS OF LARVAE EXPOSED TO VARYING RELATIVE HUMIDITIES*

This part of the present study represents a quasinatural scenario in that it mimics the situation in nature where a tick drops off its host in conditions that change from favourable to unfavourable, and vice versa. A tick will not always end up in an ideal environment and the conditions will not always remain constant for the duration of the tick's life cycle.

Group A showed that, *R. sanguineus* displayed moulting success for all temperature-humidity combinations. With results between 10 and 75%, it would be safe to say that these larvae can tolerate or survive exposure to extremely low relative humidities although with reduced success. It is also an indication of the ability of *R. sanguineus* to survive in the more arid regions of the world, although prolonged exposure will undoubtedly have deleterious consequences. This ability to survive low humidities could be due to the fact that immature engorged ticks are able to produce metabolic water (Kahl 1991). For *H. leachi*, Group A, which was first exposed to desiccation, failed to successfully moult. It should be noted that low relative humidities only affect the length of the premoult period and not the moulting success (Heath 1974). This suggests that *H. leachi* larvae do not have the ability to recover from a spell of low humidity and that exposure to low humidities results in a sharp increase in mortality. Group B, which was first placed in a high relative humidity environment, also displayed little success but managed to moult after a three- and five-day exposure to high humidity. Van Der Lingen (1995) obtained similar results in a study done on *Ixodes rubicundus*. His results showed that exposure to a low relative humidity (26%RH) directly after engorgement or just before ecdysis resulted in an increased moulting period and mortality rate. Furthermore, engorged larvae are sensitive to dehydration at any point during the moulting period but that the longer the exposure the greater the mortality rate (Van Der Lingen 1995). This suggests that (despite the fact that immature ticks often detach in an ideal environment) should the conditions change in nature so that the humidity drops dramatically, then the moulting success will drop. However, if the exposure to ideal conditions was adequate then the tick can still moult and survive. This would explain why *H. leachi* has a tendency to burrow into the soil (Fourie, Kok and Visagie-unpublished data), which possibly provides prolonged high humidity conditions. This, in turn, favours the tick's survival.

Ticks have to contend with fluctuating and unpredictable conditions in terms of temperature and relative humidity. It is even more difficult to quantify tick survival under these conditions in nature because of the overlap in terms of seasons and conditions. Limiting factors can, however, be identified through studies under controlled conditions in the laboratory.

*(d) DETERMINATION OF POSSIBLE DROP-OFF RHYTHMS OF LARVAE*

Factors that affect or influence the survival of larvae include temperature and relative humidity. The tick cannot determine these variables. This implies that the environment, where the tick lays its eggs or where engorged larvae and nymphs detach and drop off, has to be favourable for further development. According to Hadani and Rechav (1969) the rhythmic drop-off of engorged *R. sanguineus* larvae is circadian and occurs at the onset of the host's greatest activity. In the present study, *R. sanguineus* larvae that were fed on dogs displayed a diurnal drop-off rhythm, which is in accordance with the observation of Hadani and Rechav (1969).

*H. leachi* that were fed on dogs and mice mainly displayed a diurnal drop-off rhythm. In the case of *H. leachi*, which were fed on mice, there were three distinct peaks. The maximum was recorded late in the afternoon.

Endogenous circadian regulation mechanisms, entrained by exogenous signals, control the detachment of engorged ticks (Hadani and Rechav 1970; Hadani and Ziv 1974). An example of such a signal would be change in light intensity as suggested by Kitaoka (1962). However, the accuracy of the detachment depends on the endogenous rhythm that is influenced by the host's physiological and activity patterns (Rechav 1978). Factors in the host's blood and the adrenal cycle of the host have also been identified as having an influence (Amin 1970). The detachment of *R. sanguineus* larvae mainly during daytime implies that a significant proportion of the ticks will detach at the resting place of the dog, where moulting will take place. It will also maximise the chances of the nymphs to parasitize the same host. This ensures that the larvae, which moult into nymphs, will find a host.

The immature stages of *H. leachi* have a preference for small rodents, in particular the striped mouse, *Rhabdomys pumilio* (Hoogstraal 1956; De Graaff 1981). These mice are broad-niche animals and can be found in grassland areas but also in bushy and semi-dry vlei country (Perrin 1950; De Graaff 1981; Skinner and Smithers 1990). In layman terms they are found in the veld. With regard to their daily activity, they are regarded as being crepuscular with two peaks of activity. The first peak can be observed in the early morning from about 5- 8:30am. The second peak can be observed in the afternoon from about 14:30-17:30pm (De Graaff 1981; Skinner and Smithers 1990). The larvae dropped off their host during the same periods and thus ensure that the emerging nymphs may find a host. The period during which these ticks drop off represents an active period during which the mice are out foraging. This indicates that the chances of finding a different host are greater, which is important in a three-host tick.

Complex relationships have evolved between particular tick species and their natural hosts. A typical example of such a relationship is the detachment rhythm displayed by engorged *Ixodes rubicundus* ticks (Du Toit, Fourie and Horak 1994). This circadian rhythm has both physiological and ecological importance and has been shown to be the case by several authors (Amin 1970; Hadani and Rechav 1970; Hadani and Ziv 1974; Doube 1975; Rechav 1978; Guglielmone 1993). In the case of *Dermacentor reticulatus*, the fact that the larvae are vulnerable is not a problem because the larvae are always in close spatial vicinity to burrow-dwelling or ground-living animals (Zahler and Gothe 1995). This is similar to *H. leachi*, in which case the immatures, utilise rodents as hosts (Hoogstraal 1956). A typical example of a burrowing rodent is the burrowing striped mouse (*Rhabdomys pumilio*) (Perrin 1950; De Graaff 1981; Skinner and Smithers 1990). Larvae may not always be in the ideal environment but at least they are always close to their host (Zahler and Gothe 1995), which highlights the necessity for a synchronised detachment. The preference of *R. sanguineus* to infest the readily available domestic dog in all parasitic stages shortens its host-seeking time and reduces the exposure of the less tolerant stages to conditions of desiccation (Koch and Tuck 1986). This has important consequences regarding the survival of the tick and once again highlights the importance of synchrony in terms of detachment from its host.

As was already mentioned, these so-called drop-off rhythms are correlated with activity or behavioural patterns of the host (Rechav 1978). There is certainly an adaptive advantage to the dropping off of engorged ticks where the likelihood of encountering a suitable host is high. The fowl tick, *Argas miniatus*, only drops off at night when the fowl is on the roost. This makes the same host readily available for the next stage to infest (Hooker 1908 after Amin 1970). Ticks like *Dermacentor variabilis* display drop-off patterns that correspond to the host's peak activity (Amin 1970; McEnroe and Specht 1987). This implies that when the host is outside its nest, in its favourite feeding ground, that its peak activity triggers the detachment of the engorged larvae. Thus when moulting has taken place then the emerging nymphs have a better chance of encountering a host (Amin 1970). This scenario would best suit a tick like *Dermacentor variabilis* because its immature stages have a different host preference. If the tick preferred the same host then it would have been an advantage for the engorged ticks to drop off in the nest of the host. In short, for a tick to survive it has to find itself in a favourable environment, either in terms of temperature and relative humidity or in terms of host availability. This dilemma is overcome through synchronised detachment.

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# CHAPTER 5

## DEVELOPMENTAL BIOLOGY - NYMPHS

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## INTRODUCTION

As in the case of larvae, the conservation of water is crucial for the survival of nymphs. In ever-fluctuating relative humidity conditions nymphs have their own moisture requirements, which differ from that of the other stages in the life cycle of the tick (Heath 1981). The added effect of temperature further affects ticks and emphasises the importance of rapid host location in order for the next generation to be successful (Amoo and Dipeolu 1985). Finding a host is made easier when the tick drops off in an area to which the host is likely to return. In other words, if the tick's detachment rhythm is synchronised with the activity of the host then the chances are good that the tick will find a host more readily (Yeruham, Hadani, Galker, Rosen and Gunders 1995). When the tick detaches, it finds itself in a microclimate in which the conditions will determine whether or not it can survive.

Very little research has previously been done on the biology *R. sanguineus* nymphs. Even less has been done in South Africa. This is also true for *H. leachi* nymphs. Very little literature is available on their biology. Laboratory investigations provide useful information regarding the ecological requirements of a tick species (Heath 1981). A tick's survival under variable field conditions requires a suitable temperature and a high humidity (Sauer and Hair 1971) and much information on the requirements can be obtained under controlled conditions in the laboratory.

*AIMS AND OBJECTIVES*

The objectives of the present study were to determine:

(a) the extent to which temperature and relative humidity influence the survival of unfed (flat) nymphs;

(b) the extent to which the temperature and relative humidity influence the pre-moult period and moulting success of engorged nymphs:

(c) the extent to which varying periods of desiccation influence the moulting success of engorged nymphs;

(d) whether or not a circadian drop-off rhythm exists for fully engorged nymphs which detach from their host.

# MATERIALS AND METHODS

### *(a) SURVIVAL OF FLAT NYMPHS*

Eggs of *R. sanguineus* and *H. leachi* were obtained in the same way as described in Chapter 3 and were kept at 25°C and 90%RH in glass vials (10ml), the tops of which were covered with gauze to allow free air flow. Newly hatched *R. sanguineus* and *H. leachi* larvae (± 14 days old) were allowed to feed on dogs and mice, respectively. The detached engorged larvae were kept at a temperature of 25°C and a 90±2% RH until they moulted. Flat nymphs (± 18 days) were subsequently placed in groups of 20 in glass vials (10ml) as above. The glass vials were placed inside one-litre plastic containers and covered with black plastic. These one-litre bottles were then allocated to each of the different temperature and relative humidity regimes (Fig. 5.1). Observations were made every second day until all the nymphs had died. The time taken for this to happen was noted in each case.

### *(b) PRE-MOULT PERIOD AND MOULTING SUCCESS*

Flat *R. sanguineus* and *H. leachi* nymphs were fed only on dogs (Beagles) and allowed to feed to repletion. The number of nymphs needed for this section of the present study was perceived to be too large for the limited number of mice available at the time. Nymphs of both species were therefore allowed to feed on dogs. Fully engorged nymphs were collected in trays placed beneath the cages in which the dogs were kept. The collected nymphs were subsequently placed in groups of 7 or 8, in 10ml glass vials (as before) and exposed, in the dark, to various temperature and relative humidity regimes (Fig. 5.1). Observations on the number of nymphs that moulted were made every second day and continued until all the nymphs had moulted. Nymphs were regarded as being dead when they became discoloured (black), if they did not display any locomotor activity or if their palps were splayed. This was only regarded as being final after a further two weeks had passed, after the last tick had moulted (Koch and Tuck 1986).

### *(c) MOULTING SUCCESS OF NYMPHS EXPOSED TO VARYING RELATIVE HUMIDITIES*

Flat *R. sanguineus* and *H. leachi* larvae were fed on Beagle dogs and mice, respectively, to repletion. Detached engorged larvae were placed at 25°C and 90%RH in glass vials (10ml) until they moulted. The flat nymphs of both species were subsequently again fed only on dogs to become engorged. The fully engorged nymphs were divided into four groups, namely, A, B, C and D. Each group consisted of five glass vials (10ml) with 20 engorged nymphs in each. Both groups A and B were exposed to the same conditions of temperature and relative humidity but in a different order. The ticks in group A were first placed in a desiccator (25°C and 0%RH) and then transferred to a higher humidity (25°C and 90±2%RH). The period of exposure to the low humidity varied from one to five days for each sub-group. The ticks in group B were first placed in a container with a 90±2%RH and then in the desiccator. The period of exposure to the high humidity varied between one to five days for each sub-group. Observations were made every second day to determine if any engorged nymphs had moulted and also to determine the moulting success. Nymphs in group C were exposed only to 0%RH and those in group D only to 90±2%RH.

*(d) DETERMINATION OF POSSIBLE DROP-OFF RHYTHMS OF NYMPHS*

Flat nymphs of both species were bred as before. Two week old nymphs were exposed to 20°C and 70±2%RH at a photoperiod of 14L:10D for one week before feeding.

The nymphs were fed on Beagle dogs (20°C; 70±2%RH; 14L:10D). Under the supervision of a qualified veterinarian, the dogs were anaesthetised using a drug called Rompun® (Bayer; Active ingredient: Xylazine 20mg/ml). At that point the nymphs were placed on the dog and allowed to disperse on their own. The reason for the use of the drug was to prevent the dogs from scratching off any of the nymphs placed on it. A total of 150 nymphs were placed on each dog and two dogs were used per tick species. The number of engorged nymphs, which fell off, were counted and taken as a percentage of the number used to infest the dog. The cages in which the dogs were kept had a specially designed base made from wooden strips, which were positioned so as to allow the engorged nymphs to fall through into the tray below. The nymphs were prevented from escaping by a water-filled channel around the border of the tray. The engorged ticks, which had fallen through into the tray below, were collected every two hours. This was done by washing them into a bucket below and through a sieve to remove and count the engorged nymphs that had been retrieved. This process was repeated three times for each species.

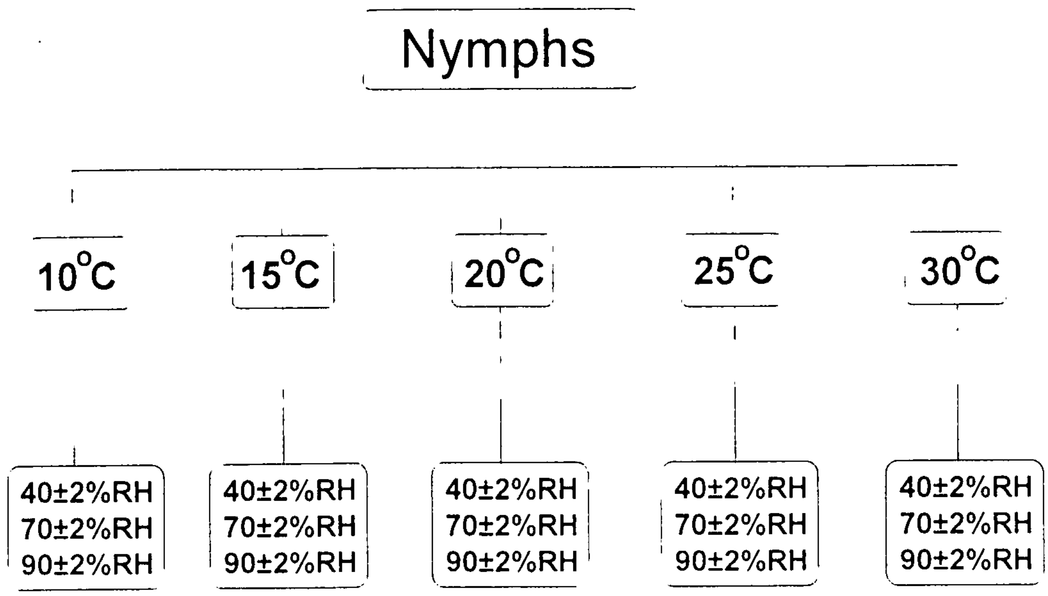


**Figure 5.1.** Different combinations of temperature and relative humidities to which flat and engorged *R. sanguineus* and *H. leachi* nymphs were exposed.

# RESULTS

*(a) SURVIVAL OF FLAT NYMPHS*

The survival times of *R. sanguineus* flat nymphs exposed to different temperatures and relative humidities are summarised in Table 5.1(a) and presented graphically in Fig. 5.2. The mean survival times, in general, decreased with an increase in temperature and increased with an increase in relative humidity, from 40 to 90%. In the case of 10°C (90±2%RH), however, the same pattern was not observed. The longest mean survival time (166.5 days) was recorded at 15°C and 90±2%RH. The shortest mean survival time (22.5 days) was recorded at both 25°C (40±2%RH) and 30°C (40±2%RH).

**Table 5.1(a).** Summary of the survival time (days) of *R. sanguineus* flat nymphs exposed to different temperatures and relative humidities.

| TEMP. (°C) | RH (±2%) | Min. no. of Days | Max. no. of Days | Mean no. of Days | n |
|---|---|---|---|---|---|
| 10 | 90 | 75 | 194 | 134.5 | 20 |
| 10 | 70 | 59 | 209 | 134.0 | 20 |
| 10 | 40 | 32 | 75 | 53.5 | 20 |
| 15 | 90 | 75 | 258 | 166.5 | 20 |
| 15 | 70 | 75 | 151 | 113.0 | 20 |
| 15 | 40 | 32 | 59 | 45.5 | 20 |
| 20 | 90 | 75 | 145 | 110.0 | 20 |
| 20 | 70 | 32 | 79 | 55.5 | 20 |
| 20 | 40 | 25 | 37 | 31.0 | 20 |
| 25 | 90 | 26 | 88 | 57.0 | 20 |
| 25 | 70 | 19 | 53 | 36.0 | 20 |
| 25 | 40 | 19 | 26 | 22.5 | 20 |
| 30 | 90 | 53 | 71 | 62.0 | 20 |
| 30 | 70 | 19 | 31 | 25.0 | 20 |
| 30 | 40 | 19 | 26 | 22.5 | 20 |

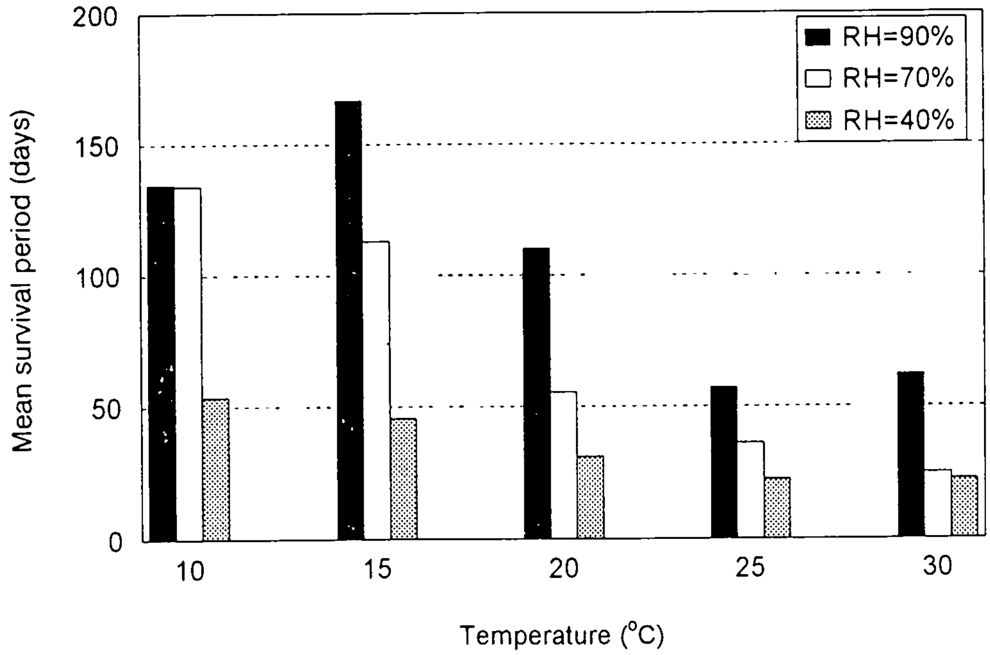


**Figure 5.2.** Mean survival times (days) of *R. sanguineus* nymphs at different temperatures and relative humidities.

The survival times of *H. leachi* flat nymphs exposed to different temperatures and relative humidities are summarised in Table 5.1(b) and presented graphically in Fig. 5.3. The mean survival times, in general, also decreased with an increase in temperature and increased with an increase in relative humidity from 40% to 90%. In the case of 10°C (90±2%RH), however, the same pattern was not observed. The longest mean survival time (221.5 days) was recorded at 15°C and 90±2%RH. The shortest mean survival time (11.5 days) was recorded at 30°C and 40±2%RH.

**Table 5.1(b).** Summary of the survival time (days) of *H. leachi* flat nymphs exposed to different temperatures and relative humidities.

| TEMP. (°C) | RH (±2%) | Min. no. of Days | Max. no. of Days | Mean no. of Days | n |
|---|---|---|---|---|---|
| 10 | 90 | 141 | 191 | 166.0 | 20 |
| 10 | 70 | 148 | 208 | 178.0 | 20 |
| 10 | 40 | 14 | 30 | 17.0 | 20 |
| 15 | 90 | 191 | 252 | 221.5 | 20 |
| 15 | 70 | 153 | 202 | 177.5 | 20 |
| 15 | 40 | 10 | 20 | 15.0 | 20 |
| 20 | 90 | 168 | 191 | 179.5 | 20 |
| 20 | 70 | 79 | 85 | 82.0 | 20 |
| 20 | 40 | 10 | 20 | 15.0 | 20 |
| 25 | 90 | 62 | 122 | 92.0 | 20 |
| 25 | 70 | 23 | 62 | 42.5 | 20 |
| 25 | 40 | 6 | 27 | 16.5 | 20 |
| 30 | 90 | 93 | 103 | 98.0 | 20 |
| 30 | 70 | 37 | 70 | 53.5 | 20 |
| 30 | 40 | 10 | 13 | 11.5 | 20 |

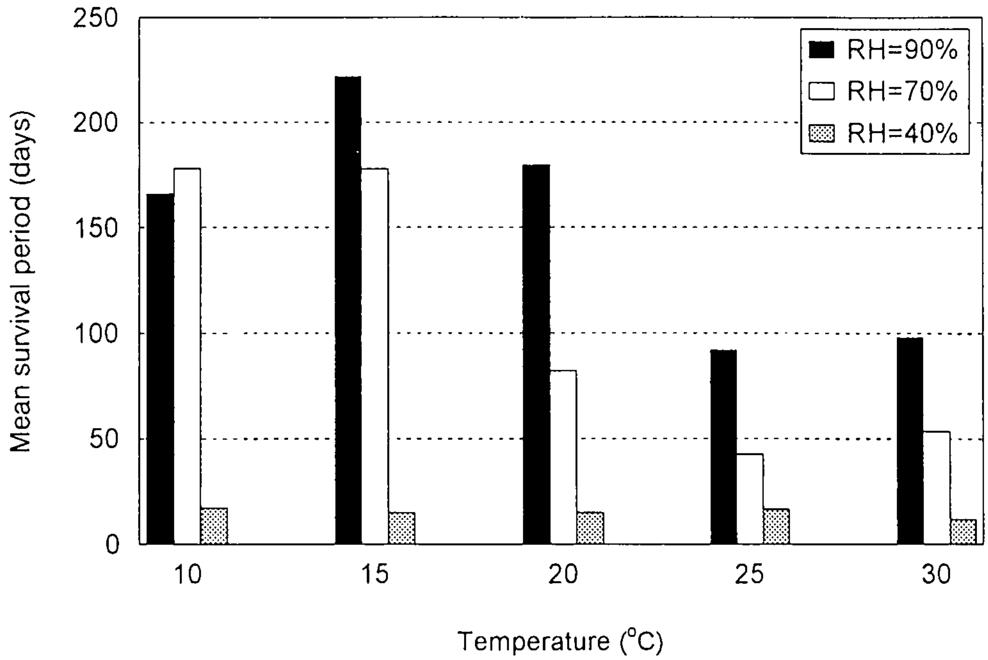


**Figure 5.3.** Mean survival times (days) of *H. leachi* nymphs at different temperatures and relative humidities.

*(b) PRE-MOULT PERIOD AND MOULTING SUCCESS*

Results on the pre-moult period and moulting success of engorged *R. sanguineus* and *H. leachi* nymphs are summarised in Table 5.2. In the case of *R. sanguineus* none of the nymphs moulted at either 10 or 15°C. At 20°C, nymphs moulted from 39 to 49 days; at 25°C from 16 to 19 days and at 30°C from 13 to 20 days. Moulting success varied between 88 to 100%. At the highest temperature (30°C) moulting success at 40 and 70%RH, respectively, was 88% (Table 5.2).

In the case of *H. leachi* none of the nymphs moulted at 10°C. At 15°C nymphs moulted from 35 to 46 days; at 20°C from 18 to 30 days; at 25 °C from 11 to 16 days and at 30°C from 13 to 19 days. Moulting success varied from 88 to 100%. At the temperature of 20°C moulting success at 40%RH was 88%.

**Table 5.2.** Summary of the pre-moult period and moulting success of engorged *R. sanguineus* and *H. leachi* nymphs which were exposed to varying temperatures and relative humidities.

| TEMP. (°C) | RH (±2%) | PRE-MOULT PERIOD (DAYS) | | MOULTING SUCCESS (%) | n |
|---|---|---|---|---|---|
| | | MIN. | MAX. | | |
| *R. sanguineus* | | | | | |
| 10 | 90 | 0 | 0 | 0 | 8 |
| 10 | 70 | 0 | 0 | 0 | 8 |
| 10 | 40 | 0 | 0 | 0 | 8 |
| 15 | 90 | 0 | 0 | 0 | 8 |
| 15 | 70 | 0 | 0 | 0 | 8 |
| 15 | 40 | 0 | 0 | 0 | 8 |
| 20 | 90 | 39 | 41 | 100 | 8 |
| 20 | 70 | 40 | 49 | 100 | 8 |
| 20 | 40 | 39 | 47 | 100 | 8 |
| 25 | 90 | 16 | 18 | 100 | 8 |
| 25 | 70 | 16 | 19 | 100 | 8 |
| 25 | 40 | 16 | 19 | 88 | 7 |
| 30 | 90 | 13 | 17 | 100 | 7 |
| 30 | 70 | 14 | 20 | 88 | 8 |
| 30 | 40 | 13 | 19 | 88 | 7 |
| *H. leachi* | | | | | |
| 10 | 90 | 0 | 0 | 0 | 8 |
| 10 | 70 | 0 | 0 | 0 | 8 |
| 10 | 40 | 0 | 0 | 0 | 8 |
| 15 | 90 | 36 | 46 | 100 | 8 |
| 15 | 70 | 35 | 43 | 100 | 8 |
| 15 | 40 | 35 | 45 | 100 | 8 |
| 20 | 90 | 18 | 30 | 100 | 8 |
| 20 | 70 | 18 | 30 | 100 | 7 |
| 20 | 40 | 18 | 30 | 88 | 8 |
| 25 | 90 | 11 | 15 | 100 | 8 |
| 25 | 70 | 12 | 16 | 100 | 8 |
| 25 | 40 | 12 | 15 | 100 | 8 |
| 30 | 90 | 13 | 18 | 100 | 8 |
| 30 | 70 | 13 | 19 | 100 | 8 |
| 30 | 40 | 14 | 16 | 100 | 8 |

The relationship between the reciprocal of the moulting time and the temperature for *R. sanguineus*, was given by the following relationship:

$y = a + bx$

where $a = -0.0524694$

$b = 0.00425366$

$y = 1$ / moulting time (days)

$x =$ temperature in °C

$R^2 = 92.63\%$

The relationship between the reciprocal of the moulting time and the temperature for *H. leachi*, was given by the following relationship:

$y = a + bx + cx^2$

where $a = -0.08292$

$b = 0.009662$

$c = -0.0001608$

$y = 1$ / moulting time (days)

$x =$ temperature in °C

$R^2 = 84.79\%$

The regression equations for the two tick species are graphically presented in Figs 5.4 & 5.5. The Critical Developmental Temperature (CDT) was 12.3°C and 10.1°C for *R. sanguineus* and *H. leachi*, respectively.

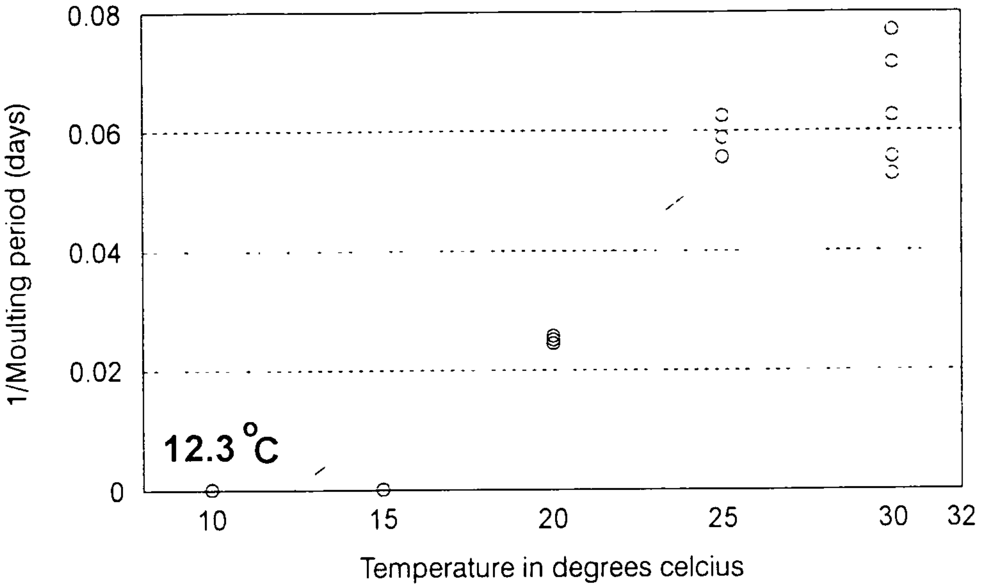


**Figure 5.4.** Graphical representation of the relationship between the reciprocal of the moulting time (days) and the temperature (°C) for *R. sanguineus* nymphs at 90±2%RH. The critical temperature is 12.3 °C.

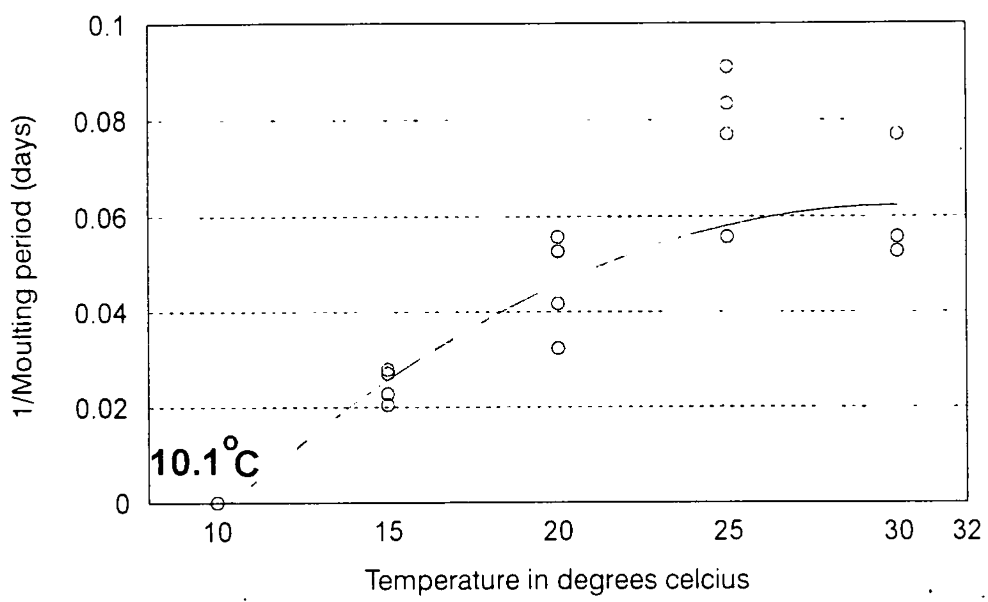


**Figure 5.5.** Graphical representation of the relationship between the reciprocal of the moulting time (days) and the temperature (°C) for *H. leachi* nymphs at 90±2%RH. The critical temperature is 10.1 °C.

*(c) MOULTING SUCCESS OF NYMPHS EXPOSED TO VARYING RELATIVE HUMIDITIES*

The group of *R. sanguineus* nymphs exposed constantly either to a low (0%, group C) or a high (90%, group D) relative humidity had a moulting success of 100 and 80% respectively (Table 5.3a). The minimum and maximum time it took for the nymphs to moult for the above two groups was 12-16 and 12-20 days, respectively. Moulting success for both groups A and B was high, ranging between 95-100%. The mean pre-moult periods for groups A and B varied between 9.5-16.5 and 15.5-16.5 days, respectively. From the results, it seems as if the relative humidity did not have any effect on the duration of the pre-moult period.

**Table 5.3(a).** Summary of the moulting time and moulting success of *R. sanguineus* nymphs which were exposed to a constant temperature of 25°C and varying relative humidities (namely, High: 90%; Low: 0%), in total darkness.

| GROUP * | Exposure period (days) | $\bar{x}$ No. of days | MIN. No. of days | MAX. No. of days | Moulting success (%) | n |
|---|---|---|---|---|---|---|
| A | 0%RH | | | | | |
| | 1 | 16.0 | 14 | 18 | 100 | 20 |
| | 2 | 16.0 | 14 | 18 | 100 | 20 |
| | 3 | 15.5 | 13 | 18 | 100 | 20 |
| | 4 | 16.5 | 14 | 19 | 95 | 20 |
| | 5 | 14.5 | 10 | 19 | 95 | 20 |
| B | 90±2%RH | | | | | |
| | 1 | 16.5 | 14 | 19 | 100 | 20 |
| | 2 | 16.0 | 13 | 19 | 95 | 20 |
| | 3 | 15.5 | 13 | 18 | 95 | 20 |
| | 4 | 15.5 | 13 | 18 | 95 | 20 |
| | 5 | 16.0 | 14 | 18 | 100 | 20 |
| C | 0%RH | 16.5 | 12 | 16 | 100 | 20 |
| D | 90±2%RH | 16.0 | 12 | 20 | 80 | 20 |

* Group A: 0%RH for 1-5 days and 90%RH until moult
B: 90%RH for 1-5 days and 0%RH until moult
C: constant 0%RH
D: constant 90%RH

With reference to *H. leachi* exposed constantly either to a low (0%, group C) or a high (90%, group D) relative humidity, the nymphs displayed a moulting success of 20 and 70%, respectively (Table 5.3b). The range of moulting time it took for the above two groups was 16-18 and 14-18 days, respectively. The mean pre-moult periods for groups A and B varied between 16.0 - 18.5 and 14.5 - 16.5 days, respectively. Moulting success for groups A and B varied between 70-90 and 30-60%, respectively (Table 5.3b).

**Table 5.3(b).** Summary of the moulting time and moulting success of *H. leachi* nymphs which were exposed to a constant temperature of 25°C and varying relative humidities (namely, High: 90%; Low: 0%), in total darkness.

| GROUP * | Exposure period (days) | $\bar{x}$ No. of days | MIN. No. of days | MAX. No. of days | Moulting success (%) | n |
|---|---|---|---|---|---|---|
| A | 0%RH | | | | | |
| | 1 | 18.5 | 15 | 22 | 90 | 20 |
| | 2 | 16.0 | 13 | 19 | 70 | 20 |
| | 3 | 17.0 | 13 | 21 | 80 | 20 |
| | 4 | 17.5 | 13 | 22 | 80 | 20 |
| | 5 | 16.0 | 13 | 19 | 70 | 20 |
| B | 90±2%RH | | | | | |
| | 1 | 16.5 | 13 | 20 | 50 | 20 |
| | 2 | 14.5 | 14 | 15 | 30 | 20 |
| | 3 | 14.5 | 12 | 17 | 30 | 20 |
| | 4 | 14.5 | 12 | 14 | 60 | 20 |
| | 5 | 14.5 | 14 | 15 | 40 | 20 |
| | | | | | | |
| C | 0%RH | 17.0 | 16 | 18 | 20 | 20 |
| D | 90±2%RH | 16.0 | 14 | 18 | 70 | 20 |

* Group A: 0%RH for 1-5 days and 90%RH until moult
B: 90%RH for 1-5 days and 0%RH until moult
C: constant 0%RH
D: constant 90%RH

*(d) DETERMINATION OF POSSIBLE DROP-OFF RHYTHMS OF NYMPHS*

In the case of both species, nymphs only started detaching after about three days. In the case of *R. sanguineus* most of the nymphs detached early morning (Fig. 5.6). The first peak was displayed at 3:00am and the second, 24 hours later, at 5:00am. The highest drop-off percentage was 14.23 and 11.17%, respectively.

In the case of *H. leachi* most of the nymphs detached during the afternoon or early evening (Fig. 5.7). The first peak was displayed at 7pm and the second at 1pm. The highest drop-off percentage was 25.64% and 19.68%, respectively.

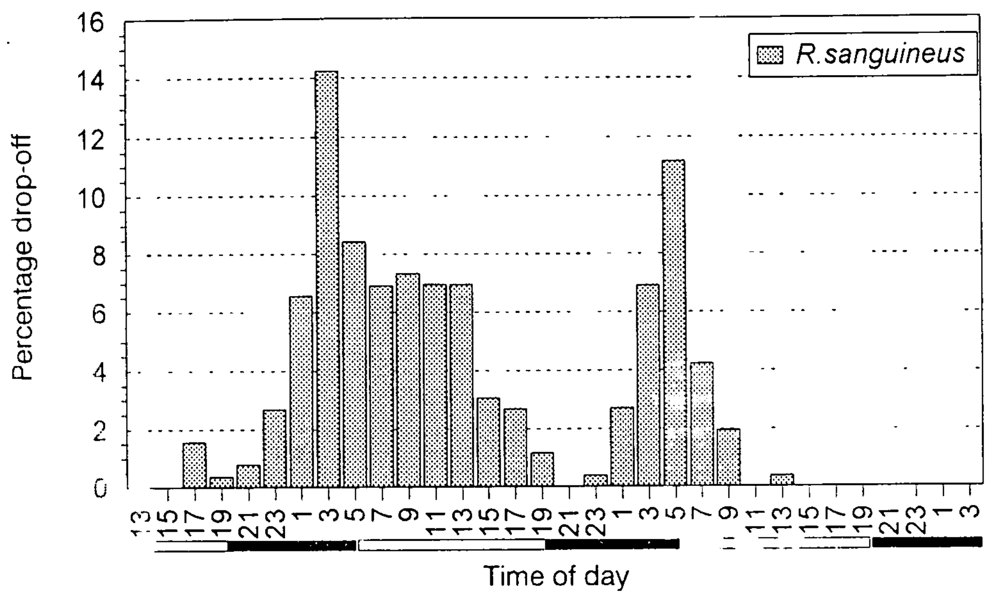


**Figure 5.6.** Histogram showing the time specific detachment pattern of *R. sanguineus* nymphs (The horizontal shaded bar indicates the photoperiod).

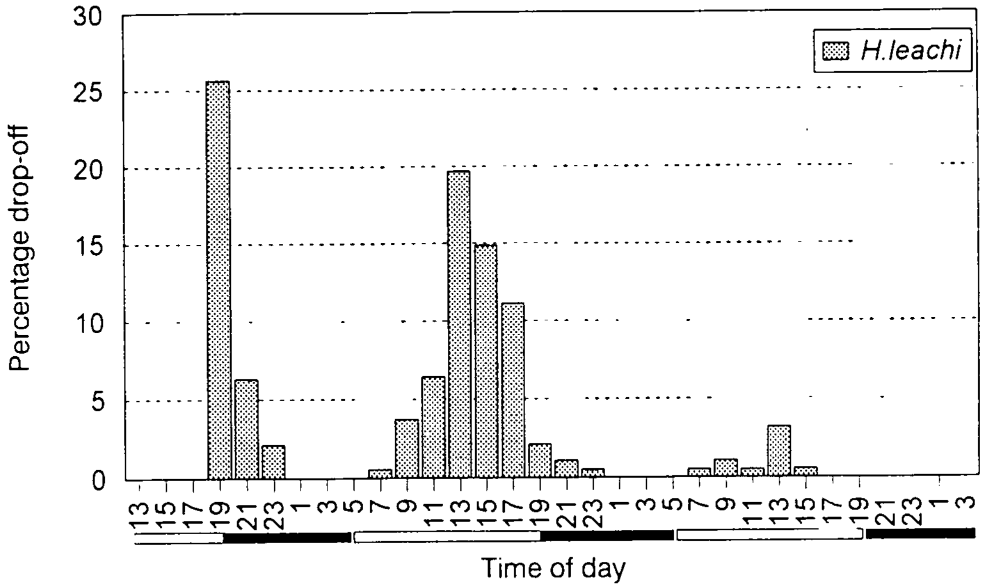


**Figure 5.7.** Histogram showing the time specific detachment pattern of *H. leachi* nymphs (The horizontal shaded bar indicates the photoperiod).

# DISCUSSION

*(a) SURVIVAL OF FLAT NYMPHS*

In general the trend for both *R. sanguineus* and *H. leachi* was an increase in survival time with a decrease in temperature and an increase in relative humidity. The maximum survival time for nymphs of both species was recorded at low temperatures and high humidities. For the long-term storage of flat nymphs in the laboratory this temperature should be used. At higher temperatures between 20-30°C and at a 90±2%RH the maximum longevity of *H. leachi* nymphs was consistently higher compared to *R. sanguineus*. Conversely, the maximum longevity of *R. sanguineus* nymphs at low relative humidities 40±2%RH and temperatures ranging from 10-30°C was, in general, substantially longer than that recorded for *H. leachi*. There was a relationship between longevity and temperature and between longevity and relative humidity in the case of both species. The higher the relative humidity and the lower the temperature, the longer the survival period (Knülle 1966; Koch and Tuck 1986). The data in the present study indicate that *R. sanguineus* nymphs were more tolerant to desiccation when exposed to low humidities than those of *H. leachi*. Many tick species can live for long periods without feeding i.e. off its host, under adverse conditions. The length of this period depends on the tick's ability to maintain its water balance and the ability to conserve energy (Needham and Teel 1986; Fielden and Rechav 1996). Water balance is an important key to understanding the survival of ticks and provides information about the tick's microclimatic requirements. The reason for this is that after having fed, most tick species detach from their host, after which their survival depends on the microclimate in which they end up (Knülle 1966).

Water balance depends on the capabilities of the tick in question. These capabilities include active absorption of water vapour from unsaturated air, an integument that reduces transpirational water loss, and spiracular valves that guard the tracheal system. Other factors include a slow metabolic rate during this non-feeding stage to conserve energy reserves and the ability of the tick's tissues to tolerate significant changes in the osmotic concentration of the haemolymph, which bathes them (Needham and Teel 1986). The active uptake of water depends on the critical equilibrium humidity (CEH). This can be defined as the lowest threshold activity at which active water uptake can still occur. This typically lies between 80-90%RH (Lees 1946, 1947). Lower relative humidities prevent the active uptake of water and hence force the tick to rely on other factors already mentioned. Under desiccating conditions these factors will determine a tick's ability to survive and implies that *R. sanguineus* is more tolerant to desiccation than *H. leachi*.

*(b) PRE-MOULT PERIOD AND MOULTING SUCCESS*

A decrease in pre-moult period with an increase in temperature is normal for ixodid tick species (Koch and Tuck 1986; Van der Lingen 1995; Zahler and Gothe 1995) and is most probably related to an increase in metabolic rate at the higher temperatures. The pre-moult periods recorded during the present study cannot be compared to any in the available literature because very little information is available in this regard. Bechara, Szabó, Ferreira and Garcia (1995) showed that the pre-moult period for *R. sanguineus* was approximately seven days whereas the present study showed it to be 13-18 days (under similar conditions of approximately 30°C and 90%RH) for both *R. sanguineus* and *H. leachi*. The hosts used were, however, not dogs. Guinea pigs and hamsters were used and therefore the results obtained cannot be compared to the findings of the present study. The results of the present study also indicated that relative humidity does not significantly influence pre-moult periods. This is also in accordance with the results of Kock and Tuck (1986).

According to the findings of Koch and Tuck (1986), engorged *R. sanguineus* nymphs are able to moult with a 98% success rate under desiccating conditions of 35°C and 35%RH. The present study has shown this to be true for *R. sanguineus* but not for *H. leachi*, although 30°C was the highest temperature used. The present study has also shown that low temperatures and high humidities favour the survival of nymphs but engorged nymphs of both species do not moult at 10°C. This can be explained by the critical developmental temperature (CDT) for these species. The CDT for *R. sanguineus* and *H. leachi* was higher than 10°C. This indicates that any temperature below these values are unfavourable with regard to moulting and therefore, as was shown in the present study, moulting will not take place. *R. sanguineus* did not moult at 15°C either. This result indicates that 15°C is too low for *R. sanguineus* and implies that the CDT is higher than what was found in the present study. It should be noted that the graph in Fig. 5.4 only represents a trend and if smaller temperature increments had been used the CDT may have been higher. Furthermore, the variation (±2°C) of the temperature-controlled cabinets is also possibly a contributing factor.

*(c) MOULTING SUCCESS OF NYMPHS EXPOSED TO VARYING RELATIVE HUMIDITIES*

In the present study the exposure of *R. sanguineus* nymphs to a desiccating environment after detachment prior to ecdysis did not affect the moulting success. This is most probably related to the ability of the nymphs to limit passive water loss (Needham and Teel 1986) and also explains the wide distribution of *R. sanguineus* in many different ecotypes. It is also possible that desiccation does not affect the moulting success because of the ability of immature engorged ticks to produce metabolic water (Kahl 1991). The results of the groups C and D indicated a lower success under high relative humidity conditions than desiccating conditions. This unexpected occurrence can possibly be explained by the fact that the high relative humidity and temperature proved to be ideal for fungal growth. The fungus resulted in the death of some of the engorged nymphs, hence the lower moulting success. Van der Lingen (1995) did a similar study on *Ixodes rubicundus* and found that low relative humidities affected the length of the pre-moult period but not the moulting success. However, it should be noted that these ticks undergo diapause and take longer to moult than *R. sanguineus*, under similar conditions. Heath (1974) found similar results for *Haemaphysalis longicornis*, which showed that low relative humidities also affected only the length of the pre-moult period and not the moulting success. Furthermore, although *R. sanguineus* has a high tolerance for desiccating conditions, its behaviour must also be taken into account. These ticks readily climb and inhabit cracks in structures during the non-parasitic stages (Fourie, Kok and Visagie-unpublished data) thus increasing its niche to include human dwellings (Koch and Tuck 1986).

In the present study the exposure of *H. leachi* nymphs to desiccating conditions prior to ecdysis resulted in a significantly lower moulting success compared to those nymphs exposed to desiccating conditions for periods ranging from 1-5 days immediately after detachment. Compared to *R. sanguineus* it was evident that *H. leachi* nymphs were more leaky and this is most probably reflected in the off-host habitat selection of the tick. Whereas engorged *R. sanguineus* larvae and nymphs may be found in cracks and crevices in brick walls (Fourie, Kok and Visagie-unpublished data), *H. leachi* will select a more protected micro-environment underneath the soil surface or debris of vegetation.

*(d) DETERMINATION OF POSSIBLE DROP-OFF RHYTHMS OF NYMPHS*

*R. sanguineus* displayed a circadian rhythm with two distinct peaks. *H. leachi* also displayed two distinct peaks. This was shown to be true for *R. sanguineus*, in the studies conducted by Hadani and Rechav (1969). *R. sanguineus* nymphs detach during the scoto phase and *H. leachi* mainly during the photo phase. Complex relationships have evolved between particular tick species and their natural hosts. An example of such a relationship is the detachment rhythm displayed by engorged *Ixodes rubicundus* ticks (Du Toit, Fourie and Horak 1994). This circadian rhythm has both physiological and ecological importance and has been demonstrated by several authors (Amin 1970; Hadani and Rechav 1970; Hadani and Ziv 1974; Doube 1975; Rechav 1978; Guglielmone 1993). Endogenous circadian regulation mechanisms, entrained by exogenous signals, control the detachment of engorged ticks (Hadani and Rechav 1970; Hadani and Ziv 1974). An example of such a signal would be a change in light intensity as suggested by Kitaoka (1962). However, the accuracy of the detachment depends on the endogenous rhythm, which is influenced by the host's physiological and activity patterns (Rechav 1978). Factors in the host's blood and the adrenal cycle of the host have also been identified as having an influence (Amin 1970).

*Ixodes rubicundus* immatures detach from elephant shrews when the host is not active, while basking in the sun (Du Toit 1993). This increases the probability of the subsequent stages finding a host and is thought to be controlled by the photoperiod. Hadani and Ziv (1974) showed that *Hyalomma excavatum* nymphs display a circadian detachment rhythm. These ticks detach form their gerbil host photoperiodically. Guglielmone (1993) also showed that *Amblyomma triguttatum triguttatum* immatures display a diurnal detachment rhythm that reflects the rhythm of it natural hosts.

The tick detaches where it is most likely to find another host after moulting and where the conditions are most favourable for the completion of its life cycle (Belozerov 1982). If it is accepted that dogs sleep during the early hours of the morning and are more active late in the afternoon, then it is easy to realise that the timing of the drop-off has a dual ecological purpose. When the dog returns to its kennel the well-timed detachment of the engorged nymphs ensures that when they moult into adults, there will be an available host which will return to its dwelling. *H. leachi* is also a three-host tick which requires small rodents for its immatures to feed on (Hoogstraal 1956). *Rhabomys pumilio*, the striped mouse, forages during the day and is most active from 5-8:30am and 14:30-17:30pm (De Graaff 1981). The detachment of *H. leachi* nymphs, which is predominantly diurnal, coincides with the animal's foraging activity. The detachment of the nymphs at this point would serve to increase the probability of finding a host. *H. leachi* adults do not utilise the small rodents but instead prefer larger animals like the domestic dog. Hence, there would be no advantage in detaching in the nests of the rodents it infests. The advantage of this type of synchronisation is the fact that it increases the probability that the following instar will find a suitable host.

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# CHAPTER 6

## SURVEYS - CLINICAL ASSESSMENT AND SEROLOGY

# CHAPTER 6

# SURVEYS - CLINICAL ASSESSMENT AND SEROLOGY

## INTRODUCTION

Certain ixodid ticks have been identified as far back as the end of the 19th century as vectors of disease to domestic animals and man (Theis and Budwiser 1974). They can transmit various kinds of microbial diseases such as tularemia, relapsing fever, and various tick bite fevers like Q fever (Spielman, Wilson, Levine and Piesman 1985; Magnarelli, Stafford, Mather, Yeh, Horn and Dumler 1995), human babesiosis, ehrlichioses (van Heerden 1992) and Lyme disease (Spielman, Wilson, Levine and Piesman 1985). In South Africa *Ehrlichia canis* and *Babesia canis* cause ehrlichiosis and babesiosis, respectively, which are important diseases of dogs (Neitz and Thomas 1938; Oberholzer and Ryke 1993). *Rhipicephalus sanguineus* can potentially transmit many of these diseases (Arthur 1962; Hoogstraal 1956). The close relationship between man and dog makes the risk of disease greater and certainly deserves attention. Many tick species directly affect the dogs they parasitise. They are capable of causing a variety of afflictions such as pain and irritation due to the bite, inflammation and secondary bacterial infections at the feeding site, tick toxicosis, tick bite allergic reactions, and also blood loss anaemia.

Clinical assessments provide important information but need to be broadened to incorporate more than the above-mentioned associated diseases. This type of approach presents a more holistic picture of the dog's health, by including other health conditions. These conditions include genital neoplasma, distemper, trauma, emaciation, demodectic mange, lameness, and many more, all of which relate to the circumstance in which the dog finds itself. Assessments, of this nature, of dogs that occur in different localities, could potentially reflect on the attitude of dog owners towards their dogs, the ability of the owners to care for their dogs and the disease or reservoir status of the dogs.

The objectives of the present study were:

- to clinically assess and compare the condition, with respect to general health, of dogs in the different study localities
- to conduct a serological survey in the different study localities with regard to *Ehrlichia* spp.
- to determine the dog owners' general perception towards the health of their dogs

# MATERIALS AND METHODS

*CLINICAL ASSESSMENT*

Two hundred dogs, 50 each in Bloemfontein, Heidedal, Botshabelo and Thaba Nchu, respectively, were each examined clinically for specific diseases or conditions. The dogs were selected purely at random, subject to the owner's approval. The following conditions were recorded: genital neoplasma (tumor resembling transmissible venereal tumors), distemper, trauma (scratch and bite marks from fighting), emaciation (thin state due to malnutrition), demodectic mange (skin condition), lameness (inability to walk), pyoderma (skin condition caused by bacteria), abscessations, keratitis / blindness (eye condition), cryptorchidism (the dropping of a single testis), flea-bite dermatitis (skin condition caused by fleas), *Stomoxys* sp. bites (bites found particularly on the ears), conjunctivitis (eye condition) and castration. All diagnoses were done in the presence of a qualified veterinarian who verified each case before it was recorded. Flea bite dermatitis is linked to the presence of fleas. For this reason, it was decided to take a token sample in those cases where fleas were observed. The accurate sampling of fleas was beyond the scope of the present study but a token sample, using a standard flea comb, was taken in order to identify flea species present.

The dogs were also subjected to condition scoring based on the method used on cattle (Van Niekerk and Louw 1990) and a variation of this method adapted for dogs (Rautenbach, Boomker and De Villiers 1991). The scores ranged from a minimum of one to a maximum of five and were used to reflect the condition of the dog. The lowest score of one indicated that the dog was extremely emaciated and a high score of five indicated that the dog was overweight. Hence, a dog with a score of three would be in a good condition and would be healthy. It should be noted that half scores were also used in cases where it was difficult to decide between two scores. The different scores were paraphrased as follows:

1 - A thin dog with ribs, spinous and transverse processes easily visible.

2 - A thin dog with ribs, spinous and transverse processes not visible but, which can be felt.

3 - A dog in a healthy condition, in which case the ribs, spinous and transverse processes can only be felt if pressed very hard.

4 - A dog in good condition in which case the ribs, spinous and transverse processes cannot be felt.

5 - A dog which is overweight, a condition which is not regarded as being a healthy condition.

The reason for using the method used on cattle was to minimise the level of subjectivity in scoring a particular dog. In a survey done by Rautenbach, Boomker and De Villiers (1991) this method also proved to be effective as it eliminated the frame size and mass variables.

*SEROLOGICAL SURVEY*

In an attempt to compare the dog populations in different communities with regard to disease prevalence, the study area was divided into four localities, namely, Bloemfontein, Heidedal, Botshabelo and Thaba Nchu. These localities were further divided, proportionally, into zones or residential areas. Bloemfontein was divided into its residential areas with the exception of Heidedal and Mangaung. Heidedal was included as a locality on its own and was divided into three subsections. Botshabelo was divided into the existing 19 zones. Thaba Nchu was divided into three zones, namely, Motlatla, Mokwena and Seloshesha.

It was decided that, statistically and based on the total number of individually owned properties, a sample size of 200 in total, i.e. 50 per locality, would provide adequate data from which to extrapolate. A property was chosen randomly before going out into the areas so as to remove any form of bias. Thereafter, it was decided that every fourth property from that point would be approached for a blood sample from the resident dog. In the event of there not being a dog, the property next to it was used and so on until a dog was found. This was done as a guideline for the present study but was not necessarily always adhered to as circumstances often dictated. E.g. some dogs were too vicious and could not be approached to collect a blood sample.

Blood was drawn from a vein on one of the dog's forelegs into sterile serology tubes (5ml vac-u-test tubes, 12 x 75mm; Radem Medical (Pty.) Ltd., Johannesburg, South Africa). Sterile 21G needles (Vacutainer needles, 0.8 x 40mm; Weil Organization Distributors, Johannesburg, South Africa) and a standard bulldog-shoulder were used. The tubes were then kept at room temperature to allow clotting. Sera were separated by centrifugation in a standard MSE Minor centrifuge at 3 000 rpm for 10 minutes. Approximately 2ml of the serum was collected from each sample and stored in 1.8ml vials (Nunc Cryo Tube Vials, Weil Organization Distributors, Johannesburg, South Africa), in an ultra-deep freezer at -70 °C. Serum samples were analysed by the Department of Microbiology at the University of the Orange Free State for *Ehrlichia canis* and *Ehrlichia chaffeensis*, using indirect fluorescent antibody (IFA) testing.

Some samples were contaminated or in some cases labeling smudged, due to the freezing process, so that it was impossible to trace their origin. Both categories totaled 39 with "spoilt" samples being one, 18, three and 17 for Bloemfontein, Heidedal, Botshabelo and Thaba Nchu, respectively. These cases were ignored and excluded from the IFA test.

*PERCEPTION OF DOG OWNERS*

While examining the dogs, the owners were asked for their opinion with regard to the dog's general health. The reason for this was to obtain a basic idea of the owner's general perception of the dog's health. This is referred to as the "habitus" and is divided into three categories:

- An owner who felt that his/her dog was chronically ill was given a score of one.
- An owner who felt that his/her dog was acutely ill was given a score of two.
- An owner who felt that his/her dog was healthy was given a score of three.

This was a variation of the method used by Rautenbach, Boomker and De Villiers (1991) in that it was not done by using a proper scientific procedure of gathering information regarding the dogs' history, analysing and drawing conclusions. The owner was only asked for his opinion. It is a fact that there are several different reasons for keeping a dog and there are different attitudes towards the well being of dogs, which may impact on the dogs' health (see Chapter 7). This survey was done purely to test for differences in opinion, regarding the dog's health, between the dog owners from the different study localities. This opinion may or may not have an impact on the efforts made to control tick infestations. Its accuracy was not tested, as this would be revealed in the clinical assessment and the serology surveys that were also conducted. The results from the condition scoring were not compared to that of the habitus scoring. The reason for this was that although there was an overlap regarding the dogs used, the two investigations were largely run separately. However, it was possible to compare the mean condition and habitus scores for the respective study localities.

*STATISTICAL TESTS*

A two-tailed t-test was used to compare the different study localities with each other. If a significant difference was found, further testing was done in the form of a one-tailed t-test. This was done using a 95% confidence level.

# RESULTS

*CLINICAL ASSESSMENT*

A summary of the number of cases for 14 conditions observed in the dogs is given in Table 6.1.

**Table 6.1.** Summary of clinical conditions observed in dogs from the various study localities (based on a sample of 50 dogs per locality where some dogs displayed more than one condition)

| | Conditions | Localities | | | |
|---|---|---|---|---|---|
| | | **Bloemfontein** | **Botshabelo** | **Heidedal** | **Thaba Nchu** |
| 1 | Genital neoplasma | - | 1 | - | 1 |
| 2 | Distemper | - | 1 | - | - |
| 3 | Trauma | 1 | 3 | 2 | 15 |
| 4 | Emaciation | - | 10 | 8 | 11 |
| 5 | Demodectic mange | - | 7 | 3 | 2 |
| 6 | Lameness | - | - | - | - |
| 7 | Pyoderma | - | 2 | - | 3 |
| 8 | Abscessations | - | - | - | - |
| 9 | Keratitus and blindness | 1 | 2 | - | - |
| 10 | Cryptorchidism | - | - | - | - |
| 11 | Flea-bite dermatitis | 3 | 13 | 5 | 6 |
| 12 | *Stomoxys* bites | 10 | 13 | 11 | 14 |
| 13 | Conjunctivitis | 1 | 3 | 1 | - |
| 14 | Castration | 1 | - | - | - |
| | Flea species – token sample | *Ctenocephalides felis sp.*, *Echidnophaga gallinacea* and *Pulex irritans* | | | |

Genital neoplasma was observed in only two dogs, one each in Botshabelo and Thaba Nchu. Only one case of distemper was observed in Botshabelo. Conjunctivitis was observed in all localities except Thaba Nchu. Lameness, abscessations and cryptorchidism were not observed at all. The conditions most frequently observed were *Stomoxys* bites (48) followed by emaciation (29), flea-bite dermatitis (27) and trauma (21). The occurrence of these conditions was also compared statistically.

With regard to trauma, there was no significant difference ($P>0.05$) between Heidedal, Bloemfontein and Botshabelo. The occurrence of trauma was however, significantly ($P<0.05$) higher in Thaba Nchu with a total of 15 trauma cases compared to the other localities. With regard to flea-bite dermatitis, there was no significant difference ($P>0.05$) between Heidedal, Bloemfontein and Thaba Nchu. Botshabelo differed significantly ($P<0.05$) from the other localities. When comparing the localities with regard to *Stomoxys* bites, it was found that there was no significant difference between the four localities ($P>0.05$). No cases of emaciation were recorded in Bloemfontein and could not be statistically tested against the other localities. The frequency of occurrence of emaciation did not differ significantly ($P>0.05$) between Botshabelo, Heidedal and Thaba Nchu.

Three flea species were sampled from the dogs, namely, *Ctenocephalides felis felis*, *Echidnophaga gallinacea* and *Pulex irritans*. The predominant species was *Ctenocephalides felis felis*, followed by *Echidnophaga gallinacea* and very few *Pulex irritans*.

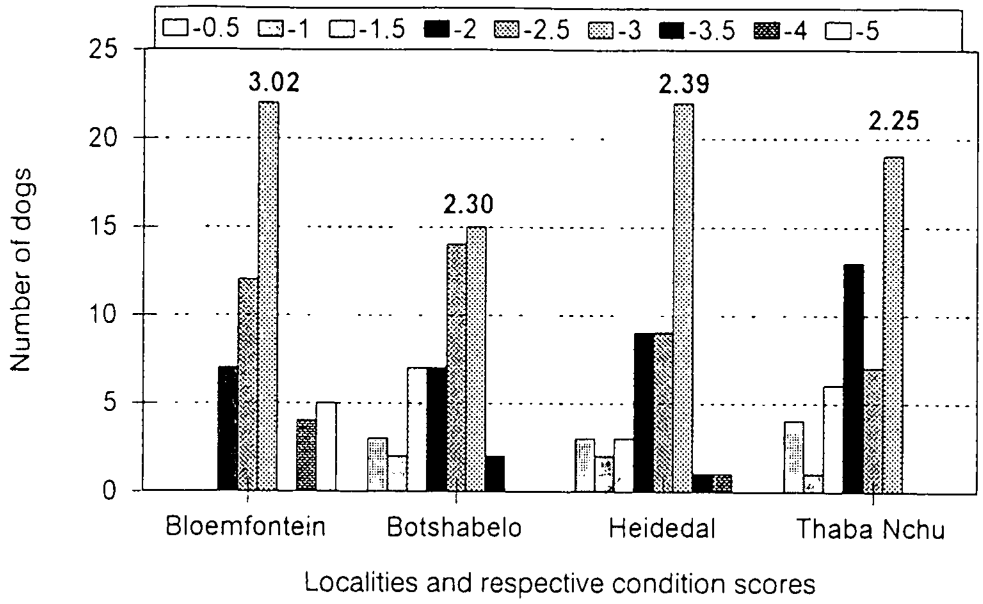


**Figure 6.1.** Graph showing the number of dogs per condition score category (see legend) in four different study localities. The mean can be seen above each respective section of the graph.

The condition scores allocated to dogs in the different study localities are graphically illustrated in Fig. 6.1. Dogs in Bloemfontein had significantly ($P<0.05$) higher condition scores compared to other localities. Condition scores of dogs in Bloemfontein varied between 2 and 5 while those in the other localities varied between 0.5 and 4 (Fig. 6.1). The mean scores for Bloemfontein, Botshabelo, Heidedal and Thaba Nchu were 3.02, 2.30, 2.39 and 2.25, respectively. The maximum and minimum scores were as follows: Bloemfontein (5 and 2), Heidedal (4 and 0.5), Botshabelo (3.5 and 0.5) and Thaba Nchu (3 and 0.5).

*SEROLOGICAL SURVEY*

The percentage of dogs seropositive for *Ehrlichia canis* and *Ehrlichia chaffeensis* in the different study localities are presented graphically in Fig. 6.2.

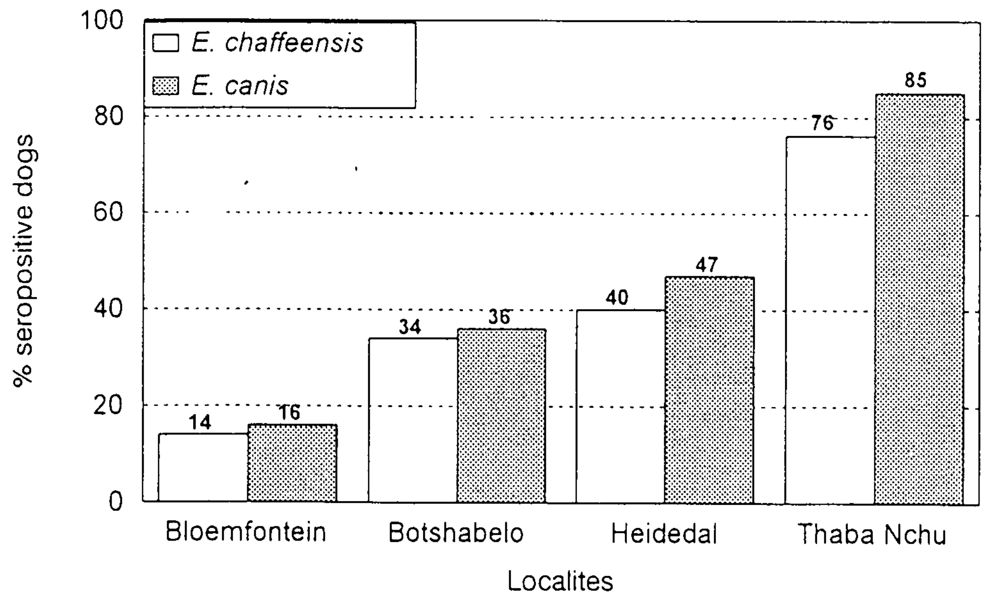


**Figure 6.2.** Graph showing the percentage dogs that tested positive for *Ehrlichia canis* and *Ehrlichia chaffeensis* antibodies, in four different localities.

Bloemfontein had significantly ($P<0.05$) less seropositive cases against *Ehrlichia* compared to the other localities. There was also a significant difference ($P<0.05$) between Botshabelo and Thaba Nchu. However, when comparing Heidedal with Botshabelo, there was no significant difference.

*PERCEPTION OF DOG OWNERS*

A summary of the habitus scores allocated to dogs in the different study localities is given in Table 6.2.

**Table 6.2.** Habitus scores allocated to the dogs in four different study localities.

| LOCALITY | HABITUS SCORE | | | |
|---|---|---|---|---|
| | 1 | 2 | 3 | $\bar{x}$ |
| Bloemfontein | - | 8 | 42 | 2.90 |
| Botshabelo | 24 | 14 | 12 | 1.76 |
| Heidedal | 40 | 6 | 4 | 1.32 |
| Thaba Nchu | 20 | 20 | 10 | 1.80 |

Bloemfontein displayed a significantly ($P<0.05$) higher habitus score when compared to other study localities. This indicated that the people in Bloemfontein generally believe that their dogs are in a healthy condition. When comparing Heidedal to Botshabelo and Thaba Nchu there was also a significant difference ($P<0.05$) but in contrast, the Heidedal scores were mostly one. This indicates that the people in Heidedal generally believed that their dogs were chronically ill. When comparing the scores of Botshabelo and Thaba Nchu no significant difference ($P>0.05$) was found. Both scores tended towards two and one, indicating that the people in these two areas believed their dogs were acutely or chronically ill.

# DISCUSSION

*CLINICAL ASSESSMENT*

In a previous study conducted by Rautenbach, Boomker and De Villiers (1991) a cross-sectional survey was done in a town called Maboloka in Bophutatswana (South Africa), in which the conditions of the dogs were assessed. This particular area is a poorer developed area with moderate to standard housing, in which shanty houses predominate, not unlike the poorer developed localities used in the present study. The mean number of dogs per household was 0.68. This ratio is used as an indicator of canine over-population but 0.68 is seen as a relatively low population density when compared to developing countries. This low ratio is explained by the high death ratio in the area. It was found that the dog population lacked longevity, which was possibly due to the poor health status and high disease rate. Chapter 7 refers to a similar study done in the greater Bloemfontein area. When comparing "urban: formal" and "urban: informal" areas the number of dogs per household was 0.82 and 1.0, respectively. When comparing Bloemfontein, Botshabelo and Thaba Nchu, the number of dogs per household was 0.81, 0.80, and 0.59, respectively. This indicates that irrespective of the differences in socio-economic levels, the number of dogs per household is low and comparable between the different study localities. This is in accordance to the results of Rautenbach *et al.* (1991).

An interesting fact that was perhaps not highlighted in the present study was the high death ratio that Rautenbach *et al.* (1991) mentioned. It would make sense that the poorer developed areas would have a high death ratio resulting in a low dog density. This argument, however, cannot be used in the case of the affluent localities as the people in these localities do have the means to treat their dogs. This fact is supported by the results of the present study where the incidents of more serious clinical conditions are very low in Bloemfontein. Hence, one explanation for this would be that the common low dog density applies but for different reasons. In the case of the poorer developed localities the death ratio is simply high and the number of dogs per household is low because these dogs die at a very early age. In the case of the affluent localities, there is probably a conscious decision to keep fewer dogs, unless breeding is the reason for keeping dogs.

In the present study the mean condition recorded in the resource poor environments were marginally higher compared to that reported by Rautenbach *et al.* (1991). Rautenbach *et al.* (1991) reported a mean condition score of 1.93, ranging from 0.5 to 3.5. In total 67.3% were judged to be healthy, 5% acutely ill and 27.5% chronically ill. They also found that the generally poor condition of the dogs in Maboloka was a result of an insufficient and unbalanced diet, heavy loads of internal and external parasites, and chronic wasting disease. The disease prevalence was high, with canine ehrlichiosis suspected of being the highest. The prevalence of genital neoplasma was also high (Rautenbach *et al.* 1991), unlike the situation found in the present study. Distemper was found to have a high prevalence but in the present study only one case was identified. This disease may have been more prevalent in the present study but its low prevalence is thought to be caused by the high turnover of the population. This situation and many of the conditions, in general, can be attributed to unrestrained movement, concomitant diseases, and because of the lack of vaccinations (Rautenbach *et al.* 1991). According to the latter authors, the dog population was found to have a high mortality rate overall which was indicated by the fact that very few cases of babesiosis were found. This disease is not fatal if treated. Hence, its absence would suggest that those dogs, which were infected by it, died. The only case of castration was found in Bloemfontein. This is an effective way of controlling reproduction in dogs but is also believed to control the behavioural characteristics of the dog (Blackshaw and Day 1994). This is an expensive alternative and it fits in with the socio-economic profile of this affluent locality.

The most prominent conditions afflicting the dogs in the present study area were trauma, emaciation, *Stomoxys* bites and flea-bite dermatitis. For the present study and for the sole purpose of testing for differences between the localities, only the four above-mentioned cases were tested statistically. Trauma was not significantly different except in the case of Thaba Nchu. Trauma in itself is due to fighting and refers to the scarring and injuries resulting therefrom. This is unavoidable except in cases where dogs are permanently restricted to a property and when there is only one dog on that property. Behavioural patterns or instinct result in dogs fighting and this has nothing to do with the socio-economic level of a locality. Thus, it makes sense that there would not be a difference between the affluent and poorer developed localities, despite the fact that the dogs in the affluent localities were fenced in and restricted to the property. Their owners often take these dogs for walks. Under these circumstances these dogs encounter other dogs, which presents a situation where fights can erupt. Also, in many cases there was more than one dog within the fenced property. The fact that Thaba Nchu differed can only be explained by the fact that the people in this area used their dogs to protect their cattle and to hunt (see Chapter 7).

Emaciation was non-existent in Bloemfontein, which is probably due to the fact that the owners were able to feed their dogs properly. The other localities did not differ significantly from each other with regard to cases of emaciation. This was an indication of a lower socio-economic level, which dictates that the family is fed first and only thereafter do the dogs get fed. Often it would seem that the dogs were left to fend for themselves, hence the fact that they are allowed to roam freely.

There was no significant difference in the *Stomoxys* bite cases for all four localities perhaps indicating the difficulty in controlling the fly that causes this condition. This indicates that the fly that causes this is more difficult to control than any of the other conditions. It is probably not an indication of poor sanitation, as would be the case with the housefly, but is related to the parasitic nature of the fly *Stomoxys calcitrans*. This problem, however, was beyond the scope of the present study and would require further investigation in order to elaborate more extensively.

The last condition considered was that of flea bite dermatitis. In the present study, the incidence of dermatitis was comparable to that of other countries. There was no significant difference between Heidedal, Bloemfontein and Thaba Nchu. Botshabelo showed a significant difference with a total of 13 cases. According to Varghese, Jagadish and Bhalerao (1994) skin diseases are the largest single group of conditions seen in small animal practice in Bombay (India) where the incidence of dermatitis ranges between 11.37% and 14.13%. This was comparable to other countries like the U.K. and Canada where the flea bite dermatitis was as high as 21.69%, tick bite dermatitis 15.66%, and pyoderma 4.82%. The most common cause of the skin diseases has been found to be ectoparasites. Flea bite dermatitis was the most prevalent among the ectoparasitic infestations in Bombay (Varghese, Jagadish and Bhalerao 1994) and as the name suggests, fleas are responsible (Dryden and

Blakemore 1989). This is a serious condition and for this reason it is necessary to look at the parasite that causes it.

In the present study three flea species were identified from the token samples taken, namely, *Ctenocephalides felis* sp., *Echidnophaga gallinacea* and *Pulex irritans*. Studies conducted by Harman, Halliwell and Greiner (1987), Rautenbach, Boomker and De Villiers (1991) and Dryden (1993), also identified *Ctenocephalides felis felis*, *Pulex irritans* and *Echidnophaga gallinacea* as being the most common flea species which infest dogs. *Ctenocephalides felis felis* seems to be the dominant flea species that infests dogs (Dryden and Blakemore 1989; Dryden 1993; Dryden and Rust 1994; Heath, Arfsten, Yamanaka, Dryden and Dale 1994; Chesney 1995). This, however, depends on its geographical distribution and may differ from area to area (Koutinas, Papazahariadou, Rallis, Tzivara and Himonas 1995). Fleas cause skin irritation and itching, as a result of their frequent bites. They also cause an allergic dermatitis in some cases (Dryden and Blakemore 1989; Gross and Halliwell 1985; Halliwell 1981; Harman, Halliwell and Greiner 1987; MacDonald 1993 after Koutinas, Papazahariadou, Rallis, Tzivara and Himonas 1995). *Ctenocephalides felis* has been identified as the main cause of this flea allergy dermatitis. The pruritus associated with this can be intense and often spread over the whole body of the dog. Chewing, licking and scratching is usually evident (Dryden and Blakemore 1989). This does not mean that the diseases they cause cannot result in fatality. Lastly, it should be noted that the cat flea, *Ctenocephalides felis felis*, in the absence of a dog or cat, would feed on humans (Angarano and Parish 1994).

From the results it was evident that the dogs from the poorer developed localities were in a worse condition than those from the affluent localities. Dogs were affected by many factors that contributed to their condition which are related to attitude, education and the availability of money. Access to veterinary services was also important as it is very often absent in the poorer developed localities. The most prominent conditions highlighted in the present study were trauma, emaciation, *Stomoxys* bites and flea-bite dermatitis. Different people have different attitudes and opinions and they believe the things they are familiar with. Ignorance and the lack of education regarding animal husbandry creates problems in the poorer developed localities. The problem of language differences adds to this because even if attempts are made to rectify or improve this, it has to be communicated in the mother tongue of those being targeted. Many speak a form of slang, which differs from place to place. This presents a mammoth task.

*SEROLOGICAL SURVEY*

Canine ehrlichiosis is a disease of the domestic dog (Van Heerden 1992). It is caused by *E. canis,* which is a rickettsial organism (Mathewman, Kelly, Mahan, Semu, Tagwira, Bobade, Brouqut, Mason and Raoult 1993; Botros, Elmolla, Salib, Calamaio, Dasch and Arthur 1995) and according to Van Heerden (1992) and Groves, Dennis, Amyx and Huxsoll (1975) it is transmitted by *R. sanguineus*. *E. canis* is not a human pathogen (Anderson, Dawson, Jones and Wilson 1991). It is also known as canine typhus, rickettsiosis, canine haemorrhagic fever, idiopathic haemorrhagic syndrome, tropical canine pancytopenia, tracker dog disease, Lahore canine fever and Nairobi bleeding disease (Greene and Harvey 1984). In South Africa, it is more prevalent in the higher rainfall regions where ticks are abundant. It is a common occurrence in dogs from the poorer communities (Van Heerden 1992). Heidedal, Botshabelo and Thaba Nchu fall into this category and the results certainly support this. The dogs in these areas are infested with ticks to a much greater extent (i.e. with much larger numbers) than the more affluent areas (see Chapter 2) and are either allowed to roam freely or are confined to a small area (van Heerden 1992). The largest contributing factor to the "under diagnosis" of this disease is the fact that its clinical signs are often atypical, inconspicuous and it often occurs concurrently with other infections (van Heerden 1992) like *Babesia canis* (Neitz and Thomas 1938; van Heerden 1989). Its diagnosis is difficult but the indirect fluorescent antibody test (IFA), which was used in the present study, has been proven to be both sensitive and specific (Mathewman *et al.* 1993).

A characteristic symptom is lateral or bilateral epistaxis (Walker, Rundquist, Taylor,Wilson, Andrews, Barek, Hogge, Huxsoll, Hildbrandt and Nims 1970). This results in death, which can be attributed to internal haemorrhaging, although some dogs do survive (Walker *et al.* 1970). It has been suggested that immunocompetent dogs may eliminate *E. canis* organisms and recover fully (Greene and Harvey 1984). This, with resultant remission of haematological and biochemical abnormalities has, however, only been shown to occur with the appropriate tetracycline therapy (Huxsoll, Amyx, Hemelt, Hildebrandt, Nims and Cochenour 1972).

As already mentioned, the vector of *E. canis* is *R. sanguineus* (Harris,Waner and Bark 1997; Maretzki, Fisher and Greene 1994). *E. equi* and *E. ewingii* are both causative agents of granulocytic ehrlichiosis and meningitis in dogs. The former is a milder form of the type of ehrlichiosis caused by *E. canis*. Clinical signs include cervical pain, epistaxis, gingival bleeding, pruritus, progressive lethargy and nonregenerative anaemia. It is transmitted transstadially and not transovarially in the tick. Thus larvae and nymphs become infected by feeding on an infected host (Harris, Waner and Bark 1997; Maretzki, Fisher and Greene 1994). Canine ehrlichiosis, caused by *E. canis*, is an acute, subacute or chronic disease (Greene and Harvey 1984). Mononuclear cells, in the blood, are infected by *E. canis*, where it multiplies and causes the destruction of blood cellular components (Van Heerden 1992). The acute phase is characterised by fever, anorexia, weight loss, weakness, pale mucous membranes, lymphadenopathy, dyspnea and edema of the limbs (Buona Voglia, Sagazio, Gravino, De Capranariis, Cerundolo and Buona Voglio 1995). Both the acute and chronic stages may be characterised by thrombocytopenia, leukopenia and anemia (Greene and Harvey 1984).

*E. chaffeensis* is the causative agent of human monocytic ehrlichiosis (Anderson, Dawson, Jones and Wilson 1991; Dumler and Bakken 1995; Magnarelli, Stafford, Mather, Yeh, Horn and Dumler 1995) and *E. equi* is responsible for human granulocytic ehrlichiosis (Dumler and Bakken 1995). Humans are infected through the bite of a tick carrying one of these pathogens. The clinical and laboratory manifestations of granulocytic and monocytic ehrlichioses are similar. Symptoms include fever, malaise, headaches, myalgia, progressive leukopenia, thrombocytopenia, anaemia and arthralgia. Patients also on occasion, show other signs like coughing and sometimes a rash (Dumler and Bakken 1995; Fingerle, Goodman, Johnson, Kurtti, Munderloh and Wilske 1997). The mean duration of the infection is about three weeks and more than 60% of all documented cases (especially those who are older) resulted in hospitalisation. Males are infected more often but women are susceptible to a more severe infection. The severe complications include renal failure, disseminated intravascular coagulation, seizures, and coma. As is the case with rickettsial infections, the longer the period between infection and treatment, the greater the chance of death (Dummler and Bakken 1995). Treatment involves the administering of a tetracycline such as doxycycline, even for children (Dummler and Bakken 1995). As mentioned earlier *E. canis* is not a human pathogen and Anderson, Dawson, Jones and Wilson (1991) go on to suggest that dogs are not reservoirs of human ehrlichiosis.. The results of the present study, however, showed that the dogs sampled do have antibodies for *E. chaffeensis*, which is the causative agent for human ehrlichiosis. It should be noted that although the pathogenesis of human ehrlichiosis is poorly understood, if the diagnosis and treatment is delayed it could become far more severe (Dumler and Bakken 1995).

With approximately 80% of the dogs being infected with both *E. canis* and *E. chaffeensis* in Thaba Nchu, it might be accurate to call Thaba Nchu a high-risk locality. Bloemfontein had a low percentage of less than 20% but this does not mean there is no risk. The close relationship between dogs and humans increases the possibility of being bitten by a tick carrying one of these pathogens and therefore the risk of infection. The dogs in these localities are affected and very often it goes unnoticed. The most important message derived from this is that these dogs carry pathogens that affect man and thus pose a threat not only to the dogs but to the owner as well. The lack of veterinary involvement in the non-affluent areas in southern Africa worsens the health status of the dogs in these areas and could thus have an effect on the human population (Rautenbach *et al* 1991).

*PERCEPTION OF DOG OWNERS*

The differences in the opinion of the dog owners in the different localities can be ascribed to many possible reasons. The fact that the dog owners in Bloemfontein generally felt that their dogs were healthy could be attributed to the fact that they are in a better financial position and were thus able to take proper care of their dogs. Knowledge regarding the possible diseases that affect dogs, parasitic infestations and the implications of not taking proper care of dogs is probably an added factor. This would imply that, since there was no significant difference between the other localities, there was a lack of finance but also of the necessary knowledge and exposure regarding diseases in dogs. This contention should, however, be tested more accurately. Problems which arose during the survey, which may have affected the results, included communication difficulties because of language differences and difficulty in obtaining an accurate history of the dogs' health. Although an interpreter was present, the combination of a lack of education and the variations within a particular language made communication difficult. Rautenbach, Boomker and De Villiers (1991) experienced similar problems. It is notable that the condition scoring indicated that the dogs in Bloemfontein received a mean score indicative of a healthy condition. The mean scores for Botshabelo, Heidedal and Thaba Nchu indicated that the dogs in these areas were not healthy but instead were thin and possibly emaciated. This was in agreement with the dog owners' opinions with regard to their dogs' health and implies that they were aware of their dog's condition. It was also in agreement with the emaciation and the serological results of the present study. Despite lack of relevant knowledge and resources it is apparent that the dog owners of all the localities were aware of their dogs' condition and appeared to have some level of concern for their dogs, the reasons for which may differ (see Chapter 7).

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# CHAPTER 7

## SURVEYS - ATTITUDES

# CHAPTER 7

# SURVEYS - ATTITUDES

## INTRODUCTION

It was Gandhi (1959) who once said: " The greatness of a nation and its moral progress can be judged by the way its animals are treated"

Since the beginning of mankind, animals have always played a major role in human relationships that are interdependent on animals and nature in general (Bergler 1988). Animals have always been an essential part of our history, culture, and existence (Rowan 1988). Domestic animals have always been a source of help and support to man in hunting, protection, stimulation and delight. The natural relationship of dog and master, man and animal, has been true for long periods in the human history (Bergler 1988). The study of the relationship between man and animal has recently been recognised as a multi-disciplined research field but is in no way a new field (Odendaal and Weyers 1990). This relationship is characterised by attitudes, patterns of behaviour and other variables shared by people in different situations. It is a symbiosis which has been long taken for granted but has been rediscovered and recognised for its role in human development and upbringing (Bergler 1988). There are large differences between different peoples and different cultures, however, and thus also in the type of animals kept and their numbers. Man's self image, socially, culturally, religious and economic is reflected in the significance he attaches to his animals (Bergler 1988).

Dogs play an important role in the everyday life of man. Dogs perform numerous tasks like protecting property and livestock, hunting and even serve as draught animals in some cultures (Bergler 1988). They also perform a deeper role by fulfilling psychological needs in being a playmate and companion for both children and adults alike. Pets, but especially dogs, are also used extensively by clinical psychologists as aids to therapy. This includes therapy for children, cardiovascular disorders, depression, loneliness and many more (Bergler 1988). As companions, dogs present a great deal. They offer a safe outlet for human needs for contact with another warm being and may also satisfy the needs for intimacy (Rowan 1988).

The ownership of dogs was also previously a form of class distinction in that only the nobility could afford to own hunting dogs of a purer breed. Hunting was a sport exclusive to the nobility because they were the only landowners (Bergler 1988). The result thereof was that the peasants (or those from a poorer socio-economic group) had to settle for crossbred mongrels (Bergler 1988). These early pet owners (i.e. the former) had something in common, namely, money and rank (Rowan 1988). The specially bred dogs were the playthings of the rich and their beneficial impact on the quality of life was beyond doubt (Bergler 1988).

Today dogs are regarded as a reflection of our own personalities and insecurities and the image we would like the world to perceive (Rowan 1988). The value placed on animals can safely be regarded as a reflection of those values that a particular society, culture or religion possesses (Bergler 1988). It has been observed that dogs often portray certain positive human characteristics, which are often absent in human counterparts. An example of such a trait is loyalty. Dogs are regarded as the epitome of loyalty (Bergler 1988). It has been debated that pet ownership is often explained in terms of a general love of animals (Bergler 1988) and giving a dog a human name indicates that an animal has a relatively high status (Rowan 1988).

The objectives of the present study were the following:

*DOG DENSITY*

- To compare dog densities in "urban: formal" and "urban: informal" areas
- To relate dog densities to the degree of socio-economic development in the different localities.

*DOG OWNERSHIP ATTITUDES*

- To determine whether there are differences in dog-ownership attitudes in the four localities selected, which present different socio-economic scenarios.
- To investigate which methods and products are used to control tick infestation in the different study localities.

# METHODS AND MATERIALS

*DOG DENSITY*

In order to determine dog density by means of a questionnaire survey, two questions were posed. The first was: "How many dogs do you own?" and the second was: "Have any of your dogs died in the past year?" If any dogs had died within the past year then it was recorded and presented as part of the data. The number of respondents per group was determined by the relative size of the localities in question. It was decided that, statistically and based on the total number of individually owned properties, a sample size of about 400 (in total) for all three regions would provide an adequate sample in terms of accuracy. This total of 400 was then divided proportionally between the areas, based on their relative size (through stratified randomization). The number of properties sampled in Bloemfontein, Botshabelo and Thaba Nchu were 135, 233 and 32 respectively. By looking at the property numbers it was possible to choose a number at random without having seen the property itself, therefore ensuring no bias. Once a number was selected it was decided that every fourth number after that would be included in the sample. This was done for all localities. Thereafter, the property numbers were matched with an address. Business addresses were not included in the present study and were skipped and replaced before visiting the property. These addresses were then visited to obtain a response to the survey. This ensured that the present study was conducted randomly and in an unbiased manner. The size of the respective localities was determined, not by geographical area, but by the number of registered municipal stands (properties). This information was obtained from the surveyor-general in Bloemfontein and the Department of Land Affairs in Thaba Nchu.

Statistical tests took the form of a one-way analysis of variance (ANOVA).

*DOG OWNERSHIP ATTITUDES*

The study area (see Chapter 1), although the same as in the dog density survey above, was divided slightly differently to the first survey. The study area was split into four different types, which will be referred to as localities, with regard to the level of development and socio-economic status. The first locality included areas such as Universitas and Willows. The second locality included Heidedal, which in turn consists of three sub-divisions of varying socio-economic levels, all of which are lower than that of Bloemfontein, the affluent locality. This justified the inclusion of Heidedal as a separate locality. The third locality was Botshabelo and the fourth locality was Thaba Nchu. Further differences between these localities were also identified in making distinctions such as the geographical positioning of the localities, which in turn affects the accessibility to resources.

A questionnaire, consisting of 16 questions was compiled (see Annexure 1). A total of 50 questionnaires were printed for each locality. Dog-owners in the respective localities were approached and interviewed with regard to the questionnaires. This was done individually and at random, by approaching any dog owner. An attempt was made to spread the questionnaires across the entire locality. Each questionnaire was given to a dog-owner, answered and retrieved immediately. In the less developed areas an interpreter was used in order to explain the questions in the preferred language of that specific area. The questions were set up mainly in a close-ended manner (Kellert 1988) so that the respondent had to choose from a set of possibilities. The reason for this was firstly, to test specific aspects of the study and secondly, to prevent the respondents from giving a large variety of possibly unrelated answers. There were, however, two open-ended questions (questions 9 and 13), which allowed the respondent to provide his/her own answers.

The "approved product" category includes all products available for the treatment of tick or flea infestations that are obtainable from approved dealerships (like pet shops) and which are registered (Act 36 of 1947) for use on dogs.

Statistical tests took the form of a Chi-square test.

# RESULTS AND DISCUSSION

*DOG DENSITY*

The minimum, maximum and total number of dogs per household in the different study localities are summarised in Table 7.1.

**Table 7.1.** Summary of the number of dogs sampled in each locality.

| REGION | No. of Dogs | | | | SAMPLE SIZE |
|---|---|---|---|---|---|
| | Min. | Max. | Total | $\bar{x}$ | |
| **Bloemfontein** | 0 | 4 | 109 | 0.81 | 135 |
| **Botshabelo** | 0 | 18 | 187 | 0.80 | 233 |
| **Thaba Nchu** | 0 | 2 | 19 | 0.59 | 32 |
| | | | | | **Tot. 400** |

In the preparation phase of the present study, it was decided that about 400 samples would be an adequate sample for accurate results regarding the study localities. The minimum number of dogs found per property was zero. The maximum number of dogs found per household in Bloemfontein, Botshabelo and Thaba Nchu was 4, 18 and 2, respectively. The maximum of 18 in the case of Botshabelo is referred to as an outlier as it is not the trend in the locality. Being an exception to the rule, it has very little weight and hence very little influence on the mean value. The mean number of dogs found per property in these three study localities were 0.81, 0.80 and 0.59 respectively, illustrated in Fig. 7.1. The mean values did not differ significantly ($P>0.05$).

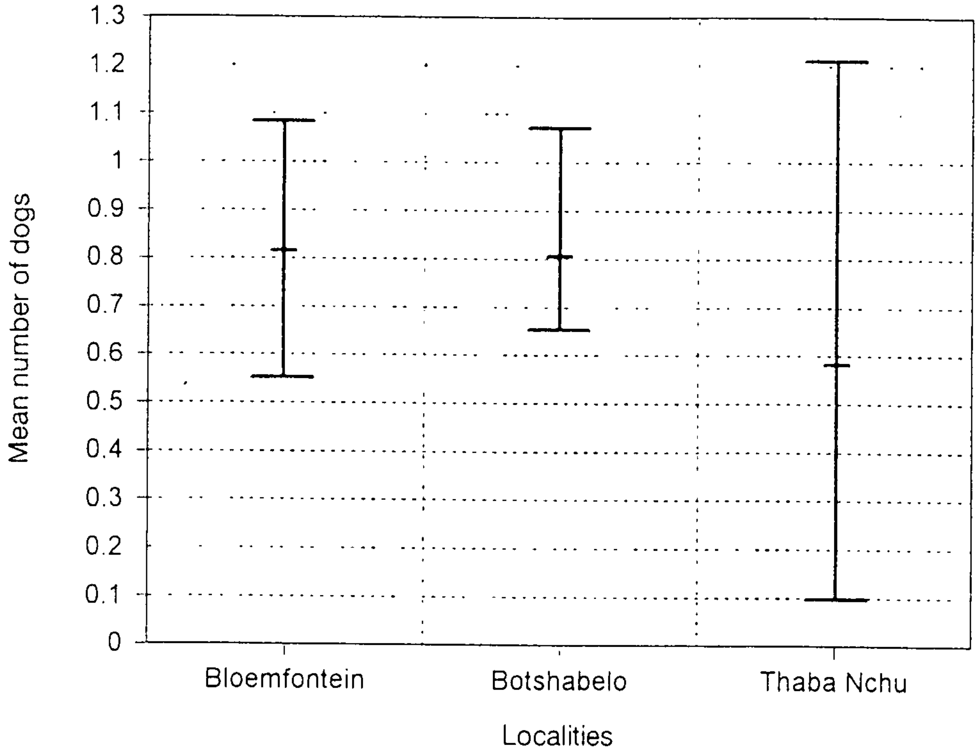


**Figure 7.1.** Mean number and 95% confidence intervals of dogs per household in the three different study localities.

The mean number and confidence intervals (95%) of dogs / household in the "urban: formal" and "urban: informal" communities were compared and are presented graphically in Fig. 7.2.

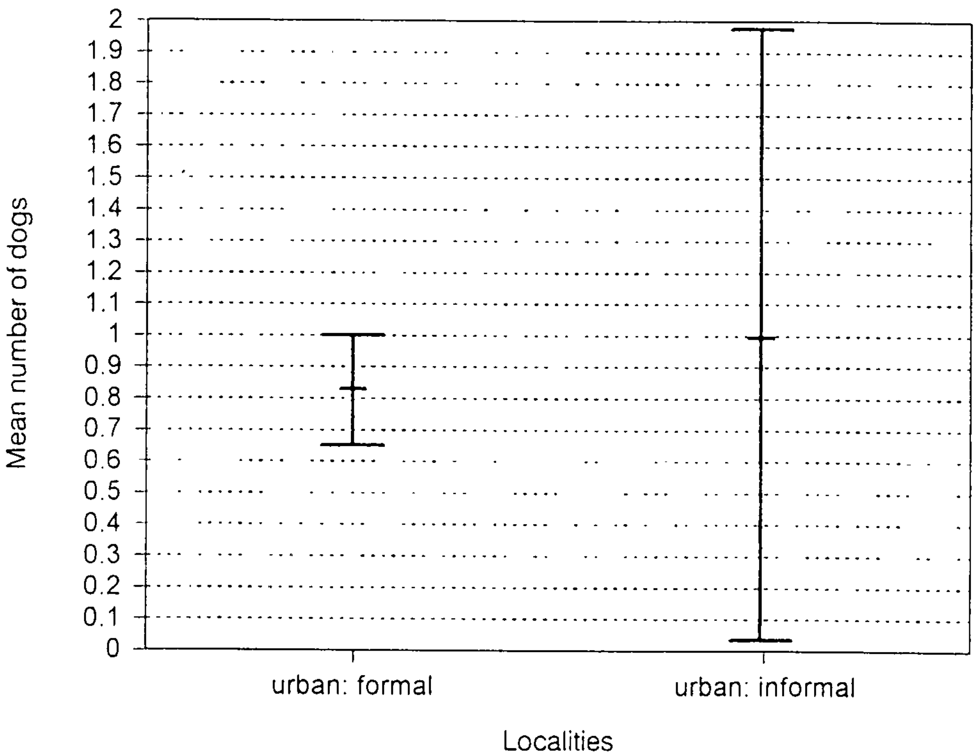


**Figure 7.2.** Mean number of dogs per household and 95% confidence intervals in urban: formal and urban: informal areas.

The "urban: formal" areas displayed the least variation as compared to the "urban: informal" areas. The means of the "urban: formal" and "urban: informal" areas were 0.82 and 1.0, respectively and were comparable but the maximum and minimum values clearly showed a greater deal of variation in the case of the "urban: informal" areas. Despite all this, statistical tests show that there was no significant difference between the mean number of dogs found in the "urban: formal" and "urban: informal" areas (P = 0.72). In reality the households in the "urban: formal" areas had at least one dog but in the case of the "urban: informal" areas there were many households with two dogs and also many with no dogs.

The present study was done to determine and compare the density of dogs found in the different localities. The information obtained from it can be used to quantify both the tick and tick-related disease problems in the respective areas. In short, in the "urban: formal" areas there appeared to be a tendency for families (per household) to own a dog. On the other hand, the situation in the "urban: informal" areas, appeared to be different. Those households that own dogs, own more than one dog. In contrast, there were households that did not have dogs at all. The reasons for this might become clear later in this chapter when the "reasons for owning a dog" are discussed. In a previous study conducted by Rautenbach, Boomker and De Villiers (1991) it was shown that the dog density, in a small town called Maboloka, was 0.68 dogs per household with a human to dog ratio of 11.1 to 1. This is low in comparison to that of developed countries. France, the United Kingdom and the USA have human to dog ratios of 6.3 to 1, 9.4 to 1 and 7.3 to 1, respectively (Schneider 1975; Singleton 1976). These countries are highly developed and have socio-economic levels much higher than South Africa. The implication of this is that the more affluent communities keep more dogs because they can afford to feed them. However, this does not mean the poorer communities do not care enough to want more dogs but that there are others factors involved. Rautenbach *et al.* (1991) indicated that the relative under population in Maboloka could be the result of a high death rate. This would be applicable to any socio-economically poor community and could mean that the poorer communities in South Africa have low dog densities simply because they cannot afford to keep dogs.

*DOG OWNERSHIP ATTITUDES*

Some of the questions posed in the questionnaire survey on the attitudes of dog-owners towards ectoparasitic infestations on their dogs, are summarised in Table 7.2(a). These only represent a part of the questionnaire and were chosen to highlight the dog owners' attitude towards ectoparasitic infestations on their dogs. The rest of the questions can be seen in Annexure 1, some of which will be dealt with later in this chapter. The results of the answers to the selected questions in Table 7.2(a) are summarised in Table 7.2(b).

**Table 7.2 (a).** Summary of a few selected questions posed in the questionnaire to determine attitudes of dog-owners to ectoparasite infestation of their dogs.

| | Questions posed in Survey (see Table 7.2(b) for findings) |
|---|---|
| 3 | Are your dogs restricted to your yard? |
| 4 | Are your dogs infested with ticks? |
| 5 | Are your dogs infested with fleas? |
| 6 | Do you regard these ticks as being a problem? |
| 7 | Have any of your dogs died of a tick related disease? |
| 10 | Are the control measures (which you use), effective? |
| 11 | Do you use the dipping facility at the SPCA? |
| 12 | Do you think there is a need for improved methods (or products) for control of ticks? |
| 14 | Did you know that ticks are vectors for diseases sometimes transmitted to humans (e.g. tick-bite fever, Q-fever)? |
| 15 | Are you aware of the seriousness of these diseases? |

**Table 7.2 (b).** Summary of responses to selected questions posed in Questionnaire for four different localities.

| | Bloemfontein | | | | Botshabelo | | | | Heidedal | | | | Thaba Nchu | | | | P values |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Ques | Yes | No | ? | n | Yes | No | ? | n | Yes | No | ? | n | Yes | No | ? | n | |
| 3 | 96 | 4 | - | 100 | 20 | 40 | - | 60 | 84 | 16 | - | 100 | 58 | 42 | - | 100 | < 0.005 |
| 4 | 38 | 62 | - | 100 | 48 | 52 | - | 100 | 72 | 28 | - | 100 | 60 | 40 | - | 100 | < 0.0001 |
| 5 | 38 | 62 | - | 100 | 60 | 40 | - | 100 | 10 | 90 | - | 100 | 66 | 34 | - | 100 | < 0.0001 |
| 6 | 50 | 38 | 12 | 100 | 52 | 40 | 8 | 100 | 90 | 4 | 6 | 100 | 80 | 20 | - | 100 | < 0.0001 |
| 7 | 8 | 86 | 4 | 98 | 50 | 44 | 6 | 100 | 40 | 52 | 8 | 100 | 44 | 42 | 14 | 100 | < 0.0001 |
| 10 | 76 | 4 | 10 | 90 | 48 | 36 | 16 | 100 | 50 | 26 | 24 | 100 | 76 | 12 | 12 | 100 | < 0.0001 |
| 11 | 0 | 88 | 12 | 100 | 16 | 60 | 24 | 100 | 18 | 46 | 36 | 100 | 8 | 90 | 2 | 100 | < 0.001 |
| 12 | 58 | 42 | - | 100 | 88 | 12 | - | 100 | 70 | 26 | - | 96 | 74 | 26 | - | 100 | < 0.01 |
| 14 | 88 | 12 | - | 100 | 20 | 80 | - | 100 | 30 | 70 | - | 100 | 38 | 62 | - | 100 | < 0.0001 |
| 15 | 86 | 14 | - | 100 | 40 | 60 | - | 100 | 18 | 42 | - | 60 | 48 | 52 | - | 100 | < 0.0001 |

The samples that did not add up to 100 resulted from respondents that did not answer a particular question. In Bloemfontein and Heidedal the percentage of households that restrict their dogs to their yards was 96% and 84%, respectively. The other localities displayed a very low percentage (< 59%). This means that most dogs in the more affluent areas were not allowed to roam freely around the locality. In the case of Botshabelo and Thaba Nchu, a very small percentage of the dogs were restricted to a property. This can be accounted for by the fact that most of the properties in these localities were not properly fenced off. The existing fences were more than often in a state of collapse (Krige 1998). This made it difficult to restrict a dog to a particular property if it is not chained.

Most of the respondents in Heidedal and Thaba Nchu believed that their dogs were infested with ticks whereas 62% of the respondents in Bloemfontein believed that their dogs were not infested with ticks. This suggests that many dog owners were aware of their animal's general condition. The same applies to fleas with the exception of Heidedal where 90% of the respondents did not believe their dogs have fleas (Table 7.2b).

Heidedal and Thaba Nchu's respondents regarded ticks as being a problem, with percentages as high as 90% and 80%, respectively. It was interesting to note that Bloemfontein and Botshabelo displayed much lower percentages. Whether this was due to being well informed with regard to control methods or simply ignorance is unknown at this stage. This was also reflected in the fact that only 8% of the respondents in the affluent areas reported that one or more of their dogs had died due to tick related diseases. In the other localities much higher percentages (> 39%) were reported. In support of this was the fact that the majority of the respondents from both Bloemfontein (76%) and Thaba Nchu (76%) felt that the control measures that they were using were effective. The other localities reported a lower percentage (< 51%). The respondents from Thaba Nchu also believed that their control measures were effective but still felt that ticks were a problem. This was a sign of concern for their dogs.

The question regarding the use of the SPCA's dipping facility was perhaps not a very fair one because the SPCA is too far and therefore inaccessible to the people who stay in Thaba Nchu and Botshabelo. The highest percentage of the respondents that claimed not to use the dipping facility at the SPCA comes from Bloemfontein (88%)

and Thaba Nchu (90%). The SPCA provides a free dipping service to the surrounding communities. In the case of Bloemfontein, it is probably due to the fact that they can afford to buy their own remedies and thus treat their dogs at home. In the case of Thaba Nchu, it was understandable that they would not be able to use the SPCA's facilities because of the long distance they would have to travel. It was interesting that some of the respondents from Thaba Nchu (which is 65km away) claimed to use it. This was also true for Botshabelo (55km away), which is also a long distance from the SPCA. This could be explained by the fact that many of the people who stay in these localities commute into Bloemfontein on a weekly or daily basis, to work. It is under these circumstances that it is possible. Alternatively, it is also possible that the question was not clearly understood. The majority of respondents from all four localities felt that there was a need for improved products. This suggests that there was a concern, on the dog owner's part, with regard to the dog's condition and well being.

High percentages ($\geq$70%) of the respondents in Botshabelo, Heidedal and Thaba Nchu felt that there was a need for improved methods of tick control. On the other hand, although all localities indicated awareness, there was a higher percentage (88%) of respondents in Bloemfontein that indicated that they were aware of the fact that ticks are vectors of diseases transmitted to humans. The same applied to the question on the seriousness of these diseases. The affluent areas displayed the highest percentage (86%) of respondents who indicated that they were aware of it (Table 7.2b). There is a general belief that people who live in affluent areas like those selected under the Bloemfontein banner, are better educated and better informed. The fact that the respondents from the other localities, although to a lesser degree, indicated that they are aware of the above-mentioned is probably due to experience. Even without detailed information it does not take much to link the death of a dog with a heavy tick infestation.

The categories of tick control products used by the respondents, at the different localities, are summarised in Table 7.3.

**Table 7.3.** Summary on the type of product used for tick control. Values expressed in percentage form.

| RESPONSE | BLOEMFONTIEN | BOTSHABELO | HEIDEDAL | THABA NCHU |
|---|---|---|---|---|
| Approved products – other than dip | 58 | 2 | 62 | 2 |
| Dip | 40 | 80 | 48 | 54 |
| Used engine oil | - | - | - | 8 |
| Jeyes Fluid | 2 | 2 | - | 20 |
| Blue death | - | - | - | 2 |

Most of the respondents in Bloemfontein (58%) and Heidedal (62%) used approved products. This was far more than in the other two localities (< 3%). The approved products refer to the products that have been registered (in accordance with Act 36 of 1947) to be used on dogs. The respondents of Bloemfontein and Heidedal used these products to a great extent whereas the other localities did not. The reason for this has to be socio-economically based because the approved products are expensive (comparatively speaking) and not everybody can afford to buy these products. Although dip is also an approved product it was separated from the other products. The reason for this was that many respondents used it in conjunction with other products. All four localities had a high percentage of respondents that use dip but the highest was in Botshabelo (80%). Used engine oil was only utilised in Thaba Nchu and was a method used on cattle in this area as well (Dreyer, Fourie and Kok 1998). The percentage of respondents who used Jeyes Fluid was also highest in Thaba Nchu. Heidedal was the only locality in which Jeyes Fluid was not used. The use of "Jeyes fluid" is troubling, as it is toxic. In fact it is absorbed through the skin of the dogs and if the concentration is too high, the dog dies (Dreyer pers comm 1997). Blue death, registered for use against ants and certainly not for ticks, was only used in Thaba Nchu. The fact that respondents, including those in Bloemfontein, actually used products like Blue death and Jeyes fluid indicates that there was an apparent lack of correct information regarding products that can be used for tick control but also that there was a concern for the dog. The use of toxic compounds not registered for use may also pose a potential hazard for the owner. This concern of the dog owners for their dogs may be so great that the dog owner would attempt to use alternative forms of control to combat the tick problem, sometimes to the detriment of the animal.

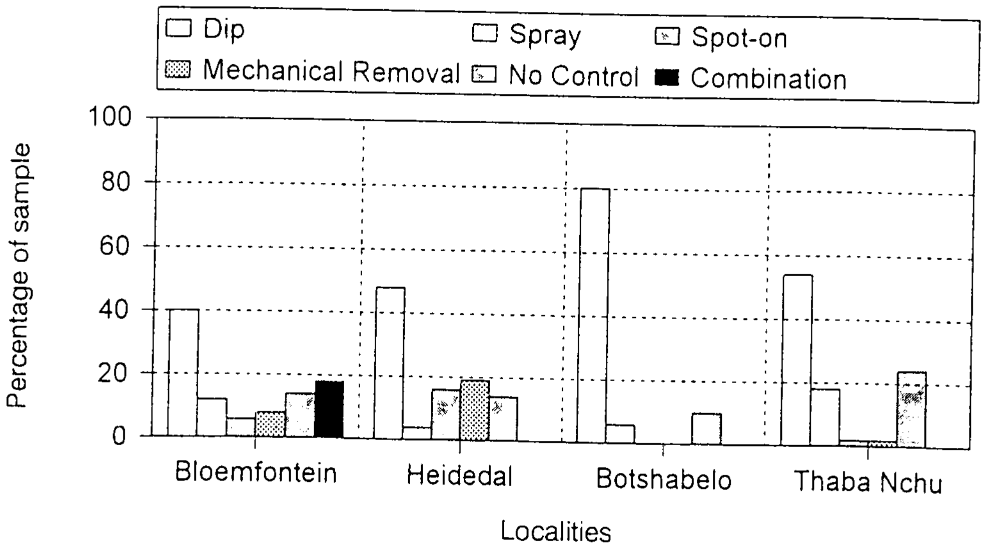


**Figure 7.3.** Method of tick control practised in the different study localities.

The different tick control methods used by the respondents in the different study localities are illustrated in Fig. 7.3. There was a significant difference between the different localities regarding different methods of tick control (P<0.0001). A submersion dip was the method most commonly used by the respondents in all four localities (between 40-80%). The second most commonly used method of control was the use of an acaricidal spray, although the percentages were generally small (< 18%). It was of particular interest that the physical removal of ticks from the dogs was less popular in Thaba Nchu and not practised in Botshabelo. This is perhaps a good thing because of the fact that the ticks found on dogs are vectors for diseases transmissible to humans. This makes physical removal an undesirable practice unless proper equipment, like forceps, is used. Another good method of control was the use of an acaricidal spot-on, a method that was more popular in Bloemfontein and Heidedal. It was also interesting that there were many people who did not take any measures of control and it is perhaps these people that need to be targeted with regard to education on the dangers of uncontrolled tick infestations.

Recommendations, made by the respondents in response to question 13, to solve the tick problem are summarised in Table 7.4.

**Table 7.4.** Summary of results on suggestions regarding tick control.

| Locality | Better products | Regular dipping | Provision of Clinic | Poison | Nothing | No response |
|---|---|---|---|---|---|---|
| **Bloemfontein** | - | 1 | - | 1 | - | 48 |
| **Botshabelo** | - | - | 10 | - | 1 | 39 |
| **Heidedal** | 2 | 2 | - | - | - | 46 |
| **Thaba Nchu** | - | - | 19 | - | - | 31 |

From the large percentage of non-responses in Table 7.4, it is evident that most respondents did not have any idea as to what should or could be done to combat the tick problem. However, in Botshabelo and Thaba Nchu, many respondents felt that a provision of a clinic would solve the problem. It was not clear what their expectations were with regard to the service the clinic should deliver e.g. dipping, veterinary services, etc, but it was clear that they associate a clinic with the solutions to their problems. It was also understandable that they would want a clinic of some sort because these localities are situated far from establishments like the SPCA and veterinarians. The fact of the matter is that they do not have access to the necessary facilities, which is a problem that needs to be addressed.

The percentage of animals possibly kept as pets, other than dogs, is summarised in Table 7.5 and gives an indication of the diversity in this regard. There was a significant difference between the localities ($P < 0.0001$).

**Table 7.5.** Summary of percentage of respondents, who have pets other than dogs.

| OTHER PETS | BLOEMFONTEIN | HEIDEDAL | BOTSHABELO | THABA NCHU |
|---|---|---|---|---|
| Cats | 26 | 20 | 12 | 28 |
| Birds | 18 | 6 | 20 | 10 |
| Cats & Birds | 0 | 4 | 8 | 10 |
| Cats & Birds & Other | 0 | 0 | 4 | 14 |
| None | 56 | 70 | 56 | 38 |

The first thing to note is that most dog-owners who responded, did not own other pets. This was true for 56% respondents in Bloemfontein, 56% in Botshabelo, 70% in Heidedal and 38% in Thaba Nchu, respectively. Pets, which are very often kept in addition to dogs, are cats and this is followed by birds. It should be noted that, as it was later discovered, in areas like Botshabelo and Thaba Nchu, the respondents often included chickens in the category of pets. Respondents in Botshabelo and Thaba Nchu often owned chickens (a food source) whereas those in Heidedal and Bloemfontein owned parrots or canaries. In the case of Botshabelo and Thaba Nchu, the percentages regarding the birds kept as pets were not necessarily accurate as a result of an oversight in the questionnaire structure.

The reasons for owning a dog, as supplied by the different groups of respondents, are summarised in Table 7.6.

**Table 7.6.** Summary of reasons for owning a dog as supplied by respondents. Values are expressed in percentages.

| REASONS | BLOEMFONTEIN | HEIDEDAL | BOTSHABELO | THABA NCHU |
|---|---|---|---|---|
| (C)ompanion | 4 | 0 | 0 | 0 |
| (G)uard dog | 6 | 52 | 96 | 82 |
| (P)et | 38 | 48 | 2 | 6 |
| (H)unting | 0 | 0 | 2 | 12 |
| (B)reeding | 4 | 0 | 0 | 0 |
| (P) & (G) | 24 | 0 | 0 | 0 |
| (C) & (P) | 12 | 0 | 0 | 0 |
| (C), (P) & (G) | 12 | 0 | 0 | 0 |

There was a significant difference ($P< 0.0001$) between the different localities with regard to reasons for keeping a dog. In Bloemfontein the respondents indicated that they kept dogs for a variety of reasons, with the exception of hunting. The main reason for keeping a dog in Bloemfontein was as a pet (38%). Botshabelo and Thaba Nchu displayed only a small percentage of respondents who used this as a reason for keeping a dog. Respondents in Heidedal kept dogs mainly for two reasons, namely, as pets (48%) and as guard dogs (52%). Companionship as a reason (or in combination with another reason) for keeping a dog was only given in Bloemfontein, although in small percentages (4 - 6%). In Botshabelo (96%) and Thaba Nchu (82%) the main reason for keeping a dog was for it to serve as a guard dog (i.e. for protecting assets) and also for hunting. In Bloemfontein the percentage was very low (6%) and suggested that these respondents have other means of protecting their assets e.g. alarm systems. It would appear that the dogs in Botshabelo and Thaba Nchu have to serve a purpose in order to "earn" their keep. This is probably due to the fact that these respondents are poorer and that anything caught by their dogs can provide a form of nourishment, e.g. rabbits. This was also coupled with the fact that some of these respondents own cattle and that the dogs accompany the cattle when they go out to graze. It is while they are out in the veld with the grazing cattle that these dogs are able to hunt. This brings up another important fact. The respondents from these localities indicated that the reason for keeping a dog was mainly for protection. The problem that many have is cattle theft (Dreyer, pers comm 1997) and this is partially overcome by allowing their dogs to accompany the cattle. This would be a good motive for keeping dogs that would otherwise be expensive to feed. It is also another reason why the dogs in these localities are less restricted, as mentioned earlier.

It was shown by Odendaal and Weyers (1990) that in South Africa, people who display affection towards their dogs do not use them for economic purposes like hunting or breeding. In the developing countries of southern Africa, dogs are used mostly for hunting, protection and sometimes as pets (Odendaal and Weyers 1990). This was certainly evident in the present study. Furthermore, the older respondents (between 55 and 64 years) kept more dogs. The reason suggested for this was that people of that age group could afford it and used the companion animals as substitutes during the "empty nest" phase of life (Odendaal 1994). This was also true for the more educated respondents and is probably due to the fact that the educated have better jobs and can thus afford to (Odendaal 1994). It is accepted that factors such as age, education, urbanisation, and social class status influence the formation of attitudes (Herzog & Burghardt 1988). There are dramatic differences in the perceptions between the highly educated and the least educated and between the various socio-economic and ethnic groups in society (Herzog & Burghardt 1988).

In a previous study, addressing the relationship of knowledge to age, sex, ethnicity and urbanisation, conducted in America by Kellert (1988), demographic groups were distinguished by age, education, income, race, and residence, among other things. In his study it was found that the respondents with limited education scored much lower than those who were highly educated. This suggested that, comparatively speaking, the least educated showed a lack of interest and affection for animals on the whole. In a previous study conducted by Blackshaw and Day (1994) in Australia it was shown that companionship and to a lesser extent, protection, were the main reasons for owning a dog. The results of the present study tend towards the contrary in respect of the interest shown by the least educated as well as the reasons for owning a dog.

Odendaal (1994) points out that the interpretation of data obtained in a study, that addresses the demographics of companion animals, is difficult because of the numerous contributing factors. These are factors (e.g. breed and sex of the animals, availability of space and facilities, community hygiene, amongst others) that were not specifically looked at in the present study and present the opportunity for an independent study. Furthermore, there were several shortcomings with regard to the present study. The first would have to be the language problem. The fact that there is such a spectrum of languages and dialects made communication difficult. The present study hinged on the use of interpreters and it was difficult to ascertain whether all parties had the same understanding of the questions posed. The time constraint was an added factor. This type of study is complex and there are many influencing factors. The same was found to be true by Odendaal (1994) who highlights the fact that the problems are too large for the scope of an ordinary questionnaire survey.

Shortcomings aside, it can be concluded that the different socio-economic circumstances of the affluent and poorer localities influence the reasons for keeping a dog, which ranges from the dependency of keeping a dog as a companion to the practicality of keeping a dog for hunting. However, whatever the reason, there appears to be a common deep concern for dogs amongst all groups. Lastly, socio-economic circumstances and possibly the lack of knowledge (or exposure) regarding the use of safe effective products appear to be factors which determine which products are used to combat the tick problem. The high percentage of respondents in all localities that attempt to control ticks, however, indicates that the dog owners do care for their dogs.

ANNEXURE I

# SURVEY / QUESTIONNAIRE

Area: ____________________

Address: ____________________

____________________

____________________

Dog Owner: ____________________

Family Size: ____________________

Unmarried / Single: Yes ☐ No ☐

Age: __________

1. **How many dogs do you own ?**

| | | Male | Female |
|---|---|---|---|
| a) | 0 | ☐ | ☐ |
| b) | 1 | ☐ | ☐ |
| c) | 2 | ☐ | ☐ |
| d) | 3 | ☐ | ☐ |
| e) | more than 3 | ☐ | ☐ |

2. **Why do you own a dog ?**

a) as a companion ☐
b) as a guard dog ☐
c) as a pet ☐
d) for hunting ☐
e) for breeding ☐

3. **Is your dog restricted to your yard ?**

a) yes ☐
b) no, the dog is free to roam ☐

4. **Are your dogs infested with ticks ?**

a) Yes ☐
b) No ☐

5. **Are your dogs infested with fleas ?**

a) Yes ☐
b) No ☐

6. **Do you regard these ticks as being a problem ?**

a) Yes ☐
b) No ☐
c) I do not know ☐

7. **Have any of your dogs died from a tick related disease ?**

a) Yes ☐
b) No ☐
c) not sure if tick related ☐

8. **What measures do you take to get rid of the ticks on your dog ?**

a) dip ☐
b) spray ☐
c) spot-on ☐
d) physical removal ☐
e) no control ☐

9. **What product do you use ?**

______________________

______________________

10. **Are the control measures (which you use) effective ?**

a) Yes ☐
b) No ☐
c) I am not sure ☐

11. **Do you use the dipping facilities at the SPCA?**

a) Yes ☐
b) No ☐
c) I did not know they had a dipping facility ☐

12. Do you think there is a need for improved methods (or products) for the control of ticks ?

a) Yes ☐
b) No ☐

13. What do you suggest should be done to control the tick problem ?

______________________________

______________________________

______________________________

14. Did you know that ticks are vectors for diseases that are sometimes transmitted to humans (e.g. tick bite fever, Q-fever) ?

a) Yes ☐
b) No ☐

15. Are you aware of the seriousness of these diseases ?

a) Yes ☐
b) No ☐

16. Do you have any other pets in the family ?

a) cats ☐
b) birds ☐
c) other ☐

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# SUMMARY

The present study was conducted in the greater Bloemfontein area and focussed on the interaction between ticks and dogs. The sampling localities included Bloemfontein, Botshabelo and Thaba Nchu, and various sub-divisions thereof. *Rhipicephalus sanguineus* and *Haemaphysalis leachi*, being dog ticks and the expected dominant species, were subject to close scrutiny. The present study was a comparative one which consisted of both field and laboratory work and which was designed to address several objectives. The first objective was to study tick diversity, prevalence and relative density. The second objective was to study the effect of various abiotic factors (temperature, relative humidity and photoperiod) on the development rate and survival of eggs, larvae and nymphs of the two dominant species. The third objective was to examine the influence that social factors (e.g. attitudes of dog owners, socio-economic levels, education and awareness, development levels in terms of infrastructure) have on the relationship between ticks and dogs.

The results of the present study indicated that the dogs in the greater Bloemfontien were parasitised by at least nine different species. The species with the highest prevalence (and relative density) were *R. sanguineus* (73.5% and 27.4) and *H. leachi* (22.4% and 5.8). Other species constituted less than 1.2% of the total sample and were regarded as being incidental. These species included *Boophilus decoloratus*, *Hyalomma truncatum*, *Rhipicephalus evertsi evertsi*, *Ixodes rubicundus*, *Rhipicephalus* sp., *Rhipicephalus gertrudae* and *Rhipicephalus follis*. There was a clear distinction between the different

attachment sites of the two dominant species and both species displayed seasonal patterns. Greater numbers of the two dominant species were found during the warm summer months and very few during the cold winter months.

Daily egg production of the two dominant species was similar to that of other ixodid ticks while the pre-oviposition and incubation periods decreased with an increase in temperature. Pre-oviposition periods for *R. sanguineus* ranged from 4.8 (25°C, 90%RH) to 21.0 (15°C, 40%RH) days and for *H. leachi* from 4.3 (30°C, 90%RH) to 12.1 (15°C, 40%RH) days. Incubation periods for *R. sanguineus* ranged from 19.0 (30°C, 90%RH) to 72.0 (15°C, 90%RH) days and for *H. leachi*, from 15.5 (30°C, 90%RH) to 66.7 (15°C, 40%RH) days. The fecundity of *R. sanguineus* ranged from 10.3 (15°C, 40%RH) to 17.2 (25°C, 90%RH), while for *H. leachi* it ranged from 7.7 (15°C, 40%RH) to 16.3 (15°C, 90%RH) eggs per mg body mass. The fecundity for both species, at a specific temperature, increased with an increase in relative humidity. Generally, unfed larvae and nymphs of both species survived for longer periods at lower temperatures. The pre-moult period of larvae and nymphs of both species decreased with an increase in temperature. Both engorged larvae and nymphs of both species displayed a circadian drop-off rhythm.

The general condition of the dogs in the affluent localities were better than those found in other localities. Dogs tested positive for *Ehrlichia canis* and *Ehrlichia chaffeensis* in all localities but was the highest in Thaba Nchu. The mean number of dogs in the different localities did not differ significantly. Generally speaking most respondents did indicate concern for their dogs and many saw ticks as being a problem. However, the level of education was a determining factor when it came to the understanding of the relevant diseases and the vectors thereof. Ignorance and a lack of necessary resources were important factors contributing to poor animal health in the greater Bloemfontein. The attitudes displayed by dog owners also contributed towards the general condition of the dogs sampled.

There is little information available on *R. sanguineus* and *H. leachi* generally and particularly in the greater Bloemfontein. The present study has provided some basic information regarding (i) the diversity, prevalence and relative density, (ii) the effects of abiotic factors on eggs, larvae and nymphs, and (iii) the influence that social factors (particularly the attitudes of dog owners) have on the relationship between these two dominant tick species and dogs. This type of information can contribute substantially towards endeavours to effectively control the tick species that parasitise our dogs.

## KEY TERMS

Ticks

Dogs

Diversity

Prevalence

Relative density

Oviposition

Fecundity

Conversion Efficiency Index (CEI)

Survival

Moulting

Circadian drop-off rhythm

Serology

Attitudes

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