Ecological Dynamics of Tick-Borne Zoonoses

Daniel E. Sonenshine
Thomas N. Mather,
Editors

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Preface

The importance of tick-borne diseases to public health appears to have grown greatly during the 20th century. In addition, knowledge of the tick-transmitted diseases affecting livestock and other animals has expanded greatly since the seminal discovery of the Texas cattle fever agent, *Babesia bigemmina*, by Smith and Kilbourne in 1893. However, despite great progress in knowledge, most of these diseases remain and some have even increased in range. Control of these pervasive tick-borne disease problems has been hindered by significant gaps in our theoretical knowledge as well as a general lack of suitable tools and methods needed to evaluate the dynamic interplay between diverse factors regulating tick populations and the pathogens that they transmit.

The purpose of this book is to provide the reader with the knowledge necessary to understand the ecological principles that contribute to the occurrence of tick-borne disease affecting public health, livestock, pets, and wildlife, as well as to apply those principles to specific disease problems. The goal is not to treat each principle in the abstract, but rather to consider how various factors integrate dynamically. To this end, the book is divided into two parts, with the beginning contributions (Chapters 1–8) focusing on principles related to the dynamics of zoonoses and later contributions (Chapters 9–13) providing examples of how these principles integrate in relation to particular zoonoses.

In planning this volume, we asked each contributor to consider integrating the existing descriptive knowledge of biotic and abiotic factors as they affect vector and reservoir competence, population regulation, geographic dissemination, or pathogen transmission and survival with examples, or even speculations on how these factors interact to result in the enzootic and epizootic states observed in nature. Thus, these chapters offer insights into ecological processes affecting disease maintenance, disease spread, pathogen amplification, and many other attributes of the various zoonoses rather than rote, elementary descriptions.

The editors are very much indebted to the many contributors, all of whom are outstanding researchers in the field of tick-borne zoonoses. We especially appreciate their willingness to go beyond the standard review format so common in other texts, and to contribute new and often untested hypotheses. We hope that the description of the ecological principles and

detailed examination of representative tick-borne diseases will enable the reader to understand how knowledge of zoonotic disease ecology may improve the public health, and, ultimately, reduce or even eradicate these important disease problems. We also hope that this book will foster excitement and increased interest in the field of vector ecology, as well as assist in developing strategies for management of existing and emerging tick-borne zoonoses.

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October 1993

Contents

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I

PRINCIPLES

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1

Introduction

DANIEL E. SONENSHINE

1. INTRODUCTION

Identification of the infectious organisms and their arthropod vectors are the essential first steps that enable scientists to understand vector-borne diseases, i.e., disease agents that can infect vertebrate animals and (in most cases) humans. Many, perhaps most, vector-borne diseases are zoonoses, i.e., they infect a variety of vertebrates and are also transmissible to man. In the case of tick-borne zoonoses, knowledge of these diseases has accumulated gradually over the preceding 100 years. Smith and Kilbourne's (1893) report of the transmission of the malaria-like protozoan, *Babesia bigemina*, by the cattle tick, *Boophilus annulatus*, began this era of discovery, which has continued throughout the 20th century. Numerous reports of tick transmission of other diseases soon followed: examples include relapsing fever by soft ticks (Dutton and Todd, 1905), East Coast Fever by the African brown ear tick (*Rhipicephalus appendiculatus*) (Theiler, 1904), Rocky Mountain spotted fever by wood ticks (*Dermacentor andersoni*) (Ricketts, 1906, 1907), Boutonneuse fever by the brown dog tick (*Rhipicephalus sanguineus*) (Brumpt, 1932), and babesiosis of cattle, dogs, and other animals by many different tick species (summarized by Kuttler, 1988). The decades since World War II have seen equally dramatic discoveries, including tick transmission of tick-borne encephalitis, Crimean–Congo Hemorrhagic Fever (Chumakov et al., 1968, 1969; Casals, 1969), Kyasanur Forest Disease (Bhat, 1991), Lyme disease (Johnson et al., 1984) and human Ehrlichiosis (Anderson et al., 1991), to name but a few examples. These important discoveries have led to a vast literature dealing with the biology of the pathogenic agent and the pathologic response of the human or animal hosts.

Important as these discoveries were to the understanding of tick-borne diseases, they are only the beginning of the process. Scientists now recognize that the ecological relationships that exist between tick vectors and pathogens in their zoonotic cycle can profoundly influence the patterns of transmission and disease for humans, domestic animals and wildlife. Without knowledge of these ecological relationships, it is impossible to understand the epidemiology of these tick-borne diseases. Consequently, in our quest for such knowledge,

our first task is to determine the dominant ecological parameters affecting the conservation and regulation of tick-borne zoonoses, as well as the factors that influence their geographic and seasonal distribution. Here, in this introduction, we will identify these factors and examine them briefly so as to obtain an overall understanding of the ecological dynamics of tick-borne zoonotic diseases. More detailed, comprehensive discussion of these subjects will be given in the following chapters.

What are the ecological parameters that significantly affect tick-borne zoonotic diseases? Among the most important are: (1) vector competency; (2) population dynamics of the vector species; (3) seasonal and diel activity of the tick vector; (4) zoogeographic range of the vectors; (5) host specificity and other host-related factors; and (6) habitat requirements, including micrometeorologic constraints. Major changes in the relationships among these factors determine the status of the disease, i.e., whether it is enzootic (relatively stable, little or no change in disease incidence) or epizootic (unstable, with large increases in disease incidence).

2. VECTOR COMPETENCY AND RESERVOIR HOSTS

Disease transmission is not a simple process nor is it the same in all tick-borne diseases. Vector competency plays an important role. Vector competency is the ability of biting arthropods to acquire and transmit infectious agents to their vertebrate hosts. Some tick species readily pass disease-causing agents to their hosts, i.e., they are highly competent vectors while others are inefficient vectors or are incompetent. This subject is explored in depth by R.S. Lane in Chapter 3. The dynamics of zoonotic disease are strongly influenced by the mechanism of transmission. The most common routes of transmission include: (1) transovarial (i.e., from parent to progeny via the ovaries), as occurs in the case of Rocky Mountain spotted fever (caused by *Rickettsia rickettsii*); and (2) transstadial, i.e., from one life stage to the next, as occurs in the case of Lyme borreliosis (caused by *Borrelia burgdorferi*). Substantial proliferation of infected vector ticks is common in diseases transmitted by the transovarial route. Despite this apparently efficient mechanism of disease dissemination (up to 100% transmission to ova in laboratory populations of *R. ricketsii*-infected *Dermacentor andersoni* was reported by Burgdorfer and Brinton (1975), infection rates in wild-caught ticks infected with pathogenic rickettsiae are relatively low (usually less than 10%). In contrast, no proliferation occurs when transmission is via the transstadial route. Nevertheless, remarkably high rates occur in many populations of ticks infected with *Borrelia burgdorferi* (from 50% to 100% in some populations) even though transovarial transmission is unimportant in maintaining this spirochete (Burgdorfer, 1992). Pathogen transmission can also be via horizontal or vertical means, with significant impact on the occurrence of disease. Physiological and ecological factors that may contribute to the success of these transmission processes are discussed in detail in Chapter 3.

Reservoir hosts are those animals upon which ticks feed that are also capable of acquiring, maintaining, and donating infectious organisms to other ticks. Thus, the reservoir host serves as a common infectious "pool" whereby disease-causing microbes are transferred from infected to uninfected ticks. However, many animals are incapable of acquiring or maintaining infectious tick-borne disease agents, i.e., they are not reservoir competent, even though they may be excellent hosts for feeding ticks. Thus, certain species of mice, especially the white-footed mouse, *P. leucopus*, are excellent reservoir hosts for the spirochete, *Borrelia burgdorferi*, the agent of Lyme borreliosis, as well as hosts for the vector tick, *Ixodes dammini* (= *I. scapularis*).* Other species of mice, e.g., the house mouse, *Mus musculus*, are also competent reservoir hosts but support only about 10% of the tick infestation supported by white-footed mice (Sonenshine, unpublished). In contrast, white-tailed deer (*Odocoileus virginianus*), while excellent amplifier hosts for the vector tick, are reservoir incompetent for the spirochete. Similarly, in the case of Crimean–Congo Hemorrhagic Fever (CCHF), few vertebrate hosts, e.g., hares (*Lepus* spp.) develop viremia of sufficient duration or titer to enable feeding ticks to acquire infection, despite the extremely wide variety of species that show antibody to CCHF. Thus, finding antibodies to a disease agent, although indicative of exposure, does not prove that the individual is a reservoir. To determine whether a particular animal is reservoir competent, laboratory tests can be done to show: (1) that an infection is developed in the animal following inoculation of infectious microbes by needle or bite of known infected ticks; and (2) that susceptible, uninfected (so-called "clean" ticks) become infected while feeding on the inoculated individual. Isolation of the infectious agents from wild-caught individuals provides additional evidence of reservoir competency. Conclusive evidence of reservoir competency can be obtained by **xenodiagnosis**, wherein susceptible, "clean" ticks acquire infection after being allowed to feed on a wild-caught individual.

3. SEASONAL ACTIVITY

Most ticks actively seek hosts only during certain well-defined periods of the year. These well-defined seasonal activity periods are the weeks or months when ticks transmit disease. Consequently, with few exceptions, transmission of tick-borne disease is also seasonal. Knowledge of the seasonal activity patterns of vector ticks is essential for determining the periods when people or livestock are at risk of acquiring infection.

When ticks emerge from diapause (see below), they **quest** for hosts. This is done by either: (1) ambush strategy, wherein ticks climb herbaceous or woody vegetation and wait for opportunities to attach to transient hosts; or (2) the

* A recent paper by Oliver et al. (1993) suggests that *I. dammini* and *I. scapularis* are the same species. Although the final status of the taxon *I. dammini* has not been resolved, this synonymy will be used hereafter.

hunter strategy, wherein ticks walk or run across open ground to attack hosts. Questing ticks are highly responsive to host stimuli, especially CO_2, NH_3, and body heat. The questing period may vary within the zoogeographic range of the species. Moreover, different life stages may quest at different periods of the year. These periods of seasonal activity are controlled by the tick's response to changing environmental conditions. In temperate and subarctic regions, changing photoperiod, incident solar energy and ambient temperature provide the specific cues that alter tick behavior, such as host-seeking activity (Sonenshine, 1991). In tropical regions, where variations in day length and temperature are less pronounced, alternating rainy versus dry seasons can become the controlling influence on tick seasonal activity. Once questing behavior of the ambush type (see below) has been initiated, individual ticks may remain in this active state for days or even weeks until forced by changes in body water balance to abandon this behavior and seek more sheltered, humid microenvironments. Often, periods of questing behavior alternate with periods of rehydration. In some species, questing behavior exhibits a diel cycle. In contrast, individuals recruited into the population when environmental changes signal unfavorable periods enter diapause, whereupon they cease questing altogether. Questing behavior by hunter ticks (see below) is rather different. In this case (e.g., *Amblyomma hebraeum*), the ticks remain hidden in protected microhabitats and only leave in response to host stimuli. The seasonal activity periods of the different life stage of the vector ticks presents another variable that strongly influences the dissemination of disease-causing organisms.

An example of the interactions between tick seasonal activity and disease is seen in the case of Rocky Mountain spotted fever (RMSF). Transmission of the causative agent, *Rickettsia rickettsii*, is dependent upon the bite of the vector tick, primarily *Dermacentor variabilis* in the eastern US and eastern Canada, and *D. andersoni* in western North America. In the east, transmission begins in late winter or early spring, but is limited almost exclusively to small mammals. This is because the only life stage active at this time is the larva. Overwintering *D. variabilis* larvae emerge from their winter diapause in March, April or even May, depending upon latitude (Sonenshine, 1979) and attack small mammals. Soon after the onset of larval feeding, these hosts acquire the infection. Examination of white-footed mice, *Peromyscus leucopus*, trapped in a study area near Richmond, Virginia, USA, showed that the first large-scale increase in seroconversions occurred in March. Thereafter, seroconversions increased as increasing numbers of infected larvae as well as newly molted nymphs joined the population. Studies by Korch (1984) at the Aberdeen Proving Grounds in Maryland, USA, also showed an increase in seroconversions during the spring (April to June) in the meadow vole, *Microtus pennsylvanicus*. Seropositive hosts were also found in late summer and fall, presumably in response to the second surge of immature tick activity that occurs then.

Human cases of RMSF appear in rapidly increasing numbers in May and generally reach their highest numbers during June, July, and August; few cases

are reported before early May or after late September. This period is consistent with the adult *D. variabilis* seasonal activity period, with a lag of 2–3 weeks. In the northeastern US, especially New York and southern New England, peak case frequency often occurs in May or June, reflecting the early dominance of overwintering adults in the vector tick population and the tendency towards a 2-year life cycle. In the Rocky Mountain region, cases are also concentrated in spring or early summer, when adult *D. andersoni* are active. Armed with this knowledge, physicians can reasonably exclude RMSF during other periods of the year when considering a diagnosis. Ecological factors important for the understanding of Rocky Mountain spotted fever are discussed more fully by Schriefer and Azad in Chapter 10.

Knowledge of vector seasonal activity is also important in determining when to apply expensive and time-consuming control measures. Thus, park and wildlife refuge managers as well as private persons interested in disrupting active transmission of Lyme disease are advised to dispense Damminix tubes (Ecohealth, Boston, MA, USA) or pesticidal applications from May through July, when nymphal *I. scapularis* are most active. Considerable controversy has occurred regarding the efficacy of Damminix, which was ineffective when tested in field trials in New York and Connecticut (Daniels et al., 1991; Stafford, 1991).

4. HOST SPECIFICITY AND HOST UTILIZATION

The occurrence of tick-borne disease depends on the vertebrate animals on which ticks feed as well as the ticks' ability to transmit disease. Just as we have seen limitations in vectorial capacity, we also find limits in the utilization of different vetebrates as hosts. In effect, all ticks do not feed equally on all vertebrate animals. Many, perhaps most, ticks show some degree of host specificity, accepting only a limited variety of animals as a candidate blood source. Such ticks are **host specific**, i.e., they are restricted to a particular class, order, or even genus of vertebrates as hosts. True, or **physiological host specificity**, represents the result of: (1) the tick's ability to recognize and respond to specific host-originated compounds, especially odorants such as CO_2, NH_3, lactic acid, and other volatiles, reinforced by thermal and contact stimuli characteristic of the host body; and (2) pharmacologically active compounds in tick saliva that enable the ticks to suppress (or evade) host homeostatic mechanisms, thereby enhancing blood flow into the feeding pool but minimizing inflammatory responses that call attention to the tick's presence. For detailed discussions of these phenomena, the reader is referred to the book, *Biology of Ticks*, Volume 1 (Sonenshine, 1992) and the excellent review by Ribeiro (1987). Host utilization is also the result of ecological determinants. Major ecological determinants include: (1) the habitat or habitats in which the ticks quest or hunt for hosts; (2) questing height, i.e., the elevation above the ground at which ticks rest while waiting for passing hosts; (3) diel questing, i.e., the time of day when ticks actively seek hosts. Thus, ticks which climb relatively high on to grassy or weedy vegetation (e.g., 1 m) and which can tolerate substantial water

loss can quest in relatively dry, meadow environments during daylight hours. Others, less tolerant of environmental extremes, are restricted to less stressful habitats (e.g., young, second growth deciduous woodlands) or are active at night or during the early morning and evening hours. Examples include the tropical tick *Amblyomma cajennense*, which is relatively tolerant of desiccation and which quests for long periods in open, exposed environments versus the lone star tick, *A. americanum*, which is easily stressed by xeric conditions and which prefers brushy or wooded habitats (Needham and Teel, 1991).

Host specificity extends across a broad range of extremes from those that are highly host specific at one end to those that exhibit little, if any specificity at the other. The latter are termed **non-specific or opportunistic** ticks. Examples of highly host-specific species include: (1) the squirrel tick, *Ixodes marxi*, which lives in the nests of its arboreal hosts and feeds solely on sciurid rodents; and (2) the cattle tick, *Boophilus microplus*, which feeds exclusively on cattle and certain other ruminants. Less rigorous patterns of host specificity are common. Often, such ticks will feed on a particular class of vertebrates, e.g., birds, but no other vertebrate animals. Many species exhibit restricted host ranges in one or two stages of the life cycle, but a much broader range of hosts in another period of the life cycle. An example is the so-called rabbit tick, *Haemaphysalis leporispalustris*. Actually, this species is best described as a "bird–rabbit tick" since the larvae and nymphs readily attack an immense variety of ground-feeding birds (as well as rabbits), while the adults feed exclusively on lagomorphs. A similar host range is found in *Ixodes dentatus*, a proven vector of Lyme borreliosis (Anderson et al., 1990). Many of the bird- or bat-infesting *Ornithodoros* species also show strict host specificity in the adult and nymphal stages, but less specificity as larvae. Occasionally, however, such ticks will also attack man and have been responsible for instances of human illness that were difficult to diagnose or treat. An example is the pigeon tick, *Argas reflexus*. In Berlin, Germany, 22 people were reported to have developed symptoms from bites of this tick, including urticaria, bronchial obstruction, and even loss of consciousness (immediate allergic reactions) (Dautel et al., 1991). Finally, the generalists constitute the opposite end of the host specificity spectrum. Such non-specific ticks feed readily on virtually all terrestrial vertebrates, although amphibians are rarely used. Examples include: (1) deer tick, *Ixodes scapularis* (=*dammini*), which feeds on a wide variety of small mammals (including both insectivores and small rodents), medium-sized carnivores, deer, birds, reptiles and even man; (2) the European sheep tick (or castor bean tick), *Ixodes ricinus*, with a similar expansive host range; and (3) the argasid tick, *Ornithodoros hermsii*, which feeds on a wide range of small and medium-sized mammals. These ticks have been responsible for noteworthy outbreaks of tick-borne relapsing fever (caused by *Borrelia hermsii*) when humans took shelter in abandoned mountain cabins that were also used by small rodents.

Even when the host range is restricted to a particular taxonomic group, e.g., Rodentia, certain animals in the community serve as primary hosts. An example is shown in the case of immature American dog ticks, *D. variabilis*. In southern Nova Scotia, where *D. variabilis* is exceptionally abundant, virtually

all small rodents in the tick's habitats are attacked. Primary habitats, as determined by relative abundance studies, are the small, abandoned old fields (meadows) and the ecotones at the boundaries between old fields and the surrounding second growth woodlands. Only two small mammal species in these habitats are of major importance in terms of parasite yields, namely, the red-backed vole, *Clethrionomys gapperi* and the meadow vole, *Microtus pennsylvanicus*. These two vole species accounted for 76.5% of all of the larvae and 82.4% of all of the nymphs of *D. variabilis* which fed on small mammal hosts during a 3-year period (Garvie et al., 1978). Substantial tick yields were also obtained from jumping mice, *Zapus hudsonicus* and snowshoe hares, *Lepus americanus*. Further south, in Virginia, USA, two different small mammals were predominant, namely, the meadow vole and the white-footed mouse, *P. leucopus*, which together accounted for over 90% of the yield of immature ticks. *P. leucopus* was also present in the Nova Scotia site but it added little to the overall tick yield. White-footed mice are exceptionally important as hosts for the larvae and nymphs of *I. scapularis* (= *dammini*) contributing more than 90% of the yield of fed ticks in some localities (Spielman, 1988).

Knowledge of the host ranges of important tick vector species is useful in designing programs for control of tick-borne diseases, since it enables the operators to focus scarce resources on only one or two animal species, e.g., dipping of cattle in insecticidal baths or using Damminix tubes to target white-footed mice. The role of host specificity and other host-related factors in tick control will be discussed by Schmidtman in Chapter 8.

5. HABITATS

To the lay person, ticks appear ubiquitous. Actually, ticks are highly restricted in the habitats they use. Virtually all species are adapted to a single, often highly specific habitat, the **optimum habitat type**, in which they flourish. Some examples for tick-borne diseases transmitted by ixodid ticks include: (1) the oak–hickory and oak–hickory–pine forests of the southeastern and south central US where dog ticks, *D. variabilis* (primary vector of RMSF) are abundant; (2) deciduous forests and thickets in the northeastern and north central US where the deer tick, *I. scapularis* (primary vector of Lyme disease), is abundant; (3) oak forests of central and northern Europe, termed thermophilic oak forests by Daniel et al. (1976), where *I. ricinus* (primary vector of Tick-borne Encephalitis) flourishes; and (4) savannah grasslands, tall grass prairies and montane forest communities in areas of southern and eastern Africa with relatively high rainfall (*c.* 50–100 cm/annum) where the brown ear tick, *Rhipicephalus appendiculatus* (primary vector of East Coast Fever) is abundant.

Despite erratic reporting, questions regarding reliability of diagnosis and other pitfalls affecting statistics on the occurrences of cases of reportable diseases, there is generally good agreement between distribution of tick-borne disease cases and the primary vector. In the case of RMSF, I plotted the distribution of reported cases over a 20-year period and showed that most

cases occurred in the oak–hickory–pine dominant habitats of the southeastern and south-central US, especially in suburban areas around cities and large towns (Sonenshine et al. 1972). In suburban areas of Ohio near Cincinnati, cases were most frequent in a focus characterized by numerous old fields and small woodlots dominated by oak–hickory forest (Linemann et al., 1973).

In the case of Lyme borreliosis in the northeastern and mid-Atlantic regions of the USA, the highest incidence of human cases occurs near heavily wooded, humid coastal regions where the zoonosis is also most intense. Both the tick vector, *I. dammini* (=*I. scapularis*) and the pathogen *B. burgdorferi*, are abundant in these coastal areas where the climate reflects the maritime influence. Further inland, where continental climatic conditions prevail, human cases of disease diminish in the plateau regions and are virtually absent in the cooler mountainous zones, despite the abundant deer and mouse hosts. *I. dammini* (=*I. scapularis*) is also much less abundant in the plateau regions and essentially absent in the mountains (Amerashinghe et al., 1992).

One of the best examples of the correlation between the distribution of a tick-borne disease and its primary vector is found in the case of the cattle disease, theileriosis, caused by the protozoan parasite, *Theileria parva* and its African vector, the brown ear tick, *Rhipicephalus appendiculatus*. With the aid of satellite mapping, vegetation, temperature and rainfall data were integrated using geographic information systems technology to develop a climate matching model. One such model, CLIMEX, was used to predict areas suitable for tick survival and development. The model proved to be a good indicator of distribution in most areas (but not of tick abundance) (Perry et al., 1990). Comparison of the distributions of East Coast Fever (ECF) caused by *T. parva* and *R. appendiculatus* also showed good agreement, i.e., outbreaks of ECF were most prevalent where habitats were most suitable for the vector tick. This principle and others affecting the distribution of tick-borne diseases of livestock are discussed further by Norval and Perry in Chapter 9.

6. ZOOGEOGRAPHY OF TICK-BORNE ZOONOSES

Infectious diseases transmitted by air, water, contact, or other non-mandatory biological routes tend to be more or less global in distribution. In contrast, vector-borne diseases, including those that are tick-borne, are more limited with distributions that reflect the geographic ranges of the primary vector as well as the environmental tolerances of the pathogenic agent. This is discussed in detail by Korch in Chapter 6. Here, we will examine the subject briefly and note how this phenomenon affects our understanding of disease occurrence.

From a global perspective, the distribution of tick-borne diseases can be categorized into three major groups: regional, continental, and inter-continental. Examples of the regional type are:

1. Powassan encephalitis in the northern US and southern Canada, caused by a flavivirus antigenically similar to other members of the Tick-borne

Encephalitis (TBE) group. The primary vectors are *Ixodes cookei* in eastern North America and *Dermacentor andersoni* in the western US and Canada. Both are opportunistic species that attack a wide variety of hosts, including man. The virus has also been reported from ticks in the former USSR (Artsob, 1989).

2. Kyasanur Forest Disease, found in certain mountainous areas of India, is also caused by a flavivirus similar to TBE. The disease is transmitted by *Haemaphysalis spinigera.*
3. Tick-borne epizootic abortion, caused by a spirochete, *Borrelia coriaceae*, and transmitted by the argasid tick, *Ornithodoros coriaceus* in western North America.

Examples of the continental type are:

1. Rocky Mountain spotted fever, caused by *Rickettsia rickettsii*, which is distributed throughout most of the USA, and occasionally in southern Canada, Mexico, and sporadically, in Central and South America. The primary vector in eastern North America, where more than 90% of the cases occur, is *D. variabilis*, and *D. andersoni* is the primary vector in the western region of the continent.
2. Tick-borne encephalitis (TBE), caused by a flavivirus, which is found throughout most of Europe and some areas of northern Asia where the primary vectors are *I. ricinus* (Europe) and *I. persulcatus* (eastern Europe and Asia).

Finally, the inter-continental type includes disease agents that have adapted to a relatively large number of wide-ranging tick vectors. Examples include:

1. Lyme disease, caused by *Borrelia burgdorferi*, which is now known to occur throughout the holarctic regions of the earth. The primary vectors are all species of the subgenus *Ixodes* (so-called *I. ricinus* group), although there is evidence of transmission by *Ixodes* species of other subgenera, e.g., *I. hexagonus* (Gern et al., 1991). These are mostly ticks with very broad geographic ranges, e.g., *I. ricinus.*
2. Relapsing fever, caused by species of the genus *Borrelia*, e.g., *B. hermsii*, but which causes a very different clinical response in man than *B. burgdorferi.* Although a large number of *Borrelia* species have been incriminated as causative agents, the disease (in man) they produce is the same. In addition, each of the many relapsing fever *Borrelia* spp. appears to be specific for its tick vector, all of which are species of the genus *Ornithodoros.* Thus, the reason for the enormous geographic range in this case is the ability of the many distinct species of the microbe (e.g. *B. duttonii* in eastern Africa, *B. hermsii* in western North America, *B. crocidurae* in North Africa, etc., and each adapted to its own vector tick) to cause a similar disease.

The zoogeography of tick-borne diseases, as with all diseases, is a dynamic, constantly changing phenomenon. Many questions remain. Two examples are:

(1) why are there differences in the distributions of the primary vector and the disease? For example, *D. variabilis* is abundant in certain areas of southern Canada, while *D. andersoni* is prevalent in populated areas of western Canada, yet there are few if any cases of RMSF in these regions. (2) Why is Lyme borreliosis epidemic in the northeastern US but not in the southeastern US, despite the fact that white-tailed deer, *Odocoileus virginianus*, white-footed mice, *P. leucopus*, and vector-component ticks, *Ixodes scapularis* (=*I. dammini*) are abundant in both regions? It is likely that these and other similar questions concerning the distribution of tick-borne disease will be the subjects of new investigations for many years to come.

7. ENZOOTIC VERSUS EPIZOOTIC DISEASE STATES

Host–pathogen–vector interrelationships are of paramount importance in understanding the ecology of vector-borne diseases, including tick-borne diseases. Review of the evidence from many different tick-borne zoonoses suggests that these diseases tend to persist in an enzootic state (i.e., stable, low rates of infection) when transmission is effected by an opportunistic tick species to a wide variety of hosts. However, when these hosts are supplanted by one or two dominant species, i.e., when the host range is narrowed, the force of transmission is focused and increased, often leading to high rates of infection in vector and reservoir host. For example, tick-borne **louping ill** virus is transmitted to an impressive variety of mammalian and avian hosts, usually by *Ixodes ricinus*. Dispersal in this manner results in relatively low levels of infection; viral isolates range from 0.1% to 0.4% of *I. ricinus* sampled in northern Britain (Reid, 1988). Moreover, most of the small mammals, birds and other vertebrates occasionally infected by these ticks develop weak viremias, usually below the threshold necessary to infect other ticks. Sheep, however, are capable of developing relatively high viremias which can persist for many days. Sheep are also excellent hosts for *I. ricinus*, supporting all three life stages. Consequently, where human intervention through sheep herding has narrowed the host range, the disease is intensified, with catastrophic effects on the sheep population.

Another example of this principle is found in the northeastern US, where Lyme borreliosis has increased to epidemic proportions since its discovery in Connecticut (Steere et al., 1977). Although the predominant vector in this region, *Ixodes dammini*, is an opportunistic species and immatures will attack a wide array of mammalian and avian hosts, well over 90% are found on white-footed mice, *Peromyscus leucopus* (Spielman et al., 1985; Spielman, 1988). Adults feed predominantly on white-tailed deer (*Odocoileus virginianus*) which are highly efficient hosts, leading to enormous increases in tick population size (**amplifier hosts**). The vector competency and susceptibility to infection of this tick is remarkable and probably without parallel among arthropod disease vectors. Virtually all ticks feeding on borrelemic hosts acquire infection, suggesting an extremely low threshold for acquiring infection while the bite of a single tick is sufficient to infect a white-footed mouse (Donahue et al., 1987;

Lane et al., 1991). The reservoir capacity of white-footed mice for *B. burgdorferi* far exceeds that of other reservoir-competent small mammals; one white-footed mouse is estimated to be capable of infecting as many *I. scapularis* immatures as 12 chipmunks or 221 meadow voles (Mather et al., 1989). White-footed mice remain infectious for long periods, probably for their entire life span (Donahue et al., 1987). Spielman (1988) suggests that the force of transmission of *B. burgdorferi* is enhanced by the overwhelming predominance of white-footed mice as the hosts for the immature stages of *I. scapularis.* In contrast, feeding on other hosts tends to disperse the infection, diluting it and, thereby, resulting in maintenance of the infection at low levels, i.e., as an enzootic disease. Another example of this phenomenon is found in the tick-borne disease, **Crimean–Congo Hemorrhagic Fever** (CCHF) (Hoogstraal, 1979; Linthicum and Bailey, 1993). Although viral isolations have been made from at least 29 tick species, including two argasids, and virus isolations have been made from numerous mammals and birds, only a few tick species and vertebrate hosts are important in the ecology of this zoonotic disease. The most important tick vectors in Europe and Asia are *Hyalomma marginatum marginatum* and *Dermacentor marginatus*; *H. m. marginatum* is also considered to be the most important vector in Africa. Hares (*Lepus europaeus*) and hedgehogs (*Erinaceus europaeus*) are the most important hosts. These animals are competent reservoir hosts with high levels of viremias and are excellent donors of virus to ticks. Moreover, the European hare is a cyclical species, undergoing periodic rapid, massive increases in population densities, making it an excellent amplifier host. Ground-feeding birds, especially rooks (*Corvus* spp.) and doves (*Columba livida*) are also excellent amplifiers of the tick population, even though they are incompetent reservoirs of the virus. These factors are believed to have contributed to the outbreak of CCHF that occurred in the Crimean region of the USSR during the war years 1941–1944. During this period, human pressures on the local environment were relaxed, especially intensive cultivation and pasturing of livestock, allowing lush growth of wild vegetation along rivers and floodplains, and the concomitant expansions in the host and vector populations noted above (Watts et al., 1988).

The concept of zoonotic intensification by over dependence on only one or two highly competent reservoir hosts, or a single amplifier of tick abundance, is subject to challenge. In the case of Lyme borreliosis, numerous mammals besides white-footed mice serve as hosts for the vector, *I. scapularis*, and as reservoirs for *B. burgdorferi.* Ticks fed on these hosts often sustain high levels of infection, e.g., 37% infection in nymphs from short-tailed shrews (*Blarina brevicauda*) collected in Massachusetts (Telford et al., 1990). In addition, several mammals besides white-tailed deer may function as amplifiers of the vector population, even if they are not as important as donors of infection. In southern New York state, Fish and Daniels (1990) showed that a single raccoon can produce as many fed larvae as six white-footed mice.

Although zoonotic intensification is common where only one or two competent reservoir and amplifier hosts support the vector species, some diseases remain enzootic with low levels of natural infection even where such

conditions exist. An example is seen in the case of Rocky Mountain spotted fever. In the eastern US, the predominant vector to man is the American dog tick, *Dermacentor variabilis*. This tick is also the primary reservoir for maintaining the zoonosis in the natural environment. Although other tick species have been found infected with spotted fever group (SFG) rickettsiae, e.g., *Haemaphysalis leporispalustris* and *Ixodes brunneus* (Clifford et al., 1969) from cottontail rabbits and birds, as well as *Amblyomma americanum*, *I. cookei*, *I. dentatus*, *I. scapularis*, *I. texanus*, and *Rhipicephalus sanguineus* from a variety of mammal and bird hosts, these have been mostly non-pathogenic rickettsial species (*R. montana*, *R. bellii*, and *R. rhipicephali*) (Burgdorfer, 1975, 1988). Numerous mammal species are susceptible to the disease, but white-footed mice, meadow voles (*Microtus pennsylvanicus*) and red-backed voles (*Clethrionomys gapperi*) are the primary hosts for the immature tick population, while raccoons are the most important hosts of the adults in the eastern US (Sonenshine, 1975). Studies in eastern North America (Sonenshine et al., 1966; Garvie et al., 1978; Smart and Caccamise, 1988) have shown that well over 90% of the larvae feed on white-footed mice and meadow voles (or meadow voles and red-backed voles). Similarly, despite a wide spectrum of carnivores and larger mammals, most adults feed on raccoons (*Procyon lotor*) (Sonenshine, 1975), although porcupines (*Erethizon dorsatum*) and black bears (*Ursus americanus*) are also important hosts in the more northern regions (Campbell, 1979). Thus, one would expect zoonotic intensification to be the natural outcome of this highly focused vector–reservoir relationship, with epidemic occurrence of the disease. However, tick infection rates are relatively low, usually below 10% in most vector populations that have been assessed (see Chapter 10 by Schriefer and Azad). A major limiting factor in this zoonosis is the brief period of rickettsial infection in the vertebrate hosts. White-footed mice, meadow voles and other small mammals are rickettsemic, i.e., circulate rickettsiae in their blood, for only 3–10 days following infection by tick bite (Burgdorfer, 1988). Thus, uninfected ticks can acquire rickettsiae from donor (i.e., infectious) hosts for only a brief interval in the hosts' life span. In my studies in Virginia, we observed a rapid increase in seroconversions in mice in early spring. Thereafter, the frequency of seropositive mice remained fairly constant, declined in the summer, but rose again in the fall with the emergence of a new cohort of juvenile mice. Similar findings were noted by Korch (1984) in his study site in Maryland. In both cases, large proportions of the host population were no longer serving as efficient donors to feeding ticks.

In terms of overall ecological value, the reservoir competency of a particular host for a zoonotic infection is as important as tick vector competence. This is because the circulation of arthropod-borne pathogenic agents becomes significant when they infect vertebrate animals and cause disease.

Another important question affecting the role of ticks in disease transmission concerns the number of pathogenic organisms necessary to establish infection. Simply stated, how many microbes must be present in the host to initiate infection in a feeding tick? Similarly, how many microbes must be transmitted from an infected tick to establish infection in a susceptible host?

In some cases, the numbers required are extremely small, i.e., the threshold for transmission is very low. An example is found in the case of Lyme borreliosis, where the bite of a single *B. burgdorferi*-infected tick is sufficient to infect susceptible mice. This extreme susceptibility to spirochetal infection is undoubtedly one of the major contributing factors to the high incidence of infection, frequently exceeding 50%, in some populations of the tick vector, *Ixodes scapularis*. Much more common, however, are tick-borne zoonoses where the threshold levels are relatively high, i.e., where there are substantial barriers to initiating infection in ticks and hosts. For example, in Britain, the malaria-like organisms *Babesia microti* in voles, *Clethrionomys glareolus*, are transmitted by the rodent-feeding tick, *Ixodes trianguliceps*. Ticks are highly competent vectors for transmitting these babesias. However, tick infection occurs only during a very brief period of 5 days or less when the level of parasitemia in the voles has risen to at least 2% (Randolph, 1992). Similarly, high infection thresholds are necessary to infect the African brown ear tick, *Rhipicephalus appendiculatus* with the protozoan that causes East Coast Fever, *Theileria parva*. In this case, the vast majority of the ingested organisms are destroyed in the midguts of feeding ticks and many ticks escape infection. Thus, high levels of parasitemia develop only in about 10% of the tick population, although this level of infection is sufficient to spread the disease when ticks become abundant (Young et al., 1992). Much the same phenomenon is found with tick-borne arboviruses. Infection of ticks is "dose dependent; the higher the blood titer of virus, the greater the proportion of ticks that become infected" (Nuttall et al., 1992).

The ecological status of tick-borne disease is also influenced by the variety of host–vector systems, termed ecological cycles, within which it circulates. Some tick-borne diseases are expressed in two (or, rarely, even more) ecological cycles, one comprising a tick vector that conveys the disease agent to man or his livestock, the other (or others) comprising a tick vector that circulates the pathogen in a cryptic cycle among wild hosts. Pathogens are exchanged between the two cycles when the tick vectors share the same hosts. An example of this phenomenon, discussed in detail by Lane in Chapter 3, is found in the ecology of Lyme borreliosis in northern California, USA. In this case, the vector to man is the opportunistic *Ixodes pacificus*. However, this tick is a relatively inefficient vector, presumably due to the large numbers of the larvae and nymphs that feed on reservoir-incompetent lizards and birds. The second vector is *Ixodes neotomae*, a non-man-biting, host-specific tick which maintains a cryptic cycle among rodents, primarily woodrats (*Neotoma fuscipe*) and lagomorphs (Brown and Lane, 1992). In the eastern US, where Lyme borreliosis circulates in a cycle involving primarily white-footed mice, white-tailed deer, and *I. scapularis*, Telford and Spielman (1990) have postulated an enzootic cycle involving the bird–rabbit tick, *Ixodes dentatus* and cottontail rabbits (*Sylvilagus floridanus*). These authors suggested that this cycle operates independently of the zoonotic cycle between mice and deer, but "that infection may occasionally be exchanged between these two cycles." Yet another example is found in the ecology of the rickettsiosis, boutenneuse fever (= Mediterranean

tick-bite fever). The pathogen agent, *Rickettsia conorii*, circulates in a highly focused "domestic" cycle comprising dogs and the brown dog tick, *Rhipicephalus sanguineus*, in suburban areas, farms and other densely populated, settled communities. The rickettsiae also circulate in a so-called "natural" cycle involving small rodents, insectivores, and various wild carnivores. *R. sanguineus* also feeds on these animals, but other ixodid ticks, e.g., *I. ricinus*, *Dermacentor reticulatus*, *Haemaphysalis leachi*, *Rhipicephalus evertsi*, and others are also regarded as important vectors in the various regions where they occur (Rehacek and Tarasevich, 1988). Interaction between these two disparate cycles occurs when the tick-infected wild hosts periodically invade the settled areas, dropping rickettsia-infected vectors (or dogs leave the settled areas, dropping rickettsia-infected *R. sanguineus* in the reverse direction).

8. REFERENCES

Amerasinghe, F.P., Breisch, N.L., Azad, A.F., Gimpel, W.F., Greco, M., Niedhart, K., Pagac, B., Piesman, J., Sandt, J., Scott, T.W., and Sweeney, K. (1992) Distribution, density and Lyme disease spirochete infection in *Ixodes dammini* (Acari: Ixodidae) on white-tailed deer in Maryland. *J. Med. Entomol.* **29**, 54–61.

Anderson, B.E., Dawson, J.E., Jones, D.C., and Wilson, K.H. (1991) *Ehrlichia chaffeensis*, a new species associated with human ehrlichiosis. *J. Clin. Microbiol.* **29**, 2838–2842.

Anderson, J.F., Magnarelli, L.A., and Stafford, K.C., III (1990) Bird-feeding ticks transstadially transmit *Borrelia burgdorferi* that infect Syrian hamsters. *J. Wildl. Dis.* **26**, 1–10.

Artsob, H. (1989) Powassan encephalitis. Vol. 4, pp. 29–49. *In:* Monath, T.P. (ed.) *The Arboviruses. Epidemiology and Ecology.* CRC Press, Inc., Boca Raton, FL.

Bhat, H.R. (1991) Tick ecology in relation to Kyasanur Forest Disease. Vol. 1, pp. 11–33. *In:* Channabasavanna, G.P. and Viraktamath, C.A. (eds) *Progress in Acarology.* IBH Publishing Co., Oxford, New Delhi.

Brown, R.N. and Lane, R.S. (1992) Lyme disease in California: a novel enzootic transmission cycle of *Borrelia burgdorferi*. *Science* **256**, 1439–1442.

Brumpt, E. (1932) Longevité du virus de la fièvre boutenneuse (*Rickettsia conorii*, n. sp.) chez la tique *Rhipicephalus sanguineus*. *C. R. Soc. Biol.* **110**, 1119. [not seen in original; cited in Rehacek and Tarasevich (1988).]

Burgdorfer, W. (1975) A review of Rocky Mountain spotted fever (tick-borne typhus), its agent and its tick vectors in the United States. *J. Med. Entmol.* **12**, 269–278.

Burgdorfer, W. (1988) The spotted fever group diseases. pp. 279–301. *In:* Steele, J.H. (ed) *CRC Handbook in Zoonoses. Section A. Bacterial, Rickettsial and Mycotic Diseases. Vol. 2.* CRC Press, Boca Raton, FL.

Burgdorfer, W. (1992) Vector–spirochete relationships of arthropod-borne borrelioses. pp. 111–120. *In:* Munderloh, U.G. and Kurtti, T.J. (eds) *First International Conference on Tick-borne Pathogens at the Host–Vector Interface: An Agenda for Research.* University of Minnesota, St. Paul.

Burgdorfer, W. and Brinton, L.P. (1975) Mechanism of transovarial transmission of spotted fever rickettsiae in ticks. *Ann. New York Acad. Sci.* **244**, 61–72.

Campbell, A. (1979) Ecology of the American dog tick, *Dermacentor variabilis*, in southwestern Nova Scotia. pp. 135–143. *In:* Rodriguez, J.G. (ed) *Recent Advances in Acarology II*. Academic Press, New York.

Casals, J. (1969) Antigenic similarity between the virus causing Crimean hemorrhagic fever and Congo virus. *Proc. Soc. Exp. Biol. Med.* **131**, 233.

Chumakov, M.P., Belyaeva, A.P., Voroshilova, M.K., Butenko, A.M., Shalunova, N.V., Semashko, I.V., Mart'yanova, L.I., Smirnova, S.E., Bashkirtsev, V.N., Zanvodova, T.I., Rubin, S.G., Tkachenko, E.A., Karymsheva, V.Ya., Reingol'd, V.N., Popov, G.V., Kirov, I., Stolbov, D.N., and Perelatov, V.D. (1968) Progress in studying the etiology, immunology, and laboratory diagnosis of Crimean hemorrhagic fever in the USSR and Bulgaria. *Mater. 15. Nauchn. Sess. Inst. Polio. Virus. Entsefalitov* (Moscow, October 1968) **3**, 100 (in Russian). (In English, NAMRU3-T613.)

Chumakov, M.P., Smirnova, S.E., and Tkachenko, E.A. (1969) Antigenic relationships between the Soviet strains of Crimean hermorrhagic fever virus and the Afro-Asian Congo virus strains. *Mater. 16. Nauchn. Sess. Inst. Polio. Virus. Entsefalitov* (Moscow, October 1969) **2**, 152 (in Russian). (In English, NAMRU3-T614.)

Clifford, C.M., Sonenshine, D.E., Atwood, A.L., Robbins, C.S., and Hughes, L.E. (1969) Tests on ticks from wild birds collected in the eastern United States for rickettsiae and viruses. *Am. J. Trop. Med. Hyg.* **18**, 1057–1061.

Daniel, M., Czerny, V., Dusbabek, E., Honzakova, E., and Olejnicek, J. (1976) Influence of microclimate on the life cycle of the common tick *Ixodes ricinus* (L.) in thermophilic oak forest. *Folia Parasitologica (Praha)* **23**, 327–342.

Daniels, T.J., Fish, D., and Falco, R.C. (1991) Evaluation of host-targeted acaricide for reducing risk of Lyme disease in southern New York state. *J. Med. Entomol.* **28**, 537–543.

Dautel, H., Kahl, O., and Knulle, W. (1991) The soft tick *Argas reflexus* (F.) (Acari: Argasidae) in urban environments and its medical significance in Berlin (West). *Appl. Ent.* **111**, 380–390.

Donahue, J.G., Piesman, J., and Spielman, A. (1987) Reservoir competence of white footed mice for Lyme disease spirochetes. *Am. J. Trop. Med. Hyg.* **36**, 92–96.

Dutton, J.E. and Todd, J.L. (1905) The nature of tick fever in the eastern part of the Congo Free State, with notes on the distribution and bionomics of the tick. *Br. Med. J.* **2**, 1259–1260.

Fish, D. and Daniels, T.J. (1990) The role of medium-sized mammals as reservoirs of *Borrelia burgdorferi* in southern New York. *J. Wildl. Dis.* **26**, 339–345.

Garvie, M.A., McKiel, J.A., Sonenshine, D.E., and Campbell, A. (1978) Seasonal dynamics of American dog tick, *Dermacentor variabilis* (Say), populations in southwestern Nova Scotia. *Can. J. Zool.* **56**, 28–39.

Gern, L., Toutoungi, L.N., Hu, C.M., and Aeschlimann, A. (1991) *Ixodes* (*Pholeoixodes*) *hexagonus*, an efficient vector of *Borrelia burgdorferi* in the laboratory. *Med. Vet. Entomol.* **5**, 431–435.

Hoogstraal, H. (1979) The epidemiology of tick-borne Crimean–Congo hermorrhagic fever in Asia, Europe and Africa. *J. Med. Entomol.* **15**, 307–417.

Johnson, R.C., Schmid, G.P., Hyde, F.W., Steigerwalt, A.G., and Brenner, D.J. (1984) *Borrelia burgdorferi* sp. nov.: etiological agent of Lyme disease. *Internat. J. Syst. Bacteriol.* **34**, 496–497.

Korch, G.W. (1984) The ecology of the American dog tick, *Dermacentor variabilis*, at a site with endemic spotted fever group rickettsia. Unpublished Ph.D. dissertation, Johns Hopkins University, Baltimore, 423 pp.

Kuttler, K.L. (1988) World-wide impact of Babesiosis. pp. 1–22. *In:* Ristic, M. (ed) *Babesiosis of Domestic Animals and Man.* CRC Press, Boca Raton, FL.

Lane, R.S., Piesman, J., and Burgdorfer, W. (1991) Lyme borreliosis: relation of its causative agent to its vectors and hosts in North America and Europe. *Annu. Rev. Entomol.* **36**, 587–609.

Linneman, C., Jr, Jansen, P., and Schiff, G.M. (1973) Rocky Mountain spotted fever in Clermont County, Ohio. Description of an endemic focus. *Am. J. Epidemiol.* **97**, 125–130.

Linthicum, K.J. and Bailey, C. L. (1993) Ecology of Crimean–Congo Hermorrhagic Fever, pp. 392–437. *In:* Sonenshine, D.E. and Mather, T.N. (eds) *Ecological Dynamics of Tick-borne Zoonoses.* Oxford University Press, New York.

Mather, T.N., Wilson, M.L., Moore, S.I., Ribeiro, J.M.C., and Spielman, A. (1989a) Comparing the relative potential of rodents as reservoirs of the Lyme disease spirochete (*Borrelia burgdorferi*). *Am. J. Epidemiol.* **30**, 143–150.

Needham, G.R. and Teel, P.D. (1991) Off-host physiological ecology of ixodid ticks. *Annu. Rev. Entomol.* **36**, 659–681.

Nuttall, P.A., Jones, L.D., Labuda, M., and Kaufman, W.R. (1992) Interaction between arboviruses and their tick vectors. pp. 37–41. *In:* Munderloh, U.G. and Kurtti, T.J. (eds) *First International Conference on Tick-Borne Pathogens at the Host–Vector Interface: An Agenda for Research.* University of Minnesota, St Paul.

Oliver, J.H., Jr, Owsley, M.R., Hutcheson, H.J., James, A.M., Chen, C., Irby, W.S., Dotson, E.M., and McLain, D.K. (1993). Conspecificity of the ticks *Ixodes scapularis* and *I. dammini* (Acaris Ixodidae). *J. Med. Entomol.* **30**, 54.

Perry, B.D., Lessard, P., Norval, R.A.I., Kundert, K., and Kruska, R. (1990) Climate, vegetation and the distribution of *Rhipicephalus appendiculatus* in Africa. *Parasitol. Today* **6**, 100–104.

Randolph, S.E. (1992) Natural transmission of *Babesia microti* by *Ixodes trianguliceps.* p. 303. *In:* Munderloh, U.G. and Kurtti, T.J. (eds) *First International Conference on Tick-Borne Pathogens at the Host–Vector Interface: An Agenda for Research.* University of Minnesota, St Paul.

Rehacek, J. and Tarasevich, I.V. (1988) *Acari-borne Rickettsiae and Rickettsioses in Eurasia.* Veda Publishing House, Slovak Academy of Sciences, Bratislava, 343 pp.

Reid, H.W. (1988) Louping Ill. Vol. III, pp. 117–135. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology.* CRC Press, Inc., Boca Raton, FL.

Ribeiro, J.C. (1987) The role of saliva in blood-feeding by arthropods. *Annu. Rev. Entomol.* **32**, 463–478.

Ricketts, H.T. (1906) The transmission of Rocky Mountain spotted fever by the bite of the wood tick (*Dermacentor occidentalis*). *JAMA* **47**, 358.

Ricketts, H.T. (1907) The role of the wood tick (*Dermacentor occidentalus*) in Rocky Mountain spotted fever and the susceptibility of local animals to this disease. *JAMA* **49**, 24.

Smart, D.L. and Caccamise, D.F. (1988) Population dynamics of the American dog tick (Acari: Ixodidae) in relation to small mammal hosts. *J. Med. Entomol.* **25**, 515 522.

Smith, T. and Kilbourne, F.L. (1893) Investigators into the nature, causation and prevention of Texas or southern cattle fever. *Bull. Bur. Anim. Ind., U.S. Dept. Agric.* **1**, 301 pp.

Sonenshine, D.E. (1975) Influence of host–parasite interactions on the population dynamics of ticks. *Misc. Publ. Entomol. Soc. Am.* **9**, 243–249.

Sonenshine, D.E. (1979) Zoogeography of the American dog tick, *Dermacentor variabilis*. *Adv. Acarol.* **2**, 123–134.

Sonenshine, D.E. (1991) *Biology of Ticks*, Vol. 1. Oxford University Press, New York.

Sonenshine, D.E., Atwood, E.L., and Lamb, J.T. (1966) The ecology of ticks transmitting Rocky Mountain spotted fever in a study area in Virginia. *Ann. Entomol. Soc. Am.* **59**, 1234–1262

Sonenshine, D.E., Peters, A.J., and Levy, G.F. (1972) Rocky Mountain spotted fever in relation to vegetation in the eastern United States, 1951–1971. *Am. J. Epidemiol.* **96**, 59–69.

Spielman, A. (1988) Lyme disease and human babesiosis: evidence incriminating vector and reservoir hosts. pp. 147–165. *In:* Englund, P.T. and Sher, A. (eds) *The Biology of Parasitism. A Molecular and Immunological Approach.* Alan R. Liss, New York..

Spielman, A., Wilson, M.L., Levine, J.F., and Piesman, J. (1985) Ecology of *Ixodes dammini*-borne human babesiosis and Lyme disease. *Annu. Rev. Entomol.* **30**, 439–460.

Stafford, K.C., III (1991) Effectiveness of host-targeted permethrin in the control of *Ixodes* (Acari: Ixodidae). *J. Med. Entomol.* **28**, 611–617.

Steere, A., Malawista, S.E., Snydman, D.R., Shope, R.E., Andiman, W.A., Ross, M.R., and Steele, F.M. (1977) Lyme arthritis; an epidemic of oligoarticular arthritis in children and adults in three Connecticut communities. *Arthritis Rheum.* **20**, 7–17.

Telford, S.R., III and Spielman, A. (1990) Enzootic transmission of the agent of Lyme disease in rabbits. *Am. J. Trop. Med. Hyg.* **41**, 482–490.

Theiler, A. (1904) Rhodesian tick fever. *Transvaal Agric. J.* **2**, 421–438.

Watts, D.M., Ksiazek, T.G., Linthicum, K.J., and Hoogstraal, H. (1988) Crimean–Congo Hemorrhagic Fever. Vol. 2, pp. 177–222. *In:* Monath. T.P. (ed) *The Arboviruses: Epidemiology and Ecology.* CRC Press, Inc., Boca Raton, FL.

Young, A.S., Shaw, M.K., Ochanda, H., Morzaria, S.P., and Dolan, T.T. (1992) Factors affecting the transmission of African *Theileria* species of cattle by ixodid ticks. pp. 65–70. *In:* Munderloh, U.G. and Kurtti, T.J. (eds) *First International Conference on Tick-Borne Pathogens at the Host–Vector Interface: An Agenda for Research.* University of Minnesota, St Paul.

2

Population Ecology of Tick Vectors: Interaction, Measurement, and Analysis

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1. INTRODUCTION

The analysis of tick populations, as with of all living organisms, requires information on three fundamental processes that govern population change: birth, death, and migration. Populations of ticks increase with reproduction or immigration and decrease with deaths or emigration. This simple framework serves as an organizational base for asking questions about how, when, and why populations change, and whether one might successfully intervene in that process. Underlying this simplified perspective, however, are myriad factors that interact in sometimes complex ways to contribute to population change. The importance of each of these variables will differ among tick species, development stages, vertebrate hosts, associated parasites or predators, and environmental conditions. Ultimately, population and community analyses aim to define the role and pattern of these factors as they determine the dynamics of interaction. For ticks that act as vectors of disease agents of public or animal health importance, their capacity to do so becomes an extension of such population analysis.

In the interest of brevity, the basic biological factors underlying these events will not be discussed here. Excellent reviews detail current knowledge of tick natural history (Hoogstraal, 1985a), systematics (Oliver, 1989), various aspects of physiology (Obenchain and Galun, 1982), water balance (Needham and Teel, 1991), reproductive biology (Oliver, 1983), host specificity (Hoogstraal and Aeschlimann, 1982), and pathogen transmission (Hoogstraal, 1981). Other chapters of this volume discuss similar issues and particular examples. Rather, the goal here is to examine aspects of tick biology as they influence the manner in which population dynamics are studied. In particular, we shall examine how populations of ticks are measured and analyzed in the context of the objectives, study design, and intended applications of the results that are sought. It will be argued that a critical approach to population measurement and analysis is needed as many biological processes are involved and most interactions are richly complex, sometimes indirect, and frequently inapparent. Thus, rigor

becomes essential in gathering, summarizing and interpreting data. Thereby, the validity and utility of results are likely to be improved.

Why study populations? Firstly, although many characteristics of ticks that permit them to vector pathogens have been described through studies of individual organisms, our understanding of these observations acquires meaning solely when analyzed in the context of interacting groups. Descriptions of feeding, growth, development, and reproduction, for example, are biologically significant solely in an ecological milieu where populations develop, interact, and evolve. Indeed, the transmission of disease agents involves not only populations of vectors, but those of various other species including the vector's pathogens, vertebrate reservoirs, occasional or accidental hosts, parasites, and predators. Moreover, the likelihood of transmission is dependent, at least in part, on subpopulations of competent, infected vectors and susceptible, non-immune hosts.

Secondly, analyses of phenomena at the population level encourage examination of change in space and time, the essence of ecological dynamics and of pathogen transmission. Populations of individuals are distributed differentially in space among various habitats and biomes, just as demographic events vary over time. Such habitat- or season-specific activity is likely to play an important role in the capacity of a vector population to maintain transmission of a pathogen. Otherwise simple descriptions of state emerge as dynamic processes when viewed in the context of interactions among populations of individuals. By extension, multiple populations of different species interact as complex, evolving, biological communities. Ultimately, any attempt to interpret vector-borne disease transmission must be based on such a spatio-temporal, population perspective.

Thirdly, interactions among demographic subgroups may play a crucial role in the evolution of pathogen transmission systems, and their study requires a population perspective. Recognition of different developmental stages, subclasses, sexes, age cohorts, and generations allow us to differentiate responses to pathogen exposure, development of immunity, changing social behavior, and disease among these functional parts of a population. By analyzing interactions of different subgroups, which implies treating populations as more than simple ensembles of individuals, we gain insight into the mechanisms that perpetuate multispecies interactions like the vector–pathogen–host systems that we recognize as tick-borne zoonoses (see Mather and Ginsburg, Chapter 4).

Finally, the integration of levels of analysis becomes important. Knowledge of processes at the molecular, cellular, or organ levels has provided insight into developments within individual organisms. One goal of a population perspective, however, should be to analyze how these processes, in turn, influence the ecology of infection and disease. This task is complicated by the fact that important biological events may behave non-linearly, interacting at different rates of change. Furthermore, there may be thresholds beyond which outcomes are quite different. Only with accurate description, conscientious sampling and thoughtful analysis will we be able to gain insight into these complex interactions.

The following discussion is organized into four topics: (1) concepts and

measures in population studies; (2) sampling techniques applied to tick populations; (3) ecological factors influencing population dynamics; and (4) rationale and implications of population analysis in vector-borne disease research.

2. DESCRIPTION AND MEASUREMENT OF POPULATIONS

The foundations of studies on the population dynamics of any species must rest on concepts and measures that are clearly defined and carefully employed. Various reviews of sample design, concepts, methods and analysis employed in population studies from entomology (Morris, 1960; Southwood, 1978; Barr, 1979; Taylor, 1984; Kuno, 1991), parasitology (Margolis et al., 1982), and ecology (Pollock et al., 1990; Eberhardt and Thomas, 1991) have been consulted in preparing this chapter. The goal here is to address particular problems that arise while planning and undertaking studies of tick vector population ecology.

A population may be characterized qualitatively or quantitatively. Qualitative measures, such as whether a tick is "present" or "absent," or whether encounters are "frequent" or "rare" may be employed in describing the state of a species or its present distribution. Such simple descriptions, however, are not useful in analyzing changes in population density. From these qualitative measures, little other information can be derived concerning ecologically important population characteristics such as trends in vector survival or reproduction, transience or permanence, distribution across habitats, or associations with hosts. The qualitative presence of a species in a region should serve as impetus to undertake further study aimed at quantifying the distribution of its population in time and space.

2.1. Quantitative Measures

The number of individuals in a population is usually measured by estimation which is based on sampling the population in a given area. As distinct from collecting, which typically provides only qualitative or relative measures of a population, sampling attempts to count a defined subset of individuals that is representative of a larger population. Both sampling and collecting may be intensive, covering a small area and/or a short time period, or extensive, involving a large region and/or many years. Estimates vary in accuracy, and hence utility, depending on the extent and method of sampling.

The alternative to estimation, which rarely is attempted in practice, involves complete enumeration wherein the sample is designed to include all individuals of the population under study. Not only are there formidable practical difficulties in such a task, but estimates based on carefully planned samples usually are adequate to answer most population-based questions. Obviously, the objectives of each study strongly influence the extent and nature of sampling; these objectives must be carefully planned in advance. Indeed, it is essential that the measures and methods of sampling and estimating a

population be clearly defined and conscientiously applied throughout the course of study.

Abundance is the measure often used to describe the number of individuals in one population as compared to those in another. Expressed in unknown units, abundance is a dimensionless, *relative* quantity that carried inherent implications and popular appeal. Samples that produce information on relative abundance acquire meaning when compared to those similar measures from other demographic groups, physical locations, or time periods. **Density**, as distinguished from abundance, refers to the number of individuals per unit area, a dimensioned, **absolute** measure. Tick samples from vegetation or soil, for example, are readily calculated as density per unit area.

Samples from hosts, often obtained for convenience or biological relevance, represent a complicated measure. The dimension of reference is all or part of the surface of a vertebrate, whose own size, behavior, and population density vary. Here, the term **population intensity** has been proposed (Southwood, 1978), although the objective of estimating tick density is often to analyze population dynamic processes that usually require knowledge of absolute density. Also termed the **mean density** of parasites per host, this measure is calculated as the total number of ticks divided by the total number of hosts examined. Other host-associated measures of tick abundance are the **prevalence of infestation**, a percentage representing the number of infested hosts divided by the total number of hosts examined, and the **intensity of infestation**, which is the number of ticks divided solely by the number of *infested* hosts. These measures may not be used to estimate density without considering, and perhaps adjusting for, the sample denominator (discussed below). As abundance measures, they permit relative comparisons associated with various inherent errors.

Ironically, host-associated measures such as these both confound estimates of tick density and help explain the dynamics of population change. For example, similar estimates of ticks per host at different sites could represent highly dissimilar population densities if the density of hosts at these sites also differs; such information, however, might additionally suggest host-density limits to tick feeding, survival or reproduction. Likewise, unreliable estimates could result if the mean density of ticks per host was based on low infestation intensity or high infestation variation (Davidar et al., 1989); this might indicate, however, important spatial, immunologic, or demographic differences among vertebrate hosts. Knowledge of such caveats and their biological implications must be employed in designing, analyzing and interpreting tick population samples. These potential complications illustrate the importance of clearly defining the nature of samples and measures being used.

2.2. Temporal Variation in Samples and Estimates

As has been noted often, variation in tick activity may influence estimates of their abundance. Whether monitored as free-living or host-associated, **seasonal**

changes in host-seeking activity, typically regulated by photoperiod (Belozerov, 1982), alter the availability of ticks to samples. For this reason, information on seasonal patterns in activity is crucial to subsequent interpretation of observations and calculation of population changes. Armed with such knowledge, samples may be restricted to certain activity periods or seasonally adjusted to permit comparisons among sites. Similarly, **diurnal** variation in host-seeking, which may be photoperiod-dependent and temperature- or humidity-sensitive, influences observations of each free-living stage. Samples obtained during different times of day or under dissimilar environmental conditions must be evaluated cautiously. Numerous observations may be needed if accurate parameter estimates intended for population analysis are sought.

Temporal variation in the **duration of attachment** also influences tick counts obtained from hosts. The number of ticks on a host at any one time is the product of two rates: those of host-finding/attachment, and detachment or death. Depending on various stage- and species-specific factors, ticks may remain attached to a host for hours, days, weeks, or months (discussed below). Other factors being equal, population estimates based on the number of ticks per host will increase proportionally with the duration of attachment. That is, a tick that typically remains attached for 6 days will produce an equilibrium number of individuals per host which is twice that of a different species or stage detaching after only 3 days. Fortunately, comparisons among the same species or stage are not hampered by this type of sampling bias. Efforts to construct life tables or to model development-specific population changes, however, should consider such confounding interactions. For example, the ratio of host-associated larvae to nymphs, or that of males to females may be invalid without appropriate adjustment that accounts for duration of attachment. In a similar manner, **diurnal patterns of detachment** or "drop-off" from hosts might significantly influence estimates, depending on the time of day that hosts are examined. Since most ticks exhibit such diurnal drop-off patterns, efforts should be made to compare only those samples obtained during the same time of day.

2.3. Tick Feeding Patterns and Population Sampling

Certain characteristics of the frequency of detachment/reattachment profoundly influence host-associated sampling methods and the interpretation of population data Ticks of the two major families, Ixodidae and Argasidae, all require blood-meals from vertebrate hosts to survive, mature, and reproduce; however, the number and duration of blood meals differ markedly between the two groups. The "hard" ticks (Ixodidae) typically feed once during each of three motile stages (larva, nymph, adult) for periods that last for a few days to more than a week. Engorged larvae and nymphs typically drop from their hosts and subsequently molt to the next stage, whereas engorged females detach to lay one massive batch of eggs before dying. Males may remain on a host for weeks or months, mating with numerous females. Tick samples obtained from these vertebrate hosts represent a subsample of the total tick population of an area.

The principal challenge faced by researchers involves determining what proportion is being sampled, from what area, and how much of the variation is error due to this host-based trapping method.

A very different blood-feeding strategy evolved among the "soft" ticks (Argasidae). Despite differences among species, soft ticks generally feed for short periods (minutes to hours) on numerous occasions during their development. Consequently, hosts are rarely useful in sampling of soft ticks. Their brief yet frequent blood-meals oblige most argasids to remain near sites that are regularly visisted by preferred hosts (e.g., nests, burrows, or feeding areas). Samples of soft ticks thereby are facilitated through capture of free-living, "host-affiliated" individuals that tend to be clustered in these host-frequented sites (e.g., Denmark and Clifford, 1962; Butler et al., 1984; Adeyeye and Butler, 1989). This feeding behavior of argasid ticks and their consequent tendency to aggregate in space create different sampling concerns that eventually impact on population estimates. For example, the denominator of such samples may be represented by spatially heterogeneous units of habitat that complicate absolute calculations and comparisons among sites (discussed below). Careful evaluation of the habitat and prudent decisions concerning the location of samples should decrease bias and error, thereby improving the reliability and reproducibility of estimates.

The **diversity of host species** fed upon may differ among ticks in a manner that influences population sampling. Certain species or stages infest one or a few vertebrates while others seem almost indiscriminate in their feeding. This variety in hosts, thereby, partly determines the number of samples needed to obtain reliable population estimators. In general, observations from host species that are both numerous and heavily parasitized will most likely provide the best relative estimates of tick abundance, and eventually density. This may not be true, however, if host populations fluctuate extensively, or if capture or inspection is difficult.

Differences in **stage-specific host associations** further complicate this sampling problem. While argasid ticks generally use one species throughout their development, the blood-meal of an ixodid larva, nymph, and adult may be taken from a different individual or host species. Ixodid ticks that feed on the same species (usually the same individual animal) during all three ectoparasitic stages have been classified as "**one-host**" species. Immatures of such ticks molt *in situ* and then reattach to the same host. Sampling of one-host ticks that is intended to estimate stage-specific abundance, analyze cohort patterns, or construct life tables is facilitated considerably by this uniformity in host affiliation. Other hard ticks behave as "**two-host**" species in which larvae and nymphs typically feed on the same vertebrate species, while adults feed on an entirely different host. Finally, "**three-host**" ticks typically attach to and detach from three different individuals, often different species. For two- and three-host ticks, stage-specific sampling may differ for each particular stage–host association. For example, trapping of small mammals is often the only practical method for sampling larvae of some species, e.g., *Dermacentor variabilis*, while adults can be captured by flagging. In contrast, for other species such as *Ixodes ricinus*,

flagging can effectively capture all active life stages. Depending on the species, complications in calculation and comparison result. As described above, these sampling concerns may be further exacerbated by the number of individuals and the diversity of species exploited by each stage.

2.4. Longevity and the Estimation of Density

As compared to most other arthropods, **tick longevity** is notoriously lengthy and variable, a life-history characteristic that may influence sampling design and population analysis. Punctuated, stage-specific activity during development and low-energy expenditure during periods of quiescence permit individuals to survive months or years, and in some cases a decade or more. Typically, life cycles of most three-host ixodids last one to many years (Balashov, 1972). Such longevity, when combined with overlap among tick generations and survival exceeding that of most hosts, poses additional difficulties in population sampling and estimation. The construction of accurate life tables, thereby, is hampered. For example, during periods of seasonal diapause (Belozerov, 1982), the inactivity of ticks prevents sampling except where they congregate (nests or burrows); even in such sites, the small size and immobility of most species makes recognition and capture difficult. Furthermore, variation in longevity among individual ticks may be considerable, depending largely on the availability of hosts. Successful attachment to a host advances development, whereas limited host abundance may delay feeding and maturation for months or years. If such variation is ignored during calculation of average survival, biologically and epidemiologically important features of population dynamics may not be appreciated.

2.5. Spatial Variation and Sampling Methods

To be meaningful, measures of absolute density or relative abundance require knowledge of the spatial distribution of organisms. Indeed, the size of an area that is necessary for accurate and reliable population estimates depends principally on the manner in which ticks, whether free-living or host-associated, are distributed in space. Free-living ticks tend to be found in particular microhabitats, typically clustered in highly non-random or non-uniform patterns. Such spatial heterogeneity within and among sampling units (e.g., nests, types of vegetation) will influence density estimates. For host-based samples, spatial variation among age- or sex-classes of hosts, differences in infestation of various body parts, and associations of individuals with certain habitats must be considered in determining the type and magnitude of samples needed. In particular, the size of the sampling unit should be small enough and the number of samples sufficiently large to evaluate spatial variation within the population. Here, both previous experience and the particular objectives underlying a sample are important considerations. These and other general sampling concepts

associated with the examination and analysis of tick populations will now be illustrated using concrete examples.

3. TECHNIQUES FOR MEASURING TICK ABUNDANCE

Two life-history characteristics of ticks that distinguish them from most other arthropod vectors directly determine the methods used to study their population ecology: ticks are relatively immotile and comparatively long lived. Many hematophagous arthropods actively seek hosts by oriented movement; most ticks, however, generally wait passively in particular microhabitats during certain diel and seasonal periods that correspond to movement of their preferred hosts. A few species of "hunter" ticks actively seek hosts by rapidly crawling several meters in response to host-originated stimuli. Nevertheless, such largely passive host seeking or questing, perhaps better termed "**ambushing**," produces temporally and spatially heterogeneous population distributions that influence sampling methods and population estimates. Furthermore, the life cycles of most tick species are measured in years rather than months or weeks, with extended periods of inactivity interrupted by brief periods of blood-feeding.

Except for certain stages or species that concentrate in animal dwellings, ticks in nature are virtually impossible to capture or count during diapause or other physiologically determined developmental events such as blood-meal digestion or eclosion. Thus, most sampling methods depend upon activity associated with host seeking, feeding, or mating. During these periods, ticks may be obtained from vertebrates or attracted to devices that mimic particular host characteristics. Techniques that sample ticks which are **questing** for hosts, **attached** to them, or **concentrated** in specific microhabitats are discussed below.

3.1. Questing Tick Sampling Techniques

Samples of questing ticks may be subdivided into those which (1) **elicit the attachment of stationary individuals** to a sampling unit moved past them, or (2) **stimulate their movement** toward an olfactory attractant associated with a mechanical trap. The former approach essentially relies on human activity occurring precisely at sites where ticks are resting, while the latter involves the passive attraction of ticks from an area surrounding a stationary, baited collecting device. Technical details of these methods have been reviewed by Sonenshine (1993); here our focus is on the manner in which various methods influence the population estimates that subsequently are derived.

"**Flagging**," "**dragging**" or "**sweeping**" are terms referring to techniques that forcefully move a piece of fabric through vegetation for a given distance or period of time; questing ticks that attach are periodically removed and counted. Human or domestic animal "**walking**" collections function similarly, except that the ticks attaching to clothing or animal fur are enumerated. These techniques sample from the questing subset of a particular stage of the tick population

that is active during the prevailing diel, vegetational, climatic, and seasonal conditions of the sample. Since each method extracts ticks from vegetation differently, details of the technique must be carefully defined. The extent and validity of samples will vary with the rate of movement through the vegetation, the frequency of tick removal and counting, and the surface area, form and consistency of the fabric. Differences among habitats in the physical structure of vegetation dramatically alter the effectiveness of these methods. With such constraints in mind, relative measures of abundance may be derived and compared using common denominators such as meters walked, person-hours sampled, or ground surface covered. Depending on the question being asked, results will acquire meaning when contrasted among comparable places or times.

Olfactory attractants represent the other main class of techniques for sampling questing ticks. This approach depends upon the sensitivity, motility and oriented movement of ticks toward an attractant-emitting trap that employs stimuli which simulate either the presence of a host or of a potential mate. Particular tick species and stages will respond differently to various attractants during specific development periods. Since these sampling devices are stationary, the physical–behavioural limitation that ticks must crawl to the trap in order to be detected and enumerated influences the utility of this approach. However, the proportion of a population that is either in diapause, immotile, molting, or otherwise not host- or mate-seeking will be overlooked by these methods. Furthermore, factors such as the concentration of the attractant, microclimatic conditions (wind, humidity), and the species-specific sensitivity to the stimulus combine to influence the effectiveness and area of the sample.

Various tick species exhibit movement toward increasing concentrations of carbon dioxide, presumably because of the association with respiring animals. Accordingly, CO_2 traps that usually employ dry ice surrounded by a sticky substance have been used to attract and capture questing ticks. Similarly, live **sentinel vertebrates**, which emit both CO_2 and other attractive volatile compounds, also sample questing ticks. Unlike wild hosts or animals that are intentionally "walked" as part of a sample, sentinel vertebrates remain caged or leached to one location; ticks that crawl on to them are enumerated. Finally, **pheromone traps** are based on the attractiveness of species-specific pheromones (Leahy, 1979; Sonenshine, 1985; Norval et al., 1989) that have been exploited in traps like those using CO_2. Here, various semiochemicals are intended to deceptively lure ticks seeking mates or congregations of other conspecifics.

Although many individual ticks may be captured with little effort using attractant traps, the area being sampled is difficult to determine; in practice, it usually remains unknown. Estimates of the effectively sampled surface can be obtained by releasing marked individuals at various distances from the trap or sentinel animal. This is time and labor intensive, however, and results vary considerably as environmental conditions change. Thus, population estimates from attractant traps generally provide relative measures of abundance whose accuracy will depend on details of the method, the nature of comparisons, and the stage and species under study.

3.2. Attached Tick Sampling Techniques

A second major class of sampling evaluates naturally **attached** ticks found feeding and/or mating on hosts. All ticks feed and most ixodids mate on vertebrates (Oliver, 1989). These host animals, whether domestic or wild, in essence function as natural, moving traps. Such living sample units collecting questing ticks at rates and coverage which vary with the age, sex, size, social class, and immune states of each animal. Differences in these factors represent a potential source of variation and possible error if not carefully considered in the design, collection, and analysis of samples. As noted elsewhere, tick species- and stage-specific patterns of activity, which vary with season and habitat, also determine the effectiveness of free-ranging vertebrates as sampling devices.

This natural "**trap animal**" approach measures the subpopulation of ticks that recently quested, found a host, and had begun a blood-meal. Such collections represent tick activity during a few days within an area corresponding to the home range of the trap animal. This approach allows comparisons of relative abundances of ticks among sites and over time, and is invaluable in studies comparing the role that different vertebrate species play in the natural history of the tick. Samples from "trap animals" avoid entirely error due to hour-to-hour changes in tick activity, as they were obtained from ticks that were questing during many days preceding capture. Unfortunately, the area over which ticks were collected is usually unknown. The investigator must select sites where mechanical traps will sample the "trap animals" that have sampled the ticks. Estimates of tick density, however, cannot be calculated from these samples without extensive knowledge about home range use by the sampled host.

Once a "trap animal" is captured, a second set of sampling problems arises: ectoparasites must be found, removed, identified, and counted. Various factors represent potential sources of sampling error including: (1) host size and type of skin cover; (2) time available for inspection, i.e., whether the animal is dead, anesthetized, or alert; (3) size of the tick (species and stage); (4) magnitude of infestation; and (5) experience of the investigator. Depending on these and other factors, the percentage of attached ticks that is actually detected will vary. This complication has been addressed by subsampling procedures that are intended to reduce the time expended or the body surface examined. Thus, subsamples that examine particular body parts or a prescribed surface area, or that last for a predefined period of time may be used to produce relative measures of infestation. Because the distribution of ticks among hosts typically is highly non-normal (Randolph, 1975; Davidar et al., 1989), these subsamples would tend to underestimate ticks on heavily infested individuals. More complete and accurate counts of infestations may be obtained by holding hosts in cages for a few days and collecting the ticks that drop into water trays or a sticky surface. Most engorged female ticks detach from their hosts following a blood-meal, however, male ticks and immature stages of one-host or two-host species may be underestimated by this technique. This caged-host method also

may impact severely on the local population of hosts by disrupting territories, denying maternal care to young, or accidentally killing animals.

The sampling unit of these approaches is usually the individual vertebrate; tick abundance, thereby, is expressed qualitatively or quantitatively as the prevalence of infestation (percentage of hosts with one or more ticks) or magnitude of infestation (number of ticks per host). Comparisons among sites using these measures may be valid if habitat and host densities are comparable. Subsamples of attached ticks (e.g., number of ticks per 10-minute examination) may be evaluated similarly using estimates of relative abundance if the host species and season studied are the same. Obviously, the rate of successful host-acquisition and feeding, which represents information crucial to the analysis of development and reproduction, can only be obtained from samples of vertebrates.

Estimates of tick density per unit area are difficult to develop using data from trap animals. Depending on the frequency and coverage of hosts sampled, the subpopulation of attached ticks may be calculated. Species-specific densities of hosts must then be combined with the number of ticks per host to derive density estimates of ticks per habitat or site. The movement of hosts is unlikely to be either random or uniform, thereby complicating calculation of the area being sampled. In some cases, changes in host density across a region may correlate inversely with counts of ticks per host (Deblinger et al., 1993). Similarly, host densities may vary during the activity season of the tick, warranting adjustment for host density (Wilson and Spielman, 1985). Many ticks express little host-specificity (Hoogstraal and Aeschlimann, 1982), compounding each of these problems by increasing the number of different hosts that may be parasitized and should be examined. In summary, sampling methods involving ticks attached to hosts allow for easy, "natural" samples that also provide other biological information about hosts; results from such observations, however, must be applied cautiously to analyses of tick population dynamics.

3.3. Sampling Natural Concentrations of Ticks

A third category of techniques samples **concentrations** of ticks in particular microhabitats where they normally abound. These methods include **aspiration** of ticks from animal burrows or nests and **artificial nest-box traps** that passively capture engorged ticks as they detach from hosts that nest therein. Aspiration, sometimes following nest or burrow excavation (Logan et al., 1993), is most useful in capturing argasids which essentially reside permanently in their hosts' dwellings. Here, relative abundance (ticks per burrow or nest) is easily and accurately estimable. If the tick species is highly host-specific and nidicolous, and if its host's dwellings are easily found and recognized, one can estimate true population density of these ticks through complete enumeration of a particular area during a defined time period. Ectoparasites of birds often present such a situation that can thus be exploited (Thompson, 1957; Duffy, 1983). Tick

species that rest attached to hosts for extended periods or tend to disperse from host nests cannot thus be sampled.

Recently, an artificial nest-box trap has been developed and applied in collecting larval and nymphal *Ixodes dammini* that detach from rodent hosts (Pollack and Spielman, in preparation). This method samples a subset of those ticks that fed and dropped from hosts while inhabiting the nest-box; ticks are held captive in a sticky substance until removed by the investigator. This method rests on the laboratory finding that most immature *I. dammini* detach from their hosts during daylight hours (Mather et al., 1987b), and field observations indicating this tick's principal rodent host, *Peromyscus leucopus*, is nocturnally active and remains nest-bound during the day. Relative abundance estimates of immature ticks may be compared among nest-box traps, although adjustment for the number of rodents using each device would increase the accuracy of such comparisons. Absolute densities cannot be calculated, however, as not all *P. leucopus* of an area may be sampled and numerous other vertebrates host a percentage of feeding larvae and nymphs.

3.4. Effectiveness and Efficiency of Sampling Techniques

The effectiveness of various sampling methods has been evaluated in terms of the number of ticks captured per unit effort (Ginsberg and Ewing, 1989; Falco and Fish, 1992; Solberg et al., 1992). Each technique is more or less effective in producing a large sample depending on the species, stage, and state of ticks being sought. The reliability of each technique, as suggested by the repeatability of multiple samples from the same site, also may vary if non-tick factors (microclimate, host behavior) are influential. The most desirable method will depend on numerous characteristics of the tick and the particular information being sought. In studies designed to measure population change, methods should be evaluated by their "**efficiency**" in reliably estimating stage-specific density. An effective method that captures many ticks per unit effort may not be efficient in reflecting the population characteristics under study. While it is generally agreed that larger sample sizes produce more accurate estimates, population analyses require counts of individuals that can be evaluated from a demographically meaningful point of reference. Thus, an efficient sampling method produces results of both quantity and quality.

3.5. General Features of Sampling Design

Regardless of which sampling methods are chosen, they must be appropriate to the question(s) being asked. Existing knowledge of the natural history of the organisms involved should be considered. For example, the timing and location of samples may be planned in relation to known or suspected diurnal, seasonal, and habitat variation in tick activity. More frequent samples may be needed if tick or host abundance varies in time. Samples obtained principally during

peak seasonal activity may produce a greater number of ticks but may not lead to the most accurate density estimate. Similarly, samples should be less dispersed in space where there is variation in habitat that is important to the tick. Alternatively, habitat-specific samples of particular environments may be deemed biologically or socially important.

Trap placement may be uniform if maximal coverage of a homogeneous habitat is sought, **random** if habitat and density are believed to be heterogeneous, or **overdispersed** (clumped) in particular habitats of interest. These considerations exist whether sampling is directed at questing, concentrated, or attached ticks. Transect or grid trapping of tick-infested vertebrates may allow estimates of host density, but assumptions of tick associations with habitat might be misleading if host movement is an important concern. Various approaches to the analysis of dispersion (Lloyd, 1967) have proven useful in characterizing the spatial pattern of a population (Taylor, 1984).

Most techniques remove the sampled subpopulation, creating potential impact on future demographic processes of interest. This may represent a particular problem if removal is either intensive in a particular time or place, or if a stage that contributes significantly to reproduction is especially affected. Estimates of potential impacts of the sampling methodology on natural demographic processes should be evaluated when designing studies that are intended to prospectively monitor population change.

4. ECOLOGICAL FACTORS AFFECTING TICK POPULATION DYNAMICS

Thorough and thoughtful reviews of the ecological patterns in tick–host–environment interactions (Hoogstraal and Aeschlimann, 1982; Hoogstraal, 1985a, 1985b) provide information on the salient features of ixodid and argasid tick ecology. Various detailed accounts covering particular taxa (Hair and Bowman, 1986; Fish, 1993) or regions (Hoogstraal, 1956; Camicas et al., 1990; Norval et al., 1992) also exist. Rather than duplicate these efforts, the following section is intended to illustrate how such ecological characteristics shape tick population interactions, as well as our efforts to measure and analyze population change. Examples, admittedly biased by my own knowledge and interests, are used to describe some of the important resources and risks that influence tick reproduction, distribution, and survival.

4.1. Host Abundance and Availability

Tick population analyses are likely to be more productive if viewed from the perspective of multiple-species interactions such as the predator–prey or parasite–host models which treat resources as interacting variables rather than constants. Considered from this perspective, the populations of ectoparasites, their hosts, and perhaps other organisms interact in a dynamic ensemble,

increasing or decreasing the rates of survival and reproduction of each other. Despite considerable evidence that most demographic events involving ticks are strongly influenced by their principal vertebrate hosts, few observations indicate reciprocal, direct impact on vertebrate hosts at a population level that could be considered density dependent. Only the heaviest infestations of individual animals appear to reduce host survival (Glines and Samuel, 1989). While measurable effects on host populations may occur (Keith and Cary, 1990), they seem rare. However, naturalistic studies may not readily detect these interactions as only surviving, active hosts are available for sample. Furthermore, the causal links underlying correlations between host health and the magnitude of infestation are extremely difficult to ascertain in nature.

Even where hosts are abundant, their importance may be influenced by the suitability of the blood-meals that they provide. Host immune responses to components of the tick's saliva (Willadsen, 1980; Wikel, 1982) have been shown to reduce blood-feeding success in certain circumstances. Reduced blood uptake and ultimately inability to complete a blood-meal would diminish reproductive success, thereby influencing population dynamics. Such acquired resistance (Trager, 1939), which has a physiological basis in tick–host interactions (Wikel and Allen, 1982), seems to depend on host reactions to each tick species (Davidar et al., 1989), and apparently is under genetic influence (George et al., 1985), and may influence population dynamics (Randolph, 1979). Equally important are the "external" influences on host populations operating via nutrition, climate, predation, or disease that may directly alter their availability to dependent ectoparasites.

4.2. Reproduction, Survival, and Immigration

The potential contribution of each female tick to growth of the population varies among species by perhaps two orders of magnitude. In general, it is enormous (Oliver, 1989). Fully engorged females of certain *Hyalomma* and *Amblyomma* species may each produce tens of thousands of eggs, while *Ixodes* or *Dermacentor* ticks generally produce 3–6,000 eggs. Egg production varies with the extent of engorgement (e.g., Wilson et al., 1990b). Fecundity of this magnitude would lead to explosive increases in density or distribution if mortality were not similarly intense. Indeed, the combined mortality rates of all stages for most species must exceed 99.9% of eggs laid if these tick populations are to remain stable.

Stage-specific mortality rates and their associated underlying causes remain one of the major unknowns in population studies of most tick species. The few studies of sufficient depth to permit life-table analysis of mortality rates suggest elevated mortality during the larvae to nymph transition (Sonenshine, 1972; Gray, 1985; Yuval and Spielman, 1990). An estimated two-thirds (Schultze et al., 1986) to four-fifths (Carey et al., 1980) of larval *I. dammini* die before becoming nymphs. Our lack of knowledge concerning stage-specific mortality rates has seriously hindered analysis of most tick populations. Despite generally

high mortality rates of populations, a few individuals of most tick species exhibit remarkably lengthy longevity as a result of seasonal diapause and behavioral arrest in the absence of blood-meals. Although certain one-host species (e.g., *Boophilus*) in the tropics may achieve a few generations each year, most ticks require a year or more to produce the next generation. Hard tick life cycles lasting many years (Balashov, 1972) and survival of soft ticks that may encompass a decade or more (Hoogstraal, 1985b) have been reported. Laboratory studies, however, may artificially increase estimates of survival rates (Daniels et al., 1989) by creating optimal conditions. Field studies of survival (e.g., Stafford, 1992) would provide more realistic estimates.

Tick movement to or from a defined area may alter that species' local population density. Lacking wings, however, movement occurs by crawling or passive transport on hosts. Typically, size and anatomy limit crawling to short distances (tens of meters). Consequently, since most questions of tick density involve larger areas, the principal observations of movement involve hosts to which they are attached (Wilson et al., 1990a; Telford et al., 1993). In this manner, tick density may increase where feeding ticks detach from infested hosts that migrate through a region or simply as they move within their home ranges. In North America, immature *Haemaphysalis*, *Dermacentor*, and *Ixodes*, are known to parasitize birds (Clifford et al., 1969; Sonenshine and Stout, 1970; Sonenshine and Clifford, 1973), offering the opportunity for local and perhaps long-distance transport. *Ixodes dammini*, in particular, has been removed from at least 60 bird species in the northeastern US (Anderson et al., 1986; Schulze et al., 1986; Battaly et al., 1987). In eastern Africa, Hoogstraal and colleagues (1961, 1963, 1964) have demonstrated the long-distance transport of ticks by birds migrating to and from central Eurasia.

Hosts such as small mammals, however, usually move within home ranges measuring tens or hundreds of square meters; migration of greater distances is infrequent. Various rodent species in particular may harbor numerous immature ticks but appear to have restricted home ranges. Most epidemiologically important ticks also feed as adults on medium-sized or large mammals, especially ungulates. For example, *I ricinus* adults primarily parasitize sheep (Milne, 1950a, 1950b; Gray, 1982, 1984) making movement of attached adult ticks possible. In North America, adult *I. dammini* feed principally on white-tailed deer, a host able to distribute this tick over somewhat greater distances (Wilson and Deblinger, 1993). The role of mammal movement in dispersion of tick populations has not been adequately studied.

4.3. Environmental Influences on Development and Survival

Tick density-independent factors that comprise the non-host, physical environment strongly influence tick activity, development, reproduction, survival, and hence demography (see Chapter 5 by Daniel and Dusbabek). Unlike more complex, interactive host-dependent events, abiotic environmental effects appear to be largely unidirectional, with little or no feedback. One result is that they

often are more easily monitored and evaluated. While dense aggregations of ticks might produce altered microclimate, most often ticks select from among existing environmental conditions by oriented movement or changing diapause.

Macroclimatic conditions may limit distributions of ticks by posing temperature or humidity extremes exceeding those that a particular species has evolved to tolerate (e.g., MacLeod, 1934; McEnroe, 1977; Gardiner et al., 1981; Camicas et al., 1990; Perry et al., 1990). Laboratory observations confirm the existence of optimal physical conditions permitting development or increasing longevity of host-independent ticks (Needham and Teel, 1991). These limits are more or less narrow, and have evolved for tropical (Wilson et al., 1993), temperate (MacLeod, 1934, 1935), and arctic (Lee and Basut, 1987) species. At the limits of contemporary distributions, hosts presumably carry ticks into climatic conditions that are unfavourable, allowing for selection that influences macrogeographic distributions (see Chapter 6 by Korch). Climate directly influences tick overwintering (Daniels et al., 1989), reproduction (MacLeod, 1935) and spatial distributions (Milne, 1950b).

Climate partly determines the nature and extent of vegetation, which in turn influences environment by contributing to microhabitat, and thereby influences tick population dynamics. Whether directly (Semtner et al., 1971; Sonenshine and Levy, 1972) or indirectly through host distributions (Milne, 1950; Wilson et al., 1985, 1990a), variation in vegetation may alter microhabitat and influence survival and questing (Adler et al., 1992). Where extreme fluctuations exist, local extinction of tick populations may be observed (Norval and Perry, 1990).

Experimental field studies that have altered vegetation and hence modified microhabitat further demonstrate impact on tick population abundance. Cutting or mowing grass and shrubs has reduced abundance of questing *Dermacentor albipictus* (Drew et al., 1985), *Amblyomma americanum* (Hoch et al., 1972), and *Ixodes dammini* (Wilson, 1986). Implications for tick control are evident.

4.4. Predators, Parasitoids, and Pathogens

Interactions among ticks and their predators, parasites or pathogens have been reported from diverse settings (Jenkins, 1964; Wilson and Deblinger, 1993); little is known, however, concerning impacts on population dynamics. Although numerous vertebrates and invertebrates exploit ticks under natural conditions, few studies have addressed whether these interactions alter longevity or reproduction in a manner or magnitude that would influence population densities.

The known **predators** of ticks include other arthropods and various vertebrates. Most reports of predation, however, are largely anecdotal without quantitative measurement of effects on abundance. Insects such as ants have been observed to prey on ixodid and argasid eggs or engorged ticks. "Fire ants" (*Solenopsis invecta*) effectively reduced the local abundance of *Amblyomma americanum* in the southern US (Harris and Burns, 1972; Burns and Melancon,

1977; Fleetwood et al., 1984), while *S. geminata* diminished the number of replete female *Boophilus microplus* in Mexico (Butler et al., 1979). Similarly, spiders have been reported to prey on argasid (Ault and Elliott, 1979; Clifford et al., 1980) and ixodid (Wilkinson, 1970a) ticks; however, in-depth analyses are lacking. Spider predation effectively reduced the abundance of immature and adult *Rhipicephalus sanguineus* in Corsica (Sautet, 1936), suggesting a possible role in stabilizing populations of this tick under certain conditions. Theoretically, such generalist predators as ants and spiders, whose populations would not depend upon those of any particular prey, should be independent of those of the tick. As such, they may serve to stabilize tick populations without fluctuating out of synchrony with their prey (Levins and Wilson, 1980).

Vertebrate predators, including reptiles, birds and mammals also have been observed to prey on ticks (Jenkins, 1964); again, systematic observations are lacking. For example, lizards were seen feeding on *Ornithodoros* ticks (Clifford et al., 1980) and a skink was reported as a predator of gorged female *Amblyomma hebraeum* in Zimbabwe (Norval and McCosker, 1983). Birds such as African oxpeckers (Moreau, 1933; Van Someren, 1951) and cattle egrets (Sergent et al., 1945) are well-known consumers of large numbers of ticks. Chickens reportedly feed on engorged ticks detaching from cattle (Hunter and Bishopp, 1911) and predation of engorged *Dermacentor andersoni* by robins has been observed (Wilkinson, 1970b). Mammals such as shrews have been seen consuming adult *Ixodes ricinus* in England (Milner, 1950a) and *Rhipicephalus appendiculatus* in southern Africa (Short and Norval, 1982). Unfortunately, the impact of such generalist predators on tick population dynamics is virtually unstudied.

Parasitoids that deposit their eggs in or on ticks, effectively kill such individuals; their long-term impact on tick population density, however, remains to be evaluated. Certain adult wasps (Chacidae) oviposit on engorging ticks, producing larvae that consume the tick during development. One such cosmopolitan species, *Hunterellus hookeri* (=*Ixodiphagus caucurteri*) was released on to Naushon Island, Massachusetts, USA, in an attempt to suppress populations of *Dermacentor variabilis*, however, no impact was observed (Larousse et al., 1928; Smith and Cole, 1943). Similar studies on the impact of *H. hookeri* on *D. andersoni* produced no measurable effect on the tick population (Cooley and Kohls, 1934). Indeed, this wasp parasitizes *I. ricinus* and *I. persulcatus* in Europe (Jenkins, 1964), and was recently rediscovered in *I. dammini* populations from Massachusetts coastal islands. Despite prevalent parasitism, *I. dammini* abundance appeared similar to that of surrounding sites where this wasp was absent (Mather et al., 1987a). Studies of other related *Hunterellus* parasitoids of *Hyalomma* ticks in Africa (Hoogstraal and Kaiser, 1961) and *Haemaphysalis* ticks in India (Geevarghese and Sreenivasan, 1973) should be revealing.

While the wasp may be tick-density dependent, under certain conditions, a large proportion of ticks could be affected. For example, more than 90% of *Rhipicephalus sanguineus* were found to be parasitized by *H. hookeri* in one Nigerian study (Philip, 1931). Further research, both theoretical and

experimental, is needed to understand how parasitoids might influence tick populations.

Pathogens of ticks remain poorly studied, although various protozoa, bacteria and fungi have been described (Lipa, 1971; Hoogstraal, 1977). Certain fungi infect *Ixodes ricinus* and other ixodid ticks in Europe either as obligate or facultative parasites, or as saprophytes (Samsinakova et al., 1974). Infection is usually non-lethal, but varies by stage and season, and may influence reproduction or survival in less obvious ways. Bacterial (Brown et al., 1970) and rickettsial (Burgdorfer et al., 1973) infections of *Dermacentor andersoni* have been reported, but no evidence of impact on natural populations exists. Tick vectors of vertebrate pathogens such as *Babesia bigemina* (Gray, 1982b) and *Babesia bovis* (Davey, 1981) may exhibit reduced reproduction or survival when infected. A curious observation suggests that *Babesia microti* infection of *Ixodes trianguliceps* may actually increase feeding success and survival (Randolph, 1991). Unfortunately, of the many known tick infections, few have been studied in nature, and virtually never for their population-level impacts.

5. IMPLICATIONS FOR TICK-BORNE DISEASE RESEARCH

Accurate, though not precise, estimates of population processes are necessary to evaluate the role of a vector in pathogen transmission. Ultimately, any effort to reduce human risk of infection is directly linked to our knowledge of these principal population interactions. Aside from analyzing interactions and understanding dynamics, our ability to predict population changes in light of changing environments or purposeful intervention demands particular information about a variety of demographic events. The utility of that information partly depends on the manner in which it was collected, but also the skill with which it is integrated. Various approaches to the analysis of such data using statistical, life-table, simulation, and spatial models are discussed in Chapter 7 by Kitron and Mannelli.

Most of the concerns addressed in this chapter are equally relevant to population interactions when a vertebrate pathogen is introduced. The type of transmission system, including its complexity, the variety and competence of vectors and reservoirs, and the spatial/temporal patterns of contact will all influence the efficiency and stability with which transmission is maintained (see Chapter 4 by Mather and Ginsberg). Essentially all tick-borne pathogens that cause disease in humans are zoonoses whereby transmission is maintained in a tick–vertebrate–tick cycle; some tick-borne diseases of animals may be maintained enzootically. The number of tick vectors that are highly competent for any given pathogen is usually small; often only one plays a major role in transmission. But the capacity of a tick to vector pathogens is a complex function of its intrinsic competence and many other population parameters involving interactions between potential reservoirs and other vectors (Mather et al., 1989; Wilson et al., 1990b).

Foremost among the factors that influence transmission of arthropod-borne disease agents are the number of times a vector feeds and the diversity of vertebrate hosts that are used. The three, stage-specific, blood meals of ixodid ticks limit the possibility of infection by pathogens normally transmitted horizontally from tick to vertebrate to tick. (Vertical or transovarial transmission, in which the next generation of ticks is directly infected by their parents, simplifies this relationship.) The particular host-feeding pattern, a function of most of the population parameters discussed in this chapter, becomes extremely important in maintaining stable transmission. For example, infected nymphal ticks that feed on vertebrate species different from those used by larvae cannot participate in horizontal transmission of a disease agent. Furthermore, whether ticks are one-, two-, or three-host species directly influences their ability to become infected and transmit. These and other such factors play important roles in the ecology of transmission systems, which represent an extension of the vector population concepts that have been outlined here. Applying these concepts and techniques with care and thought, we shall be better able to gain insight into the complex interactions of vector-borne pathogen dynamics. Our ability to influence their direction in a manner that reduces human or animal disease depends on such a perspective.

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7. REFERENCES

Adeyeye, O.O. and Butler, J.F. (1989) Population structure and seasonal intraburrow movement of *Ornithodoros turicata* (Acari: Argasidae) in gopher tortoise burrows. *J. Med. Entomol.* **26**, 279–283.

Adler, G.H., Telford, S.R., III, Wilson, M.L., and Spielman, A. (1992) Vegetation structure influences the burden of immature *Ixodes dammini* on its main host, *Peromyscus leucopus*. *Parasitol.* **105**, 105–110.

Anderson, J.F., Johnson, R.C., Magnarelli, L.A., Hyde, F.W., and Myers, J.E. (1986) Involvement of birds in the epidemiology of the Lyme disease agent Borrelia burgdorferi. *Infect. Immun.* **51**, 394–396.

Ault, S.A. and Elliot, K.D. (1979) Spider predatation (Aranaea: Salticidae) on *Ornithodoros coriaceus* (Acarina: Argasidae), with a survey of the predators of the genus *Ornithodoros*. *J. Med. Entomol.* **15**, 570.

Balashov, Y.S. (1972) Bloodsucking ticks (Ixodoidea)—Vectors of diseases of man and animals. *Misc. Publ. Entomol. Soc. Am.* **8**, 1–367.

Barr, A.R. (1979) Epidemiological concepts for entomologists. *Entomol. Soc. Am. Bull.* **25**, 129.

Battaly, G.R., Fish, D., and Dowler, R.D. (1987) The seasonal occurrence of *Ixodes dammini* and *Ixodes dentatus* (Acari: Ixodidae) on birds in a Lyme disease endemic area of southeastern New York state. *J. New York Entomol. Soc.* **95**, 461–468.

Belozerov, V.N. (1982) Diapause and biological rhythms in ticks. pp. 469–500. *In:* Obenchain, F.D. and Galun, R. (eds) *Physiology of Ticks.* Pergamon, Oxford.

Brown, R.S., Reichelderfer, C.F., and Anderson, W.R. (1970) An endemic disease among laboratory populations of *Dermacentor andersoni* (=*venustus*) (Acarina: Ixodidae). *J. Invertebr. Pathol.* **16**, 142.

Burgdorfer, W., Brinton, L.P., and Hughes, L.E. (1973) Isolation and characterization of symbiotes from the Rocky Mountain wood tick, *Dermacentor andersoni. J. Invertebr. Pathol.* **22**, 424.

Burns, E.C. and Melancon, D.G. (1977) Effect of imported fire ant (Hymenoptera: Formicidae) invasion on lone star tick (Acari: Ixodidae) populations. *J. Med. Entomol.* **14**, 247–249.

Butler, J.F., Camino, M.L., and Perez, T.O. (1979) *Boophilus microplus* and the fire ant, *Solenopsis geminata.* Vol. 1, pp. 469–472. *In:* Rodriguez, J.G. (ed) *Recent Advances in Acarology.* Academic Press, New York.

Butler, J.F., Holscher, K.H., Adeyeye, O.A., and Gibbs, E.P.J. (1984) Sampling techniques for burrow dwelling ticks in reference to potential African swine fever virus vectors. Vol. 2, pp. 1065–1074. *In:* Griffiths, D.A. and Bowman, C.E. (eds) *Acarology VI.* Ellis Horwood, Chichester.

Camicas, J.L., Wilson, M.L., Cornet, J.P., Digoutte, J.P., Calvo, M.A., Adam, F., and Gonzalez, J.P. (1990) Relative potential of ticks as vectors of Crimean–Congo hemorrhagic fever virus in Senegal: ecological and epidemiological implications. *Archiv. Virol.* (Suppl. 1), 303–322.

Carey, A.B., Krinsky, W.L., and Main, A.J. (1980) *Ixodes dammini* (Acari: Ixodidae) and associated ixodid ticks in south-central Connecticut, USA. *J. Med. Entomol.* **17**, 89–99.

Clifford, C.M., Sonenshine, D.E., Atwood, E.T., Robbins, C.S., and Hughes, L.E. (1969) Tests on ticks from wild birds collected in the eastern United States for rickettsiae and viruses. *Am. J. Trop. Med. Hyg.* **18**, 1057–1061.

Clifford, C.M., Hoogstraal, H., Radovsky, F.J., Stiller, D., and Keirans, H.E. (1980) *Ornithodoros* (*Alectorobius*) *ambius* (Acarina: Ixodoidea: Argasidae): identity, marine bird and human hosts, virus infections, and distribution in Peru. *J. Parasitol* **66**, 312–323.

Cooley, R.A. and Kohls, G.M. (1934) A summary of tick parasites. *Proc. 5th Pacific Sci. Cong.* **5**, 3375–3397.

Daniels, T.J., Fish, D., and Falco, R.C. (1989) Seasonal activity and survival of adult *Ixodes dammini* (Acari: Ixodidae) in southern New York State. *J. Med. Entomol.* **26**, 610–614.

Davey, R.B. (1981) Effects of *Babesia bovis* on the ovipositional success of the southern cattle tick *Boophilus microplus. Ann. Entomol. Soc. Am.* **74**, 331–333.

Davidar, P., Wilson, M., annd Ribeiro, J.M.C. (1989) Differential distribution of immature *Ixodes dammini* (Acari: Ixodidae) on rodent hosts. *J. Parasitol.* **75**, 898–904.

Deblinger, R.D., Wilson, M.L., Rimmer, D.W., and Spielman, A. (1993) Reduced abundance of immature *Ixodes dammini* (Acari: Ixodidae) following incremental removal of deer. *J. Med. Entomol.* **30**, 144–150.

Denmark, H.A. and Clifford, C.M. (1962) A tick of the *Ornithodoros capensis* group established on Bush Key, Dry Tortugas, Florida. *Florida Entomol.* **45**, 139–142.

Drew, M.L., Samuel, W.M., Lukiwski, G.M., and Willman, J.N. (1985) An evaluation of burning for control of winter ticks, *Dermacentor albipictus*, in Central Alberta. *J. Wildl. Dis.* **21**, 313–315.

Duffy, D.C. (1983) The ecology of tick parasitism on densely nesting Peruvian seabirds. *Ecology* **64**, 110–119.

Eberhardt, L.L. and Thomas, J.M. (1991) Designing environmental field studies. *Ecol. Monogr.* **61**, 53–73.

Falco, R.C. and Fish, D. (1992) A comparison of methods for sampling the deer tick, *Ixodes dammini*, in a Lyme disease endemic area. *Exp. Appl. Acarol.* **14**, 165–173.

Fish, D. (1993) Population ecology of *Ixodes dammini*. pp. 25–42. *In:* Ginsberg, H. (ed) *Ecology and Environmental Management of Lyme Disease*. Rutgers University Press, New Brunswick.

Fleetwood, S.C., Teel, P.D., and Thompson, G. (1984) Impact of imported fire ant on lone star tick mortality in open and canopied pasture habitats of east central Texas. *Southwest. Entomol.* **9**, 158–162.

Gardiner, W.P., Gettinby, W., and Gray, J.S. (1981) Models based on weather for the development phases of the sheep tick, *Ixodes ricinus* L. *Vet. Parasitol.* **9**, 75–86.

Geevarghese, G. and Sreenivasan, M.A. (1973) Observations on *Hunterellus* sp. (Hymenoptera: Encyritidae) parasitizing *Haemaphysalis bispinosa* Neumann, 1897 (Acari: Ixodidae). *Indian J. Med. Res.* **61**, 992–997.

George, J.E., Osburn, F.L., and Wikel, S.K. (1985) Acquisition and expression of resistance by *Bos indicus* × *Bos taurus* calves to *Amblyomma americanum* infestation. *J. Parasitol.* **71**, 174–182.

Ginsberg, H.S. and Ewing, C.P. (1989) Habitat distribution of *Ixodes dammini* (Acari: Ixodidae) and Lyme disease spirochetes on Fire Island, New York. *J. Med. Entomol.* **26**, 183–189.

Glines, M. and Samuel, W. (1989) Effect of *Dermacentor albipictus* (Acari: Ixodidae) on blood composition, weight gain, and hair coat of moose, *Alces alces*. *Exp. Appl. Acarol.* **6**, 197–213.

Gray, J.S. (1982a) The development and questing activity of *Ixodes ricinus* (L) (Acari: Ixodidae) under field conditions in Ireland. *Bull. Entomol. Res.* **72**, 263–270.

Gray, J.S. (1982b) The effects of the piroplasm *Babesia bigemina* on survival and reproduction of the blue ticks, *Boophilus decoloratus*. *J. Invert. Pathol.* **39**, 413–415.

Gray, J.S. (1984) Studies on the dynamics of active populations of the sheep tick, *Ixodes ricinus* L. in Co. Wicklow, Ireland. *Acarologia* **25**, 167–178.

Gray, J.S. (1985) Studies of the larval activity of the tick *Ixodes ricinus* L. in Co. Wicklow, Ireland. *Exp. Appl. Acarol.* **1**, 307–316.

Hair, J.A. and Bowman, J.L. (1986) Behavioral ecology of *Amblyomma americanum* (L.). pp. 406–427. *In:* Sauer, J.R. and Hair, J.A. (eds) *Morphology, Physiology, and Behavioral Biology of Ticks*. Ellis Horwood, Chichester.

Harris, W.G. and Burns, E.C. (1972) Predation on the lone star tick by the imported fire ant. *Environ. Entomol.* **1**, 362–365.

Hoch, A.L., Semtner, P.J., Barker, R.W., and Hair, J.A. (1972) Preliminary observation on controlled burning for lone star tick (Acarina: Ixodidae) control in woodlots. *J. Med. Entomol.* **9**, 446–451.

Hoogstraal, H. (1956) *African Ixodoidea*, Vol. 1, *Ticks of the Sudan*. US Naval Medical Research Unit No. 3. Cairo, Egypt, pp. 1–1101.

Hoogstraal, H. (1977) Pathogens of acarina (ticks). pp. 337–342. *In:* Roberts, D.W. and Strand, M.A. (eds) *Pathogens of Medically Important Arthropods*. World Health Organization, Geneva.

Hoogstraal, H. (1981) Changing patterns of tickborne diseases in modern society. *Annu. Rev. Entomol.* **26**, 75–99.

Hoogstraal, H. (1985a) Ticks, pp. 347–370. *In:* Gaafar, S.M., Howard, W.E., and Marsh, R.E. (eds) *Parasites, Pests and Predators.* Elsevier, Amsterdam.

Hoogstraal, H. (1985b) Argasid and Nuttalliellid ticks as parasites and vectors. *Adv. Parasitol.* **24**, 125–238.

Hoogstraal, H. and Kaiser, M.N. (1961) Records of *Hunterellus theilerae* Fiedler (Encyrtidae, Chalcidoidae) parasitizing *Hyalomma* ticks on birds migrating through Egypt. *Ann. Entomol. Soc. Am.* **54**, 616–617.

Hoogstraal, H. and Aeschlimann, A. (1982) Tick-host specificity. *Bull. Soc. Entomol. Suisse* **55**, 5–32.

Hoogstraal, H., Kaiser, M.N., Traylor, M.A., Gaber, S., and Guindy, E. (1961) Ticks (Ixodoidea) on birds migrating from Africa to Europe and Asia. *Bull. WHO*, **24**, 197–212.

Hoogstraal, H., Kaiser, M.N., Traylor, M.A., Guindy, E., and Gaber, S. (1963) Ticks (Ixodidae) on birds migrating from Europe and Asia to Africa, 1959–61. *Bull. WHO* **28**, 235–262.

Hoogstraal, H., Traylor, M.A., Gaber, S., Malakatis, G., Guindy, E., and Helmy, I. (1964) Ticks (Ixodidae) on migrating birds in Egypt, spring and fall 1962. *Bull. WHO* **30**, 355–367.

Hunter, W.D. and Bishopp, F.C. (1911) The Rocky Mountain spotted fever tick. *USDA Bur. Entomol. Bull.* No. 105.

Jenkins, D.W. (1964) Pathogens, parasites and predators of medically important arthropods. *Bull. WHO Suppl.* **30**, 1–150.

Keith, L. and Cary, J. (1990) Interaction of the tick (*Haemaphysalis leporisplaustris*) with a cyclic snowshoe hare (*Lepus americanus*) population. *J. Wildl. Manag.* **26**, 427–434.

Kuno, E. (1991) Sampling and analysis of insect populations. *Annu. Rev. Entomol.* **36**, 285–302.

Larousse, F., King, A.G., and Wolbach, S.B. (1928) The overwintering in Massachusetts of *Ixodiphagus caucurteri. Science* **67**, 351–353.

Leahy, M.G. (1979) Pheromones of argasid ticks. Vol II, pp. 297–308. *In:* Rodriguez, J.G. (ed) *Recent Advances in Acarology.* Academic Press, New York.

Lee, R.E. and Basut, J.G. (1987) Gold-hardiness in the Antarctic tick, *Ixodes uriae. Physiol. Ecol.* **60**, 499–506.

Levins, R. and Wilson, M.L. (1980) Ecological theory and pest management. *Annu. Rev. Entomol.* **25**, 287–308.

Lipa, J.J. (1971) Microbial control of mites and ticks. pp. 357–373. *In:* Burges, H.D. and Hussey, N.W. (eds) *Microbial Control of Insects and Mites.* Academic Press, New York.

Lloyd, M. (1967) Mean crowding. *J. Anim. Ecol.* **36**, 1–30.

Logan, T.M., Wilson, M.L. and Cornet, J.P. (1993) Association of ticks (Acari: Ixodidae) with rodent burrows in northern Senegal. *J. Med. Entomol.* **30**, 799–801.

MacLeod, J. (1934) *Ixodes ricinus* in relation to its physical environment. The influence of climate on development. *Parasitol.* **26**, 282–305.

MacLeod, J. (1935) *Ixodes ricinus* in relation to its physical environment. III. Climate and reproduction. *Parasitol.* **27**, 489–500.

Margolis, L., Esch, G.W., Holmes, J.C., Kuris, A.M., and Schad, G.A. (1982) The use of ecological terms in parasitology (report of an ad hoc committee of the American Society of Parasitology). *J. Parasitol.* **68**, 131–133.

Mather, T.N., Piesman, J., and Spielman, A. (1987a) Absence of spirochaetes (*Borrelia burgdorferi*) and piroplasms (*Babesia microti*) in deer ticks (*Ixodes dammini*) parasitized by chalcid wasps (*Hunterellus hookeri*). *Med. Vet. Entomol.* **1**, 3–8.

Mather, T.N., Ribeiro, J.M.C., and Spielman, A. (1987b) Lyme disease and babesiosis: acaricide focused on potentially infected ticks. *Am. J. Trop. Med. Hyg.* **36**, 609–614.

Mather, T.N., Wilson, M.L., Moore, S.I., Ribeiro, J.M.C., and Spielman, A. (1989) Comparing the relative potential of rodents as reservoirs of the Lyme disease spirochete (*Borrelia burgdorferi*). *Am. J. Epidemiol.* **130**, 143–150.

McEnroe, W.D. (1977) The restriction of the species range of *Ixodes scapularis* Say, in Massachusetts by fall and winter temperature. *Acarologia* **18**, 618–625.

Milne, A. (1950a) The ecology of the sheep tick *Ixodes ricinus* L.: microhabitat economy of the adult tick. *Parasitol.* **40**, 14–34.

Milne, A. (1950b) The ecology of the sheep tick *Ixodes ricinus* L.: spatial distribution. *Parasitol.* **40**, 35–45.

Moreau, R.E. (1933) The food of the red-billed oxpecker, *Buphagus erythrorhynchus* (Stanley). *Bull. Entomol. Res.* **24**, 325–335.

Morris, R.F. (1960) Sampling insect populations. *Annu. Rev. Entomol.* **5**, 243–264.

Needham, G.R. and Teel, P.D. (1991) Off-host physiological ecology of ixodid ticks. *Annu. Rev. Entomol.* **36**, 359–381.

Norval, R.A.I. and McCosker, P.J. (1983) Tick predation by a rainbow skink. *Zimbabwe Vet. J.* **13**, 53.

Norval, R.A.I. and Perry, B.D. (1990) The introduction, spread, and subsequent disappearance of the brown ear-tick (*Rhipicephalus appendiculatus*) from the southern lowvelt of Zimbabwe. *Exp. Appl. Acarol.* **9**, 103–111.

Norval, R.A.I., Butler, J.F., and Yunker, C.E. (1989) Use of carbon dioxide and natural or synthetic aggregation–attachment pheromone of the bont tick, *Ambyomma hebraeum*, to attract and trap unfed adults in the field. *Exp. Appl. Acarol.* **7**, 171–180.

Norval, R.A.I., Perry, B.D., and Young, A.S. (1992) *The Epidemiology of Theileriosis in Africa.* Academic Press, London.

Obenchain, R.D. and Galun, R. (1982) *Physiology of Ticks.* Pergamon, Oxford.

Oliver, J.H., Jr (1983) Chromosomes, genetic variance and reproductive strategy among mites and ticks. *Bull. Entomol. Soc. Am.* **29**, 9–17.

Oliver, J.H., Jr (1989) Biology and systematics of ticks (Acari: Ixodida). *Annu. Rev. Ecol. Syst.* **20**, 397–430.

Perry, D.B., Lessard, P., Norval, R.A.I., Kundert, K., and Kruska, R. (1990) Climate, vegetation and the distribution of *Rhipicephalus appendiculatus* in Africa. *Parasitol. Today* **6**, 100–104.

Philip, C.B. (1931) Occurrence of a colony of the tick parasite, *Hunterellus hookeri* Howard, in West Africa. *Publ. Health Rep.* **46**, 2168–2172.

Pollack, R.J. and Spielman, A. A sensitive method for assessing risk of infection by the agent of Lyme disease. *Am. J. Trop. Med. Hyg.* (in preparation).

Pollock, K.H., Nichols, J.D., Brownie, C., and Hines, J.E. (1990) Statistical inference for capture–recapture experiments. *Wildl. Monogr.* **107**, 1–98.

Randolph, S.E. (1975) Patterns of distribution of the tick *Ixodes trianguliceps* Birula on its hosts. *J. Anim. Ecol.* **44**, 451–474.

Randolph, S.E. (1979) Population regulation in ticks: the role of acquired resistance in natural and unnatural hosts. *Parasitol.* **79**, 141–156.

Randolph, S.E. (1991) The effect of *Babesia microti* on feeding and survival in its tick vector, *Ixodes trianguliceps*. *Parasitol.* **102**, 9–16.

Samsinakova, A., Kalalova, S., Daniel, M., Dusbabek, F., Honzakova, E., and Cerny, V. (1974) Entomogenous fungi associated with the tick *Ixodes ricinus* (L.). *Folia Parasitol.* **21**, 39–48.

Sautet, J. (1936) Invasion domicilaire de *Rhipicephalus sanguineus* et de *Teutana triangulosa*. Role ixodiphage des araignées. *Ann. Parasit. Hum. Comp.* **14**, 126–129.

Schulze, T.L., Bowen, J.K., Lakat, M.F., Parkin, W.E., and Shisler, G.S. (1986) Seasonal abundance and hosts of *Ixodes dammini* (Acari: Ixodidae) and other ixodid ticks from an endemic Lyme disease focus in New Jersey, USA. *J. Med. Entomol.* **23**, 105–109.

Semtner, P.J., Howell, D.E., and Hair, J.A. (1971) The ecology and behavior of the lone star tick (Acarina: Ixodidae): I. The relationship between vegetative habitat type and tick abundance and distribution in Cherokee Co., Oklahoma. *J. Med. Entomol.* **8**, 329–335.

Sergent, E., Donatien, A., Parrot, L., and Lestoquard, F. (1945) Etudes sur les piroplasmoses bovines, Alger, Instit. Pasteur d'Algérie, 816 pp. *Rev. Appl. Entomol. B* **37**, 54.

Short, N.J. and Norval, R.A.I. (1982) Tick predation by shrews in Zimbabwe. *J. Parasitol.* **68**, 10–52.

Smith, C.N. and Cole, M.M. (1943) Studies of parasites of the American dog tick. *J. Econ. Entomol.* **36**, 569–572.

Solberg, V.B., Neidhardt, K., Sardelis, M.R., Hildebrandt, C., Hoffmann, F.J., and Boobar, L.R. (1992) Quantitative evluation of sampling methods for *Ixodes dammini* and *Ambylomma americanum* (Acari: Ixodidae). *J. Med. Entomol.* **29**, 451–456.

Sonenshine, D. (1972) Ecology of the American dog tick, *Dermacentor variabilis*, in a study area in Virginia. I. Studies on population dynamics using radioecological methods. *Ann. Entomol. Soc. Am*, **65**, 1164–1175.

Sonenshine, D.E. (1985) Pheromones and other semiochemicals of the Acari. *Annu. Rev. Entomol.* **30**, 1–28.

Sonenshine, D.E. (1993) *Biology of Ticks*, Vol. 2. Oxford University Press, New York.

Sonenshine, D.E. and Stout, I.J. (1970) A contribution to the ecology of ticks infesting wild birds and rabbits in the Virginia–North Carolina piedmont (Acarina: Ixodidae). *J. Med. Entomol.* **7**, 645–654.

Sonenshine, D.E. and Levy, G.F. (1972) Ecology of the American dog tick, *Dermacentor variabilis*, in a study area in Virginia. Distribution in relation to vegetative types. *Ann. Entomol. Soc. Am.* **65**, 1176–1182.

Sonenshine, D.E. and Clifford, C.M. (1973) Contrasting incidence of Rocky Mountain spotted fever in ticks infesting wild birds in eastern U.S. piedmont and coastal areas, with notes on the ecology of these ticks. *J. Med. Entomol.* **10**, 597–602.

Southwood, T.R.E. (1978) *Ecological Methods*, 2nd edn. Chapman and Hall, London.

Stafford, K.S., III (1992) Oviposition and larval dispersal of *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **29**, 129–132.

Taylor, L.R. (1984) Assessing and interpreting the spatial distributions of insect populations. *Annu. Rev. Entomol.* **29**, 321–357.

Telford, S.R., III, Urioste, S.S., and Spielman, A. (1993) Clustering of host-seeking nymphal deer ticks (*Ixodes dammini*) infected by Lyme disease spirochetes (*Borrelia burgdorferi*). *Am. J. Trop. Med. Hyg.* **47**, 55–60.

Thompson, G.B. (1957) The parasites of British birds and mammals. XXXI. Summary of the records of ticks of sea-birds. *Entomol. Monthly Mag.* **93**, 1120.

Trager, W. (1939) Acquired immunity to ticks. *J. Parasitol.* **25**, 57–81.

Van Someren, V.D. (1951) The red-billed oxpecker and its relation to stock in Kenya. *E. Afr. Agric. J.* **17**, 1–11.

Wikel, S.K. (1982) Immune response to arthropods and their products. *Annu. Rev. Entomol.* **27**, 21–28.

Wikel, S.K. and Allen, J.R. (1982) Immunological basis of host resistance to ticks. pp. 169–196. *In:* Obenchain, F.D. and Galun, R. (eds) *Physiology of Ticks.* Pergamon, Oxford

Wilkinson, P.R. (1970a) Factors affecting the distribution and abundance of the cattle tick in Australia: observations and hypotheses. *Acarologia* **12**, 492–508.

Wilkinson, P.R. (1970b) A preliminary note on predation on free-living engorged female Rocky Mountain wood ticks. *J. Med. Entomol* **7**, 493–496.

Willadsen, P. (1980) Immunity to ticks. *Adv. Parasitol.* **18**, 293–313.

Wilson, M.L. (1986) Reduced abundance of adult *Ixodes dammini* (Acari: Ixodidae) following destruction of vegetation. *J. Econ. Entomol.* **79**, 693–696.

Wilson, M.L. and Spielman, A. (1985) Seasonal activity of immature *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **22**, 408–414.

Wilson, M.L. and Deblinger, R.D. (1993) Vector management to reduce the risk of Lyme disease. pp. 126–156. *In:* Ginsberg, H. (ed) *Ecology and Environmental Management of Lyme Disease.* Rutgers University Press, New Brunswick.

Wilson, M.L., Adler, G.H., and Spielman, A. (1985) Correlation between abundance of deer and that of the deer tick, *Ixodes dammini* (Acari: Ixodidae). *Ann. Entomol. Soc. Am.* **78**, 172–176.

Wilson, M.L., Telford, S.A., III, Piesman, J., and Spielman, A. (1988) Reduced abundance of immature *Ixodes dammini* (Acari: Ixodidae) following elimination of deer. *J. Med. Entomol.* **25**, 224–228.

Wilson, M.L., Ducey, A.M., Litwin, T.S., Gavin, T.A., and Spielman, A. (1990a) Microgeographic distribution of immature *Ixodes dammini* (Acari: Ixodidae) correlated with that of deer. *Med. Vet. Entomol.* **4**, 151–160.

Wilson, M.L., Litwin, T.S., Gavin, T.A., Capkanis, M.C., Maclean, D., and Spielman, A. (1990b) Host-dependent differences in feeding and reproduction of *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **30**, 467–471.

Wilson, M.L., Dykstra, E.A., and Schmidt, E.A. (1993) Temperature- and humidity-dependent longevity of unfed adult *Hyalomma truncatum* (Acari: Ixodidae). *J. Med. Entomol.* **27**, 945–954.

Yuval, B. and Spielman, A. (1990) Duration and regulation of the developmental cycle of *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **27**, 196–201.

3

Competence of Ticks as Vectors of Microbial Agents with an Emphasis on *Borrelia burgdorferi*

ROBERT S. LANE

1. INTRODUCTION

Following the epochal discovery that the causative agent of Texas cattle fever is transmitted by *Boophilus* ticks (Smith and Kilbourne, 1893), the subsequent pioneering works of Dr. Howard T. Ricketts on the transmission of Rocky Mountain spotted fever laid the foundation for ensuing vector competence studies of tick-borne microbial agents. In several carefully designed and well-controlled studies, Ricketts (1906a, 1906b, 1907a, 1907b, 1909) demonstrated that the Rocky Mountain wood tick, *Dermacentor andersoni* (reported as *Dermacentor occidentalis*), is a major vector of the agent of Rocky Mountain spotted fever (unknown to Ricketts) in the northwestern United States. He proved experimentally that *D. andersoni* could transmit the spotted fever agent to susceptible laboratory and local animals by the bite, and that infected ticks occur in nature. Some of the susceptible local rodents were infested abundantly by subadults of the vector tick. Ricketts also established that the agent was disseminated in the tissues of infected ticks, and that it was passed transstadially and transmitted transovarially within populations of ticks.

Thus, over 80 years ago Ricketts fulfilled all four basic criteria that have been used to implicate the vector(s) of an arthropod-borne disease agent (DeFoliart et al., 1987). These four criteria are: (1) isolation of the agent from naturally infected vectors; (2) experimental demonstration that the vector can acquire infection by feeding on an infected host; (3) demonstration that experimentally infected vectors can transmit the agent while feeding; and (4) evidence that the suspected vector feeds on suspected vertebrate hosts under natural conditions.

Vector competence refers to the ability of arthropods to acquire, maintain, and transmit microbial agents. Species of arthropods that exhibit high vector competence for a particular agent readily acquire it while feeding on an infected vertebrate host, maintain it in their tissues, and transmit it efficiently while feeding on a susceptible host. Arthropods that are incompetent vectors may acquire the agent as they feed on an infected host but are incapable

of maintaining it in their tissues or transmitting it to another host. Although the term has come into wide use recently (DeFoliart et al., 1987), various authors differ somewhat as to the criteria for vector competence or effectiveness (e.g., Harwood, 1981; Matuschka and Spielman, 1986; DeFoliart et al., 1987; Hardy, 1988; Mather and Mather, 1990). On the other hand, the related and more inclusive term, vectorial capacity, comprises vector competence plus any behavioral or environmental factors that may influence the spread of a pathogen by the vector (Matuschka and Spielman, 1986).

Irrespective of one's definition, vector competence should always be viewed in the context of the interactive effects of populations of three groups of organisms, i.e., the host, the agent, and the vector (Harwood, 1981). Reservoir competence and vector competence are mutually dependent, though they are sometimes treated as independent phenomena in the literature. To complicate matters further for the vector ecologist, characteristics of different isolates of a microbial agent, such as genetically determined infectivity, may significantly affect the reservoir and vector competence of its associated vertebrates and arthropods, respectively. Therefore, to elucidate the enzootiology of an arthropod-borne zoonosis in a particular biome or landscape, all studies of vector or reservoir competence should be conducted with locally derived populations of vertebrates, agents, and vectors as much as possible.

The vector competence of arthropods is influenced by both intrinsic and extrinsic factors. The purpose of this chapter is to evaluate certain intrinsic and extrinsic factors, including various modes of transmission, that affect the vector competence of ticks for microbial disease agents. Emphasis is placed on the Lyme disease spirochete, *Borrelia burgdorferi*, which is by far the most important tick-borne agent afflicting people globally. In the United States, for instance, the Lyme disease spirochete currently accounts for approximately two-thirds of all arthropod-borne diseases reported to the United States Centers for Disease Control. In 1989 and 1990 alone, a provisional total of $>16{,}500$ cases of Lyme disease was reported by 46 states (D.T. Dennis, personal communication). Most of the literature search for this chapter was completed by late 1991, though some pertinent references were added subsequently. One notable recent development is that Oliver et al. (1993) synonymized *Ixodes dammini* with *Ixodes scapularis*. I use the former specific epithet *I. dammini* below only to avoid confusion since this chapter was completed long before publication of Oliver et al. (1993).

2. INTRINSIC FACTORS AFFECTING VECTOR COMPETENCE

Intrinsic vector competence includes internal physiological factors that govern infection of a vector and its ability to transmit an agent, as well as innate behavioral traits (DeFoliart et al., 1987). Several intrinsic factors affecting the vector effectiveness of ticks are presented in Table 3.1.

To secure a blood-meal, subadult and adult ticks must first locate a suitable

Table 3.1. Some intrinsic and extrinsic factors affecting the competence of ticks as vectors of microbial agents

Intrinsic factors	Extrinsic factors
Host-seeking and related behaviors (e.g., host preference, seasonality of tick-feeding by different life stages, degree of tick–host contact)	Host-population phenomena (abundance, home range, seasonality, diel activity patterns)
Duration of attachment	Susceptibility of preferred hosts to the agent
Enhanced tick-feeding success due to presence of other microbes in host blood or to pharmacologically active substances in tick saliva	Host immunity ("allergic klendusity")
Transstadial passage	Genetically determined variation in the infectivity of the agent
Transovarial transmission	Interference phenomenon (e.g., competition between microbes within tick tissues)
Genetically determined physiologic conditions	Environmental conditions (e.g., climate)

vertebrate host. Although a detailed assessment of behavioral factors that precede and contribute to the acquisition of a blood-meal by ticks is beyond the scope of this chapter, a few of them are discussed briefly below with regard to variables that enhance or attenuate vector competence.

In the case of the Lyme disease spirochete, the proportion of vector ticks that acquire infection is positively correlated with the duration of attachment. Most larvae of *Ixodes dammini*, the primary vector of *B. burgdorferi* in the northeastern and upper midwestern United States, ingest spirochetes within 2 days of attachment to infectious hamsters (Nakayama and Spielman, 1989; Piesman, 1991). Only 36% of uninfected *I. dammini* that were attached for 24 hours became infected, whereas 68% of ticks attached for 48 hours became infected (Piesman, 1991). Some larval ticks removed forcibly from infected hamsters 18 hours post-attachment reattached successfully to rodents, fed to repletion, molted, and were capable of transmitting spirochetes while feeding as nymphs (Piesman, 1991). Since attached ticks may detach from naturally infested vertebrates that die or while they feed on immune hosts, questing infected larvae caught in nature cannot be considered a priori to have acquired their infections transovarially (Piesman, 1991).

The feeding success and survival of a vector tick, and hence its ability to subsequently transmit an agent transstadially, may be augmented by infection in the vertebrate host (Randolph, 1991). The presence of the protozoan *Babesia microti* in the blood-meal of larval *Ixodes trianguliceps* resulted in a small increase in the engorged weight and a significant increase in the survival of this tick, particularly when larvae fed on naive hosts later in the parasitemic cycle.

Pharmacologically active substances present in the saliva of vector ticks

also may promote feeding success and, concomitantly, the transmission of parasites (Ribeiro et al., 1985; Ribeiro, 1987, 1989). The saliva of *I. dammini* contains salivary apyrase, a kininase, an anaphylatoxin destroying activity, an anti-complement, and vasodilatory prostaglandins that enhance feeding (Ribeiro et al., 1985; Ribeiro, 1987). These substances can be viewed as anti-edema components that prevent natural hosts, like the white-footed mouse, from rejecting immature *I. dammini* (Ribeiro, 1989). Furthermore, the saliva of this tick has immunosuppressive properties that may aid the transmission of *B. burgdorferi* and *Babesia microti* by depressing neutrophil function (Ribeiro et al., 1985).

Following acquisition of an infectious blood-meal, ticks may or may not be able to maintain a microbial agent in their tissues and pass it transstadially. However, ticks are far more efficient than insects in maintaining viruses, rickettsiae, bacteria, and protozoa in their bodies. In ticks, ectodermal derivatives and some muscle groups undergo histolysis but only the salivary gland alveoli are completely replaced during molting (Hoogstraal, 1980). Because most internal tissues change slowly during the lifetime of a tick, transstadial survival of most microorganisms occurs commonly and is one of several factors contributing to the high vector potential of many species of ticks. In contrast, the extensive internal changes that occur during molting of holometabolous insects (e.g., fleas) seem to have a deleterious effect on most microorganisms that cause human disease (Hoogstraal, 1980).

Transstadial development, as well as transovarial transmission, of many tick-associated bacteria, rickettsiae, and viruses were reviewed by Burgdorfer and Varma (1967). Recent experimental investigations of the vector competence of ticks for *B. burgdorferi* demonstrated that most tick species that feed on infected hosts can acquire the spirochete to varying degrees, but only certain ticks in the genus *Ixodes* pass the agent transstadially (e.g., Burgdorfer, 1984; Levine et al., 1985; Aeschlimann et al., 1986; Burgdorfer and Gage, 1986; Stanek et al., 1986; Burgess and Patrican, 1987; Donahue et al., 1987; Lane and Burgdorfer, 1987; Piesman et al., 1987a, b; Piesman, 1988, 1989, 1991; Piesman and Sinsky, 1988; Telford and Spielman, 1989; Mather and Mather, 1990; Gern et al., 1991; Piesman and Stone, 1991). Field evidence, in contrast to laboratory studies, suggests that certain populations of *Amblyomma americanum* can pass *B. burgdorferi* transstadially and transovarially (Schulze et al., 1986).

Transstadial passage of *B. burgdorferi* in *Ixodes pacificus* ticks having generalized tissue infections was 100% efficient among all F1 progeny (larva to nymph to adult) following transovarial transmission (Lane and Burgdorfer, 1987). Efficient and prolonged transstadial survival of *B. burgdorferi* also has been reported in *I. dammini* nymphs that acquired spirochetes as larvae (Piesman, 1989). Twenty-five unfed nymphs were examined 36–52 weeks after the date they dropped off a hamster as replete larvae; all 25 nymphs were found to contain spirochetes. Transstadial passage of *B. burgdorferi* also has been proven for *Ixodes ricinus* (Aeschlimann et al., 1986; Krampitz, 1986; Stanek et al., 1986), *Ixodes scapularis* (Burgdorfer and Gage, 1986), *Ixodes dentatus* (Telford and Spielman, 1989), *Ixodes hexagonus* (Gern et al., 1991), and *Ixodes*

neotomae (Brown and Lane, 1992), but the efficiency of this process for maintaining spirochetes in these ticks requires further study.

Another intrinsic factor is transovarial transmission, the passage of a microbial agent via the eggs of an infected arthropod from one generation to the next. Transovarial transmission yields two infection rates which may be unrelated to each other: (1) the transovarial infection rate, the percentage of females that pass microorganisms to their progeny; and (2) the filial infection rate, the percentage of infected progeny derived from an infected female (Burgdorfer and Varma, 1967). This phenomenon reportedly occurs with variable efficiency among many tick-associated agents under laboratory conditions, but its efficiency for maintaining and distributing such agents in nature remains to be determined in most cases.

According to Fine (1981), no evidence exists that any infectious disease agent of animals or plants can be maintained indefinitely in an insect vector by vertical (=transovarial) transmission alone. However, some species of tick-borne borreliae and spotted fever group rickettsiae may be passed ovarially for several generations without apparent detrimental effects to them or to their vectors (Burgdorfer and Varma, 1967). The SAWTOOTH female-2 strain of *Rickettsia rickettsii* was maintained through 12 generations in *D. andersoni* and still produced 100% transovarial and filial infection rates (Burgdorfer and Brinton, 1975). Commencing with the fifth filial generation, however, continued transovarial passage seems to have an adverse effect on the reproductive processes of *D. andersoni.* Increasing percentages of female ticks died within 1–2 weeks after repletion, and surviving females laid only about one-third to half as many eggs as non-infected females.

Similar findings were obtained for the American dog tick, *Dermacentor variabilis*, following infection with the same strain and two other strains of *R. rickettsii* (Burgdorfer and Brinton, 1975). It would be of interest to know whether less virulent strains of spotted fever group rickettsiae, such as *Rickettsia rhipicephali*, can be maintained in perpetuity in their vector ticks solely by transovarial and transstadial passage. Laboratory studies of continuous transovarial passage of another avirulent rickettsia of the spotted fever group, *R. montana*, in *D. andersoni* revealed that rickettsial infections decreased in intensity over time and resulted in fewer ticks passing rickettsiae via eggs (Burgdorfer and Brinton, 1975).

Repeated ovarial passage of *Borrelia duttonii* in its vector tick, *Ornithodoros moubata*, led to a total loss of the spirochetes' pathogencity by the fifth filial generation (Geigy and Aeschlimann, 1964). A similar phenomenon may account for the occurrence in ticks of spotted fever group rickettsiae that are apparently non-pathogenic for humans and other animals (Burgdorfer, 1981).

Transovarial transmission, and other aspects of the relationship of *B. burgdorferi* to its arthropod vectors, was the subject of a workshop held at the IVth International Conference on Lyme Borreliosis (Burgdorfer et al., 1991). It was concluded that, although transovarial transmission has been documented for *I. dammini* (Bosler et al., 1983; Piesman et al., 1986; Magnarelli et al., 1987; Burgdorfer et al., 1988), *I. pacificus* (Lane and Burgdorfer, 1987), and

I. scapularis (Magnarelli et al., 1986) in North America, and for *I. ricinus* in Europe (Burgdorfer et al., 1983; Stanek et al., 1986), it does not represent an important mechanism for maintaining spirochete-infected ticks in nature.

Furthermore, none of the larval *I. dammini* infected via the eggs of four experimentally infected females passed spirochetes transstadially (Burgdorfer et al., 1988). F_1 filial infection rates among these four females ranged from 7% to 100% for the eggs, 2.5% to 40% for the larvae, and 0% for the nymphs. Thus, it appears that horizontal (transstadial) passage of *B. burgdorferi* is an efficient mechanism for maintaining spirochetes in populations of *I. dammini* when larval ticks acquire spirochetes by feeding on infective vertebrate hosts (see above), but not when larvae obtain spirochetes ovarially. The latter phenomenon may be related to the number of spirochetes passed ovarially; if too few are passed via the eggs, then permanent infections cannot be established among F_1 filial ticks (Burgdorfer et al., 1988). There also is evidence that eggs heavily infected with *B. burgdorferi* fail to develop (Burgdorfer et al., 1989).

The efficiency of transovarial transmission for maintaining *B. burgdorferi* in other ticks that have been found infected naturally also should be investigated. A field study in New Jersey provided evidence that transovarial passage of *B. burgdorferi* occurs in some populations of the lone star tick, *Amblyomma americanum* (Schulze et al., 1986). Five (15.6%) of 32 clusters of questing *A. americanum* larvae collected from investigators' clothing were found to be infected with the Lyme disease spirochete. Also, transovarial passage of a spirochete that reacted with *Borrelia* (genus)-specific monoclonal antibodies, but not with *B. burgdorferi*-specific monoclonal antibodies, has been observed in the rabbit tick, *Haemaphysalis leporispalustris*, from California (Lane and Burgdorfer, 1988). Two-thirds of the F_1 (larval) progeny of a female tick removed from a black-tailed jackrabbit contained spirochetes.

Several behavioral factors that affect the capacity of vectors to transmit microorganisms include their host preferences, abundance on different hosts (=vector–host contact), and seasonality of biting activities. These principles can be illustrated by a comparison of the enzootiology of *B. burgdorferi* in the northeastern and far-western United States (reviewed by Lane et al., 1991). In the Northeast, many surveys have demonstrated that the white-footed mouse, *Peromyscus leucopus*, a highly competent reservoir of *B. burgdorferi*, is the most commonly captured and heavily parasitized host of immature *I. dammini*. Furthermore, nymphal *I. dammini* feed on *P. leucopus* before the larvae do in each transmission season, which serves to amplify the prevalence of infection in vector ticks and to increase the risk for human infection (Spielman et al., 1985; Matuschka and Spielman, 1986). Thus, it is not surprising that spirochetal infection rates in questing nymphal and adult *I. dammini* are high, averaging roughly 25% and 50%, respectively, in enzootic areas (Lane et al., 1991).

On the other hand, an expanding body of field and laboratory evidence has implicated the dusky-footed woodrat, *Neotoma fuscipes*, as a potent reservoir host of *B. burgdorferi* in northwestern California (Lane and Brown, 1991; Brown and Lane, 1992). The Lyme disease spirochete is maintained, at least in part, in an enzootic cycle involving woodrats and two of several tick

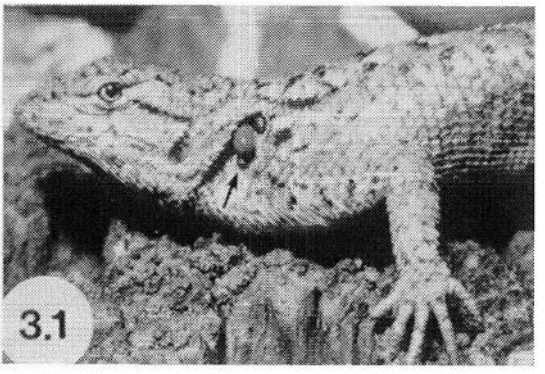

Fig. 3.1. Lateral view of a western fence lizard (*Sceloporus occidentalis*) showing immatures of the western black-legged tick (*Ixodes pacificus*) (arrow) attached to the lateral nuchal pocket. Reprinted with permission from the Entomological Society of America.

species that parasitize it, i.e., *Ixodes neotomae* and *I. pacificus* (Brown and Lane, 1992). Although it is not a member of the *I. ricinus* complex, *I. neotomae* seems to be a more efficient enzootic vector of *B. burgdorferi* than *I. pacificus* (Brown and Lane, 1992). *Ixodes neotomae* feeds primarily on rodents and lagomorphs but does not bite people. In contrast, *I. pacificus* infests approximately 80 species of reptiles, birds, and mammals including humans (Arthur and Snow, 1968; Keirans and Clifford, 1978; Furman and Loomis, 1984). Circumstantial and epidemiologic evidence have implicated *I. pacificus* as the primary vector of *B. burgdorferi* to humans in this region (Naversen and Gardner, 1978; Burgdorfer et al., 1985; Lane and Lavoie, 1988; Lane, 1990a).

In parts of northern California, immatures of *I. pacificus* feed abundantly on the western fence lizard, *Sceloporus occidentalis* (Fig. 3.1), with the peak of larval feeding activities preceding that of the nymphs by several weeks (Lane and Loye, 1989; Lane, 1990b). Indeed, in some areas coinhabited by lizards, birds, rodents, lagomorphs, and deer, the prevalence and intensity of feeding by immature *I. pacificus* on lizards in spring is an order of magnitude (or more) greater than it is on these other groups of vertebrates (Westrom et al., 1985; Lane and Burgdorfer, 1988; Lane, 1990b, c; Lane and Loye, 1989, 1991; Manweiler et al., 1990). Although *I. pacificus* larvae take, on average, about 2.4–3 times longer to feed to repletion on *S. occidentalis* than they do on sylvatic rodents under laboratory conditions (i.e., ~12 days versus ~4–5 days), lizards are still far more significant hosts of *I. pacificus* in certain habitats because of their sheer abundance and heavy tick burdens.

Further, the reservoir competence of *S. occidentalis* for *B. burgdorferi* is low (Lane, 1990b), and the vector competence of *I. pacificus* for *B. burgdorferi* is not particularly high and may be lower than that of the enzootic vector *I. neotomae* (Brown and Lane, 1992). Considered together, these biological phenomena may explain the low spirochetal infection rates reported for nymphal and adult *I. pacificus* (Lane et al., 1991).

Intrinsic physiological factors that are genetically determined may influence the vector competence of ticks also. Piesman (1989) speculated that growth-promoting factors may be present in *Ixodes* vectors of *B. burgdorferi* but are absent in other tick genera or, conversely, growth-inhibiting factors may occur in other tick genera that are lacking in *Ixodes* spp. In fact, some unidentified factor produced during molting by *I. dammini* appears to have a strong borrelicidal action (Piesman et al., 1990). *Borrelia burgdorferi* multiplied rapidly in replete larval and nymphal *I. dammini*, reaching respective mean densities of 2,735 and 61,275 spirochetes/tick on days 15 and 75 post-repletion. Recently molted nymphs and adults, however, contained 10-fold fewer spirochetes. The breakdown and reformation of the tick's exoskeleton during molting may be connected in some way to the observed decrease in the abundance of spirochetes (Piesman et al., 1990).

It has been postulated that the ability of soft ticks to acquire, maintain, and transmit their own spirochetes is genetically determined (Burgdorfer, 1981). In North America, vector specificity apparently exists for at least four species of *Borrelia*. The soft ticks *Ornithodoros coriaceus*, *O. hermsi*, *O. parkeri*, and *O. turicata* transmit spirochetes that are specific to them, i.e., *Borrelia coriaceae*, *B. hermsii*, *B. parkeri*, and *B. turicatae*, respectively. A recent claim that *B. hermsii*, *B. parkeri*, and *B. turicatae* are conspecific because of their similar DNA homologies (Hyde and Johnson, 1984; Johnson et al., 1984a) awaits confirmation or refutation following additional molecular biological studies. In this regard, the writer has recently obtained seven fresh borrelial isolates from *O. parkeri* ticks collected in Monterey County, California. Antigenic and genetic characterizations of these isolates, and comparison with strains of *B. hermsii* and *B. turicatae*, should help to resolve this controversy.

Finally, geographically divergent and genetically heterogeneous populations of a tick species may vary in their intrinsic competence to acquire and transmit a specific microbial agent. Although several laboratory experiments involving *A. americanum* demonstrated that it is an incompetent vector of the Lyme disease spirochete (Piesman, 1988; Piesman and Sinsky, 1988; Burgdorfer, 1989a; Mather and Mather, 1990), field-derived data indicate that it may be a competent vector of this spirochete in some parts of the United States. Spirochete-infected *A. americanum* unfed or replete adults, subadults, or both have been collected in Alabama, Missouri, New Jersey, North Carolina, Texas and Virginia (Magnarelli et al., 1986; Rawlings, 1986; Schulze et al., 1986; Feir and Reppell, 1990; Levine et al., 1991; Luckhart et al., 1991; Teltow et al., 1991). In New Jersey, for instance, 15.6% of 32 clusters of larvae, 3.4% of 289 nymphs, and 5.4% of 467 adults of *A. americanum*, all collected in an unfed state from investigators' clothing, were found to contain spirochetes (Schulze et al., 1986). These findings establish that transstadial and transovarial passage of *B. burgdorferi* can occur in some populations of this tick.

Recent studies of the ceratopogonid fly, *Culicoides variipennis*, the principal vector of bluetongue virus in the United States, underscore the need to elucidate the genetic and environmental factors that affect the vector competence/capacity of insects and acarines (Tabachnick, 1992). A single genetic locus was

found to control the oral susceptibility of *C. variipennis sonorensis* to infection with this virus, which offers researchers the opportunity to discover genetic markers to characterize natural populations of this vector fly. The effect of the environment on vector genotypes, once understood, can be used to predict the environmental and genetic conditions under which vector populations are hazardous (Tabachnick, 1992).

3. EXTRINSIC FACTORS AFFECTING VECTOR COMPETENCE

Numerous extrinsic factors, such as vertebrate host density and climatic conditions, affect vector competence. Vertebrates introduce other extrinsic factors including dispersal, seasonal breeding patterns, attractiveness to vectors, response to infection, activity patterns, and immune status (DeFoliart et al., 1987). The microbial agent represents another extrinsic factor that is often overlooked, i.e., variation in the infectivity of different populations of the agent for diverse populations of vectors and vertebrate hosts.

Several factors extrinsic to ticks that affect their effectiveness as vectors are listed in Table 3.1. These include various host-related factors, genetic attributes of microorganisms, intra-tick competition between microorganisms or between a micro- and a macroorganism ("interference phenomenon"), and biotic and abiotic environmental variables (e.g., climatic and edaphic factors).

The susceptibility of the preferred hosts of a tick to a microbial agent can affect its vector competence. As mentioned above, the western fence lizard is a major, if not the preferred, host of immature *I. pacificus* in some endemic foci for Lyme disease, but it is an incompetent reservoir of *B. burgdorferi.* Abundant feeding by a vector tick on a reservoir-incompetent host serves a natural zooprophylactic function by diverting potentially infected ticks from reservoir-competent hosts (Spielman et al., 1985). Therefore, zooprophylaxis lessens the effectiveness of an otherwise competent vector while simultaneously reducing the relative risk of transmission of a zoonotic disease agent to humans.

The immune status of the host may interfere with the ability of ticks to acquire or transmit microorganisms, as reviewed recently by Brown (1988a) and Jones and Nuttall (1988) (much of the ensuing discussion is based on information contained in these reviews). After having been fed upon by ixodid ticks, a host may acquire resistance to further tick infestation that involves complement dependent cellular and antibody-mediated effector mechanisms. Ticks that feed on resistant vertebrates suffer increased mortality while feeding, reduced engorgement weights, and retarded development. Moreover, vertebrates manifest an innate grooming response when ticks feed on them that is enhanced by previously acquired resistance to tick feeding. Brown (1988a) showed experimentally that the grooming response of guinea pigs following infestation by *Rhipicephalus appendiculatus* had a significant negative impact on the feeding success of this tick. For instance, tick mortality as a result of grooming by naive guinea pigs or by ticks dropping from the host was 56% (Brown, 1988a).

Host-acquired resistance to vector ticks may interfere with the transmission of animal disease agents, apparently by impairing the tick's engorgement process (Francis and Little, 1964; Campbell, 1978; Bell et al., 1979; Wikel, 1980; Brown, 1988a; Jones and Nuttall, 1988). The protective effect of host hypersensitivity against transmission of tick-borne agents has been demonstrated for the protozoan *Babesia argentina*, the bacterium, *Francisella tularensis*, and two viruses, Thogoto and Tick-Borne Encephalitis. Thus, *Dermacentor variabilis* ticks infected with the agent of tularemia, *F. tularensis*, frequently failed to infect New Zealand white rabbits that had been exposed previously to feeding by the same tick species (Bell et al., 1979), a phenomenon these authors refer to as "allergic klendusity." Transmission was reduced substantially in three of four controlled experiments leading to the conclusion that allergic klendusity must have considerable impact on the prevalence of infection of tick-borne tularemia in nature (Bell et al., 1979).

In some host–parasite systems, however, infection of the host (e.g., rabbits with the protozoan *Trypanosoma congolense*) may lessen their acquired immune response to ticks (*Rhipicephalus appendiculatus*) and thereby actually promote tick-feeding success (Heller-Haupt et al., 1983).

Just as different populations of vector ticks or vertebrate hosts belonging to a single species can vary strikingly in their susceptibilities to microorganisms, distinct populations (isolates, strains) of a specific microbial agent can vary markedly as to their infectivity or pathogenicity. In studies of vector or reservoir competence, the triad of vector, host, and agent should be characterized as to geographic origin and, whenever possible, the agent also should be distinguished antigenically and genetically. Also, one cannot assume that data gleaned from laboratory studies can be extrapolated to the field or, for that matter, to other populations of the vector, host, or agent in the absence of confirmatory studies.

To illustrate how the agent itself may affect investigations of vector/reservoir competence, recent studies in northern California have shown that there can be significant phenotypic variation among local isolates of the Lyme disease spirochete. The protein profiles (as determined by SDS-PAGE) of several tick-derived isolates of *B. burgdorferi* from discrete natural foci differed conspicuously (Lane and Pascocello, 1989). Such isolates also were found to vary in their infectivity for the deer mouse, *Peromyscus maniculatus*, a natural host of *B. burgdorferi* (Lane, 1990c; Lane and Loye, 1991; C.A. Peavey and R.S. Lane, unpublished data). Additionally, the infectivity of isolates of *B. burgdorferi* for vertebrates may be lost as a result of continuous cultivation in artificial medium (Schwan et al., 1988). Therefore, we are now using more than one strain of *B. burgdorferi* in most experimental studies of vector or reservoir competence to take into account the variation observed among different spirochetal isolates. In so doing, we are trying to obtain a more realistic picture of what may be occurring in nature than would be possible if our conclusions were based solely on the behavior of a single strain of *B. burgdorferi* in its vector ticks/vertebrate hosts.

Competition between microorganisms, or between a micro- and a macroorganism, within tick tissues could be categorized either as an intrinsic or

extrinsic factor. I chose to classify the "interference phenomenon" as an extrinsic factor because the competing organisms typically originate apart from the tick, i.e. they are acquired while ticks feed, or as a result of the direct attacks of hymenopterous parasites on ticks. In contrast, tick symbionts, which are passed transstadially and ovarially from one generation of ticks to the next, could be considered to be an intrinsic factor.

A classic example of the interference phenomenon is that of a non-pathogenic rickettsia of the spotted fever group (the so-called "East side agent") that limits the distribution of *Rickettsia rickettsii* in *Dermacentor andersoni* ticks on the east side of the Bitterroot Valley in western Montana (Burgdorfer et al., 1981). This rickettsia resembles *R. rickettsii* ultrastructurally, but it is non-pathogenic for guinea pigs and meadow voles. The East side agent is present in several tissues including the ovaries of *D. andersoni* on both the east and west sides of the Valley, but its prevalence (up to 80%) is much higher on the east side. When *D. andersoni* ticks are infected with the East side agent, virulent *R. rickettsii* cannot become established in the ovarian tissues and therefore transovarial infection is prevented from occurring. Similar findings were obtained in experiments involving two non-virulent serotypes of spotted fever group rickettsiae (*Rickettsia montana*, *R. rhipicephali*) in opposition to virulent *R. rickettsii* (Burgdorfer et al., 1981).

Interference in ticks may provide a logical explanation as to why virulent strains of *R. rickettsii* are seldom or never encountered in some localities besides the Bitterroot Valley (Burgdorfer et al., 1981). In foci of Rocky Mountain spotted fever in the central and north coastal regions of California, the prevalence of *R. rickettsii*-like strains may be quite low in populations of the Pacific Coast tick, *Dermacentor occidentalis*, that have a high prevalence of infection with *R. rhipicephali* (Philip et al., 1981). Thus, 77 (96%) of 80 rickettsial isolates obtained from 737 adult *D. occidentalis* that were collected in Mendocino (Hopland area) and Monterey (Hastings Natural History Reservation) counties were serotyped as *R. rhipicephali* and the remaining three isolates as *R. rickettsii*-like strains (=unclassified 364D isolates).

Interference between arboviruses has been reported for Thogoto virus in one of its ixodid vector ticks, *Rhipicephalus appendiculatus* (Davies et al., 1989). Thogoto virus, which is related to the influenza viruses (family Orthomyxoviridae), infects a wide range of mammalian species including humans. The wild-type virus was unable to replicate in nymphal ticks that had been exposed orally as larvae to a temperature-sensitive mutant of the same virus. Interference was observed inter- and intrastadially, was complete in 78% of dually infected nymphs, and prevented transmission of the virus to hamsters.

Another interesting case in which the effectiveness of a vector tick was reduced substantially by the competing interference of another organism involves the wasp *Hunterellus hookeri*, and the agents of Lyme disease and babesiosis on Naushon Island, Massachusetts (Mather et al., 1987). *Hunterellus hookeri* parasitizes and, in the process, eventually kills nearly one-third of questing nymphal *I dammini* ticks on the island. In sites where wasps are present, the prevalence of both agents in *I. dammini* is about 40% less than they

are in wasp-free sites. Lyme disease spirochetes were not detected in wasp-infected ticks and babesial parasites infrequently parasitized such ticks. However, the presence of one agent in a tick significantly increased the likelihood of infection with the other agent. The authors concluded that the occurrence of *H. hookeri* on Naushon Island may have reduced the risk of human infection with both pathogens by as much as a third.

Finally, few studies of the macroecological and microecological environmental factors (e.g., climatic and edaphic conditions) that limit the distribution of ticks and tick-borne disease agents have been undertaken, but the requirement for high humidity by members of the *I. ricinus* complex prevents them from becoming established in more arid regions (Lane et al., 1991). Contradistinctively, transmission of Crimean–Congo hemorrhagic fever (CCHF) virus in Senegal, West Africa, is greatest in the northern arid, sparsely vegetated, bioclimatic zone where *Hyalomma* ticks abound, and least in the southern, moister, forested zone where such ticks are less abundant (Wilson et al., 1990). Bioclimatic zones not only influence the distribution and abundance of the tick vectors of CCHF virus, but climate also affects the presence and abundance of potential reservoir hosts that may influence horizontal transmission of the virus (Wilson et al., 1990).

4. TRANSMISSION OF MICROBIAL AGENTS BY TICKS

Ticks are capable of transmitting microbes by several different routes including salivary secretions (e.g., Colorado tick fever virus, *B. burgdorferi*, spotted fever group rickettsiae), coxal fluid (some species of relapsing fever borreliae), regurgitation (e.g., *Cowdria reminantium* and possibly *B. burgdorferi*), and feces (*Coxiella burnetii*). Some agents may be transmitted by more than one route by their vector ticks, though only one route may be of enzootic or epidemiologic significance. *Borrelia coriaceae*, the putative agents of Epizootic Bovine Abortion in the far-western United States (Lane et al., 1985; Johnson et al., 1987), is transmitted by the bite of the soft tick *Ornithodoros coriaceus*. This spirochete also is voided in coxal fluid secretions, but only after the tick has fed to repletion, detached, and fallen from the host (Lane and Manweiler, 1988). Therefore, transmission of *B. coriaceae* via coxal fluid secretions is highly improbable.

Tick transmission of viruses may be practically instantaneous if high viral concentrations are present in salivary gland tissues. Although the organotropism(s) of Colorado tick fever virus in its primary vector, *Dermacentor andersoni*, is (are) not known, mere piercing of the host's skin by infected ticks results in prompt transmission (Burgdorfer, 1977). This finding suggests that the salivary glands become heavily infected with the virus, whereas the absence of transovarial transmission of Colorado tick fever virus in *D. andersoni* suggests that the virus may not invade the germinal cells of the ovaries (Eklund et al., 1962; Burgdorfer, 1977).

In contrast, the etiologic agent of Rocky Mountain spotted fever, *R. rickettsii*, which produces a generalized infection in the tissues of *D. andersoni*

once the tick's gut barrier has been overcome, is usually not transmitted to a vertebrate host until the attached tick has been feeding for at least 10 hours (Spencer and Parker, 1923). The delay in transmission is due to the fact that *R. rickettsii* seems to be in an avirulent state in unfed ticks; the rickettsiae become virulent only after prolonged attachment of the tick to its host or following ingestion of blood by ticks (Spencer and Parker, 1923). The reactivation phenomenon has been shown to be correlated with reversible structural modifications of the microcapsular and slime layers of *R. rickettsii* that are linked to physiological changes in the tick host (Hayes and Burgdorfer, 1982).

Although transmission of *R. rickettsii* to humans normally occurs through the bite of an infected ixodid tick, transmission may occur occasionally by contamination of abraded or even intact skin with fresh tick feces, by crushing infected ticks between fingers while manually deticking dogs, or by handling rickettsial infected or tick-infested wildlife (Burgdorfer, 1975, 1977).

Irrespective of the mode(s) of transmission of *B. burgdorferi* (see below), experimental studies have established that the efficacy of spirochete transmission is associated closely with the duration of attachment by nymphal and adult *I. dammini*. Nymphs of *I. dammini* transmitted *B. burgdorferi* to one of 14 rodents exposed for 24 hours, five of 14 rodents exposed for 48 hours, and 13 of 14 rodents exposed for $\geqslant 72$ hours (Piesman et al., 1987b). Similarly, females of *I. dammini* transmitted *B. burgdorferi* to none of eight rabbits exposed for 24–36 hours, two of three rabbits exposed for 48 hours, and five of five rabbits exposed for >120 hours (Piesman et al. 1991).

Transmission of *B. burgdorferi* by its various arthropod vectors was reviewed by Piesman (1989), who concluded that members of the *Ixodes* (*Ixodes*) *ricinus* complex are competent vectors of the Lyme disease spirochete while species belonging to other tick genera are inefficient vectors. It has been proposed that transmission of *B. burgdorferi* by *I. dammini* occurs via saliva following prolonged attachment (Benach et al., 1987; Ribeiro et al., 1987; Zung et al., 1989) or by regurgitation of tick-midgut contents (Burgdorfer, 1984, 1989b).

In unfed *I. dammini* ticks infected with *B. burgdorferi*, the distribution of spirochetes is usually restricted (i.e., in $>95\%$ of adult ticks) to the midgut diverticula where the organism accumulates near the microvillar brush border and in the interstitial spaces between epithelial cells (Burgdorfer et al., 1982, 1988). After attachment to hosts, spirochetes present in some ticks penetrate the gut wall and basal lamina, enter the hemocoel, invade various tissues, and are present in saliva within about 3 (nymphs) or 4 days (adults) (Benach et al., 1987; Ribeiro et al., 1987; Zung et al., 1989). These findings, along with similar ones for the related Eurasian vector, *I. ricinus* (Gern et al., 1990), provide strong support for the salivary route of transmission of *B. burgdorferi*.

Alternatively, recent experimental evidence demonstrates that infection of salivary glands in adult *I. dammini* is not a prerequisite for successful transmission of *B. burgdorferi* to New Zealand white rabbits (Burgdorfer, 1989b). Four rabbits became infected with *B. burgdorferi* after each of them had been fed upon by four female *I. dammini* that were subsequently determined to have had

midgut-restricted spirochetal infections. Perhaps some strains of *B. burgdorferi* have an organotropism for midgut tissues and as such are transmitted solely by regurgitation, whereas other strains having more generalized tissue tropisms are transmitted mainly or exclusively via saliva. To test this hypothesis, researchers would have to repeat some of the earlier experimental studies using a single (hence, a standardized) colony of ticks and well-characterized strains of *B. burgdorferi.*

Regurgitation of midgut contents as a possible means of transmission of tick-borne microbial agents is not a new concept (Cowdry, 1925). Several different experimental methods have been used to demonstrate that both argasid (soft) and ixodid (hard) ticks, such as the East African relapsing fever tick, *Ornithodoros moubata*, the lone star tick, *Amblyomma americanum*, and the castor bean or sheep tick, *I. ricinus*, are capable of regurgitating gut material while feeding (Hesse, 1981; Brown, 1988b; M. Brossard in Burgdorfer, 1989b; Connat, 1991). A radiolabeling technique utilizing tritiated ecdysteroid showed that *O. moubata* could regurgitate 1.3% of its midgut contents, and defecate an equivalent amount of radiolabel during and after the blood-meal (Connat, 1991). These findings suggest that the spirochete *Borrelia duttonii*, in addition to being transmitted via the saliva or coxal fluid of *O. moubata* ticks (Burgdorfer, 1951), also may be transmitted by regurgitation. Transmission of *B. duttonii* by infected tick feces seems far less likely because borreliae perish quickly when subjected to desiccative conditions.

5. COMPETENCE OF TICKS AS VECTORS OF *BORRELIA BURGDORFERI*: A SUMMARY

The absence of vector specificity of *B. burgdorferi* is unique among the tick-borne borreliae. Whereas the vector specificity of most of the ~21 described species of tick-borne borreliae is high, i.e., they are usually associated with one or at most a few tick species (Johnson et al., 1984b, 1987; Kelly, 1984; Baranton et al., 1992), *B. burgdorferi* or closely related spirochetes have been isolated from or detected in at least 23 species of ticks in two families (Argasidae, 2 spp.; Ixodidae, 21 spp.). This total includes 12 species of *Ixodes*, four species of *Dermacentor*, two species each of *Amblyomma* and *Haemaphysalis*, and one species each of *Argas*, *Ornithodoros*, and *Rhipicephalus*. Fourteen of these ticks were mentioned in a recent review (Burgdorfer et al., 1991); nine species that can be added to this list are *Ixodes acuminatus* (Doby et al., 1990), *Ixodes cookei* (Levine et al., 1991; Magnarelli and Swihart, 1991), *Ixodes ovatus* (Miyamoto et al., 1992), *Ixodes trianguliceps* (Doby et al., 1990), *Dermacentor albipictus* (Magnarelli et al., 1986), *Dermacentor occidentalis* (Lane and Lavoie, 1988), *Amblyomma maculatum* (Teltow et al., 1991), *Haemaphysalis bispinosa* (Zhang et al., 1990), and *Ornithodoros coriaceus* (Lane and Manweiler, 1988).

The vector competence of only 11 of these 23 tick species for *B. burgdorferi* has been investigated so far (Table 3.2). Four members of the *Ixodes ricinus* complex (*dammini*, *pacificus*, *ricinus*, *scapularis*), and three *Ixodes* ticks which

Table 3.2. Vector competence or incompetence of ticks for the Lyme disease spirochete, *Borrelia burgdorferi.* See text for pertinent references

Tick species	Acquisition during feeding	Transstadial passage	Transmission	
			By the bite	Ovarially
Ixodes dammini	+	+	+	+
I. dentatus	+	+	+	?
I. hexagonus	+	+	+	+
I. holocyclus	+	–	NA	NA
I. neotomae	+	+	+	?
I. pacificus	+	+	+	+
I. ricinus	+	+	+	+
I. scapularis	+	+	+	+
Amblyomma americanum	+	–[a]	NA	NA[a]
Dermacentor occidentalis	+	+/–	?	?
D. variabilis	+	—	NA	NA

[a] Field evidence, however, strongly suggests that both transstadial and transovarial transmission of *B. burgdorferi* occurs in *A. americanum* from New Jersey (Schulze et al., 1986).
NA, not applicable.

do not belong to this complex, *I. dentatus*, *I. hexagonus*, and *I. neotomae*, have been shown to be efficient vectors of the Lyme disease spirochete (see review by Burgdorfer et al., 1991; Gern et al., 1991; Brown and Lane, 1992). In contrast, field and laboratory studies indicate that at least some populations of *A. americanum*, *D. occidentalis* and *D. variabilis* are inefficient vectors of *B. burgdorferi* (Piesman, 1988; Piesman and Sinsky, 1988; Burgdorfer, 1989a; Mather and Mather, 1990; Lindsay et al., 1991; Brown and Lane, 1992).

Additionally, the Australian paralysis tick *Ixodes holocyclus* is an incompetent experimental vector of the JD1 strain of *B. burgdorferi* from North America (Piesman and Stone, 1991). Thus, among the eight *Ixodes* spp. whose vector competence has been studied to date, only *I. holocyclus* has been demonstrated to be an incompetent vector of *B. burgdorferi.* However, *I. holocyclus* may still be an efficient vector of Australian strains of *B. burgdorferi* or related spirochetes (Piesman and Stone, 1991).

The vector competence of other ticks that are either found to be infected naturally with spirochetes or associated circumstantially with human cases should be evaluated to determine what ticks are serving as primary or secondary vectors of *B. burgdorferi* in each geographic region. During epidemiologic studies, priority should be given to determining the vector effectiveness of abundant human-biting ticks, particularly those species that are found to contain spirochetes while host seeking. Tick species that are occasionally infected while feeding on their natural hosts may or may not be efficient vectors. Almost any ixodid tick species that imbibes a sufficient quantity of blood from a spirochetemic host may acquire spirochetes, but only competent vectors can maintain, pass transstadially, and transmit such spirochetes later by the bite.

6. CONCLUSIONS

Vector competence refers to the capacity of arthropods to acquire, maintain, and transmit microbial agents. A variety of intrinsic and extrinsic factors that affect the vector competence of argasid (soft) and ixodid (hard) ticks for microbial agents are discussed, with an emphasis on the relation of the Lyme disease spirochete, *Borrelia burgdorferi*, to its ixodid vector ticks. It cannot be overemphasized that vector competence and vertebrate reservoir competence are mutually dependent phenomena. Furthermore, vector competence must be viewed in the context of the interactive effects of populations of three groups of organisms, i.e., the vector(s), the agent(s), and the reservoir host(s). Host immunity to ticks and competition between microorganisms within tick tissues, for example, may abolish the potential of an otherwise competent vector to acquire or to transmit a microbial agent.

Competent vector ticks feed abundantly on vertebrate reservoir hosts at a time of year when the reservoirs are in an infective state, readily acquire and preserve the microbial agent in their tissues for prolonged periods, efficiently pass the agent transstadially (larva to nymph to adult), and effectively transmit the agent while feeding later on susceptible vertebrates. The degree of competence may vary even within populations of an efficient vector inasmuch as not all infected ticks, even those that are heavily infected with an agent, are capable of transmitting it. Transovarial transmission, the passage of a microbial agent from an infected female via eggs to her progeny, may augment the vector competence of some ticks. Repeated transovarial passage of some agents, however, may have an inimical effect on the reproductive capacity of their vectors (e.g., virulent strains of *Rickettsia rickettsii* in *D. andersoni* ticks) or reduce, or even eliminate, the pathogenicity of an agent (e.g., *Borrelia duttonii* in *Ornithodoros moubata* ticks), if the agent is not rejuvenated periodically by passage through a susceptible vertebrate host.

The vector competence of only ten (*Amblyomma*, one; *Dermacentor*, two; *Ixodes*, seven) of the ~23 species of ticks that have been found infected naturally with the Lyme disease spirochete has been evaluated so far. Of these, all seven *Ixodes* spp. have been determined to be efficient vectors of *B. burgdorferi*, whereas three species of *Amblyomma* or *Dermacentor* appear to be inefficient vectors. Additionally, the Australian paralysis tick, *I. holocyclus*, was found to be an incompetent vector of a North American isolate of the spirochete. These findings suggest that, in general, *Ixodes* ticks may possess physiological properties that are conducive to the growth and multiplication of Lyme disease spirochetes, while ticks in other genera may possess physiologic attributes that are less propitious, if not inhospitable, to *B. burgdorferi*. Vector ecologists are urged, whenever possible, to employ locally derived ticks and spirochetes in future studies of vector competence since genetic differences between diverse populations of vectors, agents, or both may account for some of the discrepant results reported in the literature.

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8. REFERENCES

Aeschlimann, A., Chamot, E., Gigon, F., Jeanneret, J.-P., Kesseler, D., and Walther, C. (1986) *B. burgdorferi* in Switzerland. *Zentralbl. Bakteriol. Mikrobiol. Hyg. A* **263**, 450.

Arthur, D.R. and Snow, K.R. (1968) *Ixodes pacificus* Cooley and Kohls, 1943: its life-history and occurrence. *Parasitol.* **58**, 893.

Baranton, G., Postic, D., Saint Girons, I., Boerlin, P., Piffaretti, J.-C., Assous, M., and Grimont, P.A.D. (1992) Delineation of *Borrelia burgdorferi* sensu stricto, *Borrelia garinii* sp. nov., and group VS461 associated with Lyme borreliosis. *Internat. J. Syst. Bacteriol.* **42**, 378.

Bell, J.F., Stewart, S.J., and Wikel, S.K. (1979) Resistance to tick-borne *Francisella tularensis* by tick-sensitized rabbits: allergic klendusity. *Am. J. Trop. Med. Hyg.* **28**, 876.

Benach, J.L., Coleman, J.L., Skinner, R.A., and Bosler, E.M. (1987) Adult *Ixodes dammini* on rabbits: a hypothesis for the development and transmission of *Borrelia burgdorferi*. *J. Infect. Dis.* **155**, 1300.

Bosler, E.M., Coleman, J.L., Benach, J.L., Massey, D.A., Hanrahan, J.P., Burgdorfer, W., and Barbour, A.G. (1983) Natural distribution of the *Ixodes dammini* spirochete. *Science* **220**, 321.

Brown, R.N. and Lane, R.S. (1992) Lyme disease in California: a novel enzootic transmission cycle of *Borrelia burgdorferi*. *Science* **256**, 1439.

Brown, S.J. (1988a). Evidence for regurgitation by *Amblyomma americanum*. *Vet. Parasitol.* **28**, 335.

Brown, S.J. (1988b) Vertebrate immune-mediated responses to arthropod feeding and their potential effects on pathogen transmission. *Misc. Publ. Entomol. Soc. Am.* **68**, 37.

Burgdorfer, W. (1951) Analyse des Infektionsverlaufes bei *Ornithodorus moubata* (Murray) und der natürlichen Uebertragung von *Spirochaeta duttoni*. *Acta Trop.* **8**, 193.

Burgdorfer, W. (1975) A review of Rocky Mountain spotted fever (tick-borne typhus), its agent, and its tick vectors in the United States. *J. Med. Entomol.* **12**, 269.

Burgdorfer, W. (1977) Tick-borne diseases in the United States: Rocky Mountain spotted fever and Colorado tick fever. *Acta Trop.* **34**, 103.

Burgdorfer, W. (1981) The arthropod vector as a selective transport system of animal and human pathogens. pp. 44–46. *In:* McKelvey, J.J., Jr, Eldridge, B.F. and Maramorosch, K. (eds) *Vectors of Disease Agents: Interactions with Plants, Animals, and Man.* Praeger Publishers, New York.

Burgdorfer, W. (1984) Discovery of the Lyme disease spirochete and its relation to tick vectors. *Yale J. Biol. Med.* **57**, 515.

Burgdorfer, W. (1989a) Vector/host relationships of the Lyme disease spirochete, *Borrelia burgdorferi* Vol. 15, pp. 775–787. *In:* Johnson, R.C. (ed) *Rheumatic Disease Clinics of North America.* Saunders, Philadelphia.

Burgdorfer, W. (1989b) *Borrelia burgdorferi*: its relationship to tick vectors. pp. 8–13.

In: Stanek, G., Flamm, H., Barbour, A.G. and Burgdorfer, W. (eds) *Lyme borreliosis II.* Zbl. Bakt. Suppl. 18. Gustav Fischer Verlag, Stuttgart, New York.

Burgdorfer, W. and Brinton, L.P. (1975) Mechanisms of transovarial infection of spotted fever rickettsiae in ticks. *Ann. New York Acad. Sci.* **266**, 61.

Burgdorfer, W. and Gage, K.L. (1986) Susceptibility of the black-legged tick, *Ixodes scapularis*, to the Lyme disease spirochete, *Borrelia burgdorferi. Zentralbl. Bakteriol. Mikrobiol. Hyg. A* **263**, 15.

Burgdorfer, W. and Varma, M.G.R. (1967) Trans-stadial and transovarial development of disease agents in arthropods. *Annu. Rev. Entomol.* **12**, 347.

Burgdorfer, W., Hayes, S.F., and Mavros, A.J. (1981) Nonpathogenic rickettsiae in *Dermacentor andersoni*: a limiting factor for the distribution of *Rickettsia rickettsii.* pp. 585–594. *In:* Burgdorfer, W. and Anacker, R.L. (eds) *Rickettsiae and Rickettsial Diseases.* Academic Press, New York.

Burgdorfer, W., Barbour, A.G., Hayes, S.F., Benach, J.L., Grunwaldt, E., and Davis, J.P. (1982) Lyme disease—a tick-borne spirochetosis? *Science* **216**, 1317.

Burgdorfer, W., Barbour, A.G., Hayes, S.F., Peter, O., and Aeschlimann, A. (1983) Erythema chronicum migrans—a tickborne spirochetosis. *Acta Trop.* **40**, 79.

Burgdorfer, W., Lane, R.S., Barbour, A.G., Gresbrink, R.A., and Anderson, J.R. (1985) The western black-legged tick, *Ixodes pacificus*: a vector of *Borrelia burgdorferi. Am. J. Trop. Med. Hyg.* **34**, 925.

Burgdorfer, W., Hayes, S.F., and Benach, J.L. (1988) Development of *Borrelia burgdorferi* in ixodid tick vectors. *Ann. New York Acad. Sci.* **539**, 172.

Burgdorfer, W., Hayes, S.F., and Corwin, D. (1989) Pathophysiology of the Lyme disease spirochete, *Borrelia burgdorferi*, in ixodid ticks. *Rev. Infect. Dis.* **11**, S1442.

Burgdorfer, W., Anderson, J.F., Gern, L., Lane, R.S., Piesman, J. and Spielman, A. (1991) Relationship of *Borrelia burgdorferi* to its arthropod vectors. *Scand. J. Infect. Dis.* (Suppl. 77), 35.

Burgess, E.C. and Patrican, L.A. (1987) Oral infection of *Peromyscus maniculatus* with *Borrelia burgdorferi* and subsequent transmission by *Ixodes dammini. Am. J. Trop. Med. Hyg.* **36**, 402.

Campbell, R.S.F. (1978) The use of resistant cattle in the control of ticks and vector-borne diseases. pp. 251–257. *In:* Wilde, J.K.H. (ed) *Tick-borne Diseases and Their Vectors.* Proceedings of an International Conference, Centre for Tropical Veterinary Medicine, University of Edinburgh, Scotland..

Connat, J.-L. (1991) Demonstration of regurgitation of gut content during blood meals of the tick *Ornithodoros moubata. Parasitol. Res.* **77**, 452.

Cowdry, E.V. (1925). Studies on the etiology of heartwater. II. *Rickettsia ruminantium* (n. sp.) in the tissue of ticks transmitting the disease. *J. Exp. Med.* **42**, 253.

Davies, C.R., Jones, L.D., and Nuttall, P.A. (1989) Viral interference in the tick, *Rhipicephalus appendiculatus.* I. Interference to oral superinfection by Thogoto virus. *J. Gen. Virol.* **70**, 2461.

DeFoliart, G.R., Grimstad, P.R., and Watts, D.M. (1987) Advances in mosquito-borne arbovirus/vector research. *Annu. Rev. Entomol.* **32**, 479.

Doby, J.M., Bigaignon, G., Launay, H., Costil, C., and Lorvellec, O. (1990) Présence de *Borrelia burgdorferi*, agent de spirochétoses à tiques, chez *Ixodes* (*Exopalpiger*) *trianguliceps* Birula, 1895 et *Ixodes* (*Ixodes*) *acuminatus* Neumann, 1901 (Acariens *Ixodidae*) et chez *Ctenophthalmus baeticus arvernus* Jordan, 1931 et *Megabothris turbidus* (Rothschild, 1909) (Insectes *Siphonaptera*), ectoparasites de micromammifères des forêts dans l'Ouest de la France. *Bull. Soc. Francaise Parasitol.* **8**, 311.

Donahue, J.G., Piesman, J., and Spielman, A. (1987) Reservoir competence of white-footed mice for Lyme disease spirochetes. *Am. J. Trop. Med. Hyg.* **36**, 92.

Eklund, C.M., Kohls, G.M., and Kennedy, R.C. (1962) Lack of evidence of transovarial transmission of Colorado tick fever virus in *Dermacentor andersoni. Proc. Symp. Biology of Viruses of the Tick-borne Encephalitis Complex*, Smolenice, Czechoslovak Academy of Science, p. 401.

Feir, D. and Reppell, C. (1990) Transmission of *Borrelia burgdorferi* in the state of Missouri, USA. *IV Int. Conf. Lyme Borreliosis.* Book B Abstracts, p. 47.

Fine, P.E.M. (1981) Epidemiological principles of vector-mediated transmission. pp. 77–91. *In:* McKelvey, J.J. Jr., Eldridge, B.F., and Maramorosch, K. (eds) *Vectors of Disease Agents: Interactions with Plants, Animals, and Man.* Praeger Publishers, New York.

Francis, J., and Little, D.A. (1964) Resistance of droughtmaster cattle to tick infestation and babesiosis. *Austral. Vet. J.* **40**, 247.

Furman, D.P., and Loomis, E.C. (1984) The ticks of California (Acari: Ixodida). *Bull. Calif. Insect Surv.* **25**, 1.

Geigy, V.R. and Aeschlimann, A. (1964) Langfristige Beobachtungen über transovarielle Übertragung von *Borrelia duttoni* durch *Ornithodorus moubata. Acta Trop.* **21**, 87.

Gern, L., Zhu, Z., and Aeschlimann, A. (1990) Development of *Borrelia burgdorferi* in *Ixodes ricinus* females during blood feeding. *Ann. Parasitol. Hum. Comp.* **65**, 89.

Gern, L., Toutoungi, L.N., Hu, C.M., and Aeschlimann, A. (1991) *Ixodes* (*Pholeoixodes*) *hexagonus*, an efficient vector of *Borrelia burgdorferi* in the laboratory. *Med. Vet. Entomol.* **5**, 431.

Hardy, J.L. (1988) Susceptibility and resistance of vector mosquitoes. Vol. I, pp. 87–126. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology.* CRC Press, Boca Raton, FL.

Harwood, R.F. (1981) Criteria for vector effectiveness. pp. 5–12. *In:* McKelvey, J.J., Jr, Eldridge, B.F. and Maramorosch, K. (eds) *Vectors of Disease Agents: Interactions with Plants, Animals, and Man.* Praeger Publishers, New York.

Hayes, S.F. and Burgdorfer, W. (1982) Reactivation of *Rickettsia rickettsii* in *Dermacentor andersoni* ticks: an ultrastructural analysis. *Infect. Immun.* **37**, 779.

Heller-Haupt, A., Varma, M.G.R., Langi, A.O., and Zetlin, A. (1983) The effect of *Trypanosoma congolense* infection on acquired immunity to the tick *Rhipicephalus appendiculatus. Ann. Trop. Med. Parasitol.* **77**, 219.

Hesse, G.H. (1981) Die Regurgitation bei Arthropoden und ihre Bedeutung in Zusammenhang mit der Ubertragung von Krankheitserregern. Thesis, University of Bonn (not seen, in Connat, 1991).

Hoogstraal, H. (1980). The roles of fleas and ticks in the epidemiology of human diseases. pp. 241–244. *In:* Traub, R. and Starcke, H. (eds) *Proc. Int. Conf. Fleas.* A.A. Balkema, Rotterdam.

Hyde, F.W. and Johnson, R.C. (1984) Genetic relationship of Lyme disease spirochetes to *Borrelia*, *Treponema*, and *Leptospira* spp. *J. Clin. Microbiol.* **20**, 151.

Johnson, R.C., Hyde, F.W., and Rumpel, C.M. (1984a) Taxonomy of the Lyme disease spirochetes. *Yale J. Biol. Med.* **57**, 529.

Johnson, R.C., Schmid, G.P., Hyde, F.W., Steigerwalt, A.G., and Brenner, D.J. (1984b) *Borrelia burgdorferi* sp. nov.: etiologic agent of Lyme disease. *Internat. J. Syst. Bacteriol.* **34**, 496.

Johnson, R.C., Burgdorfer, W., Lane, R.S., Barbour, A.G., Hayes, S.F., and Hyde, F.W.

(1987) *Borrelia coriaceae* sp. nov.: putative agent of epizootic bovine abortion. *Internat. J. Syst. Bacteriol.* **37**, 72.

Jones, L.D. and Nuttall, P.A. (1988) The effect of host immunity on the transmission of tick-borne viruses. *Anim. Technol.* **39**, 161.

Keirans, J.E. and Clifford, C.M. (1978) The genus *Ixodes* in the United States: a scanning electron microscope study and key to the adults. *J. Med. Entomol. Suppl.* **2**, 1.

Kelly, R.T. (1984) Genus IV. *Borrelia* Swellengrebel 1907, 582. Vol. 1, pp. 57–62. *In:* Krieg, N.R. (ed) *Bergey's Manual of Systematic Bacteriology.* Williams & Wilkins, Baltimore.

Krampitz, H.E. (1986) In vivo isolation and maintenance of some wild strains of European hard tick spirochetes in mammalian and arthropod hosts. *Zentralbl. Bakteriol. Mikrobiol. Hyg. A* **263**, 21.

Lane, R.S. (1990a) Seasonal activity of two human-biting ticks. *Calif. Agric.* 44 (March–April), 23.

Lane, R.S. (1990b) Susceptibility of the western fence lizard (*Sceloporus occidentalis*) to the Lyme borreliosis spirochete (*Borrelia burgdorferi*). *Am. J. Trop. Med. Hyg.* **42**, 75.

Lane, R.S. (1990c) Infection of deer mice and piñon mice (*Peromyscus* spp.) with spirochetes at a focus of Lyme borreliosis in northern California, U.S.A. *Bull. Soc. Vector Ecol.* **15**, 25.

Lane, R.S. and Brown, R.N. (1991) Wood rats and kangaroo rats: potential reservoirs of the Lyme disease spirochete in California. *J. Med. Entomol.* **28**, 299.

Lane, R.S. and Burgdorfer, W. (1987) Transovarial and transstadial passage of *Borrelia burgdorferi* in the western black-legged tick, *Ixodes pacificus* (Acari: Ixodidae). *Am. J. Trop. Med. Hyg.* **37**, 188.

Lane, R.S. and Burgdorfer, W. (1988) Spirochetes in mammals and ticks (Acari: Ixodidae) from a focus of Lyme borreliosis in California. *J. Wildl. Dis.* **24**, 1.

Lane, R.S. and Lavoie, P.E. (1988) Lyme borreliosis in California: acarological, clinical, and epidemiological studies. *Ann. New York Acad. Sci.* **539**, 192.

Lane, R.S. and Loye, J.E. (1989) Lyme disease in California: interrelationship of *Ixodes pacificus* (Acari: Ixodidae), the western fence lizard (*Sceloporus occidentalis*), and *Borrelia burgdorferi*. *J. Med. Entomol.* **26**, 272.

Lane, R.S. and Loye, J.E. (1991) Lyme disease in California: interrelationship of ixodid ticks (Acari), rodents, and *Borrelia burgdorferi J. Med. Entomol.* **28**, 719.

Lane, R.S. and Manweiler, S.A. (1988) Borrelia coriaceae in its tick vector, *Ornithodoros coriaceus* (Acari: Argasidae), with emphasis on transstadial and transovarial infection. *J. Med. Entomol.* **25**, 172.

Lane, R.S. and Pascocello, J.A. (1989) Antigenic characteristics of *Borrelia burgdorferi* isolates from ixodid ticks in California. *J. Clin. Microbiol.* **27**, 2344.

Lane, R.S., Burgdorfer, W., Hayes, S.F., and Barbour, A.G. (1985) Isolation of a spirochete from the soft tick, *Ornithodoros coriaceus*: a possible agent of epizootic bovine abortion. *Science* **230**, 85.

Lane, R.S., Piesman, J., and Burgdorfer, W. (1991) Lyme borreliosis: relation of its causative agent to its vectors and hosts in North America and Europe. *Annu. Rev. Entomol.* **36**, 587.

Levine, J.F., Wilson, M.L., and Spielman, A. (1985) Mice as reservoirs of the Lyme disease spirochete. *Am. J. Trop. Med. Hyg.* **34**, 355.

Levine, J.F., Sonenshine, D.E., Nicholson, W.L., and Turner, R.T. (1991) *Borrelia burgdorferi* in ticks (Acari: Ixodidae) from coastal Virginia. *J. Med. Entomol.* **28**, 668.

Lindsay, L.R., Barker, I.K., Surgeoner, G.A., McEwen, S.A., Elliott, L.A., and Kolar, J. (1991) Apparent incompetence of *Dermacentor variabilis* (Acari: Ixodidae) and fleas (Insecta: Siphonaptera) as vectors of *Borrelia burgdorferi* in an *Ixodes dammini* endemic area of Ontario, Canada, *J. Med. Entomol.* **28**, 750.

Luckhart, S., Mullen, G.R., and Wright, J.C. (1991) Etiologic agent of Lyme disease, *Borrelia burgdorferi*, detected in ticks (Acari: Ixodidae) collected at a focus in Alabama. *J. Med. Entomol.* **28**, 652.

Magnarelli, L.A. and Swihart, R.K. (1991) Spotted fever group rickettsiae or *Borrelia burgdorferi* in *Ixodes cookei* (Ixodidae) in Connecticut. *J. Clin. Microbiol.* **29**, 1520.

Magnarelli, L.A., Anderson, J.F., Apperson, C.S., Fish, D., Johnson, R.C., and Chappell, W.A. (1986) Spirochetes in ticks and antibodies to *Borrelia burgdorferi* in white-tailed deer from Connecticut, New York State, and North Carolina. *J. Wildl. Dis.* **22**, 178.

Magnarelli, L.A., Anderson, J.F., and Fish, D. (1987) Transovarial transmission of *Borrelia burgdorferi* in *Ixodes dammini* (Acari: Ixodidae). *J. Infect. Dis.* **156**, 234.

Manweiler, S.A., Lane, R.S., Block, W.M., and Morrison, M.L. (1990) Survey of birds and lizards for ixodid ticks (Acari) and spirochetal infection in northern California. *J. Med. Entomol.* **27**, 1011.

Mather, T.N. and Mather, M.E. (1990) Intrinsic competence of three ixodid ticks (Acari) as vectors of the Lyme disease spirochete. *J. Med. Entomol.* **27**, 646.

Mather, T.N., Piesman, J., and Spielman, A. (1987) Absence of spirochaetes (*Borrelia burgdorferi*) and piroplasms (*Babesia microti*) in deer ticks (*Ixodes dammini*) parasitized by chalcid wasps (*Hunterellus hookeri*). *Med. Vet. Entomol.* **1**, 3.

Matuschka, F.R. and Spielman, A. (1986) The emergence of Lyme disease in a changing environment in North America and central Europe. *Exp. Appl. Acarol.* **2**, 337.

Miyamoto, K., Nakao, M., Uchikawa, K., and Fujita, H. (1992) Prevalence of Lyme borreliosis spirochetes in ixodid ticks of Japan, with special reference to a new potential vector, *Ixodes ovatus* (Acari: Ixodidae). *J. Med. Entomol.* **29**, 216.

Nakayama, Y. and Spielman, A. (1989) Ingestion of Lyme disease spirochetes by ticks feeding on infected hosts. *J. Infect. Dis.* **160**, 166.

Naversen, D.N. and Gardner, L.W. (1978) Erythema chronicum migrans in America. *Arch. Dermatol.* **114**, 253.

Oliver, J.H., Jr, Owsley, M.R., Hutcheson, H.J., James, A.M., Chen, C., Irby, W.S., Dotson, E.M., and McLain, D.K. (1993) Conspecificity of the ticks *Ixodes scapularis* and *I. dammini* (Acari: Ixodidae). *J. Med. Entomol.* **30**, 54.

Philip, R.N., Lane, R.S., and Casper, E.A. (1981) Serotypes of tick-borne spotted fever group rickettsiae from western California. *Am. J. Trop. Med. Hyg.* **30**, 722.

Piesman, J. (1988) Vector competence of ticks in the southeastern United States for *Borrelia burgdorferi*. *Ann. New York Acad. Sci.* **539**, 417.

Piesman, J. (1989) Transmission of Lyme disease spirochetes (*Borrelia burgdorferi*). *Exp. Appl. Acarol.* **7**, 71.

Piesman, J. (1991) Experimental acquisition of the Lyme disease spirochete, *Borrelia burgdorferi*, by larval *Ixodes dammini* (Acari: Ixodidae) during partial blood meals. *J. Med. Entomol.* **28**, 259.

Piesman, J. and Sinsky, R.G. (1988) Ability of *Ixodes scapularis*, *Dermacentor variabilis*, and *Amblyomma americanum* (Acari: Ixodidae) to acquire, maintain, and transmit Lyme disease spirochetes (*Borrelia burgdorferi*). *J. Med. Entomol.* **25**, 336.

Piesman, J. and Stone, B.F. (1991) Vector competence of the Australian paralysis tick,

Ixodes holocyclus, for the Lyme disease spirochete *Borrelia burgdorferi*. *Internat. J. Parasitol.* **21**, 109.

Piesman, J., Donahue, J.G., Mather, T.N., and Spielman, A. (1986) Transovarially acquired Lyme disease spirochetes (*Borrelia burgdorferi*) in field-collected larval *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **23**, 219.

Piesman, J., Hicks, T.C., Sinsky, R.J., and Obiri, G. (1987a) Simultaneous transmission of *Borrelia burgdorferi* and *Babesia microti* by individual nymphal *Ixodes dammini* ticks. *J. Clin. Microbiol.* **25**, 2012.

Piesman, J., Mather, T.N., Sinsky, R.J., and Spielman, A. (1987b) Duration of tick attachment and *Borrelia burgdorferi* transmission. *J. Clin. Micrbiol.* **25**, 557.

Piesman, J., Oliver, J.R., and Sinsky, R.J. (1990) Growth kinetics of the Lyme disease spirochete (*Borrelia burgdorferi*) in vector ticks (*Ixodes dammini*). *Am. J. Trop. Med. Hyg.* **42**, 352.

Piesman, J., Maupin, G.O., Campos, E.G., and Happ, C.M. (1991) Duration of adult female *Ixodes dammini* attachment and transmission of *Borrelia burgdorferi*, with description of a needle aspiration isolation method. *J. Infect. Dis.* **163**, 895.

Randolph, S.E. (1991) The effect of *Babesia microti* on feeding and survival in its tick vector, *Ixodes trianguliceps*. *Parasitol.* **102**, 9.

Rawlings, J.A. (1986) Lyme disease in Texas. *Zentralbl. Bakteriol. Mikrobiol. Hyg. A* **263**, 483.

Ribeiro, J.M.C. (1987) Role of saliva in blood-feeding by arthropods. *Annu. Rev. Entomol.* **32**, 463.

Ribeiro, J.M.C. (1989) Role of saliva in tick/host interactions. *Exp. Appl. Acarol.* **7**, 15.

Ribeiro, J.M.C., Makoul, G.T., Levine, J., Robinson, D.R., and Spielman, A. (1985) Antihemostatic, antiinflammatory, and immunosuppressive properties of the saliva of a tick, *Ixodes dammini*. *J. Exp. Med.* **161**, 332.

Ribeiro, J.M.C., Mather, T.N., Piesman, J., and Spielman, A. (1987) Dissemination and salivary delivery of Lyme disease spirochetes in vector ticks (Acari: Ixodidae). *J. Med. Entomol.* **24**, 201.

Ricketts, H.T. (1906a) The study of "Rocky Mountain Fever" (Tick Fever?) by means of animal inoculations. *JAMA* **47**, 33.

Ricketts, H.T. (1906b) The transmission of Rocky Mountain spotted fever by the bite of the wood-tick (*Dermacentor occidentalis*). *JAMA* **47**, 358.

Ricketts, H.T. (1907a) The role of the wood-tick (*Dermacentor occidentalis*) in Rocky Mountain spotted fever, and the susceptibility of local animals to this disease—a preliminary report. *JAMA* **49**, 24.

Ricketts, H.T. (1907b) Further experiments with the wood-tick in relation to Rocky Mountain spotted fever. *JAMA* **49**, 1278.

Ricketts, H.T. (1909) Some aspects of Rocky Mountain spotted fever as shown by recent investigations. *Med. Rec.* **76**, 845.

Schulze, T.L., Lakat, M.F., Parkin, W.E., Shisler, J.K., Charette, D.J., and Bosler, E.M. (1986) Comparison of rates of infection by the Lyme disease spirochete in selected populations of *Ixodes dammini* and *Amblyomma americanum* (Acari: Ixodidae). *Zentralbl. Bakteriol. Mikrobiol. Hyg. A* **263**, 72.

Schwan, T.G., Burgdorfer, W., and Garon, C.F. (1988) Changes in infectivity and plasmid profile of the Lyme disease spirochete, *Borrelia burgdorferi*, as a result of in vitro cultivation. *Infect. Immun.* **56**, 1831.

Smith, T. and Kilborne, F.L. (1893) Investigations into the nature, causation, and prevention of southern cattle fever. *USDA Bur. Animal Ind. Bull.* no. 1, 301 pp.

Spencer, R.R. and Parker, R.R. (1923) Rocky Mountain spotted fever: infectivity of fasting and recently fed ticks. *Public Health Rep.* **38**, 333.

Spielman, A., Wilson, M.L., Levine, J.F., and Piesman, J. (1985) Ecology of *Ixodes dammini*-borne human babesiois and Lyme disease. *Annu. Rev. Entomol.* **30**, 439.

Stanek, G., Burger, I., Hirschl, A., Wewalka, G., and Radda, A. (1986) *Borrelia* transfer by ticks during their life cycle. *Zentralbl. Bakteriol. Mikrobiol. Hyg. A* **263**, 29.

Tabachnick, W.J. (1992) The genetics, population genetics, and evolution of insect vectors of disease: *Culicoides variipennis* and bluetongue virus transmission in the U.S. and its international impact. pp. 263–270. *In:* Walton, T.E. and Osburn, B.I. (eds) *Bluetongue, African Horse Sickness and Related Orbiviruses.* CRC Press, Boca Raton, FL.

Telford, S.R., III and Spielman, A. (1989) Competence of a rabbit-feeding *Ixodes* (Acari: Ixodidae) as a vector of the Lyme disease spirochete. *J. Med. Entomol.* **26**, 118.

Teltow, G.J., Fournier, P.V., and Rawlings, J.A. (1991) Isolation of *Borrelia burgdorferi* from arthropods collected in Texas. *Am. J. Trop. Med. Hyg.* **44**, 469.

Westrom, D.R., Lane, R.S., and Anderson, J.R. (1985) *Ixodes pacificus* (Acari: Ixodidae): population dynamics and distribution on Columbian black-tailed deer (*Odocoileus hemionus columbianus*). *J. Med. Entomol.* **22**, 507.

Wikel, S.K. (1980) Host resistance to tick-borne pathogens by virtue of resistance to tick infestation. *Ann. Trop. Med. Parasitol.* **74**, 103.

Wilson, M.L., LeGuenno, B., Guillaud, M., Desoutter, D., Gonzalez, J.-P., and Camicas, J.-L. (1990) Distribution of Crimean–Congo hemorrhagic fever viral antibody in Senegal: environmental and vectorial correlates. *Am. J. Trop. Med. Hyg.* **43**, 557.

Zhang, F., Liang, W., and Zheng, Y. (1990) A strain of *Borrelia burgdorferi* firstly isolated from *Haemaphysalis bispinosa* (Neumann). *Chinese J. Epidemiol.* **11**, 23.

Zung, J.L., Lewengrub, S., Rudzinska, M.A., Spielman, A., Telford, S.R., and Piesman, J. (1989) Fine structural evidence for the penetration of the Lyme disease spirochete *Borrelia burgdorferi* through the gut and salivary tissues of *Ixodes dammini*. *Can. J. Zool.* **67**, 1737.

4

Vector–Host–Pathogen Relationships: Transmission Dynamics of Tick-borne Infections

THOMAS N. MATHER AND HOWARD S. GINSBERG

1. INTRODUCTION

Most, and perhaps all, of the tick-borne infections affecting man are zoonoses, that is, they are maintained to a large degree in natural cycles not involving humans. For the most part, we only become interested in those tick-transmitted zoonoses that affect humans or domestic animals, or that show potential for affecting man. However, if we look beyond disease severity or impact on health, it is apparent that the frequency of human infections caused by these zoonoses, or their **epidemiologic potency**, can vary considerably. Moreover, the intensity of transmission in natural cycles, or **zoonotic potency** (frequency of animal infections), may correlate directly, inversely, or not at all with a zoonosis' epidemiologic potency. Even the same zoonosis, when maintained in a different ecological setting, can vary significantly in the number of infections produced, and so, the degree of infection risk to humans. For instance, in North America, Lyme borreliosis has become perhaps the single most important of these tick-transmitted zoonoses; the number of human infections recorded far outnumbering that of most other tick-borne diseases. However, vectors of this infection are only focally abundant with a relatively limited distribution. Many tick-borne infections, such as those caused by the spotted fever group (SFG) rickettsias, produce far fewer human infections, even in areas where their tick vectors are quite abundant. The vectors of this group of infections are generally more common and have a much broader distribution. Clearly then, **transmission factors,** and not just tick abundance, play a large part in determining the potency as well as the infection risk of a particular tick-borne zoonosis. But what are the factors that determine transmission intensity, and how do they interact?

Transmission dynamics of tick-borne zoonoses are determined by factors related to host animals, tick vectors, and the pathogens they transmit. These factors include **physiological** and **ecological** processes, which act in concert to determine the intensity of zoonotic transmission as well as risk to humans and domesticated animals. The study of tick-borne zoonoses has largely gone

beyond enumerating the abundance of infected vectors in its quest to determine the best way to suppress the impact that these infestations cause. However, the diversity of interacting forces that shape the pattern of transmission for a particular zoonosis hampers efforts to characterize the epizootiology of tick-borne diseases. Although it may be obvious that infected vectors exist in nature at different densities and possessing different infection rates, thus resulting in different levels of transmission risk, the factors determining these differences are usually less apparent. Many of these factors have been identified for particular zoonoses; differential reservoir competence, variation in pathogen transmission efficiencies, and host immune mechanisms are examples. But are these transmission factors merely attributes of particular zoonoses, or can general rules about zoonoses be formed? In this chapter we discuss various factors that can influence maintenance and amplification of an infection in its zoonotic cycle, and that may determine infection risk, exclusive of human or animal exposure factors. Rather than considering factors related to vectors, hosts, or pathogens separately, we will discuss transmission processes as they interrelate. Our attempt here is to introduce topics, providing the reader with the more significant theoretical considerations related to transmission dynamics of tick-borne zoonoses which will hopefully serve as a framework when considering specific zoonoses, as presented in Part II of this volume. In particular, we will consider factors affecting host infectivity to blood-feeding ticks, and transmission efficiencies from vectors to hosts. In addition, we present a classification scheme of epizootiologic cycles and make limited predictions about the potency of tick-borne diseases based on ecological considerations.

2. TRANSMISSION FROM HOSTS TO TICKS

The zoonotic and epidemiologic patterns of tick-borne zoonoses depend essentially on the basic reproduction rate of the tick-transmitted pathogen or parasite, which as described by Macdonald (1950, 1957) is a key element in the transmission process, irrespective of the mode of pathogen transmission. The basic reproduction rate refers to the number of secondary infections generated by a single infective individual. The manifestation of the basic reproduction rate, in terms of infections in animal populations at a given location, is the potency of the zoonosis. On average if the basic reproduction rate is less than unity, then the infection is unable to maintain itself; if greater than unity then the infection is able to maintain itself and perhaps spread. Thus, the amplification process is perhaps the most critical element in determining an infecting agent's basic reproduction rate as well as a zoonosis' potency. Tick-borne infections can be amplified either **horizontally** by passing between reservoir and tick, or **vertically** through either an inherited or venerial infection. The basic reproduction rate is typically increased by large numbers of vectors biting reservoir-competent animals. Most vector-borne infections, even those transmitted vertically, usually require some degree of horizontal amplification to be maintained (Fine, 1981).

Horizontal amplification processes require that reservoir hosts pass their infection to, presumably, more than one susceptible vector. In turn, infected vectors must transmit the pathogen to non-infected, susceptible, reservoir-competent hosts. With ixodid ticks, which usually blood-feed just once in each developmental stage, a further requirement is that the immature stages (either larva or nymph) acquire an infection which then must pass transstadially into the next biting stage. An additional caveat is that the infectious vector instar must feed on the same host species as the instar acquiring the infection, which is not always the case with three-host ticks. The "narrowness of host range" exhibited by a vector species is critical to pathogen amplification and the basic reproduction rate (Spielman et al., 1984). Certainly, both zoonotic and probably epidemiologic potency are more dependent on the number or proportion of vectors infesting reservoir hosts than on the absolute abundance of vectors. Diversion of either the transmitting or acquiring instar to alternative hosts nullifies the effect of feeding by the other tick stage, weakening amplification and reducing the basic reproduction rate. In this way, effective horizontal amplification of Lyme disease spirochetes, certain of the tick-borne encephalitis (TBE)-group viruses, and rodent piroplasms principally involve the larval (as acquiring) and nymphal (as transmitting) instars feeding principally on a limited number of small mammal hosts. The adult stages of the ticks transmitting these infections rarely, if ever, bite the small mammals that serve as critical reservoirs.

2.1. Infection of the Tick Population

The first element in horizontal amplification is the infectivity of a host species. Host species are typically divided between **reproductive hosts**, those which provide a blood meal upon which the tick advances in its life cycle, and **reservoir hosts**, which serve to perpetuate both tick and infection. Only reservoir hosts are infective and the infectivity exhibited among host species for a particular infection is usually quite variable. Infectivity may also vary temporally within the same host species. We define infectivity as the proportion of ticks to become infected by blood-feeding on a host population (Mather et al., 1989). It would depend on both the infectiousness of individual hosts as well as the proportion of hosts infected. In practice, infectivity of a host population is determined by tick xenodiagnosis (Donahue et al., 1987; Mather, 1993); the number of ticks becoming infected is expressed as a percentage of the total number of ticks feeding on a population of hosts. Because actually determining infectivity for even one population of a particular host species can be quite time and labor intensive, it is important to focus efforts and resources first on the most important hosts of those ticks being considered. Nevertheless, an infectivity of 0.8 would mean that 80% of all ticks blood-feeding on all hosts in a population became infected. To be competent vectors, the ticks would still need to survive and molt, transstadially pass their infection, acquire another susceptible host, and effectively transmit the infection. Thus, infectivity is merely one component in the amplification process.

The rate of tick infestation on reservoir hosts is critical to horizontal amplification in two ways. The number of host infections depends on the level of infected ticks and number of ticks per host (see discussion below), while the number of infected ticks produced depends on the number of susceptible ticks per host and host infectivity. We consider the latter here. Competent reservoirs with high infectivity will contribute little to amplifying an infection if relatively few ticks infest them. Several ecological and physiological factors influence this host/vector contact not the least of which are vertebrate and tick behavioral patterns. If we consider that small rodents serve as principal reservoirs of many tick-borne zoonoses, then the distribution of larval ticks must overlap with suitable rodent habitat for frequent vector contact to be made. Where this overlap is marginal, amplification will be weakened, and the infection may not persist. A second ecological factor will be the frequency that rodents enter the questing environment of the tick. Tick-questing behavior can be both species and stage specific, and in some cases may depend upon macro- and microclimatic conditions, or vegetative type and cover. Thus, if larval ticks climb vegetation to seek hosts, they will less likely encounter highly infective rodents and insectivores scurrying in leaf litter on the forest floor, but may more likely encounter medium-sized hosts or larger hosts, perhaps possessing lower infectivity.

The population dynamics of vertebrate hosts and reservoirs also play an important role in maintenance and amplification of infections. Clearly, the number of susceptible hosts may influence the natural transmission cycle by determining the potential number of infective hosts. Likewise, the density of infective hosts can influence the density of infected ticks produced, which, in turn, may determine transmission intensity both in zoonotic cycles and to humans. The turnover rate within the host population may be critical where hosts recover from infection or eventually lose infectivity. The infectivity of a host population may be considerably higher where small mammals with relatively short life spans serve as principal reservoirs for infection, than where reservoirs are larger and usually longer-lived hosts with a high proportion of immune (and non-infective) individuals. On the other hand, in cycles where host species remain infective for long periods, rapid turnover would likely lower zoonotic potency since a greater proportion of blood-meals would be taken from individuals yet to become infected (Ginsberg, 1988).

Host infectivity, tick infestation rate, and host density are inextricably linked in finally determining the abundance of infected vectors. These critical parameters have been combined analytically to compare the relative contribution, or "reservoir potential" of a host species to infecting the tick population in a given area (Mather et al., 1989). Each animal species' contribution (RP) can be calculated by:

$$RP_i = I_i L_i D_i \Big/ \sum_s (I_i L_i D_i) \tag{4.1}$$

where I is the proportion of feeding ticks (acquiring instar) that become infected, L is the average infestation rate of that instar, and D is the host density.

Moreover, i represents an individual host species population and s all host species populations in a community of hosts. The result, (RP), expressed as a percentage, is the proportion of infected ticks derived from a particular host species. The equation provides a useful means of identifying critical reservoirs. It can also serve to illustrate the dynamic relationship between reservoir and reproductive hosts in determining the level of infection risk. Factors influencing the three principal reservoir potential parameters are likely to be the determinants of pathogen amplification and its basic reproduction rate.

Natural fluctuations in reservoir population cycles can profoundly influence transmission dynamics and risk. In transmission cycles of Lyme disease spirochetes as well as perhaps in other infections where small rodents serve as principal reservoirs, increases in reservoir density are followed in subsequent transmission seasons by increases in the abundance of infected vectors (Mather, 1991) and presumably, also in increases in risk of human infection. Similarly, decreases in reservoir abundance are followed in the subsequent season by a lowering of tick infections. Furthermore, such observations can be predicted by analyzing reservoir and reproductive host densities, tick infestations, and reservoir infectivity. Using the reservoir potential equation (Mather et al., 1989) to predict Lyme disease spirochete prevalence in nymphal *Ixodes dammini* over a 5-year period, it was observed that the density of white-footed mice (*Peromyscus leucopus*) in year y was directly correlated (Fig. 4.1) with tick infection prevalence in year $y + 1$ (Mather, 1991). This type of amplification might be expected in cases where the host range is narrow. In ecologic settings where two or more hosts contribute equally as reservoirs, amplification of infection may be more difficult to predict.

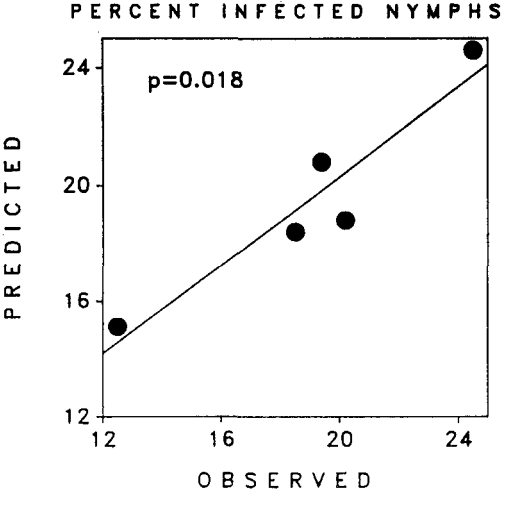

Fig. 4.1. Lyme disease spirochete prevalence in nymphal *Ixodes dammini*. Correlation between results of field observations over 5 years and those predicted using the reservoir potential equation where density of the principal reservoir host, *Peromyscus leucopus*, was the only variable.

2.2. Vector Infection Thresholds

Infection of individual ticks generally depends upon their ingestion of a threshold number of infectious units. The infection threshold will be the lowest number of infectious units needed to infect a certain proportion (say 5%) of the vector population. The higher the infection threshold, the greater the number of infectious units required to infect an individual vector, which, in turn, probably results in fewer infected vectors in a population. Thus, low infection thresholds may favor zoonotic potency, as a greater number of individual ticks are likely to become infected. Transmission thresholds vary depending on the pathogen, species of tick, and host. Pathogen virulence, co-infection, susceptibility of different tick species, and immune-modulation of infectivity by hosts may all play a role. Unfortunately, data reporting the number of infectious units required to establish a tick infection are limited.

In the eastern US, Lyme disease would appear to have both greater zoonotic and epidemiologic potency than babesiosis caused by *Babesia microti*. However, the number of infections in white-footed mice (*P. leucopus*) for Lyme disease spirochetes (*Borrelia burgdorferi*) and rodent piroplasms (*B. microti*) was the same regardless of whether the host was bitten by one, two or three infected nymphs (Mather et al., 1990). Moreover, the proportion of blood-feeding larvae that became infected after feeding on hosts bitten by either one, two or three infected ticks was similar. Thus, it would appear that the host-infecting dose had little effect on a host's eventual infectivity for either infection. In contrast, however, was the proportion of ticks that became infected with either the spirochete or piroplasm. Mouse infectivity to blood-feeding larvae for spirochetes appears to be about twice as that for *Babesia*. Furthermore, infectivity for both pathogens was somewhat reduced when ticks fed on animals co-infected with both pathogens. Differences in infectivity for these two pathogens may be due, in part, to differences in parasitemia levels (host related), tick susceptibility to infection (tick related), or some unknown virulence factor inherent in the pathogen species (pathogen related). In addition, co-infection appeared to raise the transmission threshold for both pathogens.

2.3. Duration of Infectivity

The ability of a host–vector system to maintain or amplify a zoonosis also depends on the duration or temporal presentation of pathogen infectivity in the reservoir. The duration of infectivity for various tick-borne pathogens is highly variable; for the same pathogen, it can also vary greatly between host species. Thus, animals may be infective with viral and rickettsial agents for relatively short periods of time (hours to weeks), while hosts may be infective with some of the borreliae for most of their lives. Cotton rats (*Sigmodon hispidus*) are infective for *Rickettsia rickettsii* for just 24 hours, squirrels and chipmunks may remain infective for 1–4 days, while opossums remain infective for 3–4 weeks (Boseman et al., 1967; McDade and Newhouse, 1986). In contrast,

individual white-footed mice (*P. leucopus*) may be infective for Lyme disease spirochetes (*B. burgdorferi*) over their entire lifetime (Donahue et al., 1987). However, population infectivity may still vary seasonally; in the case of Lyme borreliosis, seasonal variation in infectivity appears to be largely due to the introduction of new individuals into the reservoir population in the absence of reinfection (Tälleklint et al., 1993).

Seasonal variation in infectivity may also occur when host animals acquire immunity as a consequence of infection. Individuals usually lose infectivity, either partially or totally, through immune mechanisms related to antibody production. These same immune mechanisms may also confer protection against re-infection and re-activation of pathogen infectivity. Loss of infectivity due to recovery may occur slowly, rapidly, or not at all (although a partial loss may be seen); the former two cases may severely curtail horizontal amplification, especially where the transmitting and acquiring tick instars are active at different times of the year. Where infectivity is short-lived but tick stages overlap, amplification can still occur. Nevertheless, it would seem that hosts expressing infectivity for just short periods of time would contribute little to the basic reproduction rate. Such a horizontal amplification system may be characteristic of infections with lower potency.

Infectivity may also be influenced by episodes of alternating high and low parasitemias. Many of the tick-borne borreliae commonly exhibit such episodes. It has been suggested that periods of low and high parasitemias may be similarly reflected in the percentage of infected ticks produced from such hosts (Burgdorfer amd Schwan, 1991).

2.4. Pattern of Feeding Activity

The seasonal activity of tick feeding may be another determinant of host infectivity and the basic reproduction rate. Horizontal amplification of pathogens is often facilitated when transmission seasons are long and when the feeding activity of the infecting instar precedes that of the acquiring instar. This is especially true where the duration of infectivity is long. Such an inverted pattern of activity is usually seen in ticks requiring 2 or more years to complete their life cycle. For instance, in *I. dammini*, the activity of nymphs precedes that of larvae in any given year (Piesman and Spielman, 1979; Main et al., 1982). Larvae acquire Lyme spirochetes and rodent piroplasms (*Babesia microti*) after feeding on reservoirs infected by nymphs earlier in the summer. Nymphs derived from these larvae then carry their infection into the next transmission season. In contrast, peak larval activity of *I. pacificus*, vector of Lyme spirochetes in the western US, precedes that of nymphs (Lane, 1990), while the three stages of other *Ixodes* vectors, such as *I. ricinus* in northern Europe and *I. scapularis* in the southern US, occur nearly simultaneously (Lane et al., 1991). The absence of an inverted seasonal pattern of activity in populations of these other vectors may serve to weaken spirochete amplification and thereby reduce infection risk.

Feeding success of a tick population is critical for its maintenance but

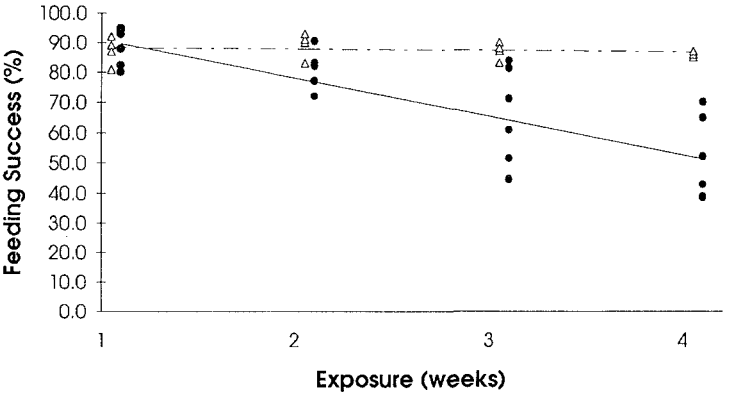

Fig. 4.2. Feeding success of larval *Ixodes dammini* on white-footed mice (*Peromyscus leucopus*) repeatedly infested with either 100 (△) or 300 (●) larvae weekly for 4 weeks.

success of the pathogen-acquiring instar is critical to the basic reproduction rate. Hosts generally exhibit some degree of innate and acquired immune-mediated resistance to tick feeding which can interfere with feeding success. These host responses often involve changes in grooming or defensive behavior associated with potent cutaneous cellular reactions at feeding sites. Acquired hypersensitivity involves antibody-sensitized mast cells that are recruited to the tick feeding site where they degranulate, releasing toxic substances that may act directly on ticks (Brown, 1988) or on other cells that inhibit tick feeding or increase grooming (Brown, 1985). Some ticks are able to evade many of these host barriers because they possess potent pharmacologic agents in their saliva (Ribeiro, 1989a, 1989b). Also, some hosts may be more permissive to tick feeding than others, perhaps because of a reduced hypersensitivity response. However, recent evidence suggests that tick-feeding success, and thus pathogen amplification, may be regulated to some degree by the density of feeding ticks even for host species considered permissive (Mather, unpublished). In preliminary trials, feeding success of larval *I. dammini* on white-footed mice (considered a permissive host for *I. dammini*) was not impaired when mice were repeatedly challenged with 100 larvae (Fig. 4.2). However, reduced feeding success did appear to develop when mice were repeatedly challenged with 250–300 larvae. Such a density-dependent mechanism, modulated by tick abundance, also may serve to apply limits to pathogen amplification.

Co-feeding or clustering of ticks at a bite site may serve to enhance infectivity, as well as provide an opportunity to introduce pathogens into alternative vector species. Tick salivary factors have largely been implicated in facilitating the accumulation of pathogens in tissues around the tick's mouthparts as it feeds. In this way, a saliva-activated transmission factor has been implicated in lowering the transmission threshold for Thogoto virus (Jones et al., 1992). In a similar manner, it has also been suggested that Lyme disease spirochetes accumulate in skin near the feeding site of *I. dammini*, perhaps in response to some chemical taxis (Nakayama and Spielman, 1989).

The anti-hemostatic, anti-inflammatory and analgesic function of *I. dammini* saliva (Ribeiro, 1989b) facilitates blood-feeding of this tick and perhaps others (namely *Dermacentor variabilis*) coincidentally infesting the same host. Tick saliva may also play a role in vector competence and infection processes.

2.5. Host Immunity Expressed Within the Vector

Infectious agents usually cause specific immune reactions within host animals. Recently, it has been suggested that the immunity that hosts develop in response to an infection may be effectively expressed within the vector, profoundly affecting infectivity (Telford and Spielman, 1993). Such a mechanism, although not without precedence, may help to explain incompetence or lowered infectivity of reservoirs despite a continued ability to isolate organisms from such hosts. Transmission-blocking immunity has been previously described in malaria (Mendis et al., 1987) and African trypanosomiasis (Murray et al., 1985). In both cases, host immunity prevented pathogen development within the vector gut. In the case of Lyme borreliae, vaccine-mediated antibodies ingested by spirochete-infected ticks "blocked" transmission by destroying the spirochetes in the tick gut (Fikrig et al., 1992). Spirochetes reside apparently dormant within the gut of vector ticks (Burgdorfer et al., 1989). Following tick attachment, the borreliae are "activated", penetrating the hemocoele and eventually migrating to salivary glands, where they are delivered via saliva (Benach et al., 1987; Ribeiro et al., 1987). This process takes a minimum of 24–36 hours following tick attachment; *B. burgdorferi* may require a period of replication in the tick's gut prior to dissemination which would make them vulnerable to destruction by ingested antibody. The generality of these observations remains to be determined. Ingestion of TBE virus-specific antibodies apparently had no effect on virus infection in *I. persulcatus* (Dumina, 1958). However, infectious agents that are confined to the tick's gut for an extended time may be particularly susceptible to this effect of reservoir immunity.

3. TRANSMISSION FROM TICKS TO HOSTS

Pathogens may be transmitted from ticks to animal hosts via saliva (borreliae, piroplasms, viruses), regurgitation (rickettsiae), coxal fluid (borreliae), and feces (rickettsiae). However, ixodid ticks lack coxal glands and so are incapable of pathogen transmission via coxal fluids. Direct inoculation of pathogens into hosts, either by saliva or regurgitation would seem to be the most efficient mode of transmission, likely requiring fewer infectious units and therefore favoring transmission, potency, and the basic reproduction rate. The various physiologic mechanisms resulting in pathogen transmission can be quite complex. The reader is referred to accounts of events leading to transmission of the rodent piroplasm, *B. microti* (Telford et al., 1993), and Lyme disease spirochete, *B. burgdorferi* (Zung et al., 1989), as examples.

3.1. Transmission Efficiencies

For most horizontally transmitted infections, one infective tick bite is all that is needed to infect a host. However, there does appear to be a minimum number of infectious units necessary to establish an infection, so that not all bites by infected ticks produce an infection. Of course, the number of infectious units necessary to infect a host likely varies, and is a component of the tick's vectorial capacity, which also includes other aspects of vector competence, the route of inoculation, conditions at the bite site, and the nature of the "media" carrying the infectious units. In transmission of *Babesia microti*, about 10,000 sporozoites are produced per sporoblast and as many as 100,000 sporozoites may form in salivary acini of nymphal *I. dammini* (Telford et al., 1993). Many thousand sporozoites are deposited in the skin around the feeding tick's mouthparts (Mehlhorn and Schein, 1987), and some 10,000–25,000 syringe-injected sporozoites appear to be needed to infect white-footed mice (*P. leucopus*) and hamsters with this piroplasm. The number of Lyme disease spirochetes necessary to infect hamsters by intraperitoneal inoculation was 10^3–10^4 bacteria while estimates of the number of spirochetes infecting nymphs are <300 bacteria (Piesman et al., 1990). Both examples suggest a lower actual transmission threshold than can be determined experimentally. The anti-inflammatory and immune-suppressive properties of tick saliva (Ribeiro et al., 1985; Ribeiro, 1989a, b) may facilitate infection of the host by inhibiting degranulation of mast cells and the activation of macrophages, neutrophils, and T cells. Moreover, since horizontal amplification of pathogens depends upon repeated host feedings, the anti-inflammatory role of *Ixodes* saliva likely favors both pathogen transmission and acquisition by ticks. Immature ticks may also inherit infection transovarially from infected females. Vertical transmission may be particularly efficient in some tick species infected with certain pathogens, whereas others may be completely refractory. The severity of infection or inoculum titer of the female tick usually determines the degree of pathogen dissemination into tick tissues, including ovarian tissues. Thus, female ticks with heavy infections of spotted fever group (SFG) rickettsiae or TBE viruses may efficiently pass their infection to high proportions of their offspring (Korenberg and Pchelkina, 1984; Burgdorfer et al., 1989). Filial infection rates can approach 100% in some cases. As long as generalized infection is not harmful to the tick, vertical transmission would presumably increase the basic reproduction rate (one infected female tick producing thousands of infected progeny). In most cycles where transovarial transmission of pathogens occurs, however, vertical transmission has been found to be an inefficient maintenance process. Pathologic effects have been observed in several species of transovarially infected ticks. Eggs of *I. dammini* heavily infected with Lyme disease borreliae failed to develop (Burgdorfer et al., 1989), and ovarially passed spirochetes appear to significantly reduce fecundity and fertility of filially infected *I. pacificus* (Lane, 1992). Following a few generations of transovarial passage, ticks infected transovarially with SFG rickettsiae suffered greater mortality soon after engorgement and those that survived produced fewer eggs that failed to develop (Burgdorfer and

Brinton, 1975). Also, pathogenic virulence may be lost through continual transovarial passage. Thus, vertical transmission may occur efficiently in some zoonotic cycles, but may lower potency in some cases through increasing tick mortality or by other yet unidentified factors. In this way, where both vertical and horizontal transmission occurs, the impact of vertical transmission may serve to reduce efficiency of the horizontal amplification processes. In most cases where it has been found to occur, the epidemiologic impact of vertical transmission must still be determined.

3.2. Tick Infection Rates

The prevalence of infection in a tick population, or the "tick infection rate" varies widely for the various tick-borne zoonoses. Endemic infection rates of $<1\%$ for spotted fever rickettsia to nearly 100% for Lyme disease spirochetes have been recorded in populations of vectors (Burgdorfer, 1975; Bosler et al., 1983). However, where transmission is stable, characteristic infection rates, or at least, characteristic ranges of infection prevalence are generally observed for a particular tick instar and zoonosis (Table 4.1). Many of the factors discussed above are likely determinants of infection prevalence in tick populations. Moreover, the reservoir potential equation (Mather et al., 1989), and now the Reservoir Potential Model (software available from Platt Systems, Manchester, CT, USA) is well suited to demonstrate how variation in host infectivity, tick infestation rates, and host density may affect the tick infection rate. In one hypothetical scenario with 12 host species (one principal reservoir, seven secondary reservoirs, four reproductive hosts), a 50% reduction in the density of the principal reproductive host, but no net reduction in the number of ticks (Wilson et al., 1984) served to increase the tick infection rate by 8% (Mather, unpublished). This increase was due to a shift in the "host mix" favoring reservoir hosts with its corresponding decrease in the dilution effect of ticks derived from the reproductive host. An even greater reduction in the density of this reproductive host (75%), but with an associated 20% decrease in tick infestations on all hosts resulted in a 22% increase in the tick infection rate. Of course, there were still 20% fewer ticks produced, both infected and non-infected, so risk for infection may have been reduced despite the higher infection prevalence in ticks. However, the example does demonstrate the complex nature of the dynamic relationships between reproductive and reservoir hosts in determining the tick infection rate.

3.3. Host Infection Thresholds

As already indicated, most host infections require just one infective tick bite. But general rules of epidemics suggest that infections do not increase linearly with increases in the number of infected contacts (Frost, 1976). Instead, the relationship between vector abundance, tick infection rate, and the probability

Table 4.1. Characteristic tick infection rates[a] for selected zoonoses in endemic areas with relatively stable transmission

Tick-borne disease	Agent	Vector	Prevalence of infection (%)		
			Larvae	Nymph	Adults
Viruses					
Colorado tick fever		*Dermacentor andersoni*		2–7	10–20
Tick-borne encephalitis		*Ixodes ricinus*		<1	<1–6
Louping Ill		*I. ricinus*		<1	
Rickettsiae					
Rocky Mountain spotted fever	*Rickettsia rickettsii*	*Dermacentor variabilis*			<1–6
		D. andersoni			1–5
		D. occidentalis			1–3
Bacteria					
Lyme borreliosis—Eastern USA	*Borrelia burgdorferi*	*Ixodes dammini*	<1	18–30	50–70
—Western USA	*B. burgdorferi*	*I. pacificus*	<1	1–3	1–3
—N. Europe	*B. burgdorferi*	*I. ricinus*	0	7–14	6–30
Relapsing fever	*B. parkeri*	*Ornithodoros parkeri*		10	10
Tularemia	*Francisella tularensis*	*Amblyomma americanum*		2	2
		Haemaphysalis leporis-palustris		6	8
Protozoa					
Babesiosis	*Babesia microti*	*Ixodes dammini*	0	11	14

[a] Estimates are rough approximations compiled from Burgdorfer (1975), Monath (1988/1989), Piesman et al. (1986), Jaenson (1991), Lane et al. (1991), Sonenshine (1993), and personal communications from C.E. Hopla, R.S. Lane, and T.G. Schwan.

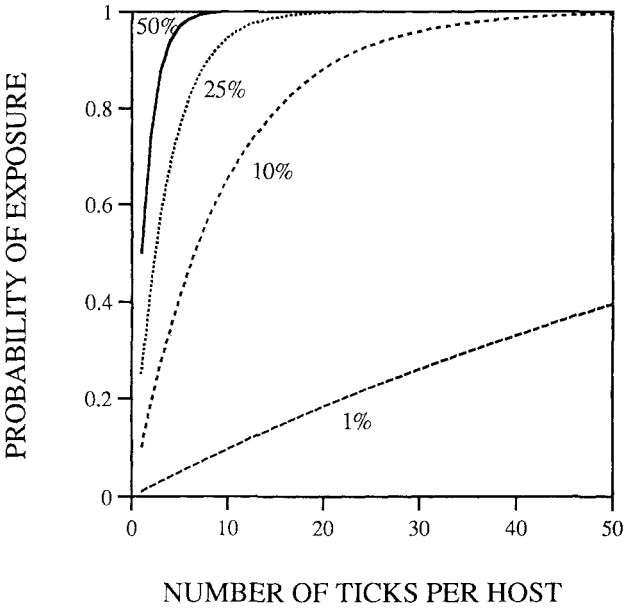

Fig. 4.3. Probability of exposure to pathogens for hosts bitten by up to 50 ticks. Percentages are overall percent of vectors infected with pathogen (tick infection rate). Modified from Ginsberg (1993), with permission from the *American Journal of Epidemiology*.

of host infection can be described by calculating the probability of being bitten by at least one infected tick (Ginsberg, 1992, 1993). Given n tick bites (where the ticks stay attached long enough to transmit their infection) and a prevalence of tick infection (k_t), then the probability that at least one of the n ticks is infected, or the probability of exposure (P_1) is given by:

$$P_1 = 1 - (1 - k_t)^n \tag{4.2}$$

Thus, it is possible to plot the probability of exposure to an infectious tick at various levels of tick contact given particular tick infection rates (Fig. 4.3). In zoonoses where tick infection rates are typically low, many more tick bites would be required to infect a host than where tick infection rates are higher. Under some circumstances, the probability of exposure increases rapidly with the number of tick bites (Fig. 4.3), which gives the appearance of a threshold number of tick bites, above which infection of hosts would be expected to increase rapidly.

Zoonotic infection thresholds may be useful indicators of zoonotic potency, as well as the probability of human exposure. As populations of ticks increase, more infections would likely occur if the tick infection rate remains the same. To be useful as an indicator of human risk it is still left to discover the relationship between frequencies of host and human tick biting, including any preferential differences exhibited by infected ticks. But to illustrate the concept

of a transmission threshold, consider the transmission dynamics of Lyme disease spirochetes along Argilla Road in Ipswich, Massachusetts (USA), during the early to mid-1980s. At that time, white-footed mice (*P. leucopus*) captured around residences were infested with an average of 1.0 ($\pm$ 1.67 s.d.) nymphal *I. dammini* (Mather, unpublished). From this, we could estimate that each mouse likely was bitten by about 40 nymphs during the summer. In 1984 and 1985, the tick infection rate for all nymphal *I. dammini* collected around Argilla Road residences was 27% and 30%, respectively (Lastavica et al., 1989; Mather, unpublished). Using Fig. 4.3, it is clear that there was a 100% probability that every mouse had likely been exposed to an infected nymph. Thus, a daily average of one nymph/mouse was well above the zoonotic infection threshold. In fact, we observed that in total, about 90% of white-footed mice (*P. leucopus*) were capable of infecting ticks as determined by tick xenodiagnosis (Mather, unpublished; Mather et al., 1989). Among these same households, about 74% of dogs (Eng et al., 1988) and between 28% and 35% of people showed evidence of infection (Eng et al., 1988; Lastavica et al., 1989). The annual incidence of Lyme disease for 1984–1985 was estimated to be 10%. Another indicator of tick exposure risk is the entomologic risk index, or the abundance of infected, host-seeking nymphs (Mather, 1993); in 1985 an average of five spirochete-infected *I. dammini* nymphs were found per hour of flagging around the same residences (Mather, unpublished). Thus, it would appear that this level of entomologic risk exceeds the epidemiological infection threshold as well. Knowledge of the infection threshold (or level of entomologic risk) below which infections are suppressed are virtually unknown for most tick-borne zoonoses, but would be invaluable in designing disease prevention strategies.

4. CLASSIFICATION OF TICK-BORNE ZOONOSES

Thus far, we have discussed many specific factors that can influence maintenance and horizontal amplification of pathogens among ticks and hosts, and the complex interactions of factors that determine zoonotic potency. The interplay of these factors can be conceptualized in general terms by examining the structure of the vector–host–pathogen system in nature. This can provide a framework with which to evaluate the implications of changes in the various factors affecting the epidemiology of tick-borne diseases.

Considerable theoretical attention has been paid to the epidemiology of tick-borne diseases, and of vector-borne diseases in general (Bailey, 1975; Fine, 1981; Goldfarb, 1986; Kitron and Alessandro, Chapter 7 of this volume). Common to virtually all of these analyses is some estimate of the rate of infection of humans with the disease. In epizootiological models, however, humans are only important if they contribute to the disease maintenance and transmission cycles; often they are merely dead-end hosts. Whether or not humans play a role in disease maintenance and transmission, the intensity of transmission to humans depends upon both zoonotic and epidemiologic potency factors.

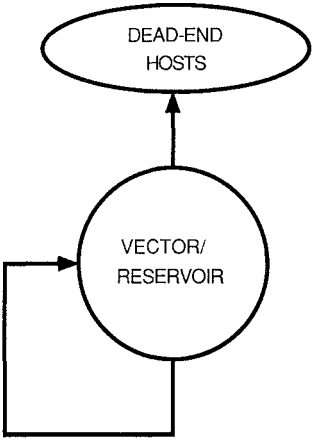

Fig. 4.4. Class I zoonosis. Tick is major vector and reservoir.

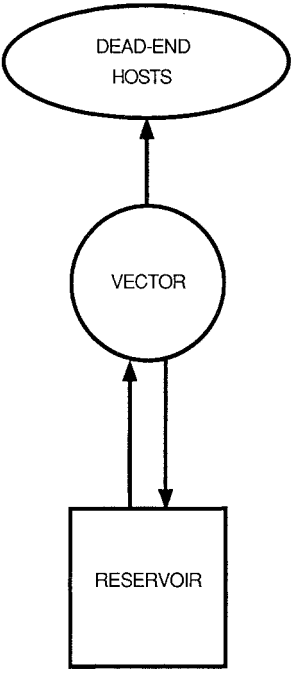

Fig. 4.5. Class II zoonosis. One tick vector and one reservoir host species.

Tick-borne diseases can be divided into five main classes, based on the structure of their epizootiological cycles (Figs 4.4–4.8):

Class I —a tick species serves as the sole (or primary) vector and reservoir.
Class II —one tick species is the primary vector and a single host species is the primary reservoir
Class III—there is one vector species, but multiple reservoir species.
Class IV—there are more than one vector species, but only one primary reservoir species.
Class V —there are multiple vector and multiple reservoir species.

The form of the epizootiological cycle has important implications for infection dynamics in nature, for pathogen prevalence in host-seeking ticks, and thus for disease epidemiology in humans. The five classes of epizootiological cycles are considered below in terms of their implications for pathogen prevalence in ticks.

4.1. Class I—Tick Species is Vector and Reservoir (Fig. 4.4)

Humans serve as dead-end hosts and have little to do with the intensity of transmission. Infection rates in host-seeking ticks depend, to a large extent, on

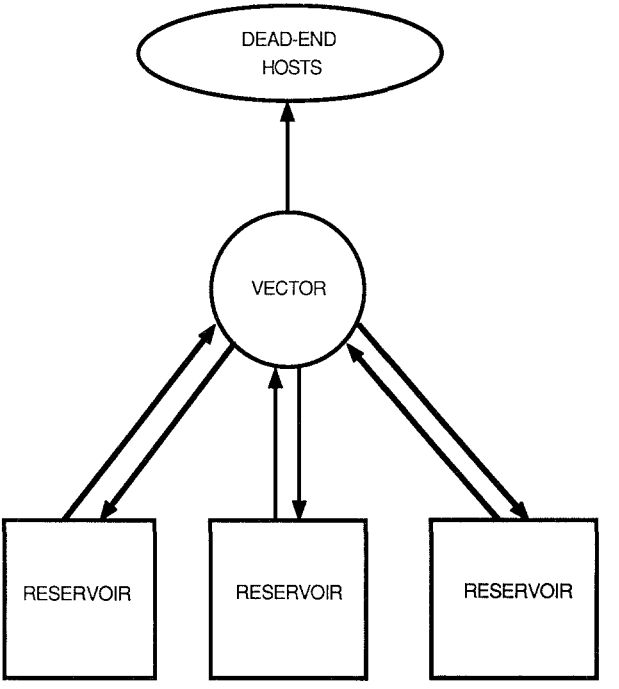

Fig. 4.6. Class III zoonosis. One tick vector and multiple reservoir species.

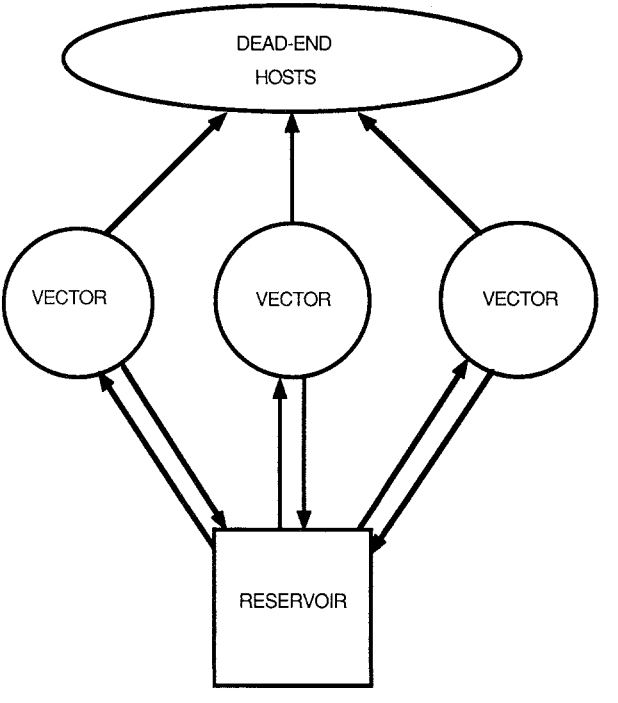

Fig. 4.7. Class IV zoonosis. Multiple tick vector species and one reservoir host species.

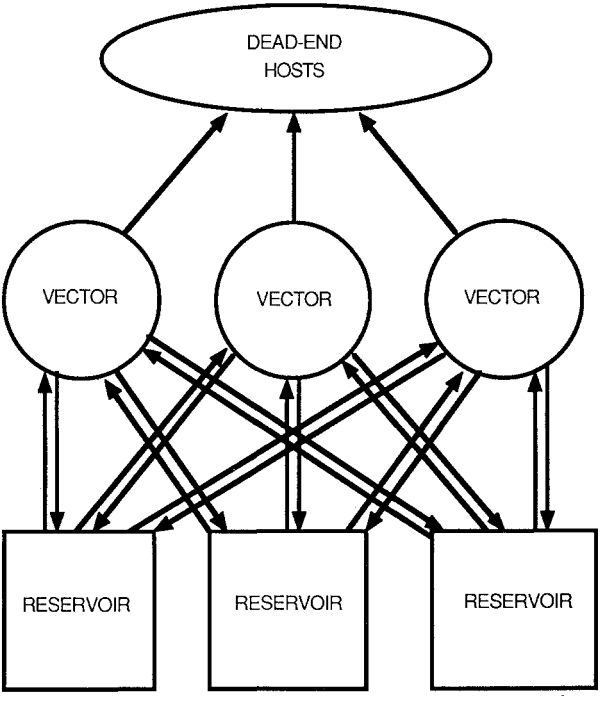

Fig. 4.8. Class V zoonosis. Multiple tick vectors and multiple reservoir host species.

the efficiency of vertical transmission. Pathogen prevalence is usually low in ticks, unless there is some mechanism for horizontal transmission among ticks (e.g., transmission among ticks feeding simultaneously on a host animal), or unless infected ticks have a reproductive advantage over uninfected ticks. An example of a disease with this type of cycle is Rocky Mountain spotted fever. The tick vectors largely maintain this infection vertically, although some horizontal amplification apparently occurs as well.

4.2. Class II—Single Tick Vector and Single Reservoir Host (Fig. 4.5)

The prevalence of infection in host-seeking ticks depends directly on the frequency of encounter between ticks and reservoir hosts, and on the efficiency of transmission between vectors and reservoirs. If the average number of tick bites per host individual is low (because ticks are rare, or differ in phenology or spatial distribution from hosts, or there are abundant non-reservoir hosts), or if transmission of pathogens between ticks and hosts is inefficient, pathogen prevalence is expected to be low. However, in cases with numerous tick bites per host animal and efficient transmission, pathogen prevalence can grow rapidly and be maintained at high levels in both ticks and hosts. The numerous tick bites per host animal result in high probabilities of transmission.

Fluctuation in relative densities and encounter frequencies of ticks and hosts can result in broad fluctuations in pathogen prevalence because of broad changes in the number of ticks per host individual and thus in the probability of transmission. Moreover, these effects could be counterintuitive. For example, lowered reservoir densities could result in fewer infected ticks if tick densities decline as a result. However, if tick densities do not decline, pathogen prevalence could increase in ticks because of larger numbers of ticks per host animal, resulting in a higher proportion of hosts infected (Porco, 1991). These fluctuations can be dampened considerably if the pathogen persists in reservoir hosts without re-infection, and if infected hosts remain infective to uninfected ticks for long periods. Examples of diseases with this type of cycle include babesiosis, some forms of tick-borne relapsing fever, and probably Lyme borreliosis at some sites.

4.3. Class III—One Tick Vector, Multiple Reservoir Species (Fig. 4.6)

Pathogen prevalence in host-seeking ticks depends, to a large extent, on the reservoir efficiencies of the various host species and on the proportion of the tick vector population that attaches to each host species (narrowness of host range), similar to the situation in Class II cycles. Therefore, host diversity, relative population densities, and preferences of ticks for each host species have a strong influence on disease transmission. However, in contrast with Class II cycles, population dynamics of single reservoir host species are unlikely to have a strong influence on pathogen prevalence in Class III cycles because of the buffering effect of other competent reservoir species (Fig. 4.6). Changes in the mix of host species toward reservoir-incompetent species (dead-end hosts) would be expected to reduce pathogen prevalence.

In Class III, changes in tick density would be expected to have a stronger influence on pathogen prevalence than in Class I and Class II cycles because the vector population is divided among multiple reservoir host species. Therefore, the average number of ticks per individual reservoir animal is lower than in the one vector/one host situation, everything else being equal. At lower tick densities on each host animal, fluctuations in tick density will have a greater effect on probabilities of exposure, and thus on transmission rates, than at higher tick densities (Ginsberg, 1992, 1993). Examples of this type of transmission cycle include Louping ill, Colorado tick fever, and Lyme borreliosis in certain ecological settings.

4.4. Class IV—Multiple Tick Vector Species, One Reservoir Species (Fig. 4.7)

The important factors are similar to those in Class II cycles, with the exception that here they apply to multiple tick species. Therefore, fluctuations in the abundance of one tick species would be unlikely to influence pathogen prevalence unless these fluctuations involved all vector species. In contrast,

fluctuations in the population of the primary reservoir host could have dramatic effects on pathogen prevalence. As in Class II cycles, the effects of fluctuations in reservoir host populations depend on their influence on tick densities, and on the number of vector ticks per host individual.

4.5. Class V—Multiple Tick Vectors and Multiple Reservoir Host Species (Fig. 4.8)

Pathogen prevalence ultimately depends upon a complex set of interactions among various vector and reservoir species. Fluctuations in populations, or other changes in any single tick or host species, are unlikely to result in large changes in potency because of the buffering effect of alternative vector and reservoir species. Pathogen prevalence is expected to be moderate, even when one or more of the tick species is an efficient vector, because of varying reservoir competence levels of the various alternative host species. Some of the tick-borne encephalitides would fit in this category.

As seen from this zoonosis classification scheme, tick-borne diseases can be expected to behave in a somewhat predictable fashion, simply by virtue of the structure of the transmission cycle in nature. Of course, particular zoonoses may fall into different categories, and thus behave differently, in different geographical areas. For example, in Lyme disease endemic areas, the behavior of pathogen prevalence levels depends, in part, on the suite of reservoir hosts. In an area with only one important reservoir host (Class II), Lyme disease potency would be subject to fluctuations in tick infection prevalence resulting from fluctuations in populations of this host. However, in areas with several host species that are competent reservoirs (Class III), Lyme disease would be more stable. Although tick infection prevalence could be influenced by changes in tick populations, it would be relatively unaffected by changes in the density of any one host species.

Except for Class I, the foregoing classification scheme deals primarily with horizontal transmission of pathogens. Vertical transmission and non-tick transmission (e.g., by contact among hosts in louping ill and African swine fever) could strongly influence epizootiology. In all cases, efficient vertical transmission among reservoirs would be expected to result in increased pathogen prevalence in host-seeking ticks because of increased prevalence in the reservoir host (in Case I, of course, the tick is the reservoir).

Non-tick transmission does not exist, by definition, in Case I. In Cases II–V, however, non-tick transmission among reservoirs would generally be expected to increase pathogen prevalence in reservoir hosts, and consequently to increase pathogen prevalence in questing ticks. In some cases, however, efficient non-tick transmission could actually reduce transmission in ticks. For example, in hosts that develop immunity to the pathogen, high proportions of immune hosts could conceivably lower overall host infectivity and thus lower the proportion of ticks infected.

Table 4.2. Critical transmission factors affecting potency of tick-borne zoonoses

Narrowness of host range
Inverted feeding pattern of transmitting and acquiring instars
Long-lived infectivity
Relatively low death rate among pathogen-acquiring instar
Properties of tick saliva
Tick density

5. CONCLUSIONS

Zoonoses produce various levels of infections driven largely by the ecological dynamics existing between vector, host, and pathogen. For zoonoses to be maintained, the basic reproduction rate of a given pathogen must be greater than one. Typically, several interacting factors combine to determine the potency of a zoonosis. Examples have been given here, and will be discussed in greater detail in Part II of this volume.

The "probability of exposure" model described above predicts that potency would be expected to increase with increasing tick density. To be sure, tick density strongly influences zoonotic potency. However, several other transmission factors also impact the potency of a zoonosis, raising potency where they do occur and perhaps suppressing it where they do not. Table 4.2 is a listing of those factors that appear to be most critical in determining potency of tick-borne zoonoses. This list is not all inclusive of the various factors that must certainly regulate the potency of particular zoonoses. Lyme borreliosis, for example, exquisitely exhibits variable zoonotic and epidemiologic potency in its different ecological settings. This variation in potency is largely regulated by the geographic manifestation of each factor as well as by the interplay among factors. Thus, Table 4.2 may serve as a useful checklist when considering interventions to suppress the risk of infection posed by various tick-borne diseases.

6. ACKNOWLEDGMENTS

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7. REFERENCES

Bailey, N.T.J. (1975) *The Mathematical Theory of Infectious Diseases*, 2nd edn. Macmillan, New York.

Benach, J.L., Coleman, J.L., Skinner, R.A., and Bosler, E.M. (1987) Adult *Ixodes dammini* on rabbits: a hypothesis for the development and transmission of *Borrelia burgdorferi*. *J. Infect. Dis.* **155**, 1300–1306.

Bosler, E.M., Coleman, J.C., Benach, J.L., Massey, D.A., Hanrahan, J.P., Burgdorfer, W., and Barbour, A.G. (1983) Natural distribution of the *Ixodes dammini* spirochete. *Science* **220**, 321–322.

Bozeman, F.M., Shirai, A., Humphries, J.W., and Fuller, H.S. (1967) Ecology of Rocky Mountain spotted fever II. Natural infection of wild mammals and birds in Virginia and Maryland. *Am. J. Trop. Med. Hyg.* **16**, 48–59.

Brown, B. (1985) Immunology of acquired resistance to ticks. *Parasitol. Today* **12**, 166–171.

Brown, B. (1988) Highlights of contemporary research on host immune responses to ticks. *Vet. Parasitol* **28**, 321–334.

Burgdorfer, W. (1975) A Review of Rocky Mountain spotted fever (tick-borne typhus), its agent, and its tick vectors in the United States. *J. Med. Entomol.* **12**, 269–278.

Burgdorfer, W. and Brinton, L.P. (1975) Mechanisms of transovarial infection of spotted fever rickettsiae in ticks. *Ann. New York Acad. Sci.* **266**, 61–72.

Burgdorfer, W. and Schwan, T.G. (1991) Lyme borreliosis: a relapsing fever-like disease? *Scand. J. Infect. Dis. Suppl.* **77**, 17–22.

Burgdorfer, W., Hayes, S.F., and Corwin, D. (1989) Pathophysiology of the Lyme disease spirochete, *Borrelia burgdorferi*, in ixodid ticks. *Rev. Infect. Dis.* **11** (Suppl. 6), 1442–1450.

Donahue, J.G., Piesman, J. and Spielman, A. (1987) Reservoir competence of white-footed mice for Lyme disease spirochetes. *Am. J. Trop. Hyg.* **36**, 92–96.

Dumina, A.L. (1958) Experimental study of the extent to which the tick *Ixodes persulcatus* becomes infected with Russian spring–summer encephalitis virus as a result of sucking the blood of immune animals. *Vop. Virusol.* **3**, 156–159.

Eng, T.R., Wilson, M.L., Spielman, A., and Lastavica, C.C. (1988) Greater risk of *Borrelia burgdorferi* infection in dogs than in people. *J. Infect. Dis.* **158**, 1410–1411.

Fikrig, E., Telford, S.R., III, Barthold, S.W., Kantor, F.S., Spielman, A., and Flavell, R.A. (1992) Elimination of *Borrelia burgdorferi* from vector ticks feeding on OspA-immunized mice. *Proc. Natl Acad. Sci. USA* **89**, 5418–5421.

Fine, P.E.M. (1981). Epidemiological principles of vector-mediated transmission. pp. 77–91. *In:* McKelvey, J.J., Eldridge, B.F., and Maramorosch, K. (eds) *Vectors of Disease Agents.* Praeger, New York.

Frost, W.H. (1976) Some conceptions of epidemics in general. *Am. J. Epidemiol.* **103**, 141–151.

Ginsberg, H.S. (1988) A model of the spread of Lyme disease in natural populations. *Ann. New York Acad. Sci.* **539**, 379–380.

Ginsberg, H.S. (1992) Ecology and management of ticks and Lyme disease at Fire Island National Seashore and selected eastern National Parks. *National Park Service Science Monograph*, NPS/NRSUNJ/NRSM-92/20. 77 pp.

Ginsberg, H.S. (1993) Transmission risk of Lyme disease and implications for tick management. *Am. J. Epidemiol.* **138**, 65–73.

Goldfarb, L.G. (1986) Epidemiological models of tick-borne infections (Acari: Ixodidiae and Argasidae). *J. Med. Entomol.* **23**, 125–131.

Jaenson, T.G.T. (1991) The epidemiology of Lyme borreliosis. *Parasitol. Today* **7**, 39–45.

Jones, L.D., Kaufman, W.R., and Nuttall, P.A. (1992) Modification of the skin feeding site by tick saliva mediates virus transmission. *Experientia* **48**, 779–782.

Korenberg, E.I. and Pchelkina, A.A. (1984) Tickborne encephalitis virus titres in engorged adult Ixodes persulcatus ticks. *Parazitol.* **18**, 123–127.

Lane, R.S. (1990) Seasonal activity of two human-biting ticks. *Calif. Agric.* **44**, 23–25.

Lane, R.S. (1992) Effect of transovarially and transstadially-passed Lyme disease

spirochetes on the development and reproductive biology of *Ixodes pacificus* (Acari: Ixodidae). pp. 282–289. *In:* Munderloh, U. and Kurtti, J.J. (eds) *Proc. First Internat. Conference on Tick-borne Pathogens at the Host–Vector Interface.* University of Minnesota, St. Paul.

Lane, R.S., Piesman, J., and Burgdorfer, W. (1991) Lyme borreliosis: relation of its causative agent to its vectors and hosts in North America and Europe. *Annu. Rev. Entomol.* **36**, 587–609.

Lastavica, C.C., Wilson, M.L., Berardi, V.P., Spielman, A. and Deblinger, R.D. (1989) Rapid emergence of a focal epidemic of Lyme disease in coastal Massachusetts. *New Engl. J. Med.* **320**, 133–137.

Macdonald, G. (1950). The anlysis of infection rates in diseases in which super-infection occurs. *Trop. Dis. Bull.* **47**, 907–915.

Macdonald, G. (1957) *The Epidemiology and Control of Malaria.* Oxford University Press, London.

Main, A.J., Carey, A.B., Carey, M.G., and Goodwin, R.H. (1982) Immature *Ixodes dammini* (Acari: Ixodidae) on small mammals in Connecticut, USA. *J. Med. Entomol.* **19**, 655–664.

Mather, T.N. (1991) The ecology of Lyme borreliosis. *Bull. Scand. Soc. Parasitol.* **1**, 15–23.

Mather, T.N. (1993) The dynamics of spirochete transmission between ticks and vertebrates. pp. 43–60. *In:* Ginsberg, H.S. (ed) *Ecology and Environmental Management of Lyme Disease.* Rutgers University Press, New Brunswick.

Mather, T.N., Wilson, M.L., Moore, S.I., Ribeiro, J.M.C., and Spielman, A. (1989) Comparing the relative potential of rodents as reservoirs of the Lyme disease spirochete (*Borrelia burgdorferi*). *Am. J. Epidemiol.* **130**, 143–150.

Mather, T.N., Telford, S.R., III, Moore, S.I., and Spielman, A. (1990) *Borrelia burgdorferi* and *Babesia microti*: Efficiency of transmission from reservoirs to vector ticks (*Ixodes dammini*). *Exp. Parasitol.* **70**, 55–61.

McDade, J.E., and Newhouse, V.F. (1986) Natural history of *Rickettsia rickettsii. Annu. Rev. Microbiol.* **40**, 287–309.

Mehlhorn, H. and Schein, E. (1987) The piroplasms: life, cycle and and sexual stages. *Adv. Parasitol.* **23**, 37–103.

Mendis, K.N., Munesinghe, Y.D., deSilva, Y.N.Y., Keragalla, I., and Carter, R. (1987) Malaria transmission-blocking immunity induced by natural infections of *Plasmodium vivax* in humans. *Infect. Immun.*, **55**, 369–372.

Monath, J.P. (ed.) (1988/1989) *The Arboviruses: Epidemiology and Ecology, Vols I–IV.* CRC Press, Boca Raton, FL.

Murray, M., Hirumi, H., and Moloo, S.K. (1985) Suppression of *Trypanosoma congolense, T. vivas* and *T. brucei* infection rates in tsetse flies maintained on goats immunized with uncoated forms of trypanosomes grown *in vitro. Parasitol.* **91**, 53–66.

Nakayama, Y. and Spielman, A. (1989) Ingestion of Lyme disease spirochetes by ticks feeding on infected hosts. *J. Infect. Dis.* **160**, 166–167.

Piesman, J. and Spielman, A. (1979) Host associations and seasonal abundance of immature *Ixodes dammini* in southeastern Massachusetts. *Ann. Entomol. Soc. Am.* **72**, 829–832.

Piesman, J., Mather, T.N., Telford, S.R., III, and Spielman, A. (1986) Concurrent *Borrelia burgdorferi* and *Babesia microti* infection in nymphal *Ixodes dammini. J. Clin. Microbiol.* **24**, 446–447.

Piesman, J., Oliver, J.R., and Sinsky, R.J. (1990) Growth kinetics of the Lyme disease

spirochete (*Borrelia burgdorferi*) in vector ticks. *Am. J. Trop. Med. Hyg.* **42**, 352–357.

Porco, T.C. (1991) A model of the enzootiology of Lyme disease in the Atlantic Northeast of the United States. *Nat. Res. Modeling* **5**, 469–505.

Ribeiro, J.M.C. (1989a) Role of saliva in tick/host interactions. *Exp. Appl. Acarol.* **7**, 15–20.

Ribeiro, J.M.C. (1989b) Vector saliva and its role in parasite transmission. *Exp. Parasitol.* **69**, 104–106.

Ribeiro, J.M.C., Makoul, G.T., Levine, J., Robinson, D.R., and Spielman, A. (1985) Antihemostatic, antiinflammatory, and immunosuppressive properties of the saliva of a tick, *Ixodes dammini*. *J. Exp. Med.* **161**, 332–344.

Ribeiro, J.M.C., Mather, T.N., Piesman, J., and Spielman, A. (1987) Dissemination and salivary delivery of Lyme disease spirochetes in vector ticks (Acari: Ixodidae). *J. Med. Entomol.* **24**, 201–205.

Sonenshine, D.E. (1993) *Biology of Ticks, Vol. 2*. Oxford University Press, New York, 464 pp.

Spielman, A., Levine, J.F., and Wilson, M.L. (1984) Vectorial capacity of North American *Ixodes* ticks. *Yale J. Biol. Med.* **57**, 507–513.

Tälleklint, L., Jaenson, T.G.T., and Mather, T.N. (1993) Seasonal variation in the capacity of the bank vole to infect larval ticks (Acari: Ixodidae) with Lyme disease spirochetes, *Borrelia burgdorferi*. *J. Med. Entomol.* **30**, 812–815.

Telford, S.R., III and Spielman, A. (1993) Reservoir immunity and vector competence. *Host Regulated Dev. Mech. Vector Arthrop.* **3**, 271–276.

Telford, S.R., III, Gorenflot, A., Brasseur, P., and Spielman, A. (1993) Babesial infections in humans and wildlife. Vol. 5, pp. 1–47. *In:* Kreier, J.P. (ed) *Parasitic Protozoa*, 2nd edn. Academic Press, New York.

Wilson, M.L., Levine, J.F., and Spielman, A. (1984) Effect of deer reduction on abundance of deer tick. *Yale J. Biol. Med.* **57**, 697–705.

Zung, J.L., Lewengrub, S., Rudzinska, M.A., Spielman, A., Telford, S.R., III, Piesman, J., and Fine, J. (1989) Fine structural evidence for the penetration of the Lyme disease spirochete *Borrelia burgdorferi* through the gut and salivary tissues of *Ixodes dammini*. *Can. J. Zool.* **67**, 737–1748.

5

Micrometeorological and Microhabitat Factors Affecting Maintenance and Dissemination of Tick-borne Diseases in the Environment

MILAN DANIEL AND FRANTIŠEK DUSBÁBEK

1. INTRODUCTION

The geographical distribution of ticks, their life history, hosts, and ability to transmit the infection are determined by both intrinsic and extrinsic factors. The intrinsic factors are the biochemical and physiological properties of the tick that determine its reaction to the external conditions of its habitat. The extrinsic factors are the abiotic and biotic components of the habitat that influence tick biology.

The main abiotic factors of the habitat correspond to the physical variables (particularly light and heat radiation) forming together the principal climatic factor—the energy balance. The circulation of the atmosphere, climate, and short-term changes in the weather all depend upon the energy balance. The energy balance is a complex of light and heat radiation falling on the active surface of the Earth. In climatological terms, the active surface refers both to: (1) the rocky, sandy, or soil-covered surface of the earth; and (2) the numerous modifications caused by vegetation cover, vertical zoning, and human activities (e.g., buildings, cities, and other man-made modifications).

Under natural conditions, climatic factors act in an integrated manner that results from their mutual physical relationships. Climate is the main factor influencing: (1) the horizontal and vertical distribution; (2) the life cycle; (3) the seasonal activities; (4) the population dynamics; and (5) the behavior of ticks at the level of both populations and individuals. This influence may be direct or indirect. In the latter case, the climate affects the complex of biotic components of the tick habitat, particularly the vegetative cover that provides a suitable microclimate for the development and survival of unattached ticks.

One of the most important of the biotic components is a suitable host as a blood source which is needed for development and reproduction. In free-living hosts, their own population dynamics, seasonality, and daily activities are also influenced by the climatic factors. This applies, to some extent, to pastured

domestic animals and even man. For instance, the peak of the spring–summer *Ixodes ricinus* activity corresponds to the main holiday period when people visit areas inhabited by ticks. Similarly, the peak of the autumn period is the main season for gathering mushrooms. Moreover, tourism, hunting, holiday excursions, and other activities are also affected by the weather. All these activities influence the epidemiology and epizootology of infections transmitted by ticks.

A detailed study of the effect of climate (especially of the microclimate) on the life history of ticks allows scientists to analyze both the parasite (vector)—host and vectors—disease-causing agent relationships. Knowledge of tick biology in relation to their habitats and microhabitats facilitates the discovery of indicators which could serve as a basis for predictive maps of the occurrence and population densities of ticks in nature. In this respect, valuable experience has been gained in mathematical modeling and remote sensing.

2. CLIMATE AND HABITAT

The climate is considered to be a long-term summation of the atmospheric elements—radiation, temperature, precipitation, humidity, and wind—and their variations. The global interactions of the climatic components are highly complex and resolve into a number of long-term meteorological cycles. A short-term variation within the climate is termed weather (Reiter, 1988).

The climate is the major component in the environment of all arthropods and all living organisms. Within their climatic limits, all the atmospheric elements constantly influence every aspect of behavior, development, and dispersal, while at the boundaries of their climatic limits relatively minor deviations from the ambient norm can be catastrophic.

2.1. Categories: Macro-, Meso-, and Microclimate

Three climate categories are recognized: (1) macroclimate, i.e., the climate of whole regions, with horizontal area of hundreds of square kilometers, with varying habitats (biotopes); (2) mesoclimate, i.e., the climate of the horizontal area which is limited by the extent of a specific habitat, e.g., pasture land, oak woodland, etc.; (3) microclimate, i.e., the climate of very small areas (microhabitats), which are the actual living environments of the ticks. Microhabitat size cannot be defined in general terms. For exophilic (= non-nidicolous) ticks, the living environment of their non-parasitic stage is mostly the lower part of vegetation, upper layers of the soil, dead leaves, litter, and upper layers of the wood humus. The microhabitat of endophilic (nidicolous) ticks is the nest of their host.

It is important to distinguish between the macroclimate of the area under study and the actual microclimate of the microhabitat, as demonstrated by Daniel and Černý (1967). Most papers attempting to analyze the influence of

meteorological factors on the existence, behavior, reproductive cycles, and other aspects of tick biology are based on measurements recorded by the nearest field stations of the public meteorological service with standard equipment and observation programs. Unfortunately, meteorological data obtained in this manner provide only general information, applicable for broad regions. However, such data do not reflect the micrometeorological features peculiar to the microrelief of the terrain, local vegetation, and other characteristics of the actual conditions in which the ticks live and develop. Relationships derived from these macrometeorological records, therefore, are of limited significance.

In view of the fact that the problems described above relate to micrometeorology, it is necessary to employ appropriate instruments and proper techniques. For example, while studying *I. ricinus* activity in an inundated forest in South Moravia, the relationship between macro-, meso-, and microclimate was analyzed (Daniel, 1978). The data obtained at the nearest station of the state observation network (about 7 km away) were taken as the macroclimate, those from a standard meteorological box placed directly in the experimental site were regarded as the mesoclimate, and data measured in the air near the ground at the experimental site were regarded as the microclimate. Statistical analysis shows that differences in the daily temperature means, maximum and minimum values are not significantly different at the micro- and mesometeorological levels, but these same values are significantly different between the meso-, macro, or the micro- and macrometeorological levels. When relative humidity is compared, all differences between the daily mean and minimal data are highly significant.

It stands to reason that the macroclimate had a leading role, and the meso- and microclimate in this case only balanced the fluctuations and maintained higher levels of humidity in particular. We attempted to determine the mathematical relation between the temperatures of the macro- and mesoclimate which would make it possible to predict the mesoclimate in the biotope under study.

Let us use for macroclimate the following designation: daily mean temperature x_1, daily temperature maximum x_2, and daily temperature minimum x_3. Analogously for mesoclimate, let us designate daily mean temperature y_1, daily temperature maximum y_2, and daily temperature minimum y_3.

The required dependence is constructed by means of a multiple linear regression equation. We assume that the dependence of the variable y_1 on the variables x_1, x_2, x_3 can be described in terms of multiple linear regression, i.e.,

$$\sum y_1 = ax_1 + bx_2 + cx_3 + d + \varepsilon \tag{5.1}$$

where ε is the residual random variable with zero expectation. The estimation of coefficients a, b, c, d is carried out by the method of least squares, i.e., by minimizing the expression

$$(y_1 - ax_1 - bx_2 - cx_3 - d)^2 \tag{5.2}$$

This condition from the mathematical aspect leads to the system of the so-called normal equations by the solution of which we obtain the values of

coefficients a, b, c, and d. An analogous procedure is used in constructing the regression equation for y_2 *and* y_3. After all calculations have been made, the following theoretical dependencies are obtained for the conditions of our experiment:

$$y_1 = 0.786x_1 + 0.157x_2 + 0.120x_3 - 0.62 \quad (5.3)$$

$$y_2 = 0.928x_1 + 0.435x_2 + 0.147x_3 - 0.027 \quad (5.4)$$

$$y_3 = 0.048x_1 + 0.119x_2 + 0.765x_3 - 1.28 \quad (5.5)$$

These mathematical relationships demonstrate the complexity of the whole problem. It should be noted, after all calculations are made, that the final version can be applied locally in a limited way in the experimental site only. In this instance it means that after the first season, which provides the required data for the calculation of coefficients, satisfactory calculations of the meso-climate can then be made from the values of the macroclimate in the following seasons, assuming that the general character of the habitat does not change.

As for the microclimate, the rich material available confirmed the fact that at a distance of several meters there are sufficient differences which can fundamentally affect the existence and development of the ticks. In this respect, e.g., the differences in their rates of development should be noted (Daniel et al., 1976, 1977a). The interrelationship between microclimatic factors is not unchangeable; indeed, it undergoes changes in individual years and between seasons of the same year.

A detailed study of the microclimate in which ticks survive is needed to understand the factors influencing the tick population under study and the reasons which may make it locally different from the tick populations developing elsewhere under different climatic regimes. This study is also important for a better knowledge of vector–pathogen relationships during individual stages of tick development and in different habitats or seasons.

Some microhabitats are virtually insulated from the effects of local climate, e.g., caves, especially ice caves in the temperate zone during the summer season. In this case the microclimate can be categorized as a cryptoclimate which has its own laws which operate independently of changes in the meso- and macroclimate.

2.2. Habitat

The term habitat has been used in the field of autoecology to characterize the place of occurrence of a certain species. At present, the term habitat is often used as a synecological concept, synonymous with biotope. The habitat can be defined as a place with characteristic living conditions, inhabited by particular types of organisms (plants, animals and microorganisms) with characteristic interrelationships. Collectively, this set of organisms is called a community or biocenosis. The biocenosis including all populations (and their cohorts), together with the abiotic environment, acts as an ecological system

or ecosystem (="geobiocenosis") (Odum, 1971). Metabolism and energy transfers between its biotic and abiotic components take place in this system.

The living conditions provided by individual habitats depend on their characteristic physical and chemical properties, especially the climate. Every habitat is locally more or less demarcated: it can be represented, e.g., by a pond, moorland, field, or wood. As concerns the size, the term habitat is not exactly defined in the ecological literature and different authors use it in different senses.

The habitat of every tick species is the place that provides suitable living conditions, within the respective community, not only for the tick itself but also for its hosts. Critical factors that control habitat suitability for ticks are: (1) an adequate blood source for the developmental stages available at the appropriate time in the life cycle (presence of hosts); and (2) favourable living conditions (particularly meteorologic factors) that enhance the survival and development of ticks and their hosts. The plant component is very important for the latter factor. On the one hand it is influenced by the local climate, while on the other it alters and modifies this local climate, creating the meso- and microclimatic conditions that are optimal for the ticks and their hosts. These relationships are pronounced in exophilic ticks and absolutely essential in the nidicolous species.

3. MICROHABITAT

The microhabitat can be defined as the smallest part of the habitat, which is the actual living environment of ticks and other small organisms, at least during a part of its life cycle, or at a time period of its living activity.

Microhabitat size varies in relation to the size of the organism, its ecological requirements and its mobility in the terrain. This definition suggests an autecological concept. However, the synecological attributes of the term must also be stressed, since it includes the links with other organisms within the community. This synecological concept is reflected in the interaction of ticks with other organisms in the community, e.g., the contact of ticks with their hosts in host-seeking stages, predator attacks on the tick resting stages, or tick infection with entomophagous fungi (Samšiňáková et al., 1974).

Because of their immense variety, only two general types of microhabitats are distinguished: (1) diffuse, dispersed without any spatial demarcation (e.g., litter layer on the surface of forest soil); and (2) restricted, which are clearly demarcated (this division corresponds with the classification of elementary foci of infections, see below). Diffuse microhabitats are mostly inhabited by exophilic ticks, while the restricted ones are inhabited by nidicolous ticks. The microhabitats created by man can include both groups.

3.1. Microhabitats of External (Exophilic) Ticks

No terrestrial habitat is homogenous as far as the abiotic factors and biocenotic composition are concerned. Thus, ticks are never evenly distributed. Rather,

the tick distribution shows a mosaic pattern. The sites where ticks are concentrated are determined by biotic and abiotic factors. In hygrophilic ticks (e.g., *I. ricinus*), these factors include the microrelief of the terrain (ravines, gorges, or shallow depressions of less than 1 m deep that provide increased soil humidity during a specific period) and abundant plant cover reflecting the increased soil humidity. This plant community influences, both directly and indirectly, the occurrence and population density of the ticks. The direct effect can be summarized as the transformation of microclimatological conditions which can favour or hamper the existence of ticks, their oviposition, development, duration of various life-cycle stages, survival, and mortality. The indirect effects are biocenotic in nature: the type and density of the vegetative cover offers shelter, resting places, and food for the tick hosts.

Ecotones are especially likely to support high tick densities. Odum (1971) defines an ecotone as a transition zone between two communities, such as between forest and meadow. Both species number and population density of many varieties of ticks are higher in the ecotone than in the adjacent communities. This ensures a wider range of potential tick hosts which in turn enhances tick concentration. One of the most important general ecotonal types is the forest margin, defined as a boundary between the forest versus the grass and shrub communities (Odum, 1971). Wherever humans settle, they maintain the communities of forest margins around their dwellings, e.g., if people settle in a forest, they transform it into a mosaic of dispersed forest areas alternating with meadows, fields, and other clearings. In open plains, however, they plant trees and, once again, create a similar landscape. In both cases, ecotones suitable for tick occurrence arise (Rosický, 1962). The phenomenon of mosaic distribution of ticks indicates that, although a suitable habitat is a necessary condition of tick existence (conditioned by climatic factors and host presence), it is not the only condition.

Horizontal migration is very limited in ticks. *Ixodes persulcatus* larvae can move to a maximum of 1.5 m from their hatching sites over a 2-week period; they become dispersed over an area of only 2.5–3.0 m^2 (Levin, 1985). *I. ricinus* exhibit a similar range of movements. Larvae labeled with [^{14}C]glycine migrated 0.5–2.5 m during 24 h and 60–80% of them did not migrate farther than 1.5 m. They were able to spread over the surface of a 1.0–5.5 m^2 area during the first 24 h and a 7.5–8.5 m^2 area in a 1-month period (Lebedeva and Filchagov, 1985). In *Ixodes dammini*, most larvae were found within 40 cm of the egg mass, but individual larvae migrated as far as 250 cm (Stafford, 1992). Larvae of this species indicate a tendency for aggregation around the oviposition site. They appear to use the ambush type of host-seeking behavior and, consequently, few actively disperse beyond 1–2 m. Arumova (1979) found radioactive *I. persulcatus* adults at a distance of 5.0–7.5 m from the site where labeled engorged nymphs had been released in the previous year. Although some adults migrated up to 40–50 m in a forest in South Primorye (western Russia) over a 50-day period, most specimens moved only 3 5 m. Similar observations were made with females of *Rhipicephalus turanicus* living in dry subtropical regions of Asia, which were found to move about 3–5 m, although

individual specimens were found as far as 10 m from their hatching sites after 15 days (Balashov, 1958, 1959). These data show that the mosaic-like distribution of ticks in the terrain is not strongly influenced by their active horizontal migration and that the ticks seek the most suitable microclimatic conditions within a rather narrow radius of horizontal movement. Nevertheless, some tick species inhabiting extreme biotopes are very mobile, which makes their survival possible. An example is the desert tick, *Hyalomma asiaticum*, which can migrate over distances of 400–500 m in a 30-day period. Most of these ticks disperse up to 50–150 m from their release sites (Balashov, 1967).

The spatial distribution of exophilic ticks is especially dependent on the distribution and migration of their hosts (Babenko and Arumova, 1985). An example of this relationship is that described by Wilson et al. (1990). These authors studied the movements of the white-tailed deer (*Odocoileus virginianus*) by means of radio-telemetry collars. They found that the average deer occupied a 0.25 ha quadrant in their Long Island, New York, study site. During the following summer they examined white-footed mice, *Peromyscus leucopus*, in the same locality for *Ixodes dammini* larval and nymphal infestations. The frequency of deer using the study area was positively correlated with the number of larval and or nymphal ticks per mouse. Similarly, Minshull and Norval (1983) found that the distribution of each developmental stage of *Rhipicephalus appendiculatus* in a park in Zimbabwe was determined primarily by the spatial distribution of ungulate hosts at the time of detachment of engorged ticks of the preceding stage. Fire influenced the distribution of all tick stages by increasing the density of grazing hosts on recently burned areas.

Habitat represents the actual living environment of tick parasitic stages, where they are exposed to the effects of the regional mesoclimate or macroclimate according to their ecological requirements and the activity of their hosts. For non-parasitic stages, however, the presence of microhabitats suitable for the development, survival and resting stages is essential. In *I. ricinus*, the optimal habitat in Central Europe is the deciduous forest, where the non-parasitic stages develop, hide, and survive in the superficial layers of soil or litter. Here they are greatly influenced by the microclimate of these microhabitats. Subsequently, when the ticks commence host-seeking activity, they use the ground-level layer of vegetation (plants or shrubs) as their microhabitats, where they are influenced by the microclimate of the ground-level air layer. In contrast, suitable habitats for the xerophilic *Hyalomma asiaticum* are the deserts and semi-deserts of South and Central Asia, where the ground surface temperature during the day may reach 70°C while the relative humidity may drop to only 20–30%. Under these conditions, adult ticks restrict their host-seeking activity at ground level to the early morning and evening hours while the developmental stages occupy more sheltered microhabitats, such as burrows, crevices, and other enclosures (Balashov, 1960).

The conceptual relation between the habitat and microhabitat is similar to that between the macroclimate (or mesoclimate) and microclimate, as discussed previously. The character of the habitat is decisive for the existence of ticks in a given locality, while the conditions of the respective microhabitats and

their microclimate influence their specific circumscribed population dynamics. Knowledge of local microhabitats and microclimate is necessary for a detailed analysis of existential factors determining the dynamics of the tick populations. Thus, the character of that part of the habitat in which the engorged tick drops from the host is a decisive factor for its success. Detached ticks which fail to shelter in a suitable microhabitat soon die or survive without further development. Detailed field studies (described below) show that there are differences between individual development stages of the same tick species, as well as differences due to the season of the year. This phenomenon becomes even more obvious when ticks are transported to new habitats, e.g., during the migration of its host in the mountains from the valleys to higher altitudes (Daniel et al., 1988).

3.2. Microhabitats of Nidicolous (Endophilic) Ticks

The same hierarchy of habitat and microhabitat relationship described for exophilic ticks is also valid for endophilic ticks. Their habitats and associated physical and biotic properties determine the basic features of tick existence and distribution, but this influence depends on the extent to which the ecological requirements of the host are met. In this case, the microhabitat of the ticks is the host's nest or burrow, where the tick finds a convenient source of blood and, at the same time, is under the direct influence of a very special microclimate affecting the community of the nest organisms. This influence is most pronounced in the nests of terrestrial mammals and birds situated in various cavities or on the ground.

The microclimate in these nests is determined primarily by: (1) the warm-blooded inhabitant of the nest; (2) the choice of nest site construction (the same species of warm-blooded host may construct its nests under the soil surface, on the ground or above the ground, see Daniel, 1988); and (3) the material used as litter. Also of great importance is the activity of the host, both directly (presence of animals in the nest, growth of their young, pollution of nests by urine and feces, etc.) and indirectly (decaying remnants of food, etc.). Finally, continuity of nest habitation, changes in host species composition, and other aspects of nest utilization by warm-blooded hosts also influences the microclimate.

The main abiotic factors (discussed in detail below) are the temperature and humidity of the immediate environment (ground air layer, soil in the vicinity, decaying wood, etc.), and the isolation of the nest. On the one hand, the microhabitat of the nest and its community of living organisms represents, to a considerable extent, a closed system separated from the surrounding habitat not only spatially but also by certain physical factors; on the other hand, the character of the nest environment and its changes are closely related with its natural surroundings, which may undergo dynamic changes caused by external factors, e.g., different seasons.

An example of these tick–host microhabitat associations is found in the

life cycle of the tick *Ixodes laguri*, which infests the nests of *Citellus* spp. These sheltered nest microhabitats have an inner structure differing in some microclimatic details. In general, such nest environments are radically different from the surrounding environments. In contrast, a very different situation is found in exposed birds' nests constructed on the ground, in shrubs or often high in tree tops. Such nests, which are only used seasonally, are only partially isolated from the surrrounding environment. A detailed description of the microclimate of this type of nest and the interaction between this microclimate and the nest inhabitants was presented by Pikula (1978, 1979).

3.3. Specific Microhabitats

A very specialized type of microhabitat used by ticks is the underground spaces, either natural (caves, rock cracks, chasms) or man-made (cellars, galleries, bunkers). The microclimate in these species is relatively stable during the whole year. Since they are dark, there is no photoperiod. The humidity is high, reaching up to 90–100% relative humidity (RH) due to undergound water. According to the temperature, three types of caves can be distinguished. (1) Ice caves of the temperate zone, which are characterized by a small entrance situated high on the hillside of a canyon or valley. The pocket-like shape of these caves, the slope to the bottom of which is very steep, facilitates entry of cold winter air but prevents penetration of the warm summer air. Due to their poor air circulation and residual ice, some of which is believed to be a remnant of the last glacial period, the temperature inside the caves is about $-5°C$ in winter and about 0°C in summer. (2) cold caves with a large entrance and a rather gradual, sloping bottom, which are characterized by temperatures that are somewhat lower or higher than those of the outer environment, depending on the season of the year. In the temperate zone, the temperatures range between 15 and 20°C in summer, but rarely drop below zero in winter. In the tropics and subtropics, the temperature in such caves may even rise above 20°C. (c) Hot caves of the subtropics and tropics (on the American continent in particular), the entrance of which is rather small, and in which the main space is separated by a narrow, sinusoid corridor preventing normal air circulation. Owing to the concentration of a large number of fructivorous and insectivorous bats (e.g., *Phyllonycteris poeyi* in Cuba), there is a large amount of guano and the temperature rises to 37°C. The air humidity is about 100% RH and the atmosphere is strongly saturated with ammonia.

Subterranean sites are almost exclusively inhabited by troglophilous tick species parasitic on bats. In cold caves, one finds, e.g., *Ixodes vespertilionis* or *I. simplex*, which develop at 10–15°C. Their population density is very low. No behavioral diapause occurs in these ticks which are frequently found to feed on hibernating bats in winter. In contrast, the hot caves are mostly inhabited by species of the genera *Antricola* and *Parantricola* or other soft ticks, e.g., *Ornithodoros viguerasi*, with extremely high population densities. Černý (1967) reported densities of *Parantricola marginatus* exceeding 2,000 specimens/m^2 in

a 2 cm layer of guano in some parts of a cave in Cuba. The number of bats present at that time inside the cave was estimated to be about 150,000. Declining bat numbers over time resulted in a gradual decrease in both air temperature and tick density, until the ticks disappeared completely (Daniel et al., 1981).

Microclimatically specific microhabitats serve as synanthropic tick shelters in building lofts. These microhabitats are usually secondarily inhabited by dendrophilous or cavernicolous tick species introduced there by their hosts. As a result of the extreme conditions in these microhabitats, the tick inhabitants are almost exclusively soft ticks. Filippova (1966) reported that the air temperature in shelters of the bat parasite *Argas vespertilionis* in the lofts reached 40°C and more in summer months. Due to the relatively high transition temperature of its cuticle (54°C), *A. vespertilionis* can maintain body water balance even under these conditions. Since the hosts periodically leave these synanthropic shelters in the winter season (migrating birds, bats), the ticks must survive a long period without any food source during their diapause. Another interesting microhabitat consists of the dry leaves of the palm, *Copernicia vespertilionum*, which serve as shelters for the Cuban bat tick, *Ornithodoros tadaridae*, and its hosts. A similar situation occurs in sea-bird nests. An example is a rocky cliff near Cuba where *Ornithodoros denmarki* was found hidden in limestone cavities and coral rocks near nests of *Sterna fuscata* and *Anous stolidus*. There were 387 specimens of *O. denmarki* hidden in the cavity ($3 \times 2 \times 1.8$ cm) of a small stone situated on the rocky surface and warmed by the tropical sun.

4. MICROCLIMATE

The microclimate is characterized by abiotic factors and represents the actual living environment of ticks. Nosek (1978) stressed its importance by calling it the "ecoclimate of ticks." Most papers dealing with the immediate influence of microclimatic changes are based on laboratory experiments studying the effect of individual factors on the tick specimen, particularly its life cycle, survival, and behavior. Field studies of the microclimate are few and mostly deal with just a small part of the tick's life history. This is caused by difficulties in the methods of study due, among other reasons, to the long development time of the ticks and the periodic alternation of non-parasitic and parasitic stages. These difficulties begin during work in the field and continue during the evaluation of results, including a large number of partial results (biotic and abiotic, often not comparable) from which generalizations must be drawn while simultaneously maintaining maximum precision in data collection.

4.1. Influence of Microclimate on an Individual Tick

The ticks are exposed to the influence of many abiotic factors decisive for their behavior, development, and survival in nature. These factors include sunlight,

photoperiod, temperature, precipitation, air relative humidity, wind and air circulation, concentration of gases, particularly CO_2, air pressure, the earth's magnetic field, solar flares, and perhaps other unknown variables. In the natural environment of the ticks, none of these factors acts in isolation. Lower temperatures are usually accompanied by increased air humidity while low air pressure follows precipitation and cooling. Increased air circulation and temperature decrease the relative air humidity. In the northern hemisphere, the low daily temperatures common in winter are accompanied by short day lengths. Some factors, such as temperature and air humidity or photoperiod, seem to be more significant than others for some tick species, and they may be decisive for their survival, reproduction or behavior. The same factors, however, are less significant for other species. Only the recent transfer of laboratory experiments to the field showed the functional complexity of all these factors influencing various activities and processes in ticks.

4.1.1. Influence of Microclimate on Tick Behavior

The activity of unfed ticks is very strongly influenced by the microclimate of the microhabitat conditions both in argasid and ixodid species. Because changes in microclimate are directly dependent on the time of day and season of the year, the changes in tick behavior show both circadian and circannual rhythms. Leonovich (1989) distinguished three main behavior patterns (excluding sexual behavior) and ten other activities or phases in adult *Ixodes persulcatus* tick behavior off the host. Two of these patterns, (1) the tick's arrival into a contact zone and (2) the tick's excitation by signals given by the potential host, are associated with host-seeking activity; the third (3), shelter-seeking behavior, is a defense reaction to unfavorable microclimate conditions. As the daily rhythms are generally considered to be influenced by exogenous signals, the changes in solar radiation, temperature and humidity appear to be the primary exogenous stimuli provoking tick activity responses (Belozerov, 1982). Recent investigation of tick behavior in the field demonstrated solar radiation to be one of the most decisive factors influencing the daily activity rhythm of some ticks. The host-seeking activity of *I. ricinus* and *I. persulcatus* unfed nymphs increased during the evening hours (4–12 p.m.) in the Moscow Region of Russia. Moreover, the number of questing ticks was correlated with solar radiation, but not with air temperature (Babenko, 1974). Host-seeking activity of adult *Dermacentor marginatus* in the Armenian steppe region was also found to be influenced by the daily light cycle. Observations revealed that the highest percentage of ticks (56–85%) recovered from the upper parts of stems were seen in the evening hours (7–8 p.m.) during August and September when air temperatures and RH values were at 12–17°C and 50–70%, respectively (Rukhkian, 1987). Host-seeking activity of the American dog tick, *Dermacentor variabilis*, larvae and adults was also correlated with the intensity of solar radiation, but not with temperature (Atwood and Sonenshine, 1967).

The daily rhythm of locomotor activity, host seeking, feeding patterns, and drop-off of ticks is influenced by photoperiod, temperature, and the interaction

of these two factors. These tick activities are precisely co-ordinated with rhythms of activity and other biological processes of the host (Balashov, 1967). Many exophilic ixodid ticks, e.g., *I. ricinus*, *I. persulcatus*, *Haemaphysalis longicornis*, *Dermacentor variabilis*, and others parasitizing pasture animals with diurnal activity rhythms have a corresponding diurnal drop-off rhythm. In nest-dwelling ticks, the drop-off rhythm may be either nocturnal, as is the case when hosts are active during the day and shelter in the nest at night (*Argas persicus*, *A. reflexus*, *Ixodes texanus*, etc.), or diurnal, when hosts shelter during the day and are active at twilight or night time (*Argas vespertilionis*, *Ixodes hexagonus*, *Haemaphysalis leporispalustris*, etc.) (Belozerov, 1982). However, Matuschka et al. (1990) recorded the detachment of *I. hexagonus*, a parasite of hedgehogs and foxes, even during the scotophase. These nocturnally detaching ticks focus their feeding on nocturnally active hosts, in spite of the possibility that such behavior might cause them to disperse from the nests of the hosts. Dispersion is provided, however, by the tendency of these ticks to detach while their host naps. Although many signals of host behavior (e.g. locomotor activity, grazing, etc.) and daily physiological changes in host metabolism influence tick detachment, the exogenous photoperiodic stimuli were recognized to be the leading factor for tick drop-off rhythm. The photoperiodic stimuli seem to be the leading exogenous signals entraining and regulating endogenous circadian mechanisms of drop-off rhythms (Belozerov, 1982).

There is strong evidence in the literature on the influence of temperature on the locomotor activity of unfed ticks and the correlation between tick activity and air temperature. Kheisin (1953) stated that *I. persulcatus* adults are capable of active movement at temperatures as low as 0.3°C. Their locomotor activity increases proportionally with temperature, reaching a maximum of 36.5 cm/min at 23°C (Babenko and Arumova, 1985). MacLeod (1935a) reported that 11°C was the temperature threshold for locomotor activity for adult *I. ricinus* in Britain. Normal activity of adults of Middle European populations of these species occurs in the range of 18–25°C at a relative humidity of 100%, although limited activity begins with temperatures as low as 5–15°C. The normal activity of *Ixodes ricinus* nymphs occurs between 10 and 22°C and that of larvae between 15 and 27°C (Nosek, 1978). However, lethargy, resting, as well as varying levels of activity and escape reactions occur in all three developmental stages at very similar temperature ranges. Shiraishi et al. (1989) demonstrated that the cattle tick, *Haemaphysalis longicornis*, became less active when the noon ambient temperature declined to 11–15°C.

The minimum activity threshold temperature for *Dermacentor variabilis* adults is 5°C (Hall and McKiel, 1961). For this species, temperature seems to be the decisive factor influencing the questing activity. In Ohio, USA, air temperatures (especially the temperature at 1 m above ground level), or interactions involving temperatures, were reported to be the most important factors controlling adult *D. variabilis* questing in analyses of 15 micro-meteorological parameters, including temperature, moisture, wind, and solar radiation (Harlan and Foster, 1990).

Daily fluctuation of atmospheric humidity is another decisive factor influencing the behavior and activity of many tick species. *I. ricinus* requires high humidity for survival and its daily movements are strongly dependent on relative humidity. In Karelia (Russia) the greater part of females migrate up and down the vegetation daily in June, July, and August, when the climate is hot and dry. In September, when the climate is colder and more humid, they remain on the top of vegetation for several days (Luta and Schulman, 1958). In Scotland, where the humid oceanic climate does not rapidly impair the tick's water balance, the ticks remain on the top of vegetation for 9 days on the average (Lees, 1951). Similar findings were made in the former Czechoslovakia (Boučková and Dyk, 1968; Dyk and Boučková, 1968) and Ireland (Gray, 1982).

Many variations in the daily rhythm of tick activity have been described. Such variations are influenced by the developmental stage and season of year both in ticks with diurnal or nocturnal activity. This reflects the adaptations of each species or developmental stage to the local climatic and microclimatic conditions. These differences often correlate with temperature and humidity preferences, which differ strongly in different tick species. Daily movement of unfed ticks on vegetation repeatedly occurs in direct response to the contemporary influence of several microclimatic elements, such as temperature, humidity, or solar radiation. In Oklahoma an increasing percentage of lone star ticks ascend vegetation as the day progresses in late May; the percentage of ticks on vegetation remains about the same throughout the day in June, and a greater percentage of ticks ascend the vegetation during the early morning and late evening hours in July (Semter and Hair, 1973). In Zimbabwe, larvae and nymphs of *R. appendiculatus* and larvae of *Boophilus microplus* ascend the vegetation daily, but larvae of *Boophilus decoloratus* and adults of *R. appendiculatus* do not migrate vertically during the hot and rainy seasons (Short et al., 1990).

Temperature, humidity, and solar radiation influence tick locomotor activity and behavior as they search for optimal microclimatic conditions. However, the degree to which they influence behavior and the hierarchical importance of these abiotic factors on behavioral patterns appears to be different in different tick species. Robertson et al. (1975) considered temperature to be greater than either relative humidity or photoperiod in its effect on adult lone star tick, *A. americanum*, activity in Oklahoma. On the other hand, Lane et al. (1985), using regression analysis revealed that temperature, relative humidity, and solar radiation, either singly or in combination, usually did not explain a significant amount of the total daytime variation in questing number of the Pacific coast tick, *Dermacentor occidentalis*, on vegetation. Study of the behavioral activity of *Rhipicephalus bursa* larvae in Spain revealed that ground temperature and sunlight intensity, but not relative humidity, had the greatest effect on the tick population (Estrada-Pena and Sanchez-Acedo, 1988). Larvae confined in experimental tubes moved daily or remained at the tips for several days at a time, depending on temperature and light intensity. Their mobility was stimulated between 19 and 24°C even when the air humidity varied between 53.0 and 90.8% RH. Their activity model for this species agrees with that

proposed by Sonenshine (1977) working with *Dermacentor variabilis* in Virginia: the progressive increase in solar intensity and temperature leads to an increase of mobility of ticks which migrate to the tops of vegetation. Ticks depart from this exposed position either when those parameters decrease or when the temperature and humidity distort the tick's water pump in such a way that they are unable to remain there. Therefore, under cold and humid conditions, light intensity plays an important role, while for hot and dry conditions, humidity is the dominant abiotic influence on locomotor activity.

4.1.2. Influence of the Microclimate on the Developmental Cycle of Ticks

Numerous reports have described the influence of solar radiation, temperature and humidity of the developmental cycle of ticks. Overall, the most important factor seems to be photoperiod, which has an impact on the mechanisms regulating diapause in ticks and the length of their developmental cycle. There are two basic types of responses to photoperiod expressed by most ticks: (1) long-day (LD) responses, which initiate active development; and (2) short-day (SD) responses, in which development is accelerated when perceived by the ticks (Belozerov, 1982). In addition, a more complex, two-step photoperiodic response is also known in certain ticks (Zaslavsky, 1972, 1975); however, in this case, development proceeds only after certain increases or decreases in day length. The LD or SD reaction and the one- or two-step reactions vary in different tick species, developmental stages or populations. Oogenesis, for instance, is regulated by the SD reaction in *I. ricinus*, *Dermacentor marginatus*, *D. reticulatus* and *Ornithodoros gurneyi*. In contrast, it is regulated by the LD reaction in *Hyalomma anatolicum* or *Argas arboreus*. Larval development is accelerated by the LD period in *I. ricinus*, *I. persulcatus* and *I. trianguliceps*, but by the two-step transition from SD to LD in *I. kazachstani*. This two-step photoperiodic reaction also occurs in the development of nymphal *I. ricinus* (Belozerov, 1982). The development of this species is strongly influenced by photoperiod rather than by temperature, humidity and food quantity, which Loew (1964) considered to be of secondary importance. The minimal induction of daily photoperiod ranges from about 14 h light and 10 h darkness in connection with a minimal light intensity of 5 lux. Larvae can be adapted partly to full darkness in the laboratory, but nymphs are unable to metamorphose under a shorter light exposure.

Temperature seems to be another limiting microclimatic factor influencing tick development. The physiological events in the tick's body can continue only in the framework of limited temperature; outside of this framework, further development is stopped. To evaluate the influence of temperature on insect or tick development, the terms **developmental zero** and **thermal constant** were established. Developmental zero, also known as **physiological zero** or the **developmental threshold**, is defined as a threshold temperature below which the ontogenic development of the species does not proceed. The sum of effective temperatures, i.e., the mean daily temperatures exceeding the developmental zero expressed in degree-days, has been termed the **thermal constant**. According

to MacLeod (1935b) the thermal constant is not a constant value, but increases with higher temperatures. The effective degree-days are therefore related to the lowest temperature values. These values vary greatly among different tick species in relation to their adaptation to cold or hot climates and appear to be one of the most important factors limiting their geographic distribution. In the thermophilic species *Argas persicus*, the developmental zero for egg embryonation is 20°C; incremental increases in temperature up to 40°C have been reported to accelerate development. At 22.5–32.5°C almost 100% of engorged females oviposited. However, at 35–40°C oviposition was interrupted and continued only after 1–2 weeks, when the temperature was reduced to 27.5°C. Paralysis appeared in females at a temperature of 45°C after 3–4 days (Frolov and Dazhiev, 1970). Temperature also influenced the number of nymphal stages of this and other species of argasid ticks (Balashov, 1963). At 20°C, 37% of *Argas persicus* stage II nymphals molted to adults versus 63% molting to nymphal stage III; 17% of stage III molted to nymphs IV. At 25°C, 90% of stage II molted directly to adults versus only 10% to nymphs III; stage IV nymphs no longer appeared. At 30°C, 3% of stage I molted directly to adults and 97% to nymphs II. Also, a majority of stage II nymphs molted to adults and only 1% to nymphs III. In addition, higher temperature can also positively influence the expression of autogeny in unfed mated females, e.g., in *Ornithodoros parkeri.* In this species autogeny occurs at a lower temperature (21°C) only among lower weight females (11–20 mg), while at a higher temperature (29°C), unfed mated females of heavier weight groups also oviposited (Pound et al., 1984).

In *Hyalomma dromedarii*, one-, two-, and three-host developmental patterns occur, with the two-host type predominating. Alternations of these developmental types are considerably influenced by microclimatic conditions and by the choice of hosts (Berdyev, 1974). This author (1969) observed a transition from one type of developmental cycle to another depending on the air temperature in Turkmenia, where all active developmental stages were feeding on camels. With rising temperature the one-host type changed to the two-, and three-host type of development. At high temperatures, *Hyalomma dromedarii* always develops in a three-host cycle, but this changes at low temperatures or when the tick feeds on atypical hosts (Bouchalová et al., 1977).

The development of many tick species is also influenced by humidity and can be stopped when ambient humidity conditions are unfavorable. In some hydrophilic species, the most humidity-sensitive developmental phases seem to be the period of embryonic development and larval eclosion. Thus, eggs of *I. persulcatus* did not hatch even at 85% RH and a temperature of 20–25°C. Shashina (1985) considers 87% RH to be the critical RH level for that species, e.g., the humidity in which 50% of the larvae hatch (Sonenshine, 1970). Egg hatching in *Ixodes ovatus* occurs only at 95–100% RH and 20–25°C (Fujimoto, 1990). On the other hand, some tropical ticks seem to be more resistant to low humidity. The eggs of *Rhipicephalus simus* and *Haemaphysalis spinigera* hatch normally inside the burrows of the Nile grass rat at 26–28°C and RH 85%. However, in the laboratory, 20% of eggs hatched even at 65% RH and 1%

hatched at 33% RH (Hussein and Mustafa, 1987). Humidity factors can also influence the number of eggs laid, the number of ovipositing females, the duration of oviposition or molting of the developmental stages in ixodid ticks (Hussein and Mustafa, 1987; Yano et al., 1988; Fujimoto, 1990). However, in the experiments of Ekpenyong and Akinboade (1991), different humidity regimes ranging from 0 to 100% RH at 25°C, did not seem to have any effect on the preoviposition, oviposition or pre-eclosion periods of *Amblyomma variegatum*. In *Dermacentor nitens*, the relative humidity (40, 61, 75, and 91% RH) had little effect on length of pre-oviposition and oviposition periods, but it strongly influenced the egg hatch (Despins, 1992). A high percentage egg hatch was observed in a low saturation deficit environment and this decreased when saturation deficit was increased.

Argasid ticks are generally no more xerophilic than ixodid ticks. Their developmental cycle is much less sensitive to low humidity because of generally better morphological and physiological adaptations to dry microclimates (Filippova, 1966). Egg mortality in *Argas reflexus* and *A. polonicus* was only 28.8% and 22.6%, respectively, at 10–30% RH. Hatching of normally developed larvae ranged from 77.5 to 78.0% at 50–75% RH in *A. polonicus* and from 66.6 to 67.2% at 30–50% RH in *A. reflexus* (Siuda, 1981; Buczek, 1988).

4.1.3. Influence of Microclimate on the Longevity of Ticks

The remarkable longevity of ticks in the absence of suitable hosts depends primarily on the ambient air humidity. The ability of ticks to maintain their water balance as well as other metabolic functions, is strongly dependent on temperature. Increasing temperatures increase cuticle water permeability and accelerates the loss of water even before the sudden increase that occurs when the transition temperature of the epicuticular lipids is reached. This transition temperature is the temperature at which water permeability of the cuticle increases abruptly due to changes in the structure in molecules of cuticular lipids (Lees, 1947; Hafez et al., 1970; Hackman and Filshie, 1982; Knülle and Rudolph, 1982). In some tropical ticks, such as *Ornithodoros savignyi* or *H. dromedarii*, however, the lethal temperature was found to be only a few degrees below their transition temperature (Hafez et al., 1971). Thus these ticks are not exposed to the danger of rapid drying by excess evaporation of water through the cuticle during a temperature rise to lethal rates. The transition temperatures of xerophilic ixodid ticks (and most argasid ticks) are higher and their cuticular waxes more waterproof than most ixodids. Needham and Teel (1986), examining the data of Hafez et al. (1970), on the effect of temperature on the cuticular permeability to water in *O. savignyi* and *H. dromedarii* confirmed that between 30 and 50°C, the ixodid integument was about 72 times more permeable than the argasid's. As apparent from studies of Hunt (1986) and Estrada-Peña and Dusbábek (1993) on three taxa of the genus *Amblyomma* and two species of the genus *Argas* (*A. polonicus*, and *A. vulgaris*) one also finds considerable differences between argasid and ixodid cuticular hydrocarbon patterns. The cuticular hydrocarbon pattern in hard ticks is characterized primarily by the

presence of *n*-alkanes and a limited number of alkenes. Argasid cuticles, however, are characterized by a higher diversity of n-alkanes as compared to the hard ticks and the complete absence of any alkenes. The long-chain hydrocarbons typical of soft ticks occur also in some arthropods inhabiting microclimatically extreme habitats, e.g., the desert scorpions (Estrada-Peña, unpublished).

It has been suggested that low winter temperatures may play an important role in determining the mortality rate of *I. ricinus* larvae and nymphs that overwinter in the engorged state (Dusbábek et al., 1971; Daniel et al., 1974, 1976, 1977a; Gray, 1981). The low temperature characteristics of the tick's water vapor pump, interacting with the winter temperature, will control overwintering survival and thus play a major role in the regulation of the average population size, variations in population size, and the species range. The winter temperature determines the geographic limits and the winter survival microenvironment for *D. variabilis.*

Low autumn and winter temperatures and their daytime variations seem to be one of the limiting microclimatic factors preventing tick survival in new biotopes when they are introduced by migrating birds or during the transport of domestic animals. Daniel et al. (1977a) described this phenomenon in his studies from South Moravia (Czech Republic). Eggs of *Hyalomma anatolicum excavatum* laid in June were unable to hatch, while females of *Rhipicephalus sanguineus* placed in the field in August did not oviposit at all. However, specimens of the same species introduced by a dog to a town flat in Warsaw after returning from a trip to the Balkans were able to survive in this microclimate for several months (Szymanski, 1979).

The microclimatic character of the microhabitat influences strongly the summer longevity and survival of different developmental stages of ticks. The negative influence of insolation and associated high temperatures accompanied by reduced relative humidity reach their maxima in the northern temperate zone in summer. These extreme conditions can be devastating for development of *I. ricinus* in exposed grassy areas (i.e., meadows) in South Moravia (Daniel et al., 1977a). The highest losses caused by microclimatic factors occur in the larval stage. Only 5.1% of engorged *I. ricinus* larvae survived between June and July in the warm, dry meadow microclimate as compared to 46.3% which molted in the cooler, more humid oak forest microhabitat. A similar condition occurs in Japan, where *Ixodes ovatus* larvae and nymphs are less tolerant to desiccating conditions than unfed adults (Fujimoto, 1990). On the other hand, adults of *Dermacentor reticulatus* survived better in the meadow microclimate than in the forest microhabitat. About 54.6% of unengorged females and 57.8% of males survived for 399 days in the meadow (including two periods of hibernation), while only 32.5% of females and 34.3% of males survived in the forest habitat in South Moravia (Černý et al., 1982). Similarly *R. sanguineus* seems to be a temperature- and desiccation-tolerant species. Unfed nymphs survive at 35% RH for 6 weeks at 20°C, 4 weeks at 30°C and even 2 weeks at 35°C. Unfed adult ticks are able to survive at 27°C for 4 weeks at 15% RH, 12 weeks at 35% RH, 16 weeks at 55%, and 28 weeks at 90% RH (Koch and Tuck, 1986).

Death of unengorged ticks at suitable temperatures is caused mainly by

desiccation. Therefore, longevity depends greatly on the ticks' ability to maintain their water reserves in a subsaturated environment. Under optimal conditions, an equilibrium exists between the tick body and ambient humidity wherein water loss and water gain are balanced (Knülle, 1966). The threshold humidity below which the ticks continuously lose water is known as the **critical equilibrium humidity** (CEH) (Knülle and Wharton, 1964). This threshold humidity varies between 75 and 94% RH for the majority of tick species and can differ also in different developmental stages or sexes of the same species. In xerophilic tropical ticks, the CEH lies at the bottom of this range, while in nest dwelling and forest ticks it is close to 90% RH (Knülle and Rudolph, 1982). It is higher in the larval and early nymphal stages. The CEH also increases with starvation and age. The CEH of unfed female *I. ricinus* has been estimated to be 86–96%. At 95% RH and 25°C, females survive for 3 months or more. At 70% RH and 25°C, they lose 5% of their original weight daily and die within 4–8 days. At 0% RH (25°C), they lose up to half of their body weight *per diem* and survive only 1 or 2 days. Under the same conditions (0% RH and 25°C), the Rocky Mountain wood tick, *D. andersoni* (CEH 86–90% RH), loses about 1–3% of its body weight daily and survives for 27 days (Lees, 1946), while the xerophilic *Ornithodoros moubata* (CEH 82–90% RH) survives at 0% RH (32.2°C) more than 96 days (Walton, 1960).

Many argasid ticks are very resistant to dehydration and are able to survive for many years, e.g., 11 years for *Ornithodoros papillipes* and up to 10 years for *Alveonasus lahorensis* (Pavlovsky and Skrynnik, 1960).

4.2. Influence of Microclimate on Populations of External (Exophilic) Ticks

The tick *I. ricinus* provides a useful model for studying the influence of microclimate. The life cycle of this species was studied during a three-year field experiment under the conditions of the South Moravian thermophilic oak forest in the Czech Republic (Daniel et al., 1976, 1977a). The study comprised three microhabitats situated close to one another, each with a distinctive microclimate, namely: (1) inside the thermophilic oak wood; (2) at its margin; and (3) in the adjoining open meadow. Microhabitat temperature and relative humidity were continuously recorded. The process of hibernation was studied simultaneously in four soil layers, i.e., on the surface and at the depths of 10, 20, and 30 cm. A total of 30,586 engorged larvae were placed in special microcages in these habitats; 9,424 unfed nymphs and 2,582 adults developed from them. These results are summarized in Figs 5.1–5.3.

4.2.1. Specific Features of the Life Cycle of Ixodes ricinus *Influenced by Microclimate of Different Microhabitats*

An average of 76.3% of larvae originating from eggs laid in the spring survived the winter period and that survival in the different habitats differed significantly

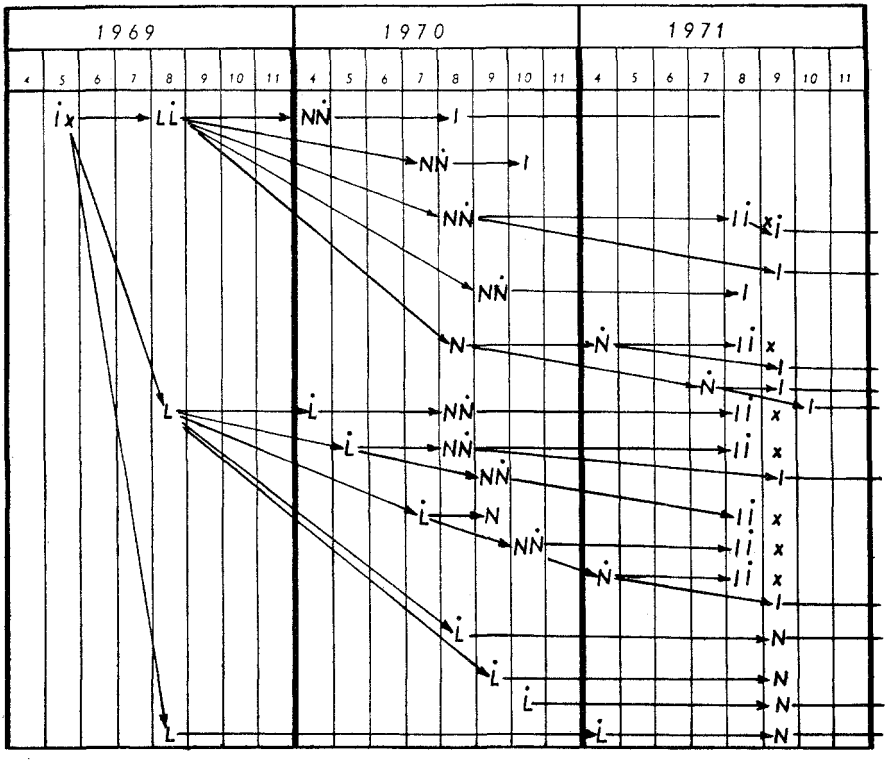

Fig. 5.1. Scheme of the development of the tick *Ixodes ricinus* in the forest habitat, depicting particular alternatives (after Daniel et al., 1976). L, N, I = unfed stages; L̇, Ṅ, İ = engorged stages; x = oviposition.

($P < 0.001$). Larval survival was lowest in the meadow (71.9%), and highest at the forest margin (83.6%). The most favorable soil layers in the forest and its margin were at depths of -10 to -20 cm, as compared to a depth of -30 cm in the meadows. The least favorable conditions in the forest and its margin were on the surface and at a depth of -20 cm in the meadow ($P < 0.001$). Some of the overwintering engorged larvae had already molted into nymphs at the beginning of the next spring when the first sample was examined. Most (4.1%) of the emerging nymphs were found in the meadow; few (0.1%) were found in the forest. These results demonstrate the importance of microclimate for the ticks and the protective value of the forest during the growing season. While the meadow habitat had less favorable conditions in the winter, its higher spring temperatures fostered more rapid tick development earlier than in the forest habitat. Overall, only 5.1% of all larvae molted successfully into nymphs during the spring and summer months in the meadow, versus 49.6% in the forest margin and 46.3% in the forest interior ($P < 0{\cdot}001$). This conclusion is also supported by a statistical assessment of the total annual production of nymphs from post-diapause larvae (in the engorged state) which showed significant differences ($P < 0.001$) between the different habitats. Tick development was

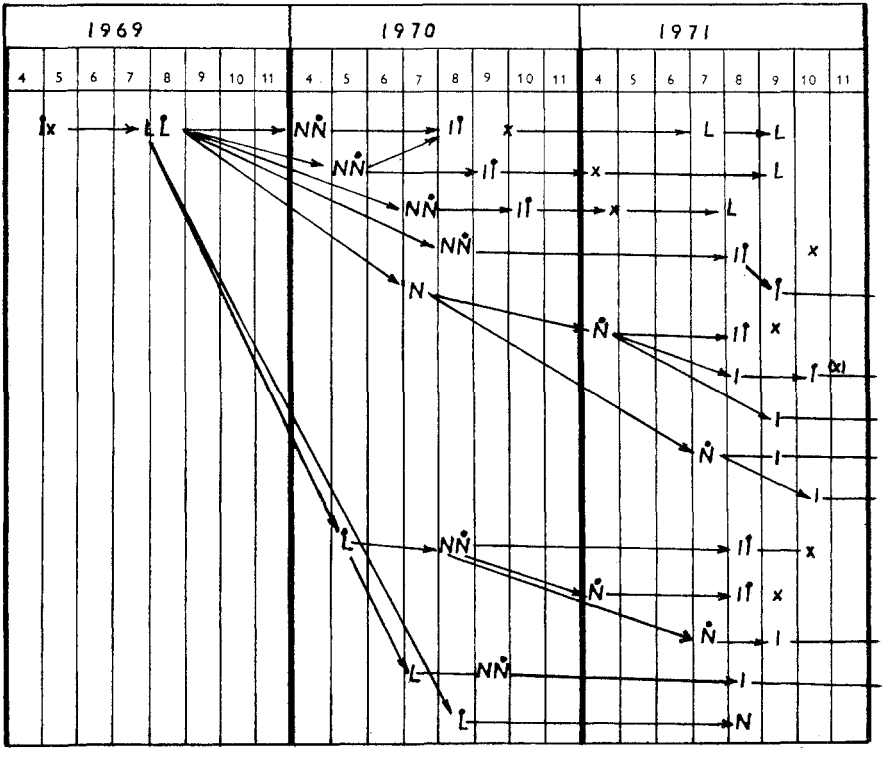

Fig. 5.2. Scheme of the development of the tick *Ixodes ricinus* in the ecotone of forest margin, depicting particular alternatives (after Daniel et al., 1976). L, N, I = unfed stages; $\dot{L}$, $\dot{N}$, $\dot{I}$ = engorged stages; x = oviposition.

most successful in the forest margin (42.2%), less so inside the forest (34.2%) and least in the meadow (only 7.6%). The negative influence of insolation, accompanied by high temperatures and low relative humidity, which reached their maxima during June and July, is clearly evident from these results.

Similar results were obtained with larvae which overwintered in the unfed state and fed in April of the following year. Again, ticks in the meadow were active earlier and capable of further development ($P < 0.001$). Highest losses in the meadow habitat were evident by late spring or early summer. In other months, habitat differences were not so pronounced. Engorged nymphs overwintered in all three habitats in a similar manner. Significant differences were found only among nymphs overwintering as unfed specimens and feeding in the following year. In this case, the nymphs from the forest were more successful as compared to those from the forest margin ($P < 0.001$); this category was not represented in the meadow. We also evaluated nymphal development over several seasonal periods. We found that the least significant losses occurred in the forest biotope. Nymphal feeding was most successful ($P < 0.001$) in this habitat.

In summary, starting with about 7,000 engorged larvae in each biotope, we observed development of: (1) 125 adults (51 males and 74 females) in the

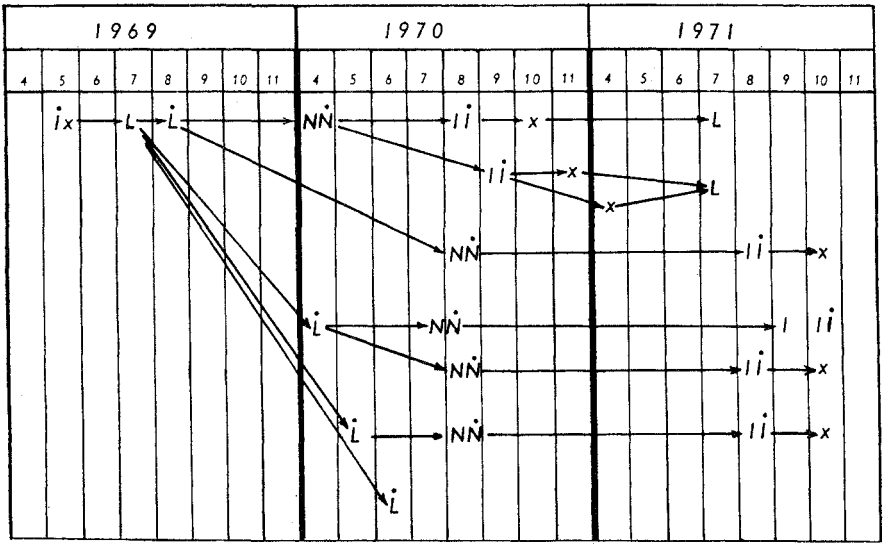

Fig. 5.3. Scheme of the development of the tick *Ixodes ricinus* in the meadow habitat, depicting particular alternatives (after Daniel et al., 1977). L, N, I = unfed stages; $\dot{L}$, $\dot{N}$, $\dot{I}$ = engorged stages; x = oviposition.

meadow; (2) 688 adults (327 males and 361 females) in the forest margin; and (3) 767 adults (376 males and 391 females) in the forest interior. The greatest losses in the larval–nymphal development in the meadow biotope occurred during the months of June and July. These results show that the grassy meadow is the least suitable habitat for *I. ricinus* development. Although development was accelerated in all phases in this habitat (a higher percentage of the experimental population completed the whole cycle in 2 years), it was characterized by higher losses during the larval to nymphal molt (Černý et al., 1974).

4.2.2. *Specific Features of the Microclimate in Different Microhabitats under Study and their Comparison*

The experiment described above dealt with microhabitats situated at distances of tens of meters from one another. Therefore, macroenvironmental changes were similar, which, in turn, enabled us to evaluate specific changes in their microenvironment. These changes were measured continuously by electroresistance thermometers and hygrometers containing bead thermistors; recordings were made at 20 s intervals during the whole three-year experiment. An example is the data for June and July of the second year of study (compare Fig. 5.4 and Fig. 5.5). As we have noted previously, there was a drastic reduction in ticks, particularly at the larval to nymphal transition, in the meadow microhabitat. Meteorological data for the three habitats, forest, forest–meadow ecotone and meadow, are summarized in Tables 5.1–5.4 and Figs 5.4–5.6 for the critical early summer period (1 June–31 July) and the second half of the summer (1 August–30 September). In addition to values measured at ground

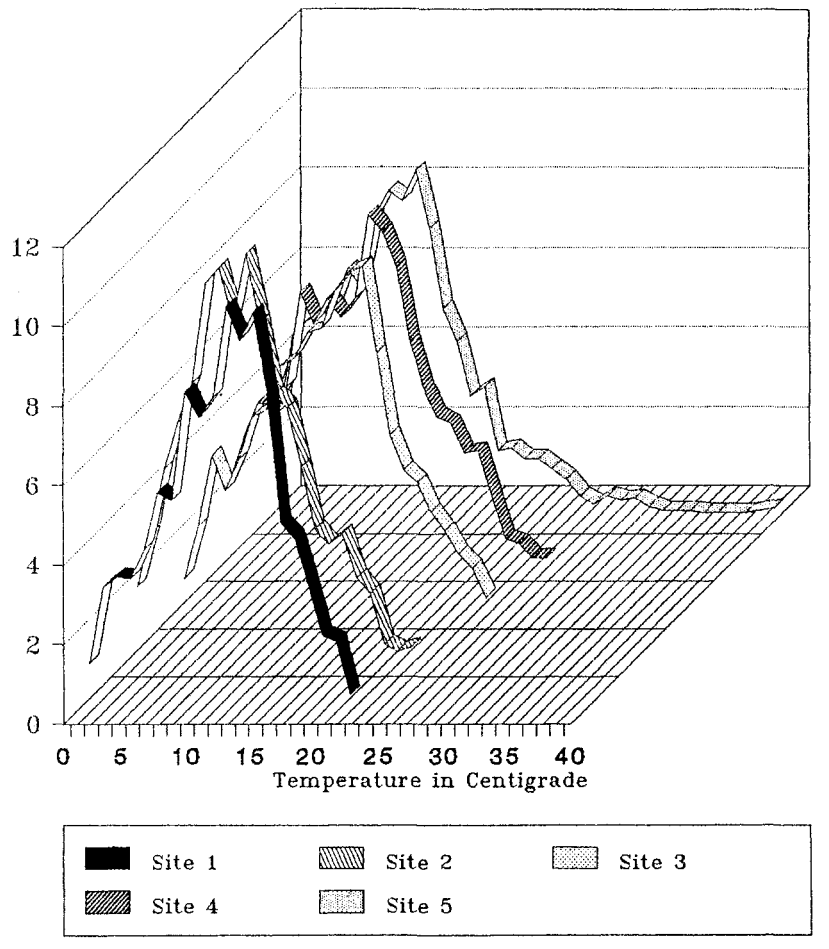

Fig. 5.4. Histogram of values of microclimate measured in the growing season (2 April–2 November 1970). *x* axis = temperature (°C); *y* axis = relative frequency of values measured (in %) during the whole vegetation period; *z* axis = sites of measurements: 1, forest (on the ground); 2, forest (20 cm above the ground); 3 = forest margin (on the ground); 4, forest margin (20 cm above the ground); 5, meadow (on the ground).

level (where tick development occurs and inactive specimens survive), measurements were also made 20 cm above the soil surface in the forest interior and forest edge.

As can be seen in Table 5.1, the average temperatures in all three habitats from June to July were even. This means that the air temperature 20 cm above the ground was only slightly decreased. The same concerns the minimum temperatures. A more marked difference was apparent in the maximum temperatures, where the surface temperatures were much higher at the forest margin and meadow as compared to the inner parts of the forest. The minimum temperatures were recorded at all places on the same day (with a variation of

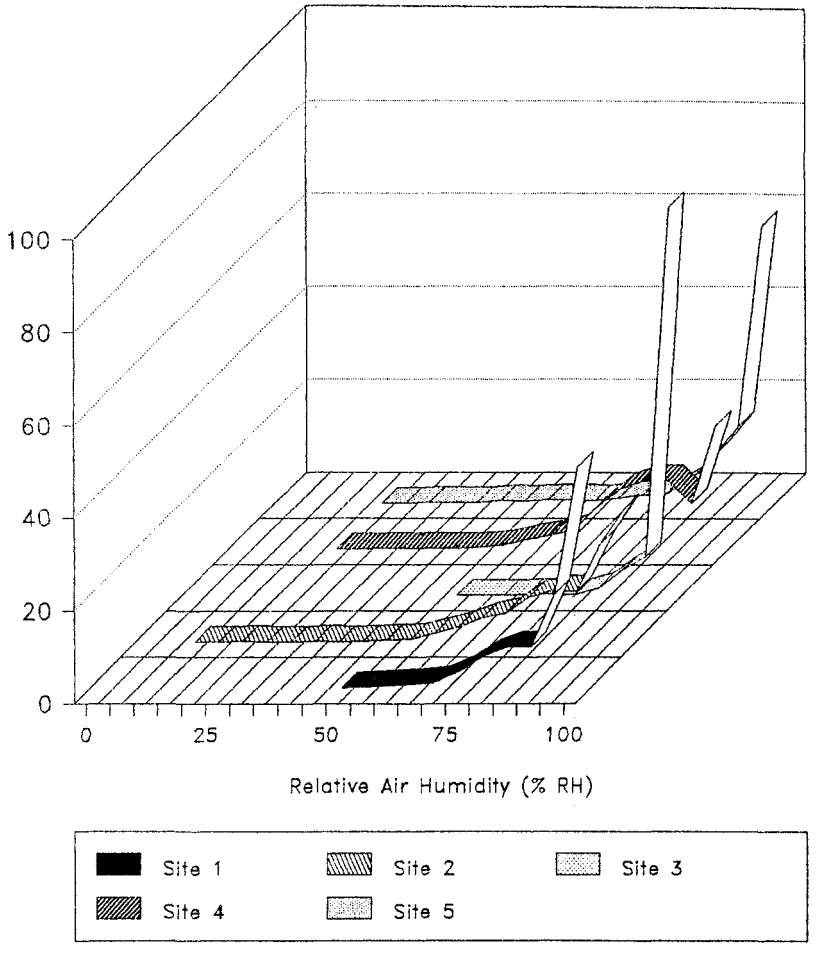

Fig. 5.5. Histogram of values of microclimate measured in the growing season (2 April–2 November 1970). *x* axis = relative air humidity (% RH); *y* axis = relative frequency of values measured (in %) during the whole vegetation period; *z* axis = sites of measurements: 1, forest (on the ground); 2, forest (20 cm above the ground); 3, forest margin (on the ground); 4, forest margin (20 cm above the ground); 5, meadow (on the ground).

only 2 h). Maximum temperatures were observed at the forest margin in June, but were not observed in the forest interior until late July (lower by 17°C).

When relative humidity is considered (Table 5.2), it is evident that the minimum relative humidity, 35% RH (the lowest value recorded in the whole set) was recorded at the meadow surface in the third week of June. In contrast, the minimum relative humidity in the forest (60% RH) was not recorded until mid-July. The relative humidity at the forest margin never dropped below 70%. Clearly, the meadow was the least optimum habitat in terms of moisture deficit stress.

Table 5.1. Basic characteristics of temperatures (in °C) in different microhabitats of *I. ricinus* in the first half of summer (1 June–31 July 1970)

Site of measurement (habitat–microhabitat)	Number of measurements made	Average	Minimum	Minimum of observations reached		Maximum	Maximum of observations reached	
				On day	At hour		On day	At hour
Forest								
Ground	705	11.7	3.0	4.6	4.00	19.1	30.7	14.00
20 cm above ground	715	9.5	1.0	4.6	4.00	21.0	14.7	14.00
Forest margin								
Ground	705	12.9	3.6	4.6	4.00	37.9	3.6	12.00
20 cm above ground	705	11.8	1.6	4.6	2.00	23.5	22.6	14.00
Meadow								
Ground	725	12.2	2.6	4.6	2.00	36.4	24.6	12.00

Table 5.2. Basic characteristics of relative air humidity (% RH) in different microhabitats of *I. ricinus* in the first half of summer (1 June–31 July 1970)

Site of measurement (habitat–microhabitat)	Number of measurements made	Average	Minimum	Minimum of observations reached		Maximum	Maximum of observations reached	
				On day	At hour		On day	At hour
Forest								
Ground	705	93.7	60.0	14.7	18.00	100.0	3.6	6.00
20 cm above ground	715	86.2	43.6	14.7	14.00	100.0	3.6	2.00
Forest margin								
Ground	705	97.3	69.6	20.6	8.00	100.0	1.6	10.00
20 cm above ground	703	86.6	46.1	14.7	8.00	100.0	1.6	2.00
Meadow								
Ground	725	91.5	35.3	24.6	12.00	100.0	1.6	4.00

Table 5.3. Basic characteristics of temperatures (in °C) in different microhabitats of *I. ricinus* in the second half of summer (1 August–30 September 1970)

Site of measurement (habitat–microhabitat)	Number of measurements made	Average	Minimum	Minimum of observations reached		Maximum	Maximum of observations reached	
				On day	At hour		On day	At hour
Forest								
Ground	662	11.1	0.4	28.9	22.00	19.7	2.8	14.00
20 cm above ground	604	8.5	0.4	8.9	6.00	17.9	2.8	14.00
Forest margin								
Ground	667	12.2	0.6	27.9	20.00	22.3	2.8	14.00
20 cm above ground	652	10.7	0.03	22.9	2.00	21.4	2.8	12.00
Meadow								
Ground	653	9.7	0.1	28.9	20.00	27.0	2.8	12.00

Table 5.4. Basic characteristics of relative air humidity (% RH) in different microhabitats of *I. ricinus* in the second half of summer (1 August–30 September 1970)

Site of measurement (habitat–microhabitat)	Number of measurements made	Average	Minimum	Minimum of observations reached		Maximum	Maximum of observations reached	
				On day	At hour		On day	At hour
Forest								
Ground	651	89.6	46.5	7.9	12.00	100.0	31.7	2.00
20 cm above ground	600	87.2	56.7	13.9	12.00	100.0	3.8	22.00
Forest margin								
Ground	667	96.7	61.7	14.9	12.00	100.0	1.8	10.00
20 cm above ground	609	79.8	33.5	2.8	12.00	100.0	18.8	8.00
Meadow								
Ground	653	95.7	48.1	19.8	12.00	100.0	1.8	2.00

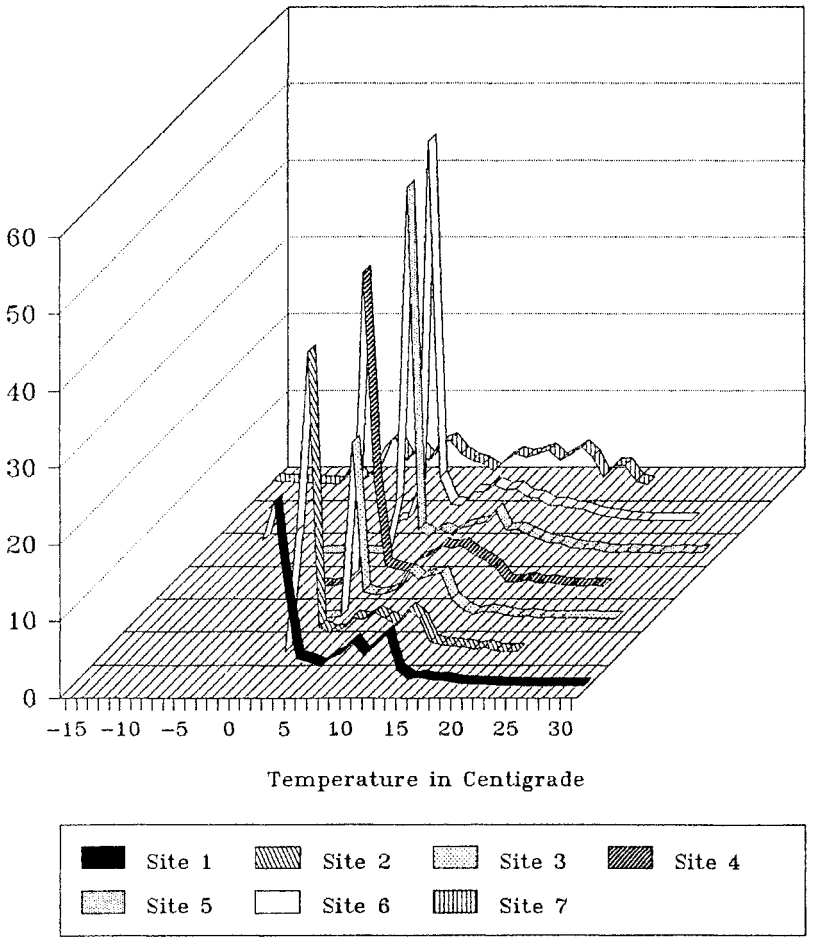

Fig. 5.6. Histogram of values of microclimate measured in the winter season (7 October 1969– 2 April 1970). *x* axis = temperature (°C); *y* axis = relative frequency of values measured (in %) during the whole winter period; *z* axis = sites of measurements: 1, forest (10 cm under the ground); 2, forest margin (10 cm under the ground); 3, meadow (10 cm under the ground); 4, forest (on the ground); 5, forest margin (on the ground); 6, meadow (on the ground); 7, experimental area (standard meteorological box, 2 m above the ground).

Micrometeorological measurements in the tick habitats in the second half of the summer period are similar to those of the earlier period (Tables 5.3 and 5.4). However, the temperature values are more uniform. Similarly, relative humidity did not drop below the early summer values (surface temperatures).

The results described above suggest that the specific nature of the effects of these basic micrometeorological characteristics of *I. ricinus* biology is not fully explained. In a more recent microclimatic study, additional features of the microhabitats and tick biology were compared using descriptive statistics,

specifically, analysis of the trend of variables, covariance and spectral analysis, analysis of relations between two bioclimatological variables based in cross-covariance and cross-spectral functions. Thus, whereas Tables 5.1–5.4 show extreme values without recording the length of their effect, the histograms in the more recent study illustrate the duration of the period that the ticks were exposed to individual temperatures (Fig. 5.4) (temperature and RH = abscissa; % time relative to the entire observation period = ordinate). The temperature values clearly show that there are great differences in the duration of the high temperatures. The first curve (site 1) in showing the percentage of time spent at 15–20°C declines precipitously and does not exceed 20°C. In contrast, the curve showing the meadow surface (site 5) declines slowly and extends towards the highest values. Concerning RH, the three-dimensional histograms (Fig. 5.5) clearly shows that the longest period of optimum high RH values occurred at the forest margin (site 3); it also shows the period of favorable humidity (80–100% RH) which is high inside the forest (site 1) versus time spent by the ticks in the unfavorable period of low RH on the meadow surface.

Winter temperatures (Fig. 5.6) in the same microhabitats as in the previous year (measured at a depth of 10 cm below ground) are also compared. Snowfall was unusually heavy and formed an even cover for almost 4 months. Under this insolating layer, the temperature at and just below ground level was approximately 0°C. However, the ground level air temperature (measured with a standard meteorological box situated directly in the experimental area, see Fig. 5.6, site 7) often dropped well below zero in winter. Occasionally, it was higher than the ground temperatures at the beginning of spring. Table 5.5 shows the temperature values in individual soil layers measured in the same locality in the habitat of thermophilic oak forest (Daniel et al., 1972). The protective role of snow cover provides additional evidence to support our hypothesis that the macroclimatic data cannot explain all of the changes in the tick's life history.

4.3. Influence of Microclimate on the Population of Nidicolous (Endophilic) Ticks

In contrast to the non-nidicolous *I. ricinus*, *I. laguri* is an example of a typical nidicolous species. Its only known host in the former Czechoslovakia is the suslik, *Citellus citellus*. Long-term field and laboratory studies were done with this species in much the same manner as that done with *I. ricinus* described

Table 5.5. Mean monthly temperatures in particular soil layers (in °C)

Month	Surface	−10 cm	−20 cm	−30 cm	−40 cm	−50 cm
December	1.57	1.65	2.11	2.67	3.31	3.59
January	−0.40	−0.29	0.14	0.71	1.42	1.50
February	−0.18	−0.42	−0.15	0.30	0.91	0.92
March	3.28	2.48	2.62	2.32	2.57	2.55

previously (Honzákova et al., 1980). Air temperatures in the suslik nests ranged from 15 to 17°C, while the RH averaged 90%.

4.3.1. *Specific Features of the Life Cycle of* Ixodes laguri *as Influenced by the Nest Microclimate of* Citellus citellus

In all stages, tick mortality during feeding on susliks was higher than in other non-nidicolous tick species reared in our laboratory. All stages were most sensitive to low humidity. Whenever the relative humidity during feeding dropped to less than 90% RH, the attached ticks desiccated and died. Greatest losses were noted in larvae; only one of 78 engorged larvae molted (1.3%). Molting success was considerably greater for engorged nymphs (30.7%). The nest environment was most favorable to engorged females. Out of 20 specimens, 16 oviposited (80%) and 13 egg batches hatched (65%). From females engorged in April, the eggs were harvested in May–June and the larvae hatched from these in August; from females engorged in July, the eggs were harvested in August and the larvae from these hatched in October–November of the same year and in May of the following year; from females engorged in August, the eggs were deposited in September, with hatching in June and July of the following year. The only nymph that molted appeared in November, originating from larvae engorged in August. From nymphs engorged in April, the adults molted in July and August of the same year. In the field experiment, eggs, hungry and engorged larvae all hibernated successfully. Just as in other tick species studied by us (*I. ricinus*, *D. reticulatus*), larvae were the most sensitive of all life stages to adverse environmental conditions. Host excrement was another factor that curtailed the developmental processes of the ticks.

These studies show that nidicolous tick, *I. laguri*, has a 2-year life cycle. Larval hatching is not limited to only one season of the year, as in *I. ricinus*, but takes place from May to November. Presumably, molting of engorged larvae follows a similar pattern.

4.3.2. *Specific Microclimatic Features of the Nest of* Citellus citellus

The basic characteristics of the nest microclimate of *Citellus citellus* are shown in Figs 5.7 and 5.8, based on data from Daniel and Albrecht (1983). Mathematical analysis of temperature and RH values made it possible to draw the following main conclusions. (1) The major factor determining the temperature regime in the nest is ground-level air and its dynamics. This influence is modified by the position of the nest in the soil: partly by the depth at which the nest is situated and partly by the character of the soil. Short-term changes (i.e., shorter than 7 days) are filtered out, and slow changes lag by 1–2 days depending on the season. In transient periods between the spring and summer season as well as in the winter season, the gradual cooling (or warming) of the soil causes different trends that can be seen in the time function of the average diurnal temperatures outside and inside the nest. The thermoinsulating

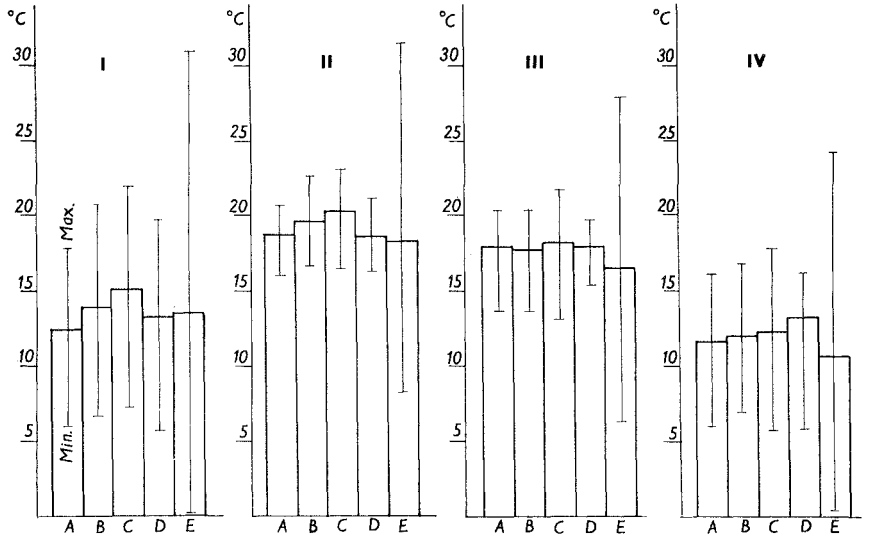

Fig. 5.7. Citellus citellus nest microclimate (after Daniel and Albrecht, 1983). Graphs of average, maximum and minimum temperature values measured in the first to fourth period of vegetation season in nest inhabited by a pair of *C. citellus* (A = bottom, B = middle, C = upper part of nest), in uninhabited nest (D) and in the ground air layer (E).

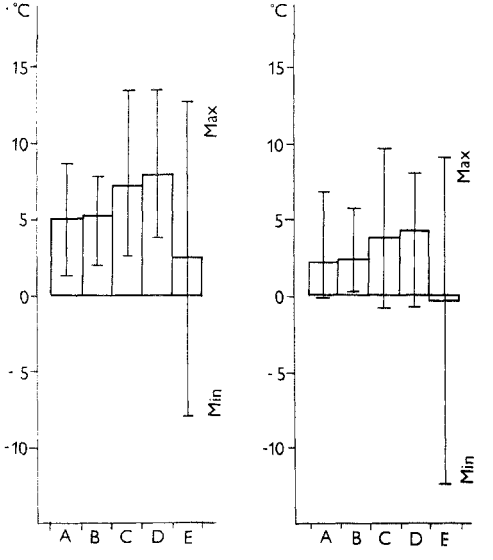

Fig. 5.8. Citellus citellus nest microclimate (after Daniel and Albrecht, 1983). Graphs of average, maximum and minimum temperature values measured in the first and second periods of winter season in the inhabited nest (A = bottom, B = middle, C = upper part of nest), in uninhabited nest (D) and in the ground layer (E).

properties of the soil moderate the extreme temperatures of the outer environment, especially when the lower temperature ranges are considered. (2) The dynamics of the temperature changes in the nest are further influenced by the activities of the nest inhabitant which manifest themselves as follows: (a) the presence or absence of the host in the nest; (b) the arrangement of the nest bedding (its supplement with fresh plant material); (c) the decaying green plant food and other plant materials; (d) the food supplies stored in the nest; and (e) the urine and feces passed in the nest.

The trend of average diurnal temperatures is not influenced by the animals' presence in the nest but the absolute level of the values measured is positively affected. Some activities of the animals, e.g., accumulation of rotting organic materials at the nest bottom, also affect the humidity regime of the nest. The higher and more constant values of the nest RH are also dependent on the position of the nest in the soil. The character of the influence as well as the intensity of effects of the activities of the nest inhabitants vary according to the season.

The nest environment is highly heterogeneous, depending on the structure of nest bedding and other components. The temperature and humidity are varied, both (1) in the vertical direction, and (2) in the centripetal direction. The nest bottom, which contains accumulated organic wastes, shows a nearly constant high RH while its temperature is governed by conditions of the surrounding soil. The middle and upper parts of the nest cavity is filled with nest bedding and represents the nest environment proper of the susliks. In the centripetal direction, the strongest influence of the susliks is in the centre of the nest, but this diminishes towards the periphery. Environmental heterogeneity is evident throughout the year but it is most striking in winter. Following the long period of minimum host activity when the nest bedding was not supplemented with fresh material and the nest was not cleaned, the bedding remained dry only in the narrow zone of direct contact with the bodies of the hibernating animals. The periphery of the nest was saturated with moisture, frequently rotten and the state of the inhabited nest became similar to that of an uninhabited (control) nest. Thus, the nest environment presents a variety of microclimatic conditions for nest parasites living in close contact with the animals (e.g., fleas, ticks, haematophagous and schizophagous mites) on the one hand and for other nidicoles associated mainly with rotting substances on the other.

4.3.3. Specific Features of the Nests of Small Terrestrial Mammals

Small terrestrial mammals are hosts of the nidicolous tick, *Ixodes trianguliceps*, in central Europe. This species behaves more like an exophilic than a nidicolous tick. The results of multi-year studies of the environment of the nests of these small mammals have contributed to our recognition of this behavior pattern.

Nests of small terrestrial mammals can be classified into three main categories. Each of them has its own specific microclimate and their position in the ground is an important criterion that differentiates them from one another.

These are as follows.

1. Underground nests (i.e., situated under the soil surface).
2. Nests situated at the ground surface or in shallow depressions. The walls and top of the nesting chamber mostly consist of wood (stumps not yet damaged by rot, or rotten wood at various stages of decomposition).
3. Nests situated above the ground level mostly in natural cavities in stumps and trunks but without a direct contact with soil.

The bank vole, *Clethrionomys glareolus*, which is a typical forest species, was chosen as a model for our long-term studies. The nests of this species can fit in all of the above-noted categories. Moreover, it is often utilized as a host by *I. trianguliceps*. Tables 5.6 and 5.7 show the basic characteristics of temperatures detected in the nest microhabitat, the habitat of the inundated forest under study and of the whole region. An analysis of these (and other) data led to the following general conclusions (for details, see Daniel, 1970, 1988).

As we have seen, ground level microclimatic conditions influenced the nest microclimate and direction of any changes. Temperature variations were balanced by the nest itself: the temperature characteristics in the ground air layer (mean temperature, mean of daily maximum and minimum temperatures) exhibited distinctly higher standard deviations than those in individual nests. In summer, the activity of small mammals either did not influence the nest temperature at all or had only a negligible effect compared to the influence of the environment.

Table 5.6. Temperature characteristics of nests of the bank vole (*Clethrionomys glareolus*) in the summer season and of the area studied (after Daniel, 1988)

Measurements taken	Daily temperature	Mean (°C)	N^a
Macroclimate of area studied	Mean	18.86	40
	Maximum	20.40	26
	Minimum	12.30	26
Mesoclimate of area studied	Mean	16.90	23
	Maximum	22.30	26
	Minimum	11.50	26
Microclimate of experimental site (air near the ground)	Mean	17.16	40
	Maximum	21.71	40
	Minimum	12.69	40
Microclimate of nest situated in a tree stump	Mean	16.28	40
	Maximum	17.86	40
	Minimum	14.71	40
Microclimate of underground nest (10 cm deep)	Mean	16.79	40
	Maximum	17.97	40
	Minimum	15.73	40
Microclimate of underground nest (20 cm deep)	Mean	15.44	40
	Maximum	16.32	40
	Minimum	14.89	40

[a] Number of days analyzed.

Table 5.7. Temperature characteristics of nests of the bank vole (*Clethrionomys glareolus*) in winter season and of the area studied (after Daniel, 1988)

Site and mode of measuring	Winter period[a]	Temperature (°C)		
		Mean	Maximum	Minimum
Macroclimate of the region (after	I	0·14	14.30	−17.30
State meteorological service)	II	2.72	19.00	−10.40
Mesoclimate of the forest	I	−0.36	13.00	−15.80
	II	2.77	18.50	−9.50
Freely on the surface	I	−0·20	6.81	−5.02
	II	0.77	11.24	−5.41
20 cm deep in the soil	I	1.71	4.55	−1.14
	II	1.03	5.80	−0.70
Microclimate of nest in tree stump	I	2.16	7.33	−2.64
(on the ground)	II	2.86	9.90	−1.36
Microclimate of nest under wood	I	2.61	6.56	−1.80
(on the ground)	II	3.46	9.90	−2.20
Microclimate of nest below ground	I	3.98	10.41	0.42
(20 cm below the surface)	II	4.06	8.34	1.39

[a] I = First half of winter season. II = Second half of winter season.

Winter temperatures in *C. glareolus* nests were very low. Extreme values in nests situated on the ground and above the ground even dropped below 0°C (Table 5.7). A correlation of time series pairs showed that there was a direct influence of the soil temperature changes in all three types of nests, without any time shift between the curves. This means that the changes in the soil temperature resulted almost simultaneously in changes in nest temperatures. An exception observed in the second half of winter was the temperature in a nest situated inside a stump, which was not in direct contact with soil. The soil temperature was lower than that in the nest. In this case it was the presence and activity of small mammals which influenced the temperature in the winter nest.

The influence of physical changes in the mesoclimate on the environment in the nests of small mammals explains the seasonal occurrence of the nidicolous tick, *I. trianguliceps*. Moreover, these mammals—the hosts of *I. trianguliceps*—are active throughout the year, without any marked period of hibernation.

5. MICROCLIMATE AND NATURAL FOCI OF DISEASES

The theory of natural foci of diseases, first proposed by Pavlovsky (1939), suggests that zoonotic diseases occur within a certain geographical territory, distinctly demarcated by certain biocenoses or biotic communities, within which a zoonotic agent circulates. The pathogenic organism is a member of the biocenose where it circulates for long periods from donor hosts via vectors to recipient hosts irrespective of man. Only when humans enter such a locality are

they exposed to attacks by vectors which transmit the pathogenic organisms previously obtained from wild reservoir hosts. When people fall ill, the natural focus becomes manifest and the focal disease becomes known to the local health services.

5.1. Biotic and Spatial Structure of Natural Foci of Diseases

Every natural focus has a definitive **biotic** and **spatial** structure. The biotic structure consists of biotic components among which the pathogenic agent circulates. For example, the tick, *I. ricinus*, the vector of Tick-borne Encephalitis (TBE) in Europe, feeds on various groups of mammals and birds. Included in the biotic structure of the natural focus are (1) those tick hosts in which viremia occurs (which promotes the circulation of the TBE virus) and (2) non-viremic hosts which allow transmission of some viruses from infected to non-infected ticks during simultaneous feeding (Jones et al., 1987; Nuttall and Jones, 1991; Labuda et al., 1993). The richer the biotic structure, the greater the number of animals that can be included in the circulation of the pathogenic agent. While TBE is transmitted to humans by only a few species (either by vector or by contact direct with the host), many groups of animal species take part in the biotic structure proper.

The biotic structure is closely connected with the spatial limits of the biocenose. The latter's importance for maintenance and circulation of the pathogenic agent is not the same throughout the entire focus. The most suitable sites were termed by Pavlovsky (1948) "elementary foci" and they are the basic unit of the spatial structure of these natural foci. They are localized at places that afford the best conditions for the food, shelter, and microclimate of both the vectors and their hosts. The elementary foci can be divided into: (1) restricted (well-demarcated foci, e.g., a burrow of a rodent); and (2) diffuse, i.e., without any exact border. Their basic element is the **nucleus**, which represents small portions of the natural focus most suitable for the existence of the vector–host system and in which the pathogenic agent can be maintained even in the interepizootic period (when the epizootic process in the remaining parts of the natural focus is depressed). In the case of infections transmitted by ticks, the elementary foci are identical with the optimal habitats of ticks and the nuclei of elementary foci include microhabitats with the most favorable microclimatic factors (Rosický and Hejný, 1961; Rosický, 1962, 1967). In cultivated landscapes, the elementary foci markedly differ from the remaining part of the natural focus, which is usually highly cultivated. Where it is surrounded by cultivated areas, the elementary focus is the only form of the natural focus (Rosický and Daniel, 1989).

5.2. Influence of Microclimatic Factors on Pathogen Development and Infection Rates in Ticks

Many tick-transmitted diseases occur only seasonally in limited climatic zones. There is clearly a wealth of evidence that climate and weather play an important

role in arboviral and other tick-transmitted disease occurrence. This can be explained by the influence of weather and microclimate on tick biology (Reiter, 1988). As we have seen in the preceding chapters, effective transmission and high vectorial capacity depends on several characteristics of the vector tick, e.g., longevity, host-seeking behavior, and mobility (Friedhoff, 1990), all of which are strongly influenced by microclimate.

The maintenance and multiplication of the parasite in the vector are influenced mostly by temperature. Fujisaki and Kamio (1988) found that constant temperatures of 15, 30, and 35°C (at 100% RH) were detrimental to the infection of salivary glands of *Haemaphysalis longicornis* with *Theileria sergenti*. High infection levels were discernible only in ticks incubated at 20 and 25°C. Apparent maturation of *T. sergenti* was not observed in salivary glands of ticks exposed to 35°C without the stimulus of a blood meal. However, Samish (1977) and Walker and McKeller (1983) found that infective *Theileria annulata* were induced in salivary glands of *Hyalomma anatolicum* without a blood-meal stimulus only by elevating the incubation temperature to 36 or 37°C. This was also observed by Irvin et al. (1981) and Young et al. (1979) in *Theileria parva* infections in the tick, *Rhipicephalus appendiculatus*. Similarly, forms of *Babesia bovis* and *B. bigemina* infective for cattle can be produced in unfed larval ticks and eggs of *Boophilus microplus* by thermal stimulation at 37°C for 3–4 days. However, a fall in temperature is necessary for *B. bigemina* parasites ingested by adult ticks to change to the forms infective for the ticks (Dalgliesh et al., 1981). Das and Sharma (1991) observed that temperature variations (4–40°C) had a significant effect on *Hyalomma anatolicum anatolicum* nymphal molting rates and transmission of *Theileria annulata* from nymphs to adults. Maximum infection levels were obtained in salivary glands of adult ticks when the engorged infected nymphs were incubated at 24–28°C.

Size and density of the vector and host populations as well as seasonal activity of the vectors also influence the epidemiology of some tick-borne diseases. Martinod and Gilot (1991) noted correlations between the occurrence of canine babesiosis, caused by *Babesia canis*, and its tick vector, *Dermacentor reticulatus*, in France. Cases of babesiosis occurred in spring and fall when adult ticks were active. Fluctuations of the *D. reticulatus* population and the onset of canine babesiosis were also correlated with climatic changes: no tick activity or clinical cases of disease were detected in winter or in summer when the ticks were in diapause.

In non-diapausing tropical ticks, the infective agents can survive for long periods. Newson et al. (1984) observed that, in Kenya, fatal cases of cattle infected with *Theileria parva* were transmitted by adult *Rhipicephalus appendiculatus* which had fasted for up to 554 days.

As regards the influence of climatic factors on the circulation dynamics of tick-borne viruses and on the diseases caused by these viruses, the relevant literature offers epidemiological analyses rather than laboratory results. An example is the paper by Bárdoš (1954), who analyzed the influence of rain on the dynamics of a TBE natural focus in the vicinity of Rožňava (Slovakia) in 1951 (after the largest TBE epidemic in Europe, with more than 660 cases).

This author suggested: (1) that an epidemic increase in human cases may occur if the rainy months (greater than the long-term mean) are followed by a decrease to normal or subnormal values during the *Ixodes ricinus* host-seeking activity period (April–August); (2) if the very moist winter–spring months (January–March) are followed by a sudden drop in precipitation in the spring months, then an increased number of human infections will occur at the beginning of the season; (3) in contrast, if the amount of precipitation is below normal during the tick season, the number of human infections will decrease.

Laboratory experiments (Danielová et al. 1983; Danielová, 1990) have provided additional evidence in support of Bárdoš's conclusions. Three experiments were performed differing from one another only in the level of viremia in the mice that served as the blood source for the ticks and in the physiological age of the ticks (Table 5.8). Infected ticks held at (1) 15°C and 75% RH, and (2) 24°C and 97% RH, respectively, were observed for virus infection for 16 weeks (more than 600 ticks were used in each experiment). The results showed that viral infection was influenced by RH to a much greater degree than by temperature. Low RH reduced the virus infection rate but it did not affect the dynamics of the infection process. The effect of temperature was analogous but less pronounced than that of relative humidity. The RH effect may have occurred in response to stress, since 75% RH presented an extremely desiccating environment, at the borderline between life and death for *I. ricinus*, whereas the temperature of 15°C used in the experiments is far less stressful; ticks readily survive much lower temperatures.

Diapause also affects virus growth in ticks. Mishaeva and Erofeeva (1979) found that the TBE virus multiplies more intensively in ticks developing without diapause than in diapausing ones. The virus titers in infected *Ixodes ricinus* nymphs developing at temperatures of 18 and 23°C without diapause were 2.5–3.9 log LD_{50} higher than those of the diapausing individuals maintained at 9°C. The TBE virus titers remained the same for 8–10 weeks in active engorged larvae and nymphs, even when they molted, but virus titers gradually diminished in diapausing ticks. The question remains as to whether these differences resulted from the effect of the very different temperatures used in the experiment rather than diapause.

6. ADAPTATIONS OF TICKS TO UNFAVORABLE MICROCLIMATIC CONDITIONS

Besides their many physiological and anatomic adaptations that enable them to feed successfully on hosts and to obtain sufficient amounts of blood, ticks have also evolved specific adaptations for survival under unfavorable microclimatic conditions. Among the most important are: (1) the protective structure and chemistry of the integument; (2) mechanisms for maintaining water balance; and (3) the ability to diapause.

The waterproofing properties of the cuticle lie in the surface wax layer

Table 5.8. Experimental infection of *I. ricinus* with TBE virus under different microclimatical conditions (orig. V. Danielová). Infection rate of *I. ricinus* nymphs and adults (%) under high and low air humidity (% RH) and temperature (°C)

Experiment	*I. ricinus* (origin of nymphs)	Viremia level during tick feeding	Infection rate of ticks (%)			
			97% RH	75% RH	24°C	15°C
1	Free living-spring (metamorphosis preceding year probably)	3–1 log LD_{50}	5.97	1.04	5.26	3.81
2	First laboratory generation (3–4 months after metamorphosis)	5–3 log LD_{50}	27.03	20.00	28.30	22.38
3	First laboratory generation (1–2 months after metamorphosis)	5–3 log LD_{50}	88.75	49.12	54.38	46.43

(Lees, 1947; Hackman and Filshie, 1982) which helps maintain water balance by resisting desiccation in unfavorable microclimate conditions. The waxy lipids forming this layer are thermosensitive and can change their structure and waterproofing properties in response to increasing temperature. These lipids, the structure of which differs in different tick species, are relatively stable, since the ambient temperature usually does not reach the transition temperature (Table 5.4), the temperature at which the lipids lose their waterproofing properties. The higher the transition temperature of the epicuticular lipids, the greater their impermeability to water loss (Knülle and Rudolph, 1982).

As noted previously in this chapter, ticks are able to maintain their water balance up to a threshold humidity (CEH) below which the tick continuously loses water (Knülle and Wharton, 1964). Several protective mechanisms have been developed in both ixodid and argasid ticks to prevent passive water loss in an unsaturated environment. Passive water loss from the body surface is limited by the effective waterproofing of the cuticle, especially by the wax layer (see above). Respiratory water loss caused by gas exchange via the tracheal system of ticks is prevented by the closing device of the spiracle. This respiratory control mechanism is CO_2 sensitive and high concentrations of CO_2 cause the spiracles to remain open. A drastic, 17-fold increase of water loss in adult *Amblyomma variegatum* was recorded by Rudolph and Knülle (1979) and Rudolph (1982) at high CO_2 concentration. However, in larvae which lack spiracles or a tracheal system, CO_2 had no effect.

The body surface and the respiratory system serve also for the passive uptake of water vapor from the atmosphere in a saturated environment. Temperature affects passive sorption and transpiration exponentially (Arlian and Veselica, 1979).

Unfed ixodid ticks extract water vapour from unsaturated air by salivating hygroscopic secretions on to the mouth parts which become water enriched when exposed to the ambient air (Kahl and Knülle, 1986). In adult *A. americanum*, the active water uptake is a solute-drive process that is dependent upon the production of a hyperosmotic fluid probably by the type I agranular acini of the salivary glands (Sigal et al., 1991). There is a temperature threshold (5–9°C) below which active uptake does not occur (Lees, 1946; Sauer and Hair, 1971). The uptake rate is exponentially faster if the temperature is increased, but the amount of water taken up is relatively constant (Knülle and Rudolph, 1982, cited in Needham and Teel, 1986). Recently, Kahl and Knülle (1986, 1988) have demonstrated that even fully engorged and detached larvae and nymphs of *Ixodes ricinus*, *I. dammini* and *Haemaphysalis punctata* consistently take up substantial amounts of atmospheric water vapor in subsaturated ambient conditions.

Diapause is another factor that enables most species of ticks to adapt to unfavourable microclimatic conditions. Their developmental cycle is adapted so that they survive in a quiescent or dormant state. Locomotor and questing activity is minimized or arrested completely, metabolic activity is reduced and metamorphosis is delayed. Diapause may be behavioral, in which unfed ticks do not seek hosts, or morphogenetic, in which developmental processes, e.g.,

molting, oviposition, etc., are delayed. In either case, energy is conserved, water loss is minimized and survival is greatly enhanced. Further discussion of the diapause process is beyond the scope of this chapter. For more detailed reviews of diapause in ticks, the reader is referred to Sonenshine (1988), Belozerov (1982), and Dusbábek (1985a, 1985b, 1988).

7. MICROHABITATS AND THEIR RELATIONS TO THE BIOINDICATORS OF TICK DISTRIBUTION

The relationship of ticks to the plant communities characterizing their habitats is well known. The specific types of vegetation in this relationship have been shown to be useful indicators of tick occurrence as well as the ecosystem that supports tick-borne diseases. An example in Europe is *I. ricinus*, which, because of its enormous epidemiological and epizootiological significance, may serve as the model species in the discussion that follows.

7.1. Vegetative Type as a Significant Bioindicator of Tick Occurrence (Based on the Example of *Ixodes ricinus*)

The macroscopically obvious expressions of the natural environment, i.e., its physical relief, vegetation, etc., are directly or indirectly associated with the hidden components of foci, i.e., vectors and causative agents of disease. The former are used as indicators of tick occurrence and existence of natural foci of diseases transmitted by them. Similarly, the specific vegetation provides a convenient bioindicator of the tick-infested ecosystem showing identical requirements for the ecological conditions of the environment. A feedback phenomenon also occurs, which is evident from the fact that a given plant community may affect the microclimate of ticks and living conditions of their hosts by providing favorable food resources and shelters. These associations have been described in many parts of the *I. ricinus* geographic range, especially in Central Europe (Rosický and Hejný, 1961; Loew et al., 1963; Hejný and Rosický, 1965; Nosek et al., 1970; Jusatz, 1978, 1981), in Finland (Ohman, 1961), Croatia (Vesenjak-Hirjan et al., 1965), Slovenia (Tovornik, 1970), and Bulgaria (Hejný and Rosický, 1962).

During their studies of TBE natural focality, Rosický and Hejný (1961) showed that specific vegetative communities are typical of *I. ricinus* habitats and TBE virus foci which make it possible to discern the detailed structure of each focus, i.e., the elementary focus and its nucleus. For example, in the Central European *Querceto–Carpinetum* lowland forest type, the elementary focus is characterized by *Stellario–Alnetum*, *Fraxineto–Alnetum* or *Cariceto remotae–Fraxinetum* associations. Rather than a homogeneous wooded undergrowth, the nucleus is divided at the plant level by its dominants and co-dominants, which change in relatively small areas, thus providing it (the nucleus) with a rather large structural variability (e.g., the *Stellario–Alnetum* association dominated

by *Aegopodium podagraria*, *Urtica dioica*, *Impatiens noli-tangere*, *Stellaria nemorum* and others).

Where the occurrence of *I. ricinus* in a given vegetation type is characteristic, a simple botanical map may seem to be sufficient for the evaluation of certain territories as appropriate tick habitat. Based on this simplified concept, maps of large areas predicting the likely distribution of this tick have already been made. This approach and the results achieved are discussed by Prokhorov (1974) and Vershinina (1985). However, such maps are unsuitable for practical utilization of plant communities as indicators of increased tick density or epidemiological analysis of pathogen circulation and disease prevention. The scales of these maps do not adequately reflect details of the landscape or other factors important for *I. ricinus* distribution and its epidemiological significance, i.e., changes caused in the landscape by human activities. Similarly, social factors are ignored, e.g., the density and proximity of human dwellings which influence the intensity of contacts between man and tick.

Mapping tick distributions on the basis of direct observations in the field (both botanical and ixodological) is time-consuming and requires good organization. Only limited areas of the territory studied can be included. Such methods cannot be used to create a generally acceptable practical map for predicting tick occurrence over a large area. A much better perspective for this purpose was provided by the medium-scale prognostic map showing suitable *I. persulcatus* habitats in the Far East, based on aerial photographs (Kuzikov et al., 1982).

7.2. Use of Satellite Data for Forecasting the Occurrence of Ticks

Remote sensing offers the best solution to the problems described above, since it makes it possible to determine the exact distribution of the respective plant communities. Using our knowledge of the tick–plant associations described previously, the remote sensing maps can be interpreted to reveal locations of potential *I. ricinus* occurrence at epidemiologically important densities. Hugh-Jones (1989), in a detailed study of this method, suggested that microclimatic characteristics can be extrapolated from remote sensing data used to describe the physical characteristics of the habitats recorded. The great attraction and value of remote sensing is that it is a digital database with excellent temporal and spatial references. Hugh-Jones (1989, 1991a, 1991b) also described the distribution of *Amblyomma variegatum* in the Caribbean region. On Guadeloupe Isle, analysis of LANDSAT-TM imaging data consistently identified a series of habitats, independently defined, using data on plant composition and environmental characteristics, which appeared to have different tick-carrying capacities. These were: (1) lightly tick-infested "dry meadows;" (2) heavily tick-infested "dry scrub" and "rocky grassland;" and (3) intermediately infested "fond" (an area of karst) and "foothills." Perry et al. (1991) discussed the role of the satellite-derived normalized difference vegetation index (NDVI) in studies of *Rhipicephalus appendiculatus* habitats in Africa. This spectral vegetation index

quantifies the level of photosynthetic activity of vegetation and is calculated from the advanced very high resolution radiometer (AVHRR) data provided by the US National Oceanic and Atmospheric Administration's meteorological satellites. Cooper and Houle (1991) used the data of LANDSAT-TM imagery to develop a map of expected tick densities of *Dermacentor variabilis*, based on the dominant vegetation type and vegetation ecotones for a study area in Orange County, North Carolina, USA.

Applying these methods to Central Europe, Daniel and Kolář (1990) constructed a predictive map of *Ixodes ricinus* occurrence on the basis of remote sensing data. The data were obtained from the MSS operating aboard LANDSAT-5. A file was selected representing a territory of 41 × 41 km. In its centre, there was an area in which large numbers of ticks were consistently present, as revealed in a 25-year continuous study of *I. ricinus*. This area proved to be a natural focus of TBE. It was taken as a model and compared with its surroundings. Six landscape categories were examined: (1) coniferous forest; (2) deciduous forest; (3) mixed forest; (4) water basins; (5) glades; and (6) housing developments. Categories 1–3 are crucial for the evaluation of the probability of tick presence, whereas the sixth category is significant in assessing the main exposure to ticks. The data were processed by supervised classification using the Bayes' decision rule of maximum likelihood. The findings were obtained both in graphic form and as statistical reviews of the presence of appropriate landscape categories.

Predictive maps obtained by this method determine not only the extent of individually suitable habitats but their dispersion into smaller, overlapping areas (the mosaic character was accurate to 80 × 80 m in real terrain). The ecotone range can be established from these data also. This makes it possible to offer a more exact prediction of *I. ricinus* occurrence. By such means, high-priority sites can be earmarked for protection of the human population against exposure to tick bites and for tick control programs.

8. REFERENCES

Arlian, L. and Veselica, M.M. (1979) Water balance in insects and mites. *Comp. Biochem. Physiol.* **64A**, 191

Arumova, E.A. (1979) Movements of *Ixodes persulcatus* ticks from the moment of nymphal engorgement to unfed adult activation. *Med. Parasitol. Parasit. Dis.* **48**, 347 (in Russian).

Atwood, E.L. and Sonenshine, D.E. (1967) Activity of the American dog tick, *Dermacentor variabilis* (Acarina: Ixodidae) in relation to solar energy changes. *Ann. Entomol. Soc. Am.* **60**, 354.

Babenko, L.V. (1974) Diurnal variations in the activity of empty nymphs *Ixodes ricinus* L. and *Ixodes persulcatus* P. Sch. (Parasitiformes: Ixodidae). *Med. Parasitol. Parasit. Dis.* **43**, 520 (in Russian).

Babenko, L.V. and Arumova, E.A. (1985) Active horizontal migration. pp. 238–239. *In:* Filippova, N.A. (ed) *Taiga Tick* Ixodes persulcatus *Schulze* (*Acarina, Ixodoidea*). Nauka Publ., Leningrad Branch (in Russian).

Balashov, Yu.S. (1958) The question of active horizontal migration of the taiga tick *Ixodes persulcatus* P. Sch. *Med. Parasitol. Parasit. Dis.* **27**, 481 (in Russian).

Balashov, Y.S. (1959) Application of mass marking of ixodid ticks for studying their mobility. *Zool. Zh.* **38**, 1028 (in Russian).

Balashov, Yu.S. (1960) Water balance and the behaviour of *Hyalomma asiaticum* in desert areas. *Med. Parasitol. Parasit. Dis.* **29**, 313 (in Russian).

Balashov, Yu.S. (1963) The effect of external factors upon the number of nymphal stages in argasid ticks. *Parazit. Sbor.* **21**, 28 (in Russian).

Balashov, Yu.S. (1967) *Bloodsucking Ticks (Ixodoidea)—Vectors of Diseases of Man and Animals.* Publ. Nauka, Leningrad (in Russian); (1972) *Misc. Publ. Ent. Soc. Am.* **8**, 159 (in English).

Bárdoš, V. (1954) The influence of rains on the dynamics of the natural focus. pp. 191–196. *In:* Blaškovič, D. (ed) *An Epidemic of Encephalitis in the Natural Focus of Infection in Rožnava.* Publishing House Slovak Academy of Sciences, Bratislava (in Slovak).

Belozerov, V.N. (1982) Diapause and biological rhythms in ticks. pp. 469–500. *In:* Obenchain, F.D. and Galun, R. (eds) *Physiology of Ticks.* Pergamon Press, Oxford.

Berdyev, A. (1969) Developmental duration of *Hyalomma dromedarii* Koch in natural conditions of Turkmenia. *Izv. Akad. Nauk Turkmen. SSR, Ser. Biol. Nauk* **2**, 54.

Berdyev, A. (1972) The duration of the life cycle of *Hyalomma asiaticum* Sch., Schl. in natural biotopes. *Parazitol.* **6**, 30 (in Russian).

Berdyev, A. (1974) Types of life cycles of ixodid ticks (Parasitiformes, Ixodidae). *Parazitol.* **8**, 227 (in Russian)

Bouchalová, J., Honzáková, E., and Daniel, M. (1977) Development and survival of the tick *Hyalomma dromedarii* Koch under laboratory conditions. *Folia Parasitol.* **24**, 55.

Boučková, I. and Dyk, V. (1968) Die sommerliche und herbstliche Tagesaktivität des gemeinen Holzbocks im Waldgebiete von Jezírko bei Brno. *Angew. Parasitol.* **9**, 36.

Buczek, A. (1988) Studies of the biology of *Argas* (*A.*) *reflexus* (Fabricius, 1974) (Acari: Ixodida: Argasidae). 1. Effect of temperature and relative humidity on embryonic development and egg hatch. *Folia biol. (Krakow)* **36**, 329.

Černý, V. (1967) Two new species of argasid ticks (Ixodoidea: Argasidae) from Cuba. *Folia Parasitol.* **14**, 142.

Černý, V., Daniel, M., and Rosický, B. (1974) Some features of the developmental cycle of the tick *Ixodes ricinus* (L.). *Folia Parasitol.* **21**, 85.

Černý, V., Szymanski, S., Dusbábek, F., Daniel, M., and Honzáková, E. (1982) Survival of unfed *Dermacentor reticulatus* (Fabr.) adults under natural conditions. *Wiadom. Parazytol.* **28**, 27.

Cooper, J.W. and Houle, J.U. (1991) Modelling disease vector habitats using thematic mapper data: identifying *Dermacentor variabilis* habitats in Orange County, North Carolina. *Prev. Vet. Med.* **11**, 353.

Dalgliesh, R.J., Stewart, N.P., and Mellors, L.T. (1981) Some effects of temperature on the life-cycle of *Babesia bovis* and *Babesia bigemina* in the tick *Boophilus microplus. FAO Anim. Health Serv., Inf. Circ. on Tick-borne Dis. Livestock AGA*: TD/81/Inf. 23.

Daniel, M. (1970) Effects of the microclimate on nest parasites of the bank vole, *Clethrionomys glareolus*, in summer season. *Acta Scientiarum Naturalium Academiae Scientiarum Bohemoslovacae*, Brno 4, 1–48.

Daniel, M. (1978) Microclimate as a determining element in the distribution of ticks and their developmental cycles. *Folia Parasitol.* **25**, 91–94.

Daniel, M. (1988) Mesostigmatid mites in nests of small terrestrial mammals and features of their environment. *Studies ČSAV, Academia, Publ. House Czechoslov. Acad. Sci., Prague*, **10**, 1–196.

Daniel, M. and Albrecht, V. (1983) Biomathematic study on the nest environment of susliks *Citellus citellus. Rozpravy ČSAV, Serie Mat. Sci. Nat., Academia, Publ. House Czechoslov. Acad. Sci., Prague*, **93**, 1–138.

Daniel, M. and Černý, V. (1967) To the methods of studying the environmental temperature of the tick *Ixodes ricinus* L. *Folia Parasitol.* **14**, 177.

Daniel, M. and Kolář, J. (1990) Using satellite data to forecast the occurrence of the common tick *Ixodes ricinus* (L.). *J. Hyg. Epidemiol. Microbiol. Immunol.* **34**, 243.

Daniel, M., Honzáková, E., and Černý, V. (1972) Overwintering of the tick *Ixodes ricinus* (L.) under conditions of a field experiment. *Folia Parasitol.* **19**, 305.

Daniel, M., Černý. V., Dusbábek, F., Honzákova, E., and Olejníček, J. (1976) Influence of microclimate on the life cycle of the common tick *Ixodes ricinus* (L.) in thermophilic oak forest. *Folia Parasitol.* 23, 327.

Daniel, M., Černý, V., Dusbábek, F., Honzákova, E., and Olejníček, J. (1977) Influence of microclimate on the life cycle of the common tick *Ixodes ricinus* (L.) in an open area in comparison with forest habitats. *Folia Parasitol.* **24**, 149.

Daniel, M., Černý, V., and de la Cruz, J., (1981) Disappearance of a giant cave tick population. *Folia Parasitol.* **28**, 28.

Daniel, M., Černý, V., Albrecht, V., and Honzáková, E. (1988) The microclimate at different altitudes of the Krkonoše Mountains and its effect on the existence of the tick *Ixodes ricinus* (L.). *Opera Coronotica* **25**, 76 (in Czech).

Daniel, M., Honzáková, E., Dusbábek, F., and Černý, V. (1974) The influence of microclimate on the developmental cycle of the tick *Ixodes ricinus* (L.). pp. 361–365. *In:* Piffl, E. (ed) *Proceedings 4th International Congr. Acarology.* Academiai Kiado, Budapest.

Danielová, V. (1990) Experimental infection of ticks *Ixodes ricinus* with tick-borne encephalitis virus under different microclimatic conditions. *Folia Parasitol.* **37**, 279.

Danielová, V., Daniel, M., Holubová, J., Hájková, Z., Albrecht, V., Marhoul, Z., and Simonová, V. (1983) Influence of microclimatic factors on the development and virus infection rate of ticks *Ixodes ricinus* (L.) under experimental conditions. *Folio Parasitol.* **30**, 153.

Das, S.S. and Sharma, N.N. (1991) Effect of temperature on transstadial transmission of *Theileria annulata* in *Hyalomma anatolicum antolicum* ticks. *Vet. Parasitol.* **40**, 155.

Despins, J.L. (1992) Effect of temperature and humidity on ovipositional biology and egg development of the tropical horse tick, *Dermacentor* (*Anocentor*) *nitens. J. Med. Entomol.* **29**, 332.

Dusbábek, F. (1985a) Identity of *Argas* (*Argas*) *polonicus* populations in Czechoslovakia and Poland. Folia Parasitol. **32**, 163.

Dusbábek, F. (1985b) Biological comparison of different populations of *Argas* (*Persicargas*) *persicus* (Oken). *Folia Parasitol.* **32**, 255.

Dusbábek, F. (1988) Comparative study of local populations of *Argas* (*Persicargas*) *persicus* (Ixodoidea: Argasidae). pp. 61–67. *In:* Channabasavanna, G.P. and Viraktamath, C.A. (eds) *Progress in Acarology*, Vol. I. Oxford and IBH Publ. Co., PVT., Ltd, New Delhi, Bombay, Calcutta.

Dusbábek, F., Daniel, M., and Černý, V. (1971) Stratification of engorged *Ixodes ricinus* larvae overwintering in soil. *Folia Parasitol.* **18**, 261.

Dyk, V. and Boučková, I. (1968) Die Temperatur-Feuchte-Relation in der Aktivität des gemeinen Holzbocks (*Ixodes ricinus* L.). *Angew, Parasitol.* **9**, 83.

Ekpenyong, G.D. and Akinboade, O.A. (1991) The effect of humidity on the reproduction of *Amblyomma variegatum* (Fabricius, 1974) (Acarina: Ixodidae). *Acarologia* **32**, 115.

Estrada-Peña, A. and Sanchez-Acedo, C. (1988) Studies on activity of *Rhipicephalus* (*Digienus*) *bursa* (Acarina: Ixodidae), under natural conditions. *Acarologia* **29**, 267.

Estrada-Peña, A. and Dusbábek, F. (1993). Cuticular hydrocarbon gas chromatography analysis of *Argas vulgaris, A. polonicus* and their hybrids. *Exp. Appl. Acarol.* **17**, 365.

Filippova, N.A. (1966) Argasid ticks (Argasidae). pp. 1–255. *Fauna of the USSR, Arachnids*, Vol. IV, no. 3. Izd. Nauka Publishers, Moscow, Leningrad (in Russian).

Friedhoff, K.T. (1990) Interaction between parasite and tick vector. *Internat. J. Parasitol.* **20**, 525.

Frolov, B.A. (1970) Studies on *Argas persicus*. II. Development of larval ticks. *Probl. Vet. Sanit., Trudy VNIIVS* **35**, 409 (in Russian).

Fujimoto, K. (1990). Ecological studies on ixodid ticks 7. The effects of humidity on oviposition development and survival of *Ixodes ovatus* Neumann (Acarina, Ixodidae). *Jap. J. Sanit. Zool.* **41**, 331.

Fujisaki, K. and Kamio, T. (1988) Effect of constant temperature on *Theileria sergenti* infection in salivary glands of nymphal *Haemaphysalis longicornis*. *Jap. J. Vet. Sci.* **50**, 529.

Gray, J.S. (1981) The fecundity of *Ixodes ricinus* (L.) (Acarina: Ixodidae) and the mortality of its developmental stages under field conditions. *Bull. Ent. Res.* **71**, 533.

Gray, J.S. (1982) The development and questing activity of *Ixodes ricinus* under field conditions in Ireland. *Bull. Ent. Res.* **72**, 263.

Hackman, R.H. and Filshie, B.K. (1982) The tick cuticle. pp. 1–42. *In:* Obenchain, F.D. and Galun, R. (eds) *Physiology of Ticks*. Pergamon Press, Oxford.

Hafez, M., El-Ziady, S., and Hefnawy, T. (1970) Biochemical and physiological studies of cerrtain ticks (Ixodoidea). Cuticular permeability of *Hyalomma* (*H.*) *dromedarii* Koch (Ixodidae) and *Ornithodoros* (*O.*) *savignyi* (Audoin) (Argasidae). *J. Parasitol.* **56**, 154.

Hafez, M., Abdel-Malek, A.A., and Guirgis, S.S. (1971) The subgenus *Persicargas* (Ixodoidea, Argasidae, *Argas*). 12. Biological studies on the immature stages of *A.* (*P.*) *arboreus* Kaiser, Hoogstraal et Kohls in Egypt. *J. Med. Entomol.* **8**, 412.

Hall, R.R. and McKeil, J.A. (1961) Occurrence of the American dog tick, *Dermacentor variabilis* (Say) in western Nova Scotia. *Can. Entomol.* **93**, 891.

Harlan, H.J. and Foster, W.A. (1990) Micrometeorological factors affecting host-seeking activity of adult *Dermacentor variabilis* (Acari: Ixodidae). *J. Med. Entomol.* **27**, 471.

Hejný, S. and Rosický, B. (1962) Biocenological–parasitological studies on the existence of a potential elementary focus of tick-borne encephalitis in the surroundings of Iskra village. *Bull. Inst. Microb., Acad. Sci. Bulg.* **14**, 9.

Hejný, S. and Rosický, B. (1965) Beziehungen der Encephalitis zu den natürlichen

Pflanzengesellschaften. pp. 341–347. *In:* Tuxen, R. (ed) *Biosoziologie.* Junk Verlag, Den Haag.

Honzáková, E., Černý, V., Daniel, M., and Dusbábek, F. (1980) Development of the tick *Ixodes laguri* Ol. in the nests of the European suslik *Citellus citellus* (L.). *Folia Parasitol.* **27**, 71.

Hugh-Jones, M. (1989) Applications of remote sensing to the identification of the habitats of parasites and disease vectors. *Parasitol. Today* **5**, 244.

Hugh-Jones, M. (1991a) LANDSAT-TM identification of the habitats of the cattle tick, *Amblyomma variegatum*, in Guadeloupe, French Windward Islands. *Prev. Vet. Med.* **11**, 355.

Hugh-Jones, M. (1991b) The remote recognition of tick habitats. *J. Agric. Entomol.* **8**, 309.

Hunt, L.M. (1986) Differentiation between three species of *Amblyomma* ticks (Acari: Ixodidae) by analysis of cuticular hydrocarbons. *Ann. Trop. Med. Parasitol.* **80**, 245.

Hussein, S.H. and Mustafa, B.E. (1987) Temperature and humidity effects on the life cycle of *Haemaphysalis spinulosa* and *Rhipicephalus simus* (Acari: Ixodidae). *J. Med. Entomol.* **24**, 77.

Irvin, A.D., Boarer, C.D.H., Doubelare, D.A.E., Mahan, S.M., Masake, R., and Ocama, J.G.R. (1981) Monitoring *Theileria parva* infection in adult *Rhipicephalus appendiculatus* ticks. *Parasitology* **82**, 13.

Jusatz, H.J. (1978) Beitrage zur Geoökologie der Zentraleuropäischen Zecken-Encephalitis. *Sitzungsber. Heidelberger Akad. Wiss., Math.-nat. Klasse* **2**, 105.

Jusatz, H.J. (1981) Epidemio-geoecological analysis of the incidence of TBE in Baden-Wurttemberg. pp. 262–264. *In:* Kunz, Ch. (ed) *Tick-borne Encephalitis.* Facultas-Verlag, Vienna.

Kahl, O. and Knülle, W. (1986) Water vapour uptake by engorged immature ixodid ticks. pp. 275–278. *In:* Vethuis, H. (ed) *Proc. 3rd Eur. Congr. Entomol.*, Part 2. University Press, Utrecht.

Kahl, O. and Knülle, W. (1988) Water vapour uptake from subsaturated atmospheres by engorged immature ixodid ticks. *Exp. Appl. Acarol.* **4**, 73.

Kheisin, E.M. (1953) Behavior of adult *Ixodes persulcatus* P. Sch. in connection with temperature and humidity of surrounding ambient. *Zool. Zh.* **32**, 77 (in Russian).

Knülle, W. (1966) Equilibrium humidities and survival of some tick larvae. *J. Med. Entomol.* **2**, 335.

Knülle, W. and Wharton, G.W. (1964) Equilibrium humidities in arthropods and their ecological significance. *Acarologia* **6**, 299.

Knülle, W. and Rudolph, D. (1982) Humidity relationships and water balance of ticks. pp. 43–70. *In:* Obenchain, F.D. and Galun, R. (eds) *Physiology of Ticks.* Pergamon Press, Oxford.

Koch, H.G. and Tuck, M.D. (1986) Moulting and survival of the brown dog tick (Acari: Ixodidae) under different temperatures and humidities. *Ann. Entomol. Soc. Am.* **79**, 11.

Kuzikov, I.V., Korenberg, E.I., Kovalevsky, Yu.V., and Rodman, L.S. (1982) The principles of average scale mapping of distribution of the ixodid ticks on the basis of aerophotomaterials. *Zool. Zh.* **61**, 1802 (in Russian).

Lane, R.S., Anderson, J.R., Yaninek, J.S., and Burgdorfer, W. (1985) Diurnal host seeking of adult Pacific coast ticks, *Dermacentor occidentalis* (Acari, Ixodidae) in relation to vegetational type, meteorological factors, and Rickettsial infection rates in California, USA. *J. Med. Entomol.*, **22**, 558.

Lebedeva, N.N. and Filchagov, A.G. (1985) New data on ecology of *Ixodes ricinus* L. larvae. *Fifth Acarol. Meeting, Frunze, Abstracts of Papers*, p. 186 (in Russian).

Lees, A.D. (1946) The water balance in *Ixodes ricinus* L. and certain other species of ticks. *Parasitology* **37**, 1.

Lees, A.D. (1947) Transpiration and the structure of the epicuticle in ticks. *J. Exp. Biol.* **23**, 379.

Lees, A.D. (1951) The seasonal and diurnal activities of individual sheep ticks (*Ixodes ricinus* L.). *Parasitology* **41**, 189.

Leonovich, S.A. (1989) The ethology of the taiga tick, *Ixodes persulcatus*, during the period of spring activity. *Parazitologia* **23**, 1 (in Russian).

Levin, M.L. (1985) Horizontal migration of empty larvae of taiga tick and the size of larval dispersal areas. *Fifth All Union Acarol. Meeting, Abstracts of Papers*, p. 187 (in Russian).

Loew, J. (1964) Uber den Einfluss der Photoperiode auf die Metamorphose von *Ixodes ricinus*. *Angew. Parasitol.* **5**, 3.

Loew, J., Radda, A., Pretzmann, G., and Groll, E. (1963) Untersuchungen in einem Naturherd der Frühsommer-Meningoencephalitis (FSME) in Niederösterreich I. Mitteilung: Ökologie und Saiosondynamik von *Ixodes ricinus*. *Zbl. Bact. Paras. Inf. Hyg., I. Abt. Orig.* **190**, 183.

Luta, A.S. and Schulman, R.E. (1958) The influence of microclimatic condition of the meadow and forest on the survival and activity of the tick *Ixodes ricinus* L. Zool. Zh. **37**, 1813 (in Russian).

MacLeod, J. (1935a) *Ixodes ricinus* in relation to its physical environment. II. The factors governing survival and activity. *Parasitology* **27**, 123.

Macleod, J. (1935b) *Ixodes ricinus* in relation to its physical environment. III. Climate and reproduction. *Parasitology* **47**, 489.

Martinod, S. and Gilot, B. (1991) Epidemiology of canine babesiosis in relation to the activity of *Dermacentor reticulatus* in southern Jura (France). *Exp. Appl. Acarol.* **11**, 215.

Matuschka, F.R., Richter, D., Fischer, P., and Spielman, A. (1990) Nocturnal detachment of the tick *Ixodes hexagonus* from nocturnally active host. *Med. Vet. Entomol.* **4**, 415.

McEnroe, W.D. (1979) Overwintering survival restriction of *Ixodes scapularis* populations in Massachusetts. *Acarologia* **20**, 214.

Minshull, J.I. and Norval, R.A.I. (1983) Factors influencing the spatial distribution of *Rhipicephalus appendiculatus* in Kyle Recrational Park, Zimbabwe. *FAO Anim. Health. Serv., Inf. Circ. Tick-borne Dis. Livestock.* AGA/83/Inf. 38.

Mishaeva, N.P. and Erofeeva, N.I. (1979) The effect of diapause of the tick *Ixodes ricinus* (Ixodidae) upon the multiplication of the tick encephalitis virus in them. *Parazitologia* **13**, 218 (in Russian).

Needham, G.R. and Teel, P.D. (1991) Off-host physiological ecology of ixodid ticks. *Ann. Rev. Entomol.* **36**, 659.

Newson, R.M., Chiera, J.W., Young, A.S., Dolan, T.T., Cunningham, M.P., and Radley, D.E. (1984) Survival of *Rhipicephalus appendiculatus* (Acarina: Ixodidae) and persistence of *Theileria parva* (Apicomplexa: Theileriidae) in the field. *Internat. J. Parasitol.* **14**, 483.

Nosek, J. (1978) The effect of microclimate on *Ixodes ricinus*. pp. 105–116. *In:* Gibson, T.E. (ed) *Weather and Parasitic Animal Disease*. World Meteorological Organisation, Geneva, Technical Note no. 159.

Nosek, J., Kožuch, O., and Grulich, I. (1970) The structure of tick-borne encephalitis (TBE) foci in Central Europe. *Oecologia* **5**, 61.

Odum, E.P. (1971) *Fundamental of Ecology*, 3rd edn. W.B. Saunders Company, Philadelphia, London, Toronto. 574 pp.

Ohman, Ch. (1961) The geographical and topographical distribution of *Ixodes ricinus* in Finland. *Acta Soc. Pro Fauna Flora Fennica*, **76**, 1.

Pavlovsky, E.N. (1939) On natural focality of infectious and parasitic diseases. *Vestnik AN SSSR* **10**, 98 (in Russian).

Pavlovsky, E.N. (1948) *A Handbook on the Parasitology of Man*, pp. 527–1022, Vol. II. Publishing House, Academy of Science, USSR, Moscow, Leningrad (in Russian).

Pavlovsky, E.N. and Skrynnik, A.N. (1960) Comparative data on the biology of some species of ticks of the genus *Ornithodorus*. *Reports of the Academy of Sciences of the USSR* (*Doklady Akademii Nauk SSSR*) **133**, 734 (in Russian).

Perry, B.D., Kruska, R., Lessard, P., Norval, R.A.I., and Kunder, K. (1991) Estimating the distribution and abundance of *Rhipicephalus appendiculatus* in Africa. *Prev. Vet. Med.* **11**, 261.

Pikula, J. (1978) Thermostatic capacity of nest, body temperature, and thermoregulation in the young of hemisynanthropic populations of *Turdus merula*, *Turdus philomelos*, and *Sylvia curruca*. *Folia Zool.* **27**, 337.

Pikula, J. (1979) Nest temperature in a hemisynanthropic population of *Turdus philomelos* in Czechoslovakia. *Acta Scientiarum Naturalium Academiae Scientiarum Bohemoslovacae*, Brno **13**, 1–37.

Pound, J.M., Oliver, J.M., Jr, and Andrews, R.M. (1984) Effect of temperature and tick weight on expression of autogeny in the argasid tick *Ornithodoros parkeri* Cooley (Acari: Argasidae). *J. Parasitol.* **70**, 279.

Prokhorov, B.B. (1974) *Mapping of Ixodid Ticks Distribution—Experience in Asian Russia.* pp. 1–83. Academy of Sciences of the USSR, Siberian Department, Irkutsk (in Russian).

Reiter, P. (1988) Weather, vector biology, and arboviral recrudescence. pp. 245–256. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology*, Vol. II. CRC Press Inc., Boca Raton, FL.

Robertson, A.S., Patrik, C.D., Semter, P.J., and Hair, J.A. (1975) The ecology and behaviour of the lone star tick (Acarina: Ixodidae). VI. Response of unfed adults to certain environmental parameters. *J. Med. Entomol.* **12**, 525.

Rosický, B. (1962) On the structure and development of elementary foci of infections in cultivated areas. pp. 134–148. *In: Advances of Biological Science.* Publ. House Czechoslovak Academy of Science, Prague.

Rosický, B. (1967) Natural foci of diseases. pp. 108–127. *In:* Cockburn, T.A. (ed) *Infectious Diseases—their Evolution and Eradication.* Ch. C. Thomas, Springfield, IL.

Rosický, B. and Hejný, S. (1961) Structure of elementary foci of tick-borne encephalitis and the possibilities of their indication by certain phytocenoses. pp. 420–422. *In:* Libikova, H. (ed) *Biology of Viruses of the Tick-borne Encephalitis Complex.* Publishing House of the Czechoslovak Academy of Science, Prague.

Rosický, B. and Daniel, M. (1989) *Lekarska Entomologie a Zivotni Prostredi* (*Medical Entomology and Environment*). pp. 1–437. Academia, Prague (in Czech).

Rudolph, D. and Knülle, W. (1979) Mechanisms contributing to water balance in non-feeding ticks and their ecological implication. pp. 385–393. *In:* Rodriguez, J.G. (ed) *Recent Advances in Acarology* Vol. 1. Academic Press, NY.

Rudolph, D. (1982) Novel uptake system for atmospheric water vapour among insects. *J. Exp. Zool.* **222**, 3212.

Rukhkian, M.J. (1987) On the vertical migration of hungry imagos of *Dermacentor marginatus*. *Parazitologia* **21**, 680 (in Russian).

Samish, M. (1977) Infective *Theileria annulata* in the tick without a blood meal stimulus. *Nature* **270**, 51.

Samšiňáková, A., Kálalová, S., Daniel, M., Dusbábek, F., Honzáková, E., and Černý, V. (1974) Entomogenous fungi associated with the tick *Ixodes ricinus* (L). *Folia Parasitol.* **21**, 39.

Sauer, J.R. and Hair, J.A. (1971) Water balance in the lone star tick (Acarina: Ixodidae): the effect of relative humidity and temperature on weight changes and total water content. *J. Med. Entomol.* **8**, 479.

Semtner, P.J. and Hair, J.A. (1973) The ecology and behaviour of the lone star tick (Acarina: Ixodidae). IV. The daily and seasonal activity patterns of adults in different habitat types. *J. Med. Entomol.* **10**, 337.

Shashina, N.I. (1985) Conditions and terms of eggs development. pp. 263–264. *In:* Filippova N.A. (ed) *Taiga Tick* Ixodes persulcatus *Schulze* (Acarina: Ixodidae). Nauka Publ., Leningrad.

Shiraishi, S., Yoshind, K., and Uchida, T.A. (1989) Studies on seasonal fluctuations of populations and overwintering in the cattle tick *Haemaphysalis longicornis*. *J. Fac. Agric. Kyushu Univ.* **34**, 43.

Short, N.J., Floyd, R.B., Norval, R.A., and Sutherst, R.W. (1989) Survival and behaviour of unfed stages of the ticks *Rhipicephalus appendiculatus*, *Boophilus decoloratus* and *Boophilus microplus* under field conditions in Zimbabwe. *Exp. Appl. Acarol.* **6**, 215.

Sigal, M.D., Machin, J., and Needham, G.R. (1991) Hyperosmotic oral fluid secretion during active water vapour absorption and during desiccation-induced storage excretion by the unfed female tick, *Amblyomma americanum*. *J. Exp. Biol.* **157**, 585.

Siuda, K. (1981) Investigation on the biology of the tick *Argas* (*Argas*) *polonicus* Siuda, Hoogstraal, Clifford et Wassef, 1979 (Acarina: Ixodides: Argasidae) 3. Effect of temperature and relative humidity on embryonic development and egg hatch. *Folia Biol.* (*Krakow*) **29**, 9.

Sonenshine, D.E. (1970) Current studies on tick biology in relation to disease in the Americas. *Misc. Publ. Entomol. Soc. Am.* **6**, 352.

Sonenshine, D.E. (1977) Radiant energy and tick activity. pp. 117–125. *In:* Gibson, T.E. (ed) *Weather and Parasitic Animal Disease*. World Meteorological Organisation, Geneva, Technical Note 159.

Sonenshine, D.E. (1988) Diapause in tick vectors of disease. pp. 219–243. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology*, Vol. I. CRC Press Inc., Boca Raton, FL.

Stafford, K.C. (1992) Oviposition and larval dispersal of *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **29**, 129.

Szymanski, S. (1979) *Rhipicephalus sanguineus* (Latreille, 1806) a new member of the Polish fauna of ticks (Parasitiformes, Ixodidae). *Bull. Acad. Pol. Sci., Ser. Sci. Biol.* **27**, 555.

Tovornik, D. (1970) *Ekosysteme von Arbovirus-Infektionen in Slowenien und in einigen anderen Gebieten Jugoslawiens*. pp. 1–81Acad. Sci. et Art. Slovenica, Ljubljana.

Vershinina, T.A. (1985) *Mapping of the Distribution and Seasonal Activity of Ixodid Ticks*. Publ. House Nauka, Novosibirsk (in Russian).

Vesenjak-Hirjan, J., Tovornik, D., and Soos, E. (1965) Geographical variety of biotopes containing foci of tick-borne encephalitis in Yugoslavia. pp. 111–119. *In:* Rosický, B. and Heyberger, K. (eds) *Theoretical Questions of Natural Foci of Diseases.* Publishing House of the Czechoslovak Academy of Science, Prague.

Walker, A.R. and McKeller, S.B. (1983) The maturation of *Theileria annulata* in *Hyalomma anatolicum anatolicum* stimulated by incubation or feeding to produce sporozoites. *Vet. Parasitol.* **13**, 13.

Walton, G.A. (1960) The reaction of some variants of *Ornithodoros moubata* Murray (Argasidae: Ixodidae) to desiccation. *Parasitology* **50**, 81.

Wilson, M.L., Ducey, A.M., Litwin, T.S., Gavin, T.A., and Spielman, A. (1990) Microgeographic distribution of immature *Ixodes dammini* ticks correlated with that of deer. *Med. Vet. Entomol.* **4**, 151.

Yano, Y., Shiraishi, S., and Uchida, T.A. (1988) Effect of humidity on development and growth in the tick *Haemaphysalis longicornis. J. Fac. Agric. Kyushu Univ.* **32**, 141.

Young, A.S., Lietch, B.L., and Omwoyo, P.L. (1979) Induction of infective stages of *Theileria parva* by exposure of host ticks to high temperature. *Vet. Rec.* **105**, 531.

Zaslavsky, V.A. (1972) Two-step photoperiodic reactions as basis for elaborating the model of photoperiodic control of arthropod's development. *Entomol. Obozr.* **51**, 217 (in Russian).

Zaslavsky, V.A. (1975) The distribution of two-step photoperiodic reactions among insects, mites and ticks. *Entomol. Obozr.* **54**, 913 (in Russian).

6

Geographic Dissemination of Tick-borne Zoonoses

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1. INTRODUCTION

The recent discovery of new tick-borne human illnesses caused by spirochetes (Lyme disease), sporozoans (human babesiosis) and rickettsia (human ehrlichiosis), as important public health threats has refocused attention on ticks, their basic ecology and the epidemiology of the diseases they transmit. In addition to newly identified tick-borne pathogens, the emergence of other infectious diseases (Legionnaire's disease, acquired immunodeficiency syndrome, etc.) has heightened the sensitivity of the general public and the media to public health issues. The question the scientific community is frequently faced with by the public and media is "where did this new disease come from?" At the outset, we can usually only provide best possible guesses, however, new technologies and advancements in such diverse fields as molecular biology, remote sensing, and geographic information systems, combined with existing disciplines, are hastening our ability to respond to these queries and may help in uncovering answers to the more complex issues involved in identifying origins.

Tick-borne diseases (TBDs) have been described as diseases of place (Pavlovsky, 1966), i.e., pathogens that are associated predictably with particular foci based on the structure of biomes or communities (biocenose). The occurrence of a particular TBD is a dynamic event, however, and often, our view represents only a temporal snapshot of processes which have culminated in the current association of an infectious agent with a particular vector in a specific setting. Analysis of the distribution of TBDs must therefore focus on the interaction, dissemination, and co-evaluation of different organisms across time and space.

The factors underlying the emergence of a novel disease, or the geographic expansion of an existing infectious agent range from events transpiring at the molecular level to global events requiring millennia to evolve. The epidemiological time-line defining the distribution of a newly recognized infectious disease typically involves several phases, beginning with the discovery of the etiologic agent, followed by the subsequent identification of additional

occurrences. Comparison of geographic strains of the pathogen hopefully produces information which can be used to elucidate its phylogeny. A number of hypotheses may be considered to account for the emergence of the disease agent: mutational events or shifts in the genetic structure of an existing infectious agent: exploitation or competition for a vacant niche or transmission route by the pathogen; altered or novel migration, importation, or dispersal by vectors or hosts; natural successional changes in biotic communities leading to open niches; increased human contact with endemic disease cycles through expansion of commerce, etc. This latter point is frequently the most important factor in our anthropocentric view of "emergent" diseases which were most likely extant before man's intrusion into the focus. As an example, the presence of relapsing fever borrelia, Asian tick typhus and tick-borne encephalitis in virgin forest areas of Azerbaijan, Tajikistan, Kazakhstan, and Siberia had a major impact on plans for development of these regions (Babenko, 1967). The basis for emergence of new TBDs can be summarized simply as: (1) the agent was endemic but unidentified; (2) the agent changes; or (3) the agent recently moved in.

This chapter will focus on aspects of dissemination of TBDs within and between habitats, the agents themselves, and the mechanisms which may have influenced the currently observed geographic patterns. Dissemination of TBDs involves the basic interaction between a tick and its host which leads to dispersal of the tick, and consequently to the infectious agents. This underlying mechanism conditions the TBD dispersal events such as those caused by bird and mammal migrations and man's influence in transporting animals. In addition to these natural events, man has directly affected geographic patterns of TBD by agricultural practices in maintaining and transporting domestic livestock, and in creating suitable habitats for endemic disease. One theme of this chapter is that fundamental distribution patterns of TBDs observed today reflect trends in mammalian evolutionary radiation and dispersal, particularly in the ungulate and rodent faunas. The gradual speciation and dispersal of these mammalian taxa over the Cenozoic era (65 million years to the present) coincident, or in response to macroclimatic change, and rearrangement of major land mass associations are the primary forces governing TBD biogeography. An excellent treatment of this hypothesis is provided by Marchette (1982) with regard to ticks and rickettsial diseases. The daily and seasonal patterns of mammal and avian movements contribute in an ongoing manner to the dispersal and mantainenance of these agents.

I first discuss Lyme disease distribution as an example of the processes involved in defining the distribution of the disease. Next, I consider several other examples of widely distributed TBDs, evaluating possible parallels in distributions of these major complexes, such as the spotted fever group rickettsia and the tick-borne flaviviruses. I thereafter provide terminology and considerations for tick dispersal follwed by the role of self-directed movement by ticks (vagility) and the influence of different host groups on the dissemination of ticks and associated infectious agents.

1.1. Lyme Disease as an Example of Tick-borne Disease, Time Scale, and Biogeographic Factors

Lyme disease serves as a model of how information gathered for modern events (past 90 years), earlier historical events, and prehistoric events suggests origin and dissemination patterns for a newly recognized TBD. This syndrome was described following an elegant epidemiological assessment of an unusual cluster of juvenile arthritis cases in a community located in the Northeastern US (Steere et al., 1977). The subsequent identification of a spirochete, *Borrelia burgdorferi*, in the tick vector, *Ixodes scapularis** (Burgdorfer et al., 1982), opened the way to assess the pathogen's geographic distribution. At present, this disease is reported from most of the "holarctic" land mass, with a disjunct focus in Australia. The apparent underlying factor for this geographic distribution is the presence of vectors of the *Ixodes ricinus* complex, of which *I. scapularis* is a member.

The wide distribution of human cases indicated that this syndrome was present as a long-standing zoonosis. Clinical recognition of human involvement could be traced back in the medical literature at least to 1909 and an etiology was proposed as early as the 1940s (Burgdorfer, 1983). Using polymerase chain reaction (PCR) techniques, Persing et al. (1990) demonstrated that *B. burgdorferi* was present in archived tick specimens of *Ixodes scapularis* from Long Island, New York, at least 30 years before the syndrome was fully resolved. Historical accounts of the distribution of the ixodid reservoirs of the agent suggest that the disease could have been continually present in New England and Ontario, Canada, during the 18th century (Spielman et al., 1985; Anderson, 1988). Evidence that the agent is expanding from these "refugia" can be inferred from demonstrated expansion in the geographic range of *I. scapularis* found on deer during surveys over the past decade in locations such as Wisconsin (French et al., 1992).

Comparative analysis of the *B. burgdorferi* genome from various geographic isolates by Rosa et al. (1991) suggest the presence of two major groups, a "western strain" encompassing North American and western European isolates, and an "eastern strain" comprising isolates from eastern Europe and Northern Asia. This general finding was confirmed by Marconi and Garon (1992), using sequence information from the 16s ribosomal RNA, and then expanded by identification of a third genomic group from spirochetes isolated from *I. persulcatus* in the Leningrad region. These analyses suggest a geographic distribution of the *Borrelia* groups as: (1) Western Europe and North America strains; (2) Central European strains; and (3) East European (Russian)/Asian (?) strains. Immunological analysis of *B. burgdorferi* strains by a variety of other investigators generally reveals a higher degree of antigenic homogeneity within North American strains than is found between European strains. These findings raise questions about the geographic origin of the pathogen. Does the greater

* *Ixodes scapularis* replaces the nomenclature for *Ixodes dammini* in this chapter based on synonymy suggested by Oliver et al. (1993).

antigenic variability observed for palearctic *B. burgdorferi* isolates reflect a longer evolutionary record or more diversity in available niches in that region? Was Lyme disease introduced within historical times into the New World from Eurasia, possibly during the colonial expansion era leading to a "founder effect" in North America, or was the North American strain introduced into Europe from the New World. Alternatively, was the disease present all along in both areas as a relict from previous epochs?

Indirect evidence suggesting a North American origin, which spread to western Europe during the historical era derives from discovery of peripheral rheumatoid polyarthritis in North American human remains dated 5,000 years BC (Rothschild et al., 1988). These investigators indicate that no such clinical findings were evident in European skeletal material prior to the 1800s, and propose that pathogens or allergens producing these effects may have been transported from the New World to the Old World. This would suggest that the agent was continuously present in eastern North America. Therefore, the spirochete and its vectors may have been widely distributed prior to destruction or alteration of the native deciduous forest habitat by the European immigrants to the region. These subsequent changes in habitat reduced the resource base for the vector species, i.e., the native rodents and artiodactyls which served as the chief blood source for the adult stages, and as the chief amplifying hosts for the pathogen. Under this scenario, small isolated refugia of the vertebrate hosts, ticks, and pathogens, however, persisted. Land-use patterns and cultural practices changed again during the 20th century, with resulting re-emergence of the deciduous forest climate in various broad areas of the original range. As suitable habitat and the vertebrate host populations re-expanded, these refugia gradually expanded and coalesced. The speed with which new disease foci were established is exemplified by the rapid emergence of a focus of human illness in a community along coastal Massachusetts, USA (Lastavica et al., 1989), where tick infestation data from deer prior to 1980 indicated an absence of *I. scapularis* (Spielman et al., 1985), but where, by 1987, there was epidemic transmission of Lyme disease (35% attack rate in humans) and an abundant deer tick population. Conditions in this community were preadapted, however, for the rapid rise of the disease, since the main hosts for the tick and spirochete were already present in a nearby nature reserve.

But this conjectural chronology begs the question of the primordial source and dissemination route for this TBD. The principal Lyme disease tick vectors are members of the *I. ricinus/I. persulcatus* group (reviewed by Keirans et al., 1992 and Filippova, 1991). These ticks are all members of the subgenus *Ixodes* s.str., and are included in an "artificial" taxonomic construct containing 13 members. The phylogenetic distances of species within this group are not well elucidated yet, therefore discussion of putative dispersal routes remains speculative. Filippova (1990) suggested that the roots of the *persulcatus* group could extend to the Paleocene and also hypothesized (Filippova, 1991) that the distribution ranges (and subsequent disjunction) of a number of these species is related to the paleological distribution of nemoral mesophilic broad-leaf deciduous forest. These forests were expansive in the holarctic land masses

during much of the Tertiary, when milder, more humid conditions prevailed until the Upper Pliocene (6 m.y.). Species such as *ricinus* and *scapularis* share common ecological niches within this forest biome. *I. persulcatus*, the taiga tick, has ecological affinities with younger, more boreal forest habitats, which likely arose during the gradual cooling of the Pliocene, culminating in the dramatic climatic changes of the Pleistocene. *I. persulcatus*, ranging east of the Ural Mountains, shows greater morphological similarity to *I. pacificus*, which is distributed on the west coast of the US, than to *I. ricinus*, an allopatric species to the west of the Urals.

These general associations suggest two different dispersal routes for the holarctic distribution of the *ricinus/persulcatus* group. An *I. ricinus–I. scapularis* association would have origins in western Laurasia and separation due to separation of the continent along the North Atlantic (i.e., pre-Eocene; Balashov, 1989). The *I. persulcatus–I. pacificus* divergence would have been due to dispersal events occasioned by the more recent Beringian route (Pliocene to Pleistocene). Additionally, this would suggest that the spirochetal agent shared a close relationships to this group throughout this geological long period. The genus *Ixodes* is considered to be one of the most primitive of the hard (ixodid) ticks. Filippova (1991) implies that the association of Lyme *Borrelia* with this more primitive tick genus is ancient, pointing out that the other major tick–*Borrelia* association, the relapsing fever diseases, is also found with the more primitive argasic ticks. Can other tick-borne disease agents be viewed with a similar regard to geological and evolutionary events to yield possible insights into the current status of geographic distributions?

The distribution of the Lyme disease agent in the Northern Hemisphere is contingent on events which transpired over the Mesozoic and Cenozoic (Table 6.1). During the mid-Cretaceous period (*c.* 110–100 m.y.), the northern hemisphere continent of Laurasia split into the North American land mass and the Eurasian land mass along the Atlantic side of these present-day continents. A North American–western European (sub-Baltic) connection via Iceland was broken in the early Eocene period (56–57 m.y.) with much more of the connection still intact above sea level up to the Miocene. A parallel dispersal route into Europe from North America continued for an undetermined interval via Greenland and Spitzbergen into the Fennoscandinavian Plate throughout the Eocene (McKenna, 1983). For most of the Tertiary Period up to the Oligocene, the land mass of Eurasia was split east of the Urals by an epicontinental sea (Turgai Strait), which likely subsided about 30 million years ago. Direct dispersal between Asia and Europe, south of the Baltic was limited and exchange between these regions may have relied on a circuitous route through Fennoscandinavia, Greenland, Proto-Iceland and into Western Europe. Similarly, movement between Asia and North America was difficult. Despite a fairly continuous overland route for movement across the Bering from the Cretaceous to the present, climatic factors have played an important filtering mechanism for this route. This area was at higher paleolatitude, being much closer to the rotational pole in the early Cenozoic. Since the Cretaceous, the region has shifted approximately 13° southward. A global cooling trend

Table 6.1. Description of major earth history and mammalian evolution which influenced dissemination of tick vectors and tick-borne pathogens

Geologic era	Time (m.y.)	Geologic events[a]	Mammalian evolution[b]	Presumptive tick evolution[c]
Early Triassic	⩾200	Pangaea—unified continents	Mammal-like reptiles	Argasids already present. Primitive *Ixodes*, *Aponnoma*, *Amblyomma* and *Haemaphysalis*
Late Triassic Early Jurassic	180	Western Laurasia (NA)/Africa rift West Gondwana (AF + SA)/IND/East Gondwana (AUS + ANT) rift	Primitive mammals (pantotheres, multituberculates, triconodonts, tilodonts)	
Early Cretaceous	135–125	AF/SA rift at southern end. NA large central continental sea. Eurasia large central continental sea. AF/EUR still connected at SW EUR. Bering at high latitude (75°N)	Marsupials and early placentals	Early *Hyalomma* possible in southwest AS
Mid-Cretaceous	110–100	AF/SA rift at mid-line. AF/Madagascar/IND rift continues. AF/IND/AUS move northward, last possible direct AUS—Gondwana route. Drying of NA inland seas (?)	Insectivora flourish (NA and EUR). Arctocyonids (pre-carnivora) in NA, EUR. Condylarths (pre-ungulates) in NA, EUR	Rhipicephalinae diverge
	90	Final AF/SA split		
Late Cretaceous	80	NA/EUR and Greenland rift begins		
	70	NA/AS contact in NW Pacific		
Early Paleocene	65	AF/EUR temporary rift (Tethys) but still connected via Spain and possible AF/AS connection through Arabia		
Mid-Paleocene Late Paleocene	60 55	Gradual split of AF into isolation	Primitive rodents and primates (*Plesiadapis*) in NA and EUR	

Geologic era	Time (m.y.)	Geologic events[a]	Mammalian evolution[b]	Presumptive tick evolution[c]
Late Paleocene	55	(NA and EUR)	Primitive rodents and primates Primitive ungulates and carnivores (NA and EUR)	
Early Eocene	52	Dispersal route switch into NA still possible by Greenland–Scotland or Fennoscand Plate.	Primitive lagomorphs (AS). Primitive rodent (*Paramys*).	*Ixodes "ricinus"* preserved in Baltic Amber
		Rise of Bering Route between AS/NA	Multituberculates, tilidonts die out	
Mid Eocene	45	IND/AS join		
		Final direct route AUS–ANT–SA	Birds achieve modern diversity	
		Probable end of NA/EUR direct route		
			Great turnover of western EUR mammals with disperses from AS and southeast EUR (Grande Coupure)	
Late Eocene	37		Perissodactyls in NA small forest dwellers.	
		Much of EUR flooded or marshy.	Early artiodactyls in NA and Eurasia expand but diverge.	
		AS/EUR separated in Baltic area	Most carnivore families present (NA and EUR).	
			Early lagomorphs	
Early Oligocene	35		Great turnover of western EUR mammals, influx of groups from AS and southeast EUR (Grande Coupure)	*Dermacentor* in Central AS origin?[d]

(*continued*)

Table 6.1. (continued)

Geologic era	Time (m.y.)	Geologic events[a]	Mammalian evolution[b]	Presumptive tick evolution[c]
Mid-Oligocene	30	Turgai Strait (east of Urals) dries up	NA extinction event for mammals Traguloids present in AS	
		Orogeny in EUR and west NA	Rodent suborders expand, enter AF	
		AF/EUR rejoin	Murids in AS, Cricetids in NA, Ground squirrels in NA and AS, Caviomorphs from NA to SA. Camelids radiate in NA. Elephants and primates enter Eurasia	
Miocene	25	Orogeny in Colorado Plateau	Cervids radiate/AS. Bovids derive from traguloids	*Dermacentor* dispersed to NA (?)
	23	AF/EUR separate again	Perissodactyls now plains dwellers	
	17	AF/EUR rejoin	Advanced Muroid rodents enter AF.	
	15	AUS/Southeast AS stepping stone island dispersal Mediterranean dries to inland desert	Bovid Eurasia center of radiation rapid cladogenesis. Primitive antelope in North AF and Southern Asia.	
	15	Upthrust of Himalayas begins	Extinction events for NA large browsers and some grazers.	
Pliocene	12		Primitive antelope in South AF	
	6	NA/SA join, Mediterranean begins refill	Sheep and goats in montane areas from Mediterranean to AS and NA. Camelids move into SA, extinct in NA. Perissodactyls move into Eurasia/AF. Ox-like bovids in AF and IND, cow-like bovids in Eurasia spread to NA. NA rodents invade SA	*Dermacentor* dispersed to AF (?)

Geologic era	Time (m.y.)	Geologic events[a]	Mammalian evolution[b]	Presumptive tick evolution[c]
Pleistocene	2	Ice sheets on most of northern land masses	Most bovids disappear from EUR but reach peak radiations in AS/AF. Camelids move into AS/AF. Modern deer flourish from Eurasian center. Secondary deer center in NA. Rodents become most diverse herbivores in NA. Camelids move into SA, extinct in NA. Perissodactyls move into Eurasia/AF. Ox-like bovids in AF and IND, cow-like bovids in Eurasia spread to NA. NA rodents invade SA	
Pleistocene	2	Ice sheets on most of northern land masses	Most bovids disappear from EUR but reach peak radiations in AS/AF. Camelids move into AS/AF. Modern deer flourish from Eurasia center. Secondary deer center in NA. Rodents become most diverse herbivores in NA	

[a] From Pielou (1979), McKenna (1983).
[b] From McKenna (personal communication), Romer (1966), Kurten (1971).
[c] From Hoogstraal (1982), Marchette (1982), Berdyev (1989). Evolutionary history of the Ixodoidea is only speculative due to lack of paleontological materials.
[d] Berdyev (1989).
NA = North America; AF = Africa; SA = South America; IND = India; AUS = Australia; ANT = Antarctica; EUR = Europe; AS = Asia.

during the Tertiary was associated with emergence of new biomes, such as grasslands, and concomitantly, major evolutionary radiations occurred within a number of mammalian orders, such as the artiodactyl ungulates and rodent faunas. This likely provides an opportunity for ixodid evolutionary radiation (particularly for the genera *Dermacentor*, *Hyalomma*, *Boophilus* and *Rhipicephalus*; Hoogstraal and Kim, 1981; Berdyev, 1986, and for associated tick-borne diseases (Traub and Jellison, 1981).

If dissemination of *B. burgdorferi* occurred from Asia to North America by way of the more recent Beringian land connections, then North American *B. burgdorferi* strains should be more closely related to "eastern" genomic strains. The relative phylogenetic distances between North American and Asian strains appears greater than that for North American and Western European strains. The presence of the "western strain" in North America would suggest either vicariance in the once contiguous distribution across the former Laurasian landmass (late Cretaceous to early Eocene), or else more recent importation of the pathogen to one or the other continent either via human-related European colonization of North America, or importation by migratory birds travelling between these continents. A more complete synthesis of these events awaits genomic characterization of additional *B. burgdorferi* isolates (or closely related pathogens) from the holarctic region as well as further genomic characterization of the important vectors.

1.2. Tick-borne Disease Complexes and Biogeography: Possible Parallels

Other tick-borne diseases with wide geographic distributions include the Tick-borne Encephalitis (TBE) flaviviruses, Congo–Crimean Hemorrhagic Fever (CCHF) viruses, species of the relapsing fever borrelias and Spotted Fever Group (SFG) rickettsias. Strains of TBE viruses and the SFG rickettsial species show some parallels in range boundaries within each group (Table 6.2), possibly suggesting common dispersal and/or isolating mechanisms.

The geographic distribution of inter-related strains of TBE viruses include central Europe TBE (TBEW—Western and Central Europe), Russian Spring–Summer Encephalitis (TBEFE—Eurasia, Asia), Louping Ill (LI—Great Britain), Kyasanur Forest Disease (KFD—India), Omsk (OM—West Siberia), Langat (LGT—Malaysia), Negishi (NEG—Japan), and Powassan (POW—North America and Primor'ye Commonwealth of Independent States). While these viruses show wide variation in virulence and antigenic markers, reflecting the high mutation rate of RNA viruses, relative evolutionary stability, resulting from maintenance by arthropod vectors (Gresikova and Calisher, 1988) allows some characteristics/mutations to be fixed which may then provide the basis for geographic classification. Genomic or antigenic relations between these virus strains suggest biogeographic events and theoretical dissemination routes for these strains.

The relative phylogenetic distance between four TBE virus strains was

Table 6.2. Comparison of geographic distributions of members of several major tick-borne zoonoses

Region	Tick-borne encephalitis strains	SFG *Rickettsia* species
British Isles	Louping Ill	
Central and East Europe	Central Europe	*slovaka*
Western Europe	Central Europe	*conorii*
Mediterranean	Central Europe	*conorii*
Southwestern Asia	—	*conorii*, Astrakhan
Africa	—	*conorii*
Central and Northeast Asia	RSSE/Omsk	*siberica*, *conorii* (?)
India	KFD	*conorii*
Southeast Asia	Langat	Thai tick agent
Australia	—	*australis*
Japan	Negishi	*japonica*
North America	Powassan	*rickettsii*
South America	—	*rickettsii*

RSSE = Russian Spring–Summer encephalitis; KFD = Kyasanur Forest Disease.

evaluated at the level of nucleotide and amino-acid sequence for the capsid-matrix proteins and for the envelope glycoproteins (Iacono-Connors and Schmaljohn, 1991). The capsid proteins, which complex with the viral RNA to form the nucleoprotein core of the virion are more highly conserved than the external envelope glycoproteins of the virion which interact with cell surface receptors and are exposed to the immunological system of the vertebrate host. The capsid proteins of LI and TBEW strains showed the smallest difference (<2%) in nucleotide sequence, followed by TBEFE (4% difference from this pair), and LGT which differed by 11% from the LI/TBEW and 8% from TBEFE. This phylogenetic dendrogram was different, however, for the envelope glycoproteins, with TBEW and TBEFE showing the greatest similarity in envelope glycoproteins followed by LI and then LGT. This may reflect more recent evolutionary pressures or opportunities for recombinant events as the former boundaries to dispersal of the vectors or vertebrate hosts of these strains have diminished. One outlying finding, however, is that LI and NEG viruses, which are widely separated geographically, shared closer nucleic-acid sequence (and deduced amino-acid sequence) homology at the envelope glycoprotein than either strain did to the other TBE viruses (Venugopal et al., 1992). Antigenically, radioimmunoassay of two epitopes on a 51-kDa polypeptide indicates greater antigenic similarity of TBEW to LI than to TBEFE (Stephenson et al., 1984). Viral isolates from widely separated sites in Western Europe showed greater similarity to each other than to eastern isolates from sites in the former Soviet Union sites as judged by reactivity patterns to a panel of monoclonal antibodies against the envelope glycoprotein (Holzmann et al., 1992). Gresikova and Sekeyova (1990) found that Powassan virus differed antigenically from a group composed of TBEW, TBEFE, and LGT virus strains

based on monoclonal antibodies raised to the Skalica strain (TBEW-related strain). As discussed below, these opposing phylogenetic results for the TBE imply different dissemination routes, and should be examined with reference to ancestral versus derived nucleic-acid (or amino-acid) sequences for these genes. A critical component to this analysis will be further evaluation of the degree of nucleotide and antigenic relatedness of POW virus to the various Eurasian strains which may resolve the biogeographic history and dissemination routes of these tick-borne viruses.

The SFG rickettsial species which are pathogenic for man have the following general distributions: *Rickettsia rickettsii* through much of the western hemisphere; *R. conorii* from southern Europe to southwest Asia, India, and Africa; *R. slovaca* in central Europe; *R. sibericus* from north central Asia, from the Ural Mountains of north central Asia, through China and northeastern Asia; the Thai tick typhus agent in southeast Asia; *R. australis* in Australia. Several recent SFG rickettsial isolates, which are still being characterized, have been obtained from the locations listed in Table 6.2. Several of the rickettsial species have overlapping boundaries or are sympatric. For example, *R. conorii* was isolated from tissues collected from Primor'ye District in northeastern Asia (Wang et al., 1987), a region which is considered the domain of *R. sibericus*. Estimates of genetic divergence within the SFG rickettsia have been made by comparing the proportion of co-migrating restriction fragments derived from PCR-amplifed products of the rickettsial citrate synthase gene (Regnery et al., 1991). An overall divergence of approximately 2.3% was observed between the SFG rickettsial human pathogens. No divergence was found between the Israeli agent and *R. conorii*, which were both highly related to *R. sibericus*. The Thai tick agent diverged slightly from this group. *R. slovaka* clustered with *R. rickettsii* in a second grouping, however, the investigators indicated that this ordering could have been a statistical artefact. If this relationship is further substantiated, however, it would suggest that the *R. rickettsii* in North America and *R. slovaka* in Europe may have diverged (or disseminated) from a common ancestor in western Laurasia, probably during the mid-Eocene. This would predate the alternative hypothesis (Marchette, 1982) or a more recent Beringian dispersal route (Late Eocene through Pleistocene) between eastern Asia and western North America, and possible derivation of *R. rickettsii* and *R. siberia* from a common ancestor.

The geographic patterns for three different pathogens, a rickettsia, virus and spirochete, potentially show similar phylogenetic distributions, and apparently reflect a separation of Asian from West European and North American faunal regions in the distant past. The Turgai strait barrier throughout the Oligocene likely produced isolating conditions which could have resulted in some of the putative relationships observed for both the SFG rickettsias and the TBE viruses. Evolutionary relationships among the relevant tick vectors should also reflect the associations seen for the pathogens. Definitive analysis of the systematics of holarctic *Dermacentor* is lacking, however, Berdyev (1989) recently proposed that this genus evolved in the Asian Oligocene with subsequent dissemination to Europe and North America around the Miocene.

If *R. rickettsii* distribution was contingent on *Dermacentor* dispersal, this would contradict the North Atlantic route hypothesis derived from analysis of the pathogens, unless *Dermacentor* evolved in North America prior to the Oligocene. An earlier ancestry for this tick genus would fit putative relationships derived from analysis of the pathogens. These data, however, are still being argued from a very tenuous position both due to the lack of a paleontological record for the ticks, and scanty genomic data for tick and pathogen. Clearly, further genomic and taxonomic information is thus necessary, from both pathogens and vectors, to construct possible geographic origins and dissemination routes for the SFG rickettsias or tick-borne flaviviruses. Additional information, such as the relationships of POW and the Tyuleniy TBE strains derived from *Ixodes uriae*, with the other strains of TBE, or the phylogenetic distances between the North American versus Eurasian *Dermacentor* species may also provide insight into the observed distributions.

1.2.1. Global Distribution of Tick-borne Arboviruses

On a still larger scale, the global distribution of viral tick-borne agents (Table 6.3) presents another interesting biogeographic pattern. Using geographic records of approximately 100 tick-borne viruses (Karabatsos, 1985) and cross-referencing these records by continent to explore overlap in distribution, South America and Australia yield the smallest total number of tick-borne viruses, while South America and Africa have the lowest level of endemicity of tick-borne viruses. These indices could either reflect the relative level of research effort devoted to these continents compared to the other land masses or could be due to the lengthy separation of these Gondwanaland-derived continents from the northern hemisphere land masses during the Tertiary. The South American tick-borne viral (TBV) flora is represented exclusively by viruses

Table 6.3. Geographic distribution of tick-borne viral agents by continent.[a] Data represent number of isolated viruses, the total proportion of the continental viral flora which is shared and the proportion shared between each continental pair. These data exclude arboviruses which are predominantly associated with dipteran vectors but may also occur in ticks

Continent	Total	% Shared	Continent					
			EUR	AF	AS	AUS	NA	SA
Europe (EUR)	28	32	—	29	14	—	4	8
Africa (AF)	23	65	35	—	43	0	9	9
Asia (AS)	41	30	10	24	—	2	5	0
Australia (AUS)	9	11	0	0	11	—	0	0
North America (NA)	19	21	5	11	11	0	—	11
South America (SA)	6	50	33	33	0	0	33	—

[a]Named viruses according to Karabatsos (1985).

Table 6.4. Geographic distribution of tick-borne viral agents by continent according to tick family and predominant host. The terrestrial host category represents mammals and primarily ticks. Data represent number of isolated viruses, the total proportion of the continental viral flora which is shared and the proportion shared between each continental pair. These data exclude arboviruses which are predominantly associated with dipteran vectors but may also occur in ticks

Terrestrial hosts for ticks								
			Continent					
Continent	Total	% Shared	EUR	AF	AS	AUS	NA	SA
Europe (EUR)								
Argasid	4	25	—	25	0	0	25	25
Ixodid	17	41	—	35	23	0	0	0
Africa (AF)								
Argasid	6	50	17	—	34	0	17	17
Ixodid	14	79	43	—	57	0	7	0
Asia (AS)								
Argasid	8	25	0	25	—	0	0	0
Ixodid	25	36	16	32	—	0	4	0
Australia (AUS)								
Argasid	0	—	—	—	—	—	—	—
Ixodid	1	0	0	0	0	—	0	0
North America (NA)								
Argasid	4	25	0	25	25	0	—	0
Ixodid	7	14	0	14	14	0	—	0
South America (SA)								
Argasid	2	50	50	50	0	0	50	—
Ixodid	0	—	—	—	—	—	—	—

associated with argasid ticks,* including a recently imported virus (African swine flu) and a group of viruses isolated from ectoparasites of pelagic birds (Table 6.4). In Australia, 89% of the endemic TBVs derived from the pelagic avifauna (Table 6.4) all of which fall into larger serogroups of viruses (Dera Ghazi Khan and Kemorovo), are shared with other continents (Table 6.5). Africa has the highest relative proportion of shared TBVs (65%—Table 6.3) suggesting a very high dessemination rate for viruses either into or out of the continent, or a high rate of evolution for viruses within the continent. The TBVs of Africa appear most closely aligned with the ixodid-associated viruses of terrestrial host faunas of Asia (57%—Table 6.4), followed by Europe (43%). This distribution pattern is likely a result of the dispersal and evolution of mammals into the continent following with the Palearctic land masses in the mid-Miocene (Table 6.1) and of dispersal due to the migrating terrestrial avifauna. In this regard, South America has yet to realize any real dissemination

* This consideration discounts the likelihood that Pichinde virus, isolated from an ixodid tick in South America, is tick borne, since the arenaviruses are primarily associated with a rodent-transmitted route.

Table 6.5. Geographic distribution and percent overlap between continents in distribution of major tick-borne virus serogroups (including ungrouped viruses). Data are presented separately for viruses isolated from ticks for all possible hosts, and viruses associated primarily with non-pelagic hosts. Total represents the number of serogroups found within a continent and the proportion which are unique to that land mass. Proportion of shared virus groups given for all pair-wise comparisons of continents

			Continent					
Continent	Total	% Unique	EUR	AF	AS	AUS	NA	SA
Europe (EUR)								
All hosts	13	0	—	62	62	31	38	23
Pelagic	4	0	—	50	100	50	75	50
Terrestrial	9	0	—	66	44	11	22	11
Africa (AF)								
All hosts	16	19	50	—	56	19	19	19
Pelagic	5	20	40	—	60	60	40	40
Terrestrial	11	18	54	—	45	0	9	9
Asia (AS)								
All hosts	19	26	42	47	—	26	26	10
Pelagic	6	16	66	50	—	66	50	33
Terrestrial	12	42	33	42	—	8	15	0
Australia (AUS)								
All hosts	9	11	44	33	56	—	44	11
Pelagic	5	0	60	60	80	—	60	20
Terrestrial	2	50	50	0	50	—	50	0
North America (NA)								
All hosts	9	11	44	33	44	44	—	33
Pelagic	4	0	75	50	75	75	—	50
Terrestrial	5	20	40	20	20	20	—	20
South America (SA)								
All hosts	4	25	75	75	50	25	75	—
Pelagic	2	0	100	100	100	50	100	—
Terrestrial	2	50	50	50	0	0	50	—

of terrestrial-associated TBVs, possibly due to the more recent (Pliocene) connection to North America and to the filtering mechanism of the Central American isthmus. It is interesting to note that one of the more successful mammalian radiations into South America has been by the cricetine rodents, with concomitant radiation of the rodent-borne arenaviruses. Bird migration into South America from North America has apparently had little impact on dissemination of the terrestrial-associated TBVs. Finally, it appears that Asia may be an important focal point for the TBVs, showing high endemicity (Tables 6.3 and 6.4) and serving as a possible source of TBVs for Europe, Africa and North America (Table 6.4).

A variety of questions concerning tick and pathogen distribution can be posed. Based on the distribution of the subgenus *Ixodes*, does *Borrelia burgdorferi* exist in South America (Need and Escamilla, 1991) and Africa (Fivaz and Petney, 1989), and will the discovery of the new human pathogen *Ehrlichia*

chaffeensis (Anderson et al., 1991; Dawson et al., 1991) result in identification of this or other human-infective *Ehrlichia* sp. in other parts of the world (Guerrero et al., 1991)? Why is the TBE complex not represented in Africa, and, why are there so few tick-borne viruses known from South America? What are the phylogenetic relationships between the geographic strains of virus which constitute Crimean–Congo hermorrhagic fever virus and would this information help discern possible dissemination routes? As suggested above, reconstruction of the dissemination routes and vehicles of tick-borne diseases through time and space will require the combined efforts of scientists contributing expertise from a wide range of disciplines.

1.3. General Considerations for the Establishment of a Tick-borne Disease

The concept of a natural nidus for transmissible diseases (Pavlovsky, 1966) suggests that disease transmission takes place in a "defined geographical landscape". Within these nidi, conditions are optimal for support of the reservoir and vector populations responsible for maintaining and transmitting the etiologic agent. Dissemination of pathogens outside these nidi, however, requires either that the pathogen becomes established in a "suboptimal" setting, or that the pathogen be transported across one or more barriers to a preadapted unoccupied site. Some of the factors which contribute to the movement or emergence of tick-borne diseases include: global events (plate tectonics, macroclimatic changes, evolutionary events); man-made alteration of natural communities; ecological changes at a local level (community succession, host population dynamics and resource availability); variation in the pathogen status (mutation, transmission cycle in a novel competent vector species, or importation of the pathogen via dispersal of infected vectors or infected hosts). Dissemination can be viewed on a microhabitat scale (as in the distribution of infected vectors across a landscape) or for greater distances. The basic conditions which must exist for the successful establishment of a tick-borne infectious organism in any new location include at a minimum the following conditions: (1) the ability of the agent to be transported to the new site; (2) the existence of a competent vector species to acquire, maintain, and potentially amplify the agent population; (3) a susceptible host for the agent; and (4) a vertebrate or vector reservoir to transmit the agent vertically. These criteria become more easily met if the vector and pathogen co-disseminate and establish at the new site, and if the vector can vertically transmit the agent. Colonization may be further enhanced if replete mated females constitute the immigrant group versus immature stages. Hence, vertebrates which disperse these females likely play a more important role in dissemination of ticks and pathogens. The parameters associated with expansion of tick-borne diseases foci may be amenable to analysis by island biogeography theory (MacArthur and Wilson, 1967) which evaluates the likelihood of a species successfully colonizing a given site based on rates of entry and loss of the species, and size and distance of the target from a source population.

Local dispersal of TBD is usually determined by the level of interaction between the various hosts and vectors on a continuous basis. Interaction of ticks with their small mammal hosts serve as a good illustration of events conditioning local dissemination. Important differences to consider in host acquisition of ticks by different small mammal species concern the tick–host contact rate, and the physiological/behavioral responses of tick and host following contact. Aggregated distribution of ticks per host are often observed in tick surveys of mammals. This may directly affect transmission dynamics, especially for horizontal transmission of the pathogen (Spielman et al., 1985). Contact rate is influenced by host and vector parameters as habitat preferences, circadian activity cycles, use of habitat during movement, and spatial dispersion of the ticks relative to host movement. Behavioral/physiological variables include grooming activity, host preferences of the tick species, sex of the host, and immunological response by the host to tick infestation (Randolph, 1979). For example, interspecific differences in observed *I. scapularis* levels between *Peromyscus leucopus* and *Microtus pennsylvanicus* and intraspecific variance for younger versus older voles for *I. scapularis* are thought to be partially based on immunological responses to the tick (Korch, 1985; Davidar et al., 1989). Female ticks may be more attractive to subadult *Hyalomma excavatum* (Rechav, 1970), while male white-footed mice may support more *I. scapularis* subadults than females (Davidar et al., 1989). For the purposes of dissemination of TBDs, however, movement parameters of the infested rodent hosts become an overriding consideration.

Animals which travel long distances as part of cyclic seasonal migrations (birds, bats, large ungulates) provide an opportunity for "saltatorial" dissemination of TBD between different biomes or faunal regions. Colonization of an area by a pathogen can be viewed from the perspective of the host, the tick, and the agent (Table 6.6). Different sets of criteria apply for dissemination episodes which place ticks and agents into entirely new biomes, with different flora, fauna, environmental conditions, and possible competitors. A TBD agent and its vector must be capable of surviving a trip, and finding alternative suitable hosts to parasitize along the route or at the other end of the journey, unless the dispersing host is regularly, or predictably available to satisfy the survival of the ectoparasites. It is beyond the scope of this discussion to focus on these important variables in detail, however, the relative potential for transporting a TBD will be a function of some combination of all these variables. Finally, we must consider whether an observation of a "new focus" of a TBD is related to a recent dissemination event, or is actually a discovery of a previously unrecognized endemic pathogen.

2. GENERAL TERMS AND CONSIDERATIONS IN TICK DISPERSAL AND DISTRIBUTION

Tick dispersal occurs either by self-locomotion of the tick (vagility), by abiotic mechanisms, or most importantly, by host-directed transport. In this latter

Table 6.6. Factors affecting dispersal and establishment of a tick-borne pathogen at novel sites from the perspective of host, tick, and agent

Host
1. Migratory vs. non-migratory habitat preferences
2. Physiological stress (reduced resistance to infestation, infection)
3. Daily rate of movement (speed, staging, etc.)
4. Population density and mixing along routes of migration
5. Age structure of population (susceptible versus resistance to infection)
6. Ectoparasite burden indices in the population (% parasitized and mean burden)

Tick
1. Host preferences (host contact rate and availability)
2. Pathogen transmission to tick (feeding relative to period of communicability in hosts)
3. Pathogen transmission in tick (transstadial, transovarial)
4. Environmental constraints to off-host survival
5. Duration of feeding relative to migration period

Agent
1. Taxonomic range for natural hosts
2. Duration, intensity and periodicity of infectious period
3. Impairment to host relative to migration events
4. Survival and transmission parameters for the tick vector

regard, there are considerable differences in the dispersive capabilities of different host species and intraspecific variability exists for dispersal potential between individuals and for a given individual over time. Host factors such as the home range size, territoriality, population density, age of host animal, colonial versus solitary social structure, and dispersion pattern of the tick species itself influence dissemination of ticks and their infectious agents.

One aspect of tick dispersal is drop-off strategy for replete ticks. For ticks which have a three-host feeding strategy, it may be critical to drop from the current host in a location favoring access to the subsequent host. The timing and location of drop-off links tick dispersal directly with the temporal movement patterns of their hosts. For instance, mammal species which tend to have non-overlapping territories, with restricted movement outside the territory, would be less likely to disseminate ticks beyond the borders of these territories than would host species with more permissive spacing strategies. Furthermore, use of space within these boundaries is not uniform, but tends to have a density function that depends on the resource distribution and host species-specific behavioral responses. The probability of a tick dropping at a specific site in the home range is proportionate to the host's use of space (i.e., the movement density function), unless drop-off following engorgement is coupled to endogenous, or circadian rhythms of the tick and host. A brief recapitulation of host movement terms and considerations, as well as tick drop-off patterning is necessary to understand the contributions of each component.

2.1. Dispersion and Drop-off Patterns

The spatial arrangement of individuals in the environment may be either uniform (under-dispersed), random, or aggregated (over-dispersed). **Dispersion** is also used to describe the frequency distribution of tick burden on hosts within a population. Many investigators have noted that the distribution of ticks on hosts exhibits an overdispersed (negative binomial) distribution with most animals having no or few ticks and a few animals supporting a large number of ticks (Sonenshine and Ziv, 1971; Nilsson and Ludqvist, 1978; Petney et al., 1991). Sonenshine (1975) pointed out that host–parasite interactions which result in the relative distribution of ticks on hosts may be viewed from either the host perspective (host-dependent phenomena) or from that of the parasite (behavioral predilection, environmental predilection, or opportunism). Similarly, the dispersion of egg masses in an environment reflects the location and activity of host(s) at the time of drop-off of the female. If hosts move randomly through the environment and drop-off pattern is random, then distribution of larval clusters will tend toward a negative Poisson distribution (i.e., a random distribution of aggregated individuals). If drop-off patterns are not random, i.e., related to endogenous factors within the tick (Rechav, 1979) or cued to exogenous host-related factors such as adrenal cycle (Amin, 1970) or activity cycle (Balashov, 1954; Camin, 1963) or to combinations of exogenous and endogenous cycles (Minshull, 1982), then the dispersion of egg masses will tend to be even more concentrated. Differential survival of egg masses due to microclimatic events will also condition the observed distribution pattern of viable hatched larvae, leading to greater relative concentration of larvae at these loci.

Spatial dispersion in subsequent life stages will also be subject to the same considerations. The likelihood that a suitable host comes into contact with a hatched cluster of larvae is a function of the movement pattern and activity level of the host in its home range, and of the host-seeking strategies of the ticks. Movement patterns of hosts will be dealt with more thoroughly in the section on small mammal influences in TBD dissemination, while host-seeking behavior by ticks is considered in greater detail in the section on vagility. These interactions will likely result in a different dispersion pattern of nymphs than larvae. The distribution pattern of nymphs should tend to be more random than that for larvae due to dispersal by this first host, unless similar mechanisms such as synchronous drop-off rhythms act to concentrate the nymphs (Mather and Spielman, 1986). Such patterns can produce aggregated dispersions of infected ticks as well as demonstrated for Lyme spirochetes in nymphal *I. scapularis* (Telford et al., 1992). However, drop-off patterns even for a particular life-history stage may range from random to aggregated based on the species of host parasitized. This difference was noted for *Ixodes scapularis* and *I. ricinus* subadult ticks, which adopt different drop-off patterns for different hosts to optimize feeding opportunities in subsequent stadia (Matuschka et al., 1991). Differential timing of drop-off between stadia such as observed for all three life stages of *Rhipicephalus appendiculatus* feeding on cattle (Minshull, 1982) would also produce varying distribution patterns of tick stadia.

2.2. Host Movement and Biogeography

Distribution and movement of hosts in dissemination of TBDs must be defined along several different time and distance scales. Daily movements within a particular area are generally devoted to foraging for resources (energy, shelter, and mating opportunities) which tend to be unevenly distributed in time and space. Temporal partitioning of resources results in behavioral strategies to reduce intra- and interspecific competition for scarce resources; strategies such as cyclic migration with seasonal shifting of foraging or mating/reproductive sites. Eventually, daily or annual movement patterns, and dispersal by individuals may expand the range of limits of the species.

Several terms are generally used to describe the spatial distribution or movement patterns of individuals. **Home range** is the area within which an individual or social group conducts most of the routine functions of daily survival, and is usually focused around one or more **centers of activity**. Home ranges are generally not fixed but may vary with season, altered resource distribution, feeding strategy, population density, population structure, and physiologic condition of the host. Ectoparasite load may contribute to these dynamics by imposing physiological stress brought about by reduced blood volume or components, or immunosuppression (Lehmann, 1993). Activity cycles within these ranges may have no regular periodicity (arrhythmic) or occur with a regular periodicity either less than 24 h (ultraradian), equal to the diel cycle (circadian), or greater than 24 h (infraradian) (Aschoff, 1981). **Territories** are areas within or coincident with the home range which are generally defended by individuals or social groups against conspecifics.

Mammalian home range size is approximately scaled on body mass (i.e., metabolic rate) as

$$\text{Area} = 8.5 \times \text{body mass}^{0.75} \tag{6.1}$$

(McNab, 1963). Browsers, however, require four-fold larger home range sizes than comparable sized grazers due to the patchy distribution of resources for the former group. Browsing species, such as white-footed mice (*Peromyscus* sp.) or deer (*Odocoileus* sp.) would thus disperse ticks over wider areas than would comparable sized grazers such as voles (*Microtus* sp.) or domestic cattle. Body size and home range size are not necessarily linearly related since larger mammals and predatory birds must forage over proportionately greater areas than would be predicted by metabolic requirements alone (Swihart et al., 1988; du Toit, 1990). Within a host population, the extent to which individuals' home ranges overlap varies by species, age, sex, population density, and resource distribution, and is another important aspect of ectroparasite dispersal. Hosts which occupy exclusive home range may limit the opportunity for the next stadia to find a different conspecific host. This would require the next stadia to either feed on the previously exposed host, possibly resulting in lower survival due to an immune response to the previous host exposure to tick salivary antigens. Strategies to avoid this potential problem include: (1) develop a mechanism to immunocompromise the host; (2) adopt a one- or two-host

feeding strategy; (3) adopt a broad host range; or (4) selectively parasitize dispersers.

Lengthy or permanent movement of the host away from an established home range (natal or otherwise) constitutes **dispersal** and is usually defined for single individuals (McCullough, 1985). Other directed movements may be cyclic and accomplished by populations, as seen with annual **migration**, which are generally associated with seasonal shifts related to resource tracking. **Nomadism** is also associated with population movements but may be more erratic or spatially random. Dispersal of hosts and vectors results in an opportunity for dissemination of pathogens. The gradual spread of hosts or vectors across space is an accumulation of these dispersal/movement activities through time.

The landscape itself may also change leading to formation or dissolution of barriers to dispersal between adjacent regions. The formation of barriers within the range of a given taxa can result in **vicariance** and establishment of new sister taxa. McKenna (1983) pointed out that vicariance can appear to look like dispersal, such as when a land form breaks apart, resulting in a passive disjunct distribution for a given taxon. Dispersal, however, is an active process whereby pre-existing barriers are crossed and ranges are expanded. The divergence in genotype between eastern and western strains of the TBE complex viruses across the Ural Mountains (Turgai Strait) may serve as an example of vicariance.

Simpson (1940) identified three basic venues for the dissemination or migration of species between regions:

1. **Corridors**—continuous stretches of land (or water), which allow non-limiting, bidirectional opportunities for movement (e.g. the land bridge which connected North and South America during the Pliocene).
2. **Filter bridges**—restrictive connections, allowing passage of only a select subset of the fauna. An example would be the Beringian land bridge, which presumably was accessible only to cold-adapted species (Hallam, 1973).
3. **Sweepstakes routes**—movement across distances or barriers which are otherwise beyond the natural unassisted capabilities of an individual, such as oceanic rafting by small vertebrates.

Pielou (1979) included both spatial and temporal components in dispersal:

1. **Jump-dispersal**—movement of an individual across great distances and inhospitable terrain within a short period of time relative to the individual's life.
2. **Diffusion**—gradual movement of populations across hospitable habitat over a period of generations of the species.
3. **Secular migration**—diffusion requiring lengthy time scales such that evolutionary changes may also take place.

Corridors, filter bridges, diffusion, and secular migration modes allow for homogeneous mixing of populations and faunas and result in a continuous distribution while the corridor remains intact. A general example of the result

of these dispersal patterns is provided by the investigation of the systematics of the *Ixodes ixodiopsis* group across the former Beringian land connection. Secular migration may partially explain the wide distribution of the Congo–Crimean hermorrhagic fever tick complex, which potentially involves as many as 30 tick species in several genera, over the geographic range of the virus. This taxonomic breadth in vector competency would suggest either a long-term association, and gradual spread and divergence of this virus within different vector species, or a highly adaptable virus phenotype. An example of recent dispersal corridors is the re-emergence of suitable habitat for deer in the deciduous forest biome of eastern North America, allowing expansion of the deer tick population.

Sweepstakes or jump-dispersal routes would result in "stepping stone" or disjunct patterns of TBD distributions. Long-distance transportation of ticks by birds or man is an example of jump-dispersal or sweepstake dispersal. Chance events, such as introduction of *Ixodes pacificus* into the Hawaiian Islands by migrating birds, demonstrate the possibility of immigration of ticks into new distant areas, while the spread of African Swine fever virus from Africa to Europe and the Caribbean, and establishment in indigenous *Ornithodoros* species is an example of accidental TBD dissemination into a preadapted setting. A "stepping stone" distribution pattern is also observed with the disjunct distribution of *Dermacentor variabilis* and SFG rickettsia along the northern US coastline and introduction into Nova Scotia (Herman and Garvie, 1985). It is likely that a combination of jump-dispersal and diffusion events have resulted in the dissemination of *Ixodes scapularis* and Lyme disease, possibly by avian hosts along the Mississippi flyway (Weisbrod and Johnson, 1989) or Atlantic flyway (Battaly et al., 1987).

Finally, mobilist geology has been identified as an underlying force in the waxing and waning of routes of dispersal as well as providing evidence for tectonic plate connections that have resulted in the present geographic distribution of tick-borne pathogens. McKenna (1983) discussed a variety of geologic mechanisms, such as escalator counterflow, Noah's Arks, back-arc spreading, and Cenozoic pole position changes which have influenced evolution and distribition of biomes.

3. DISSEMINATION MECHANISMS FOR TICKS

3.1. Vagility

Vagility refers to directed horizontal movement by a tick under its own power. These movements are mostly associated with host-seeking behavior, however, they may also occur in response to physiological stress or to enhance mating opportunities (Anderson, 1974; Oliver, 1974). Host-seeking strategies in ticks roughly fall into two categories: passive, where a stationary individual waits in ambush for a passing host, where the tick detects hosts from a distance and actively hunts or moves to the host in response to sensory cues (Waladde and Rice, 1982). Ambush-style strategists (e.g. *Ixodes dammini*) may show

regular vertical movements but little horizontal movement, while hunters (e.g. *Amblyomma variegatum*, *Hyalomma dromedarii*) will rapidly traverse substantial horizontal distances towards a host. The strategy employed by a given species is a product of evolutionary trends in feeding and reproductive mechanisms, density and distribution of hosts, and home range or movement patterns of the host. In general, the reported range of active horizontal dispersal from a known starting point by ticks does not usually exceed 20–30 m for adults (Table 6.7) and these parameters vary greatly with species and stage. Basic differences in the ability or need for horizontal movement appear due to evolutionary trends for host seeking and mating within the Ixodida (Pomerantzev, 1948). The anatomical trends in the Argasidae toward reduced chitinous support, and retention of primitive mouthparts causes certain trade-offs in both locomotory ability and length of attachment to the host. The Argasidae have evolved a generalized "wait and see" host-seeking strategy requiring close association with nests, rookeries, crevices, or other protected environments that offer proximity to hosts. Since argasid mouthparts are not adapted to long periods of attachment to hosts (compared to the ixodid ticks), feeding sessions for nymphal and adult argasids are relatively rapid (feeding to repletion within 30 min of host acquisition). The exception to this generalized pattern for argasids is the slower feeding times (3–10 days) found in species of *Argas*, especially those parasitizing birds and bats (Oliver, 1989). This exception possibly plays an important role in the world-wide dissemination of arboviruses as described above. Compared to the Ixodidae, however, several additional feeding sessions are required by argasids to accumulate sufficient energy and nutrients to reach sexual maturity.

Rapid feeding, however, lowers the net likelihood that the tick will be moved from the feeding site. This is little need to stray from the host's resting place and a premium is placed on the ability to survive long periods for the return of a suitable host. Attachment for longer periods could be disadvantageous since movements by the host away from the nest may result in accidental translocation of the tick sites with low host density and lowered chances of survival. An investigation of movement by marked relapsing fever ticks, *Ornithodoros turicata*, between gopher tortoise burrows failed to show movement by individuals away from the release site burrow, despite survival of greater than one year within the burrow (Adeyeye and Butler, 1989). Argasids must rapidly maneuver the short distances between their resting places and the host, however, and seasonal differences in site selection within these microhabitats occurs (Olusola and Butler, 1989).

Considering evolutionary trends within the Ixodidae which may ultimately influence host-seeking behavior, Pomerantzev (1948) suggested that the chief feature differentiating the host-seeking strategies of the more primitive Prostriata (Ixodinae) from the Metastriata (Amblyomminae) is the ability of the Prostriata to copulate prior to host acquisition and blood feeding. The Metastriates are sexually immature until a blood meal is taken. The adult male mouthparts of most species in the Prostriata are less developed than those of the adult female, presumably since these ticks are not reliant upon blood feeding for sexual

Table 6.7. Locomotory distances (m) observed for different tick species

	Stage	Time	Maximum range (m)	Mean range (m)	Method	Source
Amblyomma americanum	Adult		22.8			Smittle et al. (1967)
	Adult		21.3			Wilson et al. (1972)
Amblyomma hebraeum	Larva	30 days	1.5	0.5	Sentinel animals	Rechav (1979)
	Nymph	30 days	4.5	1.0	Sentinel animals	Rechav (1979)
	Adult	30 days	5.2	1.2	Sentinel animals	Rechav (1979)
Amblyomma concolor	Larva	30 min		0.05	Sentinel animal	Belan and Bull (1991)
Dermacentor andersoni	Adult	2 days		6.3	CO_2 trapping	Eads et al. (1982)
Dermacentor occidentalis	Adult	4 h	7	1.8	CO_2 trapping	Lane et al. (1985)
Dermacentor variabilis	Adult		23.4	<0.5		Sonenshine et al. (1966)
	Adult		30			McEnroe (1971)
Boophilus microplus	Larva			0.48		Wilkinson (1957)
Ixodes dammini	Larva	22 days	3	<1.0	Drag cloth	Daniels and Fish (1990)
	Adult	6 days				
	Adult	144 h	5	1.8	Dry ice bait	Falco and Fish (1991)
	Adult			<2		Falco and Fish (1988)
	Nymph			<2		Falco and Fish (1988)
	Larva	1–2 weeks	>1	0.1–0.3	Confined rings and drag cloth	Stafford (1992)
Ixodes ricinus	Adult	168 h		1.3		
Rhipicephalus appendiculatus	Larva	30 days	2	<0.5	Sentinel animals	Rechav (1979)
	Nymph	30 days	2	1.0	Sentinel animals	Rechav (1979)
	Adult	30 days	4.8	1.2	Sentinel animals	Rechav (1979)
Rhipicephalus e. evertsi	Larva	30 days	2	1.2	Sentinel animals	Rechav (1979)
	Adult	30 days	2.8	0.5	Sentinel animals	Rechav (1979)

maturation. The male mouthparts in the Prostriates may therefore be adapted more for copulatory functions, and with reduced capability or need for direct host attachment. Males can often be found on hosts with mouthparts inserted in the genital orifice of attached females. A potential consequence of this reproductive strategy is that these ticks can remain fairly nidicolous, since mating opportunities would arise just after the adult molt. This in turn may result in a more passive host-seeking strategy, at least for males, and may show more restricted host ranges. Males in several species within the Ixodinae (viz., the *ricinus* group) do have the necessary capitular morphology to allow more permanent ectoparasitism. These species show wider host ranges and have been successful at exploiting larger, more mobile hosts (such as artiodactyls) which has resulted in a broader geographic range.

Direct measurement of vagility under natural conditions usually involves marking a sample of ticks, and either placing them at various distances and directions from a collector, or positioning collectors at various distances and directions from a central release point for the ticks. The collectors used include dry ice-baited traps, sentinel animals or drag cloths. Most studies are designed to evaluate the effectiveness of the collection methods rather than to examine host-seeking behavior. Differences in experimental design make it difficult to directly compare ixodid vagility, and the stimuli used in these studies may not adequately mimic the multiple cues which ticks use to identify, orient and track potential hosts. These studies, however, provide an indicator of movement capabilities.

The data indicate that distance travelled is positively associated with life stage, and that most species are conservative in their movements (Table 6.8). Most adults in these studies did not move more than 2 m, on average, from a known starting point, while larval ticks typically remain within 1 m. The tendency for larvae to remain stationary is likely advantageous since most of the hosts of larval ticks tend to be small mammals and birds. These hosts generally occur at higher population densities and experience greater population turnover, which may increase the potential for contact with suitable hosts than is seen for the adult stages. In addition, movement away from the hatch site may place the individual into a less suitable microclimate.

Other behavioral characteristics may also be indicative of dispersal capabilities. In experimental studies, *Hyalomma dromedarii* adults demonstrate positive scototaxis, or body orientation and movement to dark-shaped objects, to host-shaped targets at the equivalent of 18 m from the tick (Kaltenrieder, 1990). This implies that the tick should be able to move this distance under an active host-finding strategy. The estimated time period necessary to reach such a target was calculated at 6.5 min based on measurements of the walking rate.

In addition to self-directed movement by ticks to seek hosts, at least two other mechanisms influence tick dissemination: environmentally induced movement and paratenic movement. Wind can convey larval ticks much greater distances than observed with self-directed movement. Lewis (1970) demonstrated the influence of wind at various times of the year and in different types of vegetation on dispersal of *Boophilus microplus* larvae. Natural vagility for this

Table 6.8. A partial listing of tick-borne pathogens associated with tick species that infest avian hosts

Tick	Bird	Agent
Ornithodoros ssp.	Marine birds	Hughes group
Ornithodoros spp.	Pigeons, swallows, gulls	Kemorovo group
Ornithodoros capensis	Marine birds	Johnston Atoll
Ornithodoros moubata		*Borrelia duttoni*
Argas cooleyi	Cliff swallows	Hughes group
Argas arboreus	Herons	Quaranfil
Argas hermanni	Pigeons	Quaranfil
Argas hermanni	Pigeons	Royal farm virus, West Nile
Argas reflexus	Pigeons	Ponteves and Grand Arbaud
Argas hermanni	Pigeons	Grand Arbaud
Argas sp.	Pigeon, swallow, gulls	Kemorovo group
Ixodes brunneus	General passerines	*Rickettsia rickettsii*
Ixodes dentatus	General passerines	*Rickettsia rickettsii*
Ixodes ricinus	General passerines	Rickettsia, Tribec, Uukuniemi
Ixodes ricinus	General passerines	*Rickettsia conorii*
Ixodes ricinus	General passerines	Uukuniemi
Ixodes dammini	General passerines	*B. burgdorferi, Babesia microti*
Ixodes hexagonus		*Rickettsia conorii*
Ixodes holocyclus		*Rickettsia australis*
Ixodes granulatis		Langat virus
Ixodes persulcatus		Langat virus
Ixodes uriae	Marine birds	Tyuleniy
Ixodes and *Haemphysalis*	General	RSSE, TBE, Louping Ill, KFD
Hyalomma m. marginatum	Rooks (*Corvus frugilegus*)	CCHF
Hyalomma m. marginatum	General	West Nile
Hyalomma m. rufipes	General	CCHF, Dhori, West Nile,
Hyalomma m. rufipes	General	Dugbe, *C. burnetti, R. conorii,*
Hyalomma m. rufipes	General	*F. tularensis, Brucella melitensis,*
Hyalomma m. rufipes	General	*Theileria annulata, Nuttalia equi*
Hyalomma rufipes	General	*Babesia caballi*
Amblyomma variegatum	General	*Rickettsia conori*
Amblyomma lepidum	General	*Rickettsia conori*
Amblyomma hebraeum	General	*Rickettsia conori*
Amblyomma nuttalli	General	*Rickettsia conori*
Amblyomma hebreum		*Cowdria ruminantium*
Rhipicephalus sanguineus		Indian tick typhus
Rhipicephalus sanguineus		*Ehrlichia canis*
Rhipicephalus sanguineus		*Babesia canis*
Rhipicephalus sanguineus		*Rickettsia conori*
Rhipicephalus simus		*Rickettsia conori*
Rhipicephalum appendiculatus		*Rickettsia conori*
Rhipicephalus evertsi		*Rickettsia conori*
Rhipicephalus turanicus		Indian tick typhus
Dermacentor variabilis		*Rickettsia rickettsii*
Dermacentor parumapertus		*Rickettsia rickettsii*
Dermacentor andersoni		*Rickettsia rickettsii*
Dermacentor nuttalli		*Rickettsia siberica*
Dermacentor silvarum		*Rickettsia siberica*
Dermacentor marginatus		*Rickettsia siberica*
Dermacentor pictus		*Rickettsia siberica*

Table 6.8. (*continued*)

Tick	Bird	Agent
Haemaphyslis leachi		*Rickettsia conorii*
Haemaphysalis leporispalustris		*Rickettsia rickettsii*
Haemaphysalis japonica		*Rickettsia siberica*
Haemaphysalis concinna	Passerines	*Rickettsia siberica*
Haemphysalis spinigera		KED

tick is reported to be approximately 45 cm (Wilkinson, 1957), however, larval ticks were recovered under controlled conditions at distances up to 78 m from the point of hatching in short grass pasture, 30 m in long grass and 5.4 m in scrub. These larvae were capable of infesting cattle as indicated by collection of adult ticks downwind from the hatch site at appropriate time intervals for full development, and by placing recovered dispersed larvae on hosts. Larvae were trapped on cloth sheets placed on sampling towers at heights of 2–3.6 m above the ground. Lewis (1970) showed in separate experiments that horses, birds, and rats could also transport these larvae, which dropped from these unsuitable hosts in viable condition at various distances from the infestation point.

3.2. Host-associated Dispersal

3.2.1. Dissemination of Ticks and Pathogens by Avian Hosts

Birds serve as exclusive hosts for approximately 75 tick species and participate in the feeding strategy of number of other species spanning the genera *Argas*, *Ornithodoros*, *Ixodes*, *Haemaphysalis*, *Rhipicephalus*, and *Amblyomma* (Hoogstraal, 1972). As a group, their biomass and population turnover provides a tremendous host source for amplifying both tick and infectious organisms. A wide variety of tick species infest birds during one or more of the feeding stages of tick development and conversely, a number of bird species serve as a blood-meal source for ticks which either exhibit non-selective host specificity in subadult stages, or are specific ectoparasites of birds as part of the natural host association in these earlier stadia. A developed tick fauna exists for the pelagic (ocean-going) avian groups that is highly associated with rookeries and colonies of these birds. The terrestrial-based bird species also have ticks that are highly host specific, but overlap more frequently in the feeding dynamics of ticks that feed on mammals and that transmit human pathogens. Terrestrial birds which feed, or spend part of their daily activities on the ground, are generally more frequently infested and support greater mean tick burdens than bird species with limited ground contact.

Local Movements and Tick Dispersal Birds are instrumental in local and long distance dispersal of ticks. The daily movement of birds provides an opportunity for short-range dispersal of ticks. Rooks (*Corvus frugilagus*) play a significant

role in the local ecology of *Hyalomma marginatum* and CCHF in Astrakhan Oblast, supporting the tick population in the immature stages, and transporting ticks locally. CCHF virus infected *H. marginatum* have been collected from this avian species in this area (Berezin, 1971). Hoogstraal (1979) indicated in his review of CCHF virus that Rusian workers found fledglings of the common tern (*Sterna hirundo*) with a mean of 78 adult *H. marginatum* ticks in a local breeding population in the Southern Caspian Sea. Sonenshine and Clifford (1973) compared the incidence of *Rickettsia rickettsii* in ticks recovered from approximately 17,000 birds in piedmont versus coastal regions of the mid-Atlantic to south-central US. The principal tick species recovered from birds were *Haemaphysalis leporispalustris* and *Ixodes dentatus*, neither of which usually attack man. *H. leporispalustris*, however, is involved in a rabbit-associated transmission cycle of Rocky Mountain spotted fever (RMSF). Rickettsial infection was not found in ticks removed from birds in the piedmont area, where human cases predominated. Rickettsial infection was paradoxically found in ticks removed from coastal ranging birds where human RMSF incidence is much lower. While a possible dispersal route for this tick-borne disease was documented, the direct impact of such findings for transmission to humans is diminished by the requirement for the agent to then enter into a *Dermacentor variabilis* transmission cycle.

One interesting aspect of the Sonenshine and Clifford (1973) survey was the lack of *Ixodes scapularis* on sampled birds in this geographic region. This could indicate that these ticks were either limited in distribution, using different hosts, or not yet abundant in the localities sampled. In the northeastern US, local wild bird populations have been shown to support infestations of subadult *Ixodes scapularis* ticks (Anderson and Magnarelli, 1984; Anderson et al., 1986; Battaly et al., 1987; Weisbrod and Johnson, 1989) which has raised the possibility that birds are partly responsible for the transport of these Lyme disease vectors within endemic disease foci as well as to other non-endemic sites in North America. Battaly et al. (1987) found 37% of 41 bird species ($N = 251$ birds) in New York parasitized with a mean of 1.31 *I. scapularis* larvae or 1.9 nymphs during a one year study. *I. dentatus*, which has also been identified as a vector of the Lyme spirochete in a rabbit-associated cycle (Telford and Spielman, 1989), was also found on 27% of bird species examined. Several birds were found concurrently infested with both tick species. Anderson et al. (1986) also found *I. scapularis* parasitizing 71% of birds (17 species) sampled in Connecticut, with some species supporting a maximum mean of 6.5 nymphs (*Cardinalis cardinalis*) or 12 larvae (*Troglodytes aedon*). In both studies, spirochete positive ticks were found, and in the latter study, *Borrelia burgdorferi* was isolated from the liver of a verry (*Catharus fuscescens*).

There are few, if any, tick mark–recapture studies performed with birds as the sampled hosts. This is an open area for research on local dispersal of ticks. An indirect example of the potential for local dispersal of ticks by birds is the distribution of itchgrass (*Rottboellia exaltata*). This grass species is native to India and was accidentally introduced into the southeastern US. Aison et al. (1984) examined the passage time of itchgrass seeds through the digestive tract

of various bird species found in the southeastern US and combined these findings with data on home range and daily movement patterns. They determined that four of 15 species of birds could contribute to local step-wise dispersal of this species over a large area. Engorgement and drop-off of ticks could be considered analogous to digestive transmit time for the grass seeds, thus local tick dispersal could operate in a similar fashion.

Long-range Movements and Tick Dispersal In addition to local dissemination of ticks and pathogens, birds operate to disperse these populations over continental and intercontinental distances in a relatively short period of time. As a class, birds are the most mobile of all land vertebrates, with a large number of temperate species making semi-annual flights over thousands of kilometers. North America has three general flyways into the Caribbean, Central and South America (Bellrose, 1976). There are eight broadly overlapping flyways from Eurasia to Africa; five major routes between northern Asia and southwest or southeast Asia; and, a number of small routes into the Malay archipelago (Baker, 1981). There are still unresolved issues, however, concerning the migration routes, and wintering and/or breeding grounds which may affect tick and pathogen dispersal, or alter our view of the relative efficiency of this dispersal mechanism. Bairlein (1985) summarized a number of unanswered research questions concerning the Euro–African flyway system, such as: do most birds cross the Mediterranean and Sahara using short stop-overs or in a great hop?; do they fly along geomorphological features across the Saharan region?; what are the ecological conditions that prevail in the wintering and staging grounds? Birds are also capable of concentrating these ectoparasites in specific staging areas or at the termini of the migration pattern. Waterfowl species, for example, are highly concentrated in over-wintering areas relative to their spatial dispersion pattern during the summer breeding season. During the process of migration in North America these dispersed local bird populations gradually concentrate along common flight patterns across Northern Canada as they converge toward the southern wintering areas. Similar processes take place along the four major migratory routes in North America, nine in Eurasia (west of the Ural Mountains), and eight in Asia.

What conditions must be present for the tick or pathogen to become successfully established as a consequence of rapid long-range migration? Some considerations for avian transport of tick-borne pathogens (in addition to those considered in Table 6.7) include: the tick must feed to repletion and be ready to drop off in a suitable habitat along the migration route; and, the tick must molt and find a suitable host at the new site. Either the new host will need to be susceptible to the pathogen, or if the pathogen is transstadially and trans-ovarially transmitted, at least one susceptible host may need to be susceptible for horizontal transmission of the pathogen. If the migrant tick is not capable of surviving in the new habitat, there must be a second susceptible tick species present and feeding during the period of parasitemia, or the host must remain infectious for a long enough period to transmit to endemic ticks. The migrating

bird may also serve as a vehicle for dissemination of the disease into new territory if it serves as a host to susceptible tick species along the migration route or at the migration terminus. If the bird species remain infectious for a long time period (as suggested for some Group B viruses) then potential for transmission is further increased. Finally, if the bird's resistance to infection is reduced due to physiological stress inherent in long distance flight, it may result in an even greater likelihood of transportation of the infectious agent, as long as the infection does not impact on the bird's flight range or survival.

Tick species which are more likely to establish new endemic sites represent the extremes of the host specificity spectrum, i.e., high host-specificity and non-specific host preference. Ticks which feed exclusively on migrating pelagic bird species in all stages of life (analogous to the strict-total category of Hoogstraal and Aeschlimann, 1982) would prosper since the principal host is also present at either end of the migration process. Theoretically, these ticks would not likely be vectors of important human tick-borne pathogens, because of their restricted host range. Examples of these ticks include *Ixodes uriae*, which are parasites of oceanic birds, and are distributed nearly from pole to pole. At least 13 different arboviruses, however, have been isolated from this tick species (Karabatsos, 1985).

At the other end of the spectrum are tick species which are generalists in their host affiliations (non-particular species of Hoogstraal and Aeshlimann, 1982). *Ixodes ricinus* and *Ixodes persulcatus* fit into this category. These two species are associated with transmission of at least 12 and six arboviruses, respectively. Gusev et al. (1962) noted a relationship between host specificity and infestation parameter frequency for migrant birds in Azerbaijan. In this study, bird specialists (*Argas persicus* and *Ixodes frontalis*) are feeding generalists (*Ixodes ricinus* and *Hyalomma marginatum*) accounted for approximately 18% and 75%, respectively, of the total number of ticks removed from 3,147 birds. The nine remaining species accounted for only 7% of the collection. These authors concluded that birds play an important role in establishing distant foci of ticks which feed specifically on birds, a secondary role in local dispersion of general feeders, and limited or unimportant role in transport of the remaining category of ticks (accidental infestations).

Several examples of transport and establishment of satellite populations of ticks have been reported (Hoogstraal, 1961, 1972, 1979) including *Amblyomma variegatum* in France, *Amblyomma hebraeum* on cattle in Bulgaria, *A. lepidum* in Azerbaijan, *Haemaphysalis wellingtoni* in Japan and *H. m. marginatum* in Sudan. Hoogstraal et al. (1967) documented an isolated established population of the European species *Ixodes arboricolis*, in Bahig, Egypt, infesting resident sparrows (*Passer domesticus*). He attributed this population to chance importation by southbound bird migrants from Europe. Berezin (1971) identified a population of the African tick, *Hyalomma marginatum rufipes* infesting cattle in the former Astrakhan Oblast, SSR, and additional specimens were found on spring migrating redstarts in the Volga basin, as well as in gull colonies in the northern Caspian Sea. In this latter instance, adult ticks were found on fledgling birds, possibly to compensate for the lack of suitable large mammal hosts.

Gusev et al. (1962) reported on a southward range extension for *I. frontalis* into a forested site in Daghestan, in the former USSR, associated with a newly established rookery. Previous multi-year flagging and ectoparasite surveys in the region established that this tick species was not represented in the acarine fauna prior to immigration of rooks (*Corvus*) into the region. Subsequent surveys determined that the tick was becoming established and expanding its local distribution.

A number of studies performed by Hoogstraal and various co-workers, as well as by Eastern European and Russian scientists during the 1950s through 1970s, demonstrate the influence of annual bird migrations on jump dispersal of tick-borne diseases. The speed and range of flight varies tremendously between species, however, the bird species which were predominantly seen in the series of investigations carried out by these workers have migration route speeds ranging from 20 to 170 km/day (Paevskii, 1973) with some species such as *Muscicapa hypoleuca* traversing 600 km over a 4-day period. Most of these data come from single point ectoparasite surveys, serological surveys, and isolation of infectious organisms from migrating birds and/or their infesting ticks. While these data reveal possible epizootiologicity, they do not directly quantify the success rate for establishment of a local population of infected ticks from a source location with known rates of endemicity. Such information would require a huge, co-operative effort to track banded bird populations and to sample associated tick ectoparasites for pathogens at the outset of a migration, along intermediate points or staging areas, and at the migration terminus.

Agent Dissemination Birds have been associated with a number of viral and bacterial pathogens of man and veterinary species, as well as pathogens which do not normally impact on these hosts, such as the tick-borne viruses of sea birds. A partial list of tick-borne diseases associated with ticks found parasitizing birds is provided in Table 6.8. The role of birds in geographic expansion of a tick-borne disease is presumably further enhanced if a given avian species is susceptible to the pathogen, and serves to amplify the pathogen as well as transport the ticks and/or pathogen. Human or veterinary tick-borne diseases which have been isolated from birds include arboviruses (Russian Spring–Summer encephalitis, European tick-borne encephalitis, louping ill, Kyasanur Forest disease, Bahig, Matruh, Dugbe, Kemorovo, Tribec, Lipovnik, Kaisodi, Tulyeni, Hughes group, Quaranfil), rickettsias (*Coxiella burnetii*, *Rickettsia rickettsii*, *Rickettsia conori*, *Rickettsia siberica*), and spirochetes (*Borrelia burgdorferi*). Birds are involved in the natural history of a total of 27 argasid-derived arboviruses (Karabatsos, 1985), many of which are associated with oceanic birds, and 11 of the ixodid-derived arboviruses. Clifford (1979) identified nine serogroups of sea-bird viruses and described the world-wide distribution of these groups relative to biogeography of their vectors and hosts. These avian-associated arboviruses provide good examples of the extent to which long-distance bird dispersal is associated with tick-borne disease.

(a) Tick-borne Virus Dissemination by Pelagic Birds

The viruses circulated by argasid ticks of sea-bird colonies (e.g., the *Ornithodoros capensis* complex) are circumglobal, especially in the lower to mid-latitudes. The Hughes (Nairovirus), Upolu (Bunyavirus-like), Nyamanini (ungrouped), Kermerovo (Orbivirus) and Quaranfil (ungrouped) serogroups are the principal viruses involved. Soldado virus (Hughes subgroup) has been isolated from sites in East Africa, the British Isles, and islands in the Indian Ocean, Pacific Ocean, and Caribbean Sea. Several of the individual virus strains have an African to mid-Pacific distribution, but most of the specifically identified viruses of this group (Table 6.3) overlap in distribution in Europe, Africa, and Asia. North America, South America, and Australia have a greater relative proportion of unique viruses within this group compared to the flora of the other land masses.

The arboviruses associated with ixodid ticks of oceanic birds are from the Group B (Flavivirus), Uukuniemi (Bunyavirus), Sakhalin (Nairovirus), and Kemorovo (Orbivirus) serogroups. This latter group is the most widespread of all the tick-borne viruses, being represented on every major land mass. Several virus strains isolated from ixodid ticks collected from colonies along the northern Pacific coastal rim of Asia and North America show a high degree of antigenic similarity, while strains of Kemorovo and Sakhalin serogroup viruses from colonies on either side of the northern Atlantic Ocean (eastern Canada and Scotland) differ antigenically from each other and from other viruses in the serogroups. These relationships are probably due to a higher rate of recent interchange across the Bering Sea versus the North Atlantic, but also likely relate to the shorter interval of temporal and spatial separation between North America and Asia across the North Pacific versus the North Atlantic. The taxonomic relationship for the virus associated with highly mobile host populations contrasts the putative pre-Miocene-derived relationships hypothesized above for the SFG rickettsias of North America and Europe which are associated primarily with less vagile host populations. However, exchange of ticks and viral agents across the North Atlantic over relatively short time periods is possible in the pelagic avifauna. Young kittiwakes (*Rissa* sp.), for example, have been shown to fly 2,000 km from Murmansk to Greenland and Newfoundland. Isolation of Uukuniemi serogroup viruses (which are typically associated with *I. ricinus* in Europe) from *I. uriae*, ticks of sea birds, in both the northwestern US and northeastern Asia, intimates a possible link between pelagic and non-pelagic bird dissemination of arboviruses. Could this dissemination mechanism provide a clue to the apparent relationship between louping ill and Negishi viruses? The ixodid ticks of sea birds, especially *Ixodes uriae*, are generally found in sea-bird colonies in both of the higher latitudes. The wide distribution of this tick species reflects the tremendous migratory distances traveled by some of their hosts (terns and petrels). Clifford (1979) noted that *I. uriae* is morphologically similar throughout its tremendous range, further suggesting that there may be a constant transfer of ticks between these greatly disparate regions of the world by these migrants.

Overall, the tick-borne arboviruses of oceanic birds accounts for a very

large proportion of the shared virus serogroups between continents. If African swine flu is excluded from consideration, the tick-borne viruses of pelagic birds account for all of the South American tick-borne viruses that are shared with other land masses. Similarly, 60–100% of the shared virus serogroups found in North America and Australia are due to this avian group. The lowest proportions are found for virus groups shared between Africa and Europe where only 29% is due to oceanic birds. This same analysis indicates that 44% and 50% of the Asian arbovirus serogroups are shared with Africa and Europe, respectively.

(*b*) *Ticks on Migrating Land Birds*

Hoogstraal and associates in Cyprus and Egypt surveyed ticks on birds during migration along the Eurasian–African migratory system (Hoogstraal et al., 1964; Clifford and Hoogstraal, 1965; Watson, 1971; Kaiser and Hoogstraal, 1974) tallied at least 83 bird species infested with 28 tick species during the autumn (southward) migration and with 15 tick species during the spring (northward) migration. *Hyalomma m. marginatum* was the most often reported species on southbound birds (58/83 bird species infested) while *Hyalomma m. rufipes* was found on 53/83 bird species during the northward migration to Europe. Nymphs of the African species *Hyalomma m. rufipes* have been recovered from migratory birds as far away as Finland (Nuoreteva and Hoogstraal, 1963) and Great Britain (Thompson, 1964). Since this species typically remains on the host for the larval and nymphal blood-meal (two-host strategy), there is a greater likelihood that the species will be transported further distances than would a three-host tick species which drops off each host after a blood-meal. The average infestation period of immature *H. m. rufipes* on birds of 16 days (Berezin, 1971) would allow sufficient time for long distance dispersal. Observed host-breadth for three-host ticks may show greater decrease as birds progress along the flyways and the ticks drop from their hosts. In Poland, for instance, Kahl (1971) reported only larval *Ixodes ricinus* on 4,848 migrating birds. In Povolzhie, in the former USSR, Borisov et al. (1969) found only *I. ricinus* and *I. persulcatus* on spring and fall migrating birds accounting for 3/22,780 acarine, mallophagan, or siphonapteran parasites recovered from 153 birds.

The bird species of greatest importance for transporting earth-bound ticks are presumably ground-dwelling or ground-feeding in habit due to the likelihood of tick–host contact. Infestation opportunities exist for other species as a result of mating, nest building, etc. Balat (1964) found no ticks on 706 adult and nesting waterfowl in the former Czechoslovakia but, however, did find *I. ricinus* on 5/41 ducklings after movement from the nests to open-water through a woodland setting. In compiling the data from the numerous African–Eurasian flyway surveys, I found no statistically significant association between groups of birds categorized generally by feeding/nesting habits and tick infestation burdens during migration. This may suggest that behavioral repertoire in these birds prior to migration should be examined for differences that may influence potential contact with the ticks.

In addition to dispersing infected ticks, birds may play an important role in horizontal transmission of pathogens to other species of ticks which parasitize birds during migratory periods. The opportunity exists to transport agents either via viremic/parasistemic birds, or via newly exposed tick species which infest these birds just before and during migration. Migrating birds infested with several tick species provide an opportunity for interspecific transfer of tick-borne pathogens, Kaiser et al. (1970) found multiple vector species infesting individuals in a migrant flock of doves (*Streptopelia turtur*) including *Argas streptopelia* (vector for Quaranfil virus), *Hyalomma m. rufipes* (vector for *Rickettsia conorii* and CCHF virus) and *Amblyomma lepidum* (vector of *Cowdria ruminantium*).

The evidence for the role of migratory birds in transporting tick-borne diseases comes from serological assays of migrating birds, recovery of infected ticks and isolation of infectious agents from these birds. Birds (especially lariforms and passerines) sampled along a south-bound migration route in Estonia, showed evidence of antibody to Uukuniemi (5%) and Kemorovo viruses (7%, Gaidanovich, 1972). Tick-borne infectious agents have been recovered from birds along migratory routes. In Egypt, Watson et al. (1972) reported an isolation rate of 7.4% of Bahig virus from white-throat warblers (*Sylvia communis*) during the fall migration from Central Europe, with consistent findings of infected birds over several migration seasons. This same virus was isolated during spring migration (tropical Africa to Europe) from a pool of larval *Hyalomma marginatum rufipes* removed from a Wheatear, *Oenanthe oenanthe* (Hoogstraal, 1978). Bahig, or other members of the Tete group were identified in migratory birds from Italy and a TBE virus was recovered from an unspecified number of migratory birds during the spring migration in Belorus (Voinov, 1978). Samoilova and Gembitsky (1974) isolated three strains of TBE virus from a sample of 183 song birds migrating along the west coast of the Baltic Sea during spring migration into northwestern Russian and Finland. These authors did qualify these findings, however, by pointing out that *Ixodes ricinus* ticks were active during this period in the resting areas along the migration route and could therefore have been the source of the viral infections in the migrants. During the autumn migration period, Brink et al. (1965) found low numbers of *Ixodes ricinus*, *I. arboricola* and *H. marginatum* on passerine birds migrating along the southeast coast of Sweden. No viruses were recovered from any of the birds, and only one bird had antibody to TBE. Berezin (1972), however, argued against an important role for migratory birds in CCHF epizootiology. He suggested that while bird importation of several mosquito-borne arboviruses (West Nile, Tahyna, and Sinbis) was likely important (owing to questionable overwintering mechanisms in vectors or hosts), CCHF foci could be maintained by local populations of *H. marginatum* and *H. anatolicum.* It was not necessary to invoke bird migration as the source of CCHF infection in this region.

Weisbrod and Johnson (1989) demonstrated Lyme disease spirochetes in 22.4% of *I. scapularis* parasitizing 58 or 9,200 sampled birds migrating along the upper Mississippi valley in Minnesota (North America). The Mississippi

flyway is one of the three major North American avian migration routes. They suggested that these birds were important in the long-distance dispersal of infected ticks to other areas of the continent. Anderson et al. (1990) have shown that spirochete-infected *I. scapularis* which fed on birds were capable of transstadial transmission of the agent, which was still infectious to hamsters. These findings support the hypothesis that ticks dropped along the flyway could molt and transmit spirochetes at a new locus. The proposition of birds expected to participate in this dispersal event would be low ($<0.6\%$), however, the large populations migrating along these routes would increase the likelihood of just such a dispersal event.

Although there is ample suggestive evidence of the role of birds in the importation of tick-borne diseases, there has been no direct demonstration of this mechanism. Hoogstraal (1961) and Work (1958) suggested that the "sudden" emergence of Kyanasur Forest disease in 1957 in a circumscribed area of Mysore State, India, was a result of importation by birds of a Russian Spring–Summer encephalitis group virus from a Central Asian focus with subsequent virus transmission to the indigenous avian, tick and mammalian fauna of the area. A number of the haemaphysalid species associated with natural transmission of this disease are frequent parasites of birds, occurring in higher infestation levels than on syntopic small mammals. Birds were shown to be experimentally susceptible to Kyasanur Forest Disease (KFD) virus (Boshell, 1969), yielding levels of viremia capable of infecting attached larval ticks. Boshell (1969) suggested, however, that KFD was a pre-existing zoonotic disease which was brought to medical discovery due to increased human activity in the zoonotic area. This view was supported by isolation of the virus from at least ten species of ticks (Boshell et al., 1968) during a period soon after the initial outbreak which could indicate that the virus was well established in the area. Serologic evidence of viral infection in a number of different bat species (Bhat et al., 1978), and isolation of the virus from bats and an *Ornithodoros* sp. tick also supports the argument that the virus was endemic, but unrecognized (Rajagopalan et al., 1969).

3.3. Small Mammals and the Dissemination of Ticks and Tick-borne Diseases

Small mammals are intimately tied to the maintenance and dissemination of tick-borne diseases, primarily at the local level. They play a crucial role in supporting the immature stages of a great number of tick species and are reservoirs of a variety of pathogens. In addition to their tremendous taxonomic and ecological diversity in the faunas of most geographic regions of the world, they contribute to disease ecology by way of their biomass and rapid pace of population turnover which provides a source for sustained, susceptible host populations for both vector and infectious agents. Due to their small size, they are able to exploit favorable microclimates in otherwise harsh environments, which further enhance opportunities for survival of ecoparasite off the host.

The term "small mammal" as defined by Bourliere (1975) constitutes ordinal groups of mammals weighing less than 3 kg as adults. This definition therefore encompasses approximately 3,300 species within the orders Monotremata, Marsupialia, Rodentia, Insectivora, Chiroptera, Lagomorpha, and Hyracoidea. Rodents account for roughly half this number, while bats account for 27%, insectivores for 12%, and lagomorphs for 2%. By comparison, there are about 640 species of larger mammals, which are represented predominantly by the Carnivora (40%) and the Artiodactyla (27%). In order of importance for tick-borne diseases, however, the rodents and lagomorphs are widely recognized for their role in tick and pathogen ecology, followed by the insectivores. The bats generally support a more specialized tick fauna and are proportionately under-represented as far as their relative importance in tick-borne diseases is concerned.

The extent of movement of TBDs by small mammals is less dramatic than that observed by birds due to the limited mobility of the group in general, but the accumulated effect of small mammal dispersal through time has mitigated the present geographic patterns of most important human pathogens. Dispersal is integral to the population ecology of many small mammals, especially rodents. It is a function of population size, spacing, composition, and productivity (Lidicker, 1975) and may have underlying genotypic bases within the population. Demographic dispersal may be triggered by lack of sufficient resources as a population approaches or exceeds saturation (saturation emigration), or may represent a behavioral adaptation in response to an inherent social organization, reproductive success, or resource tracking (presaturation emigration). This behavioral strategy may also occur in response to interspecific interactions due to competitors, predators, and, presumably, parasites. The quality or condition of individual "dispersers" will likely be different if individuals move from a location under physiological stress than if energy reserves were previously stored as a prelude to the move. Levels of physiological stress experienced as an adjunct to dispersal may have important implications regarding the degree of immunological resistance a dispenser possesses to either ectoparasite infestation and/or infectious agents (see discussion for rodents below). Finally, the amount of energy devoted to movement may alternatively extract a cost for other behaviors, such as grooming activity (Turner et al., 1983), which would further predispose the dispersing individual to transport of ticks and pathogens.

3.3.1. Evolutionary Considerations for Rodents

Rodents serve as hosts to more tick species than any other mammalian taxon, with more than 300 of the approximately 600 three-host ixodid species feeding on rodents. In addition, rodents contribute greater biomass and numbers than any other mammalian order and have become the dominant herbivores in many of the major biotas of the world as extinctions within the larger mammalian herbivorous faunas during the Pliocene and Pleistocene elevated smaller mammals to prominence. They are also the most important mammalian group

as far as the maintenance, and, perhaps, spread of TBDs is concerned. However, discussion of the evolution of the Rodentia or the geologic changes that influenced their distribution is beyond the scope of this article.

The myomorph rodents are especially tied to transmission and maintenance of a number of human pathogens including viral TBDs (West Nile, TBE complex, Dugbe, Uukuniemi, Congo–Crimean hemorrhagic fever, Bhanja, Colorado tick fever, Tribec, and Bandia), bacterial/spirochetal TBDs (Lyme borreliosis, relapsing fever borreliosis, tularemia, and tick-borne rickettsioses) and sporozoan agents (babesiosis and theileriosis). Beside dispersing infected ticks, rodents act as long-term overwintering maintenance reservoirs for agents such as *B. burgdorferi* (Levine et al., 1985), Colorado tick fever, and TBE virus (Blaskovic, 1967) and dispersal of infected rodents may introduce pathogens into new sites at the microhabitat level. The relative paucity of recognized tick-borne agents of cricetid rodents of South America illuminates the possible role of long-term events in the dissemination of TBDs. The recent entry of the Cricetidae into South America during the Pliocene (6–10 m.y.) was followed by a remendous explosion of genera in that continent as this group exploited available niches either not occupied by marsupials and caviomorph rodents, or the cricetines successfully excluded those groups through direct competition. The arenaviruses, which are transmitted by cricetid rodents, apparently expanded over this period, and novel viruses from this group are still being discovered. The spotted fever group rickettsias filtered through this land bridge (or were introduced by birds) but where are the tick-borne viruses associated with this group? Colorado tick fever and Powassan viruses are the most proximal North American tick-borne viral agents. Tularemia, an important, widely dispersed bacterial agent in the tick faunas of North America, is also apparently absent from this land mass.

(a) General Models of Rodent Dispersal and Transport of Ectoparasites

Dispersal movement by rodents is a common event at the community and population levels, and is presumably tied to patchy distribution of resources (food, shelter, and mates) in the environment. Dispersal occurs routinely for particular age and sex groups, and in association with seasonal events or changes in population density (Lidicker, 1985). This activity provides opportunities for exchange and spatial displacement of ectoparasites, and for mixing and spread of the pathogen population. In a number of microtine species, social structure forces movement of juveniles away for natal home ranges. Greater than 50% of juvenile male field voles (*Microtus agrestis*) moved more than one home range diameter (58.5 m) from their natal home ranges while only 22% of juvenile females moved from their natal range (Sandell et al., 1990). While male dispersal was tied to population density in this system, female dispersal showed no correlation with population density. Linzey (1989) found that juvenile white-footed mice (*Peromyscus leucopus*) were over-represented in marginal habitats (dispersal sinks) adjacent to the "preferred" woodland sites selectively occupied by adults. Population density effects, operating via female territoriality and variation in available forage, were directly related to the level

of transients in a study of meadow voles (*Microtus pennsylvanicus*) in Virginia (Jones, 1990).

Such events contribute to the ecological dynamics of the associated tick ectoparasites. Korch (1985) found a differential spatial distribution of adult versus juvenile meadow voles in a study of *D. variabilis* microdistribution. A large proportion of juvenile voles (75%) were captured in non-preferred woodland habitat, while most adult captures (>80%) occurred in old-field and ecotone habitats. Approximately 36% of these dispersing juvenile animals were infested with a mean of 1.5 larvae and 0.3 nymphs and had ectoparasite levels higher than "non-dispersers". Sonenshine and Stout (1968a) believed that *D. variabilis* dispersal by small mammals resulted, in part, from differences in spatial distribution of subadult versus adult American dog ticks in old-field habitats. Campbell and MacKay (1979) hypothesized that transport of larval dog ticks by woodland rodents from wooded to grassland habitats in Nova Scotia accounted for observed differences in abundance between larval and nymphal dog ticks on woodland versus old-field rodent species, respectively. In a similar approach, Laurance and Coan (1987) found that phenological changes over a single summer season in a small mammal community in southwest Idaho was the underlying factor in altered microhabitat use and population density of three rodent species which in turn influenced tick–host associations for *D. andersoni*. Seasonal shifts from xeric to mesic microhabitats for pocket mice (*Perognathus parvus*) and deer mice (*Peromyscus maniculatus*) along with recruitment of young individuals into the pocket mouse population influenced tick–host interaction, hence the distribution pattern of this tick on these hosts.

Direct evidence of dispersal of ticks by small mammals is provided by using ticks marked with an identifiable label. Radioactive tagging with ^{14}C was used by Sonenshine (1973) and by Korch (1985) to identify host-related dispersal of *Dermacentor variabilis* in endemic areas of Rocky Mountain spotted fever. Sonenshine (1973) released approximately 756,000 tagged ticks over 3 years into a 7.7 ha section of a 17.6 ha old-field–woodland site. He observed dispersal of subadult ticks up to 301 m, principally by wide-ranging white-footed mice. The mice captured in the area outside the release site, but infested with tagged ticks, were observed to have significantly greater spacing and home range size than conspecifics in the same area which were not infested with tagged ticks. Korch (1985) seeded radiolabeled larval ticks into a 1,600 m^2 old-field site, at a density of 6.4 larvae/m^2, on a 1.44 ha study area comprising a vegetation gradient from grassland to woodland. Thirty-one per cent of radiolabeled ticks were recovered on small mammals captured one or more sampling distances away from the release point. Radiolabeled larvae represented 10% of the population recovered on mammals outside of the release area. Radiolabeled larvae were recovered on white-footed mice (*Peromyscus leucopus*) in woodland settings, up to 110 m away from the release site, even though this mammal species was never captured in old-field trapping sites during a 2-year period of sampling. The distribution of the dispersal distances for labeled ticks on voles was found to be similar to the distribution of distances between subsequent

recapture for voles in general, demonstrating that transport of ticks did not necessarily differ from movement parameters of the voles.

(*b*) *Movements within Home Ranges and Infestation Level*

Movement of the rodent within its daily home range also has an influence on ectoparasite–host interactions and thus microdistribution of pathogens. The relationship between rodent host spacing or movement, and ectoparasite burden was identified as early as 1906 in studies of rats and fleas in plague sites in Bombay, India (Hirst, 1953). Milne (1949) was an early proponent that contact between *I. ricinus* and its hosts (sheep) occurred by random encounter as the host moves through its range which suggested that increased activity by the host should result in greater infestation as a host moves through space. Mohr (1961) suggested that variation between the mean ectoparasite burdens of different mammal species occupying the same habitat could be accounted for, in part, by differences in home range size, use of habitat, and surface measurements of the host affecting contact rate between the host and the vegetation. Sonenshine and Stout (1968a, 1968b) found a positive relationship between mean tick burdens on white-footed mice (*P. leucopus*) and the distance traveled by the host between successive periods of recapture, although no significant relationship was found between size of home range and infestation level. Other investigators have failed to find a strict association between movement indices and ectoparasite levels, e.g., dog tick burdens on voles (Korch, 1985) or on white-footed mice (Zimmerman et al., 1987). Lutta (1968) in fact observed more *Ixodes trianguliceps* on "settled" versus "migratory" microtines in a study in Karelia. Beside host movement, additional considerations in accounting for observed infestation levels must be given to the distribution pattern of the ticks in the habitat and to host preference by the ticks. Nilsson and Lundqvist (1978) demonstrated that for *Ixodes ricinus* in Sweden, host preference, rather than movement and activity patterns were more responsible for differences in infestation levels between the more broadly active but less infested *Clethrionomys glareolus* versus *Apodemus sylvaticus*, which had higher larval tick burdens.

The dispersion pattern of the ticks within the environment is another component in the dynamics of host movement and acquisition of ticks. For example, if the distribution pattern of egg masses (hence larval ticks) in the environment is random but egg masses are abundant, then the probability of acquiring a large tick burden may be independent of home-range size. An individual with a smaller home range which comes into frequent contact with a given cluster of larval ticks within its range could acquire an equivalent or even higher tick burden than more broadly ranging conspecifics. As these larvae feed and drop from the hosts, however, the dispersion of nymphs in the environment may change from that observed with larvae, especially if drop-off is random. This new dispersion pattern may be more sensitive to host activity levels. Korch (1985) noted that the infestation levels of larval *Dermacentor variabilis* on white-footed mice were highly overdispersed while that for the nymphs was randomly dispersed, suggesting that mice were exposed to larvae

distributed in the environment in a negative binomial dispersion pattern, and to a less aggregated distribution pattern of nymphs. These findings suggest that for this tick–host system, a few mice acquired larval ticks by contacting a newly hatched egg mass, while the distribution of nymphs in the environment is random, and is likely a function of the random drop-off pattern of engorged larvae from the previous host. Kitron et al. (1991), however, found that the overdispersed distribution of *I. scapularis* larvae of *P. leucopus* resulted from a series of non-random successful attachments of single larvae, rather than by simultaneous attachment of clumps of larvae which would be favored by greater contact rates due to highly mobile hosts.

A final important consideration in the dynamics of tick dispersal by small mammal movements may be the energy loss realized by the host due to tick feeding. This views dispersal from the small mammal's vantage point, i.e., what is the cost of the ectoparasite load on the dispersal behavior of the mammals themselves? The relative blood loss for an equivalent number of larval ticks feeding on *P. leucopus* (mean adult mass *c.* 16 g) versus *M. pennsylvanicus* (mean adult mass *c.* 40 g) may have a greater influence on movement parameters for the smaller species. Korch (1985) found up to 122 *Ixodes scapularis* larvae per *Blarina brevicauda* (short-tailed shrew, mean body mass of 18 g). Nosek and Grulich (1967) similarly observed tens to hundreds of *Ixodes ricinus* ticks of all stages on the hedgehog (*Erinaceus roumanicus*) in a focus of tick-borne encephalitis virus. The consequences to mobility for these different levels of infestation is unknown. Lehmann (1993) reviewed the empirical data on the general influence ectoparasite burden may have on fitness and pointed out that young individuals may experience more severe impact due to the ratio of accessible surface to body (i.e., blood) volume. Heavily parasitized individuals may not disperse as far as less burdened individuals. However, remembering that dispersal can be a consequence of the lowered ability of an individual to compete with conspecifics for a given home range, more heavily parasitized hosts may be forced to disperse by virtue of lowered energy to compete for space engendered by their sustained ectoparasite burden. Clearly, there would be a difficulty in identifying cause and effect in levels of ectoparasites found on dispersing individuals under a routine ectoparasite survey, and answers to such dilemmas would be best evaluated under more controlled conditions. A wide range of opportunities exists for detailed studies on the biology of interaction between ticks and hosts including host contact rates relative to dispersion patterns of ticks, interspecific differences under controlled conditions, and physiological consequences of infestation relative to the movement pattern of the host and the drop-off pattern of the tick

(c) Examples of Movement of TBD Effected by Rodents

Kharitonova et al. (1978) observed a ten-fold increase in prevalence of Omsk Hemorrhagic fever virus in migrating (dispersing) water voles in Novosibirsk, possibly associated with reactivation of latent viral infection induced by stress. Kraft (1963) noted that environmental conditions in a tularemia focus which produced fluctuation in water levels caused displacements of the *Arvicola*

terrestris population bringing these hosts into greater contact with *Dermacentor marginatus*, the principal vector for *F. tularensis* in the Tselinograd Oblast region. This enhanced the rate of tick–host contact, contributing to epizootic outbreaks of tularemia. Dissemination of Lyme disease infected ticks in a study of the transmission dynamics in a residential area was demonstrated by movement of rodent hosts into suboptimal microhabitats around human dwellings, causing an increased potential for transmission of the disease to humans.

3.3.2. Chiroptera

Bats contribute greatly in diversity and population size to the species assemblages of the tropics and temperate zone, and are among the most mobile of the mammalian taxa with numerous species migrating great distances annually. A total of 55 species of soft ticks are strict ectoparasites of bats (Oliver, 1989) and they serve as hosts for a smaller number of hard ticks. Despite their biomass, distribution and mobility, the relative contribution by bats to the distribution of the major tick-borne zoonoses of man appears to be limited. L'vov et al. (1973) isolated an arboviral agent causing human disease, Issyk-Kul virus (Bunyaviridae), from *Nyctalus noctula* (taken from a human dwelling) and subsequently isolated from other vespertilionid and rhinolophid bats and ectoparasites (*Argas vespertilionis* and *Ixodes vespertilionis*) in the Tajikistan and Kirghizia Republics (Commonwealth of Independent States). Another related agent to Issy-Kul, Keterah virus, is associated with bats and *Argas pusillus* in Malaysia (Karabatsos, 1985). Together, these agents, along with a number of bat species and argasid vectors in the *A. vespertilionis* complex, may constitute a viral tick-borne disease complex with wide distribution over the temperate and tropical regions of Eurasia, Africa and the Malaysian zoogeographic region. Other tick-borne diseases of human importance which are associated with bats or their ectoparasites include CCHF virus (reviewed in Hoogstraal, 1979), *Coxiella burnetii* (reviewed in Marchette, 1982), tick-borne encephalitis viruses (reviewed by Gresikova and Calisher, 1988) and Kyasanur Forest Disease virus (Rajagopalan et al., 1969). The direct role of bats and their ectoparasites in the transmission of TBD agents to humans is likely marginal due, in part, to host specificity of bat ectoparasites and to the specialized habitats occupied by a number of bat species. These factors limit the ability and/or frequency of contact with humans or other mammalian hosts. More studies are needed, however, in the relationship between bat migrations and the dissemination of ticks and associated pathogens.

3.3.3. Insectivores

The Order Insectivora is the most primitive taxa of eutherian mammals appearing in the fossil record of the Late Cretaceous of Asia and North America. The extent genus *Sorex* extends to the mid-Oligocene of Europe. They are contributors to the epizootiology of tick-borne viruses such as the TBE virus

complex (Nosek and Grulich, 1967), and Crimean–Congo hemorrhagic fever in Asia (Hoogstraal, 1979) and support tick vectors of CCHF in Africa (Colbo and Macleod, 1976). Shrews have recently been implicated in the ecology of Lyme disease spirochetes (Telford et al., 1990) and a new infectious spirochete was isolated from shrews (Anderson et al., 1987). Matuschka et al. (1990) suggested that the hedgehog–*I. hexagonus* association in Europe may support the epizootiology of Lyme disease in Europe. The hedgehog (*Erinaceus roumanicus*) also acts as an overwintering reservoir of TBE viruses, maintaining the virus during hibernation (L'vov, 1970).

3.3.4. Lagomorphs

The Lagomorphs (rabbits and pikas) are one of the older orders of eutherian mammals with fossil history identified to the late Paleocene of Asia which seems to be the origin of this group. The Leporidae family comprises most of the species in this order (*c.* 50 species) and are found on all major land masses except Antarctica. The Ochotonidae (14 species) are currently restricted to Asia and North America, although they are represented in the Oligocene of Europe and the Miocene of Africa.

Rabbits are implicated in a variety of tick-borne diseases, frequently participating in alternative disease cycles to those seen in co-occurring rodents, insectivores, or larger mammals. These cycles often occur in parallel and typically involve a different tick vector species. This phenomenon has been described for Rocky Mountain spotted fever rickettsia and *Haemaphysalis leporispalustris* and for Lyme disease spirochetes in *Ixodes dentatus* (Telford et al., 1989). As a group they figure prominently in supporting CCHF vectors such as the *Hyalomma marginatum* complex and *H. truncatum* (Colbo and Macleod, 1976; Hoogstraal, 1979), and *Haemaphysalis punctata.* They may also serve as alternative hosts for important vectors, sustaining these populations during population depression of the principal hosts as observed for *I. persulcatus* and mountain hare, *Lepus timidus* (Uspensky and Rubina, 1992). The distribution of rabbits and *H. leporispalustris* throughout the Nearctic is thought to be an underlying vehicle for the dissemination of RMSF within and between the continental land masses (Marchette, 1982).

3.4. The Role of Large Mammals in Support and Dispersal of Ticks and Tick-borne Agents

Large mammal (mean adult mass >3 kg) taxa which have played important roles in the dissemination of tick-borne diseases are, in order of presumed importance, the Artiodactyla, Carnivora, and Perissodactyla. This is due to: (1) their importance as a blood-meal source for adult stadia for oviposition of many species of hard ticks; (2) their greater mobility; (3) their social grouping (including domestic herds) which results in greater potential host availability; (4) their relatively stable population sizes (versus small mammals) resulting in a

more predictable blood source (although offset by the greater likelihood of an immunological rejection against the ectoparasites); and (5) the opportunity for intraspecific and interspecific horizontal transmission of pathogens by co-feeding ticks. The tremendous species diversity generated within several of the large mammal lineages during the Cenozoic probably resulted in similar radiational opportunities for ectoparasites. Finally, the activities of man in concentrating and moving large domestic animals has provided ample historical and recent opportunities for dispersing ticks and pathogens.

The major tick-borne diseases of human importance that are associated directly or indirectly with wild and domestic large mammals include spotted fever rickettsias, Lyme borreliosis, tick-borne encephalitis viruses, Crimean–Congo hemorrhagic fever virus, Colorado tick fever, and the ehrlichial diseases. A total of 22 tick-borne arboviruses have been reportedly isolated or natural infection demonstrated by serologic response from large domestic or wild ungulates, while an additional five tick-borne arboviruses have been isolated from carnivores. Besides tick-borne pathogens of human import, a variety of other rickettsial (*Cowdria ruminantium, Ehrlichia, Anaplasma*), sporozoan (*Babesia, Piroplasma, Theileria*) and bacterial (*Borrelia, Brucella*) agents are transmitted by ticks to large mammals. Some of these agents have nearly worldwide distributions, such as *Babesia equi, Babesia bovis* which have a wide distribution in South America, West Africa, Australia, and possibly Central Asia. *Theileria annulata* is endemic from North Africa, across most of Asia, and into the Sunda Shelf. Surveys in East Africa have demonstrated a wide range of piroplasma infection in large game animals (Purnell, 1980). One of the most widespread of all tick-borne viral pathogens is the argasid-borne iridovirus causing Africa Swine Fever (ASF). This agent was introduced recently into Europe, South America, and the Caribbean from Africa as a result of transportation of infected livestock and contaminated animal by-products (Wilkinson, 1988).

3.4.1. Evolutionary Considerations for Large Mammal Groups

The evolutionary history of the relevant larger mammals has strongly influenced the zoogeography of tick-borne diseases. Unfortunately, detailed discussion of these events is beyond the scope of this chapter. However, several of the more important evolutionary changes and their effects on tick-borne diseases merit consideration, if only briefly.

Within the ungulates, there has been a gradual shift in species diversity since the Paleocene from the odd-toed Perissodactyls (17 current species) to the even-toed Artiodactyls (195 current species). This shift reflected the increase in grassland and steppe habitats brought about by cooler, drier macroclimates and the concomitant changes in dentition and digestion needed to consume these new food resources. As a result, the Cervidae, Bovidae, and Camelidae are now important artiodactyl families for dissemination of ticks and TBDs, while the Perissodactyla (horses, zebras, etc.) are much less important as TBE hosts. Evolution and geographic dispersal of the Carnivora could also have

played a key role in spread of tick-borne diseases, especially by the canids (dogs). The most widely dispersed tick species of terrestrial mammals is possibly the brown dog tick, *Rhipicephalus sanguineus*, and the pathogen, *Ehrlichia canis*, shares much of this distribution. This host–pathogen–vector association probably originated in Africa during the Miocene or later, coincident with the assumed entry of canids into that continent (Marchette, 1982). The evolutionary history of the camel family probably influenced the distribution of *Hyalomma* ticks. Their absence in North and South America may have resulted from extinction of these hosts during the late Pliocene and Pleistocene. Similarly, the presumed radiation and dispersal of bovids from Asia likely influenced the phylogeny and/or dispersal of major tick genera: *Hyalomma* from Asia (Marchette, 1982) into Europe and Africa; *Dermacentor* from putative Central Asian origins (Berdyev, 1989) and *Rhipicephalus* in Africa (Kaiser and Hoogstraal, 1967). In contrast, the climatological considerations causing the depauparate bovid fauna of North America may have limited the expansion of the genus *Dermacentor* and prevented spread of *Hyalomma* ticks and CCHF virus from Asian foci. The evolution and dispersal of the bovids and the deer family (Cervidae) over geologic time are thought to have profoundly influenced the occurrence of their tick parasites and many important diseases. An example is the genus *Odocoileus*, which is an important host for the deer tick (*I. scapularis*) in the northeastern United States. If these ticks, or their ancestors, dispersed with this mammal group from North America to Asia or vice versa, *I. scapularis* should reflect a stronger Beringian relationship with the *I. persulcatus* group than with European representatives. Another concerns the African rickettsial pathogen, *Cowdria ruminantium*, transmitted to bovids solely by *Amblyomma* ticks. Its restriction to Africa (aside from a few isolated foci in the Caribbean) is believed to reflect the evolution and dispersal of these hosts and tick–host associations during earlier geologic epochs. In either case, these tick–host and disease distributions provide noteworthy new opportunities for understanding the spread of tick-borne zoonoses.

3.4.2. Movement Parameters and Tick Dispersal Potential of Large Mammals

Daily and seasonal home range size, as well as annual migration patterns, provides indications of the near-term dispersal capability of various hosts. As previously discussed, range size scales approximately with body mass, although it is generally smaller for herbivorous and carnivorous mammals. Small browsers, such as the chevrotains may have ranges of 15–50 ha, while range sizes for larger deer are proportionately greater, e.g. *Odocoileus virginianus*—30–75 ha, *Odocoileus hemionus*—11–185 ha, *Alces alces*—4–25 km^2. Grazers such as sheep, antelope, and buffalo have home ranges, such as, *Raphicerus rupicapra*—2.5 ha, *Antilope cervicapra*—7 km^2, *Tragelaphus strepsiceros*—22 km^2, *Ovis canadensis*—8–50 km^2. Examples of carnivore ranges are *Felis catus*—0.1–6 km^2, *Vulpes vulpes*—5–16 km^2, *Canis latrans*—12–80 km^2, *Felis rufus*—11–85 km^2, *Canis lupus*—30–2,580 km^2, *Felis concolor*—200–435 km^2, *Ursus*

arctos—57–769 km^2. Mobility during a single activity period or data collected over a given time period can also yield an index of dispersal by an individual of ticks or pathogens. Coyotes and wolves may travel 10–100 km during activity periods as measured by radiotelemetry.

Bloemer and Zimmerman (1988) compared mobility measurements of coyote, fox, and deer in an endemic focus of Rocky Mountain spotted fever in the southeastern US and noted that coyotes traveled between 10 and 25 km daily, and had much larger home ranges (25–50 km^2) than did deer (2.6 km^2). Coyotes were considered more important agents for dispersal of *A. americanum*, *D. variabilis*, and *I. cookei* than the other hosts examined. However, they may play a smaller role in supporting the tick populations due to lower population density.

Large-scale, seasonal transport of ticks is effected by migration of large mammal hosts. Two classes of migration patterns are generally observed for larger terrestrial mammals: altitudinal and lateral (McCullough, 1985). Altitudinal shifts between summer and winter ranges are observed for mule deer (*Odocoileus hemionus*), elk (*Cervus elaphus*), and big horn sheep (*Ovis canadensis*) in North America. Lateral migrations are commonly observed in wild bovids living in a variety of habitats in Africa. Impala (*Aepyseros melampus*) migrate short distances in acacia woodland habitats (several hundred meters) from wet to dry season ranges, whereas bovids living in semi-arid habitats, such as wildebeest (*Connocaetes taurnus*) and eland (*Taurotragus oryx*) may migrate several hundred kilometers while tracking available forage (Sinclair, 1983). Different species are observed to migrate in sequence in East Africa with Burchell's Zebra (*Equus burchelli*) and topi (*Damaliscus korrigum*) appearing first, followed by wildebeest and then Thompson's gazelle (*Gazella thompsoni*) in East Africa (Vesey-Fitzgerald, 1960; Gwynne and Bell, 1968). These dynamics may provide opportunities for interspecific shifting of ectoparasites as groups travel through a region. Although relative host preferences are reported for tick species that feed on the various African bovids (Rechav et al., 1987), there is still a broad overlap in host range for a number of common tick species, thus the possibility for pathogen transmission between host species exists.

3.4.3. Evidence for Dissemination of Diseases by Movements of the Large Mammals

Crimean–Congo hermorrhagic fever virus serves as an interesting example to speculate on the movement of larger mammals and the possible dispersal of tick-borne pathogens. The complex ecology of this virus (see Chapter 13 for a more thorough description), involves a great variety of tick species (30 identified to date); small mammals and bird hosts for the immature stages of the vectors, and larger vertebrates for the adult stages. Camicas et al. (1990) believe the occurrence of *Hyalomma* sp. vectors in a region is the key factor for virus presence. Besides the described role of birds in the movement of tick vectors of CCHF virus, a second hypothesis for the widespread dissemination of this virus involves evolution and movement of the bovids from Asia to Europe and Africa.

An assessment of the role that wild bovid or other feral large mammal species have played, or currently play, in each faunal region in support of this virus, is central to the bovid dispersal hypothesis, however, this area is poorly studied (Hoogstraal, 1979). Evidence for the epizootiological importance of the larger wild mammals is mostly restricted to serosurveys, or to isolation of virus from ticks found infesting these hosts (reviewed by Watts et al., 1989). Only slightly better data exist for the influence of the domestic large mammal species in the spread of the disease. For Nairobi sheep disease (NSD), a virus in the same genus as CCHF virus, Terpestra (1990) indicated that movement of infected ticks by wild or domestic ungulates in years with heavy rains may be responsible for outbreaks of NSD in NSD-free regions, although no direct evidence was discussed.

During the historical period, transport of ticks by camels may have been an important dispersal mechanism for infected ticks. Hoogstraal (1979) reviewed studies by Kurbanov and collaborators (1974a, 1974b) that camels likely played an important role in CCHF epidemiology of the Askhabad area, Turkmenia as hosts for the vector *H. a. asiaticum*. CCHF virus has been isolated from *H. a. asiaticum* (Smirnova et al., 1974) and *H. dromedarii* (Smirnova et al., 1978) removed from camels in Turkmenia, as well as from four additional tick species. Morrill et al. (1990) reported that 14% of camels imported into Egypt from Sudan and Kenya were seropositive for CCHF virus antibody. Since these animals are still used for long-range transport, and have likely been so used for several millennia in Asia and Africa, it is possible that new foci of endemic CCHF virus could have been established by these practices.

Movement of sheep and goat herds between diverse habitats may also influence dispersal of these pathogens. Smirnova et al. (1978) found high seropositivity rates for both sheep and goats in scattered foci in the foothill regions of Turkmenia. Movement of sheep into summer pastures from endemic CCHF regions in Tadzhikistan created opportunities for translocation of CCHF virus infected ticks (Pak and Mikhailova, 1973, as cited in Hoogstraal, 1979). Wilson et al. (1990a, 1990b) systematically sampled sheep from different bioclimatic regions in Senegal, and found that CCHF virus seroprevalence in sheep was positively correlated with the greater relative abundance of *Hyalomma* ssp. in the drier, Sahelian zone than in moister bioclimatic zones. Within this general trend, however, they indicated that spatial heterogeneity within a given bioclimatic zone is related to migration patterns of the hosts. Movement of cattle with CCHF-infected ticks (Causey et al., 1970) or of viremic but asymptomatic cattle (Swanepoel et al., 1985) also create potential dispersal opportunities for the virus.

The transovarial transmission rate of CCHF virus is, however, a crucial variable to consider in evaluating the role of large mammals for establishing a new focus of CCHF virus via dispersal of infected ticks (Burgdorfer and Varma, 1967). Transport of infected fecund females to uninfected sites provides a mode of dispersal for vertically transmitted agents. As the transovarial transmission rate diminishes, however, the relative importance of large mammal dispersal of adult ticks is also lessened. A condition of non-stable transmission results

(Spielman and Rossignol, 1984) such as seen for *Boophilus* transmission of babesioses. Kondratenko (1976) observed transovarial transmission of CCHF virus in 38–79% of pooled F_1 progeny of several Eurasian vector species (*Hyalomma m. marginatum*, *Rhipicephalus rossicus* and *Dermacentor marginatus*). Relatively low transovarial transmission rates of this virus are reported for the important vector species, *Hyalomma truncatum*, following feeding on experimentally infected sheep (Wilson et al., 1991) and rabbits (Gonzales et al., 1992). Other investigators (Shepard et al., 1989; Logan et al., 1990) have demonstrated no transovarial transmission of CCHF virus for seven species of vectors. Under conditions of low transovarial transmission rates, establishment of new foci of infection would be more dependent on horizontal transmission dynamics, especially by immature stages. Since a number of the important vector species parasitize small mammals and birds as subadults, these hosts may be more critical for the maintenance and spread of CCHF virus. Heneberg et al. (1967) felt that despite agricultural practices in eastern European foci which have caused a shifting of the host range of the adult stage of the CCHF vector, *Hyalomma m. marginatum*, from wild to domestic animals, the immature stages are still maintained by small mammals and birds, and thus should be considered as fundamental for understanding the maintenance and dissemination of CCHF virus. Camicas et al. (1990) also concluded that a small mammal–*Hyalomma truncatum* transmission cycle is critical in the maintenance cycle of CCHF virus in Senegal.

3.5. Human Influences on Large Mammal Support and Dispersal of Tick-borne Diseases

Agricultural and animal husbandry practices have created a myriad of possibilities for tick-borne pathogen dissemination. These activities create new habitats for tick vectors, opportunities for vector dispersal, and mechanisms for intercontinental transfer of infested hosts or pathogen-infected products. Habitat conversion to pasture for domestic animals with successional reversion to natural climax has been identified as the underlying process in establishment of Q fever, tick-borne rickettsioses and tick-borne encephalitis foci over large territories in the former USSR (Rosicky, 1968). The factors contributing to this process included: (1) the adaptation of ticks to conditions created by man; (2) the creation of habitat mosaics; and (3) increase in ecotonal habitat. As previously mentioned, a similar phenomenon of reversion of farmland to woodland habitat in the US is considered an important determinant in Lyme disease expansion in North America (Spielman et al., 1985).

Agricultural practices, however, have their greatest effect on tick-borne disease dissemination in the movement of domestic animal species. Examples of transport of tick-borne disease by movement of domestic animals include *Rickettsia conorii*, equine and canine babesioses, CCHF virus and *Cowdria ruminantium*. Marchette (1982) suggested that the presence of *R. conorii* in India is due to transport of domestic animals into the region from an African or

southwestern Asian biocenose in historical times since the vectors in this region are non-indigenous, and domestic mammals appear to be the chief, or only, affected population. Walker et al. (1992), however, have identified significant antigenic diversity among *R. conorii* strains, with significant antigenic differences between Indian strains and other geographic strains of *R. conorii* which suggest that this pathogen may have experienced a long period of evolutionary divergence in these regions. Although the prehistoric range of equine babesiosis (*B. equi* and *B. caballi*) is not known, these agents are widespread over Eurasia (except the British Isles and Japan), Africa and Central/South America. Equine babesiosis was documented in Australia for the first time in 1976 (Churchill, 1976) but never became permanently established. In North America, however, importation of infected horses into Florida from Cuba in 1961, along with the presence of a suitable vector (*Dermacentor nitens*) in the region, resulted in an endemic focus of *B. caballi* (Schein, 1988). Moreau et al. (1988) demonstrated an increase in the incidence of canine babesiosis in France over a 10-year period, which was ascribed in part to movement of animals by humans into the area from neighboring countries. Okaeme (1986) evaluated the risk of importation of ticks into Kainji Lake National Park, Nigeria, by examining ectoparasite burdens on infiltrating nomadic cattle. He identified seven tick species which posed a risk to indigenous wild ungulates, including three species (*H. aegyptium*, *R. simus*, and *R. appendiculatus*) which had never been recorded in the area. Five of these species collected were potential vectors of CCHF virus. Hoogstraal et al. (1969) suggested that *Haemaphysalis bispinosa*, which is widespread in the Malay Peninsula on domestic hosts, has characteristics of an introduced species. This tick was rarely found in ectoparasite surveys of indigenous wild mammals (carnivores and artiodactyls) of the region, whereas, in the adjacent Indian subcontinent this ectoparasite commonly occurs on endemic wild carnivores and artiodactyls. Heartwater (*Cowdria ruminantium*) was identified in cattle and their ectoparasites along a 600 km trek troute from endemic sites in Central Zaire to a non-endemic location in Southern Zaire (Matton and van Melckebeke, 1989). Finally, accidental importation of the heartwater vector, *A. hebraeum*, on rhinoceros from South Africa, destined for zoological parks in the US (Diamant, 1965), is analogous to "jump-dispersal" of vectors under natural conditions.

Finally, the age of rapid travel may also make it possible for humans themselves to be the agency of dissemination events for ticks and diseases. Halliday and Sutherst (1990) reported accidental introduction of a living female *Dermacentor variabilis* into Canberra, Australia, by an infested air passenger. These authors concluded that climatic conditions in this part of Australia were suitable for colonization of the area.

4. CONCLUSION

I have described a variety of processes that have impacted on the distribution and dissemination of tick-borne diseases. The location of a given pathogen

can be described from a variety of viewpoints, encompassing spatial events as grandiose as the collision of continents to the seemingly mundane activity pattern of single ticks and their hosts. Between these extremes, evolutionary pressures on host, vector, and pathogen create new patterns through time, but the evidence of past relationships may emerge by combining new technologies and theories with old observations.

5. REFERENCES

Adeyeye, O.A. and Butler, J.F. (1989) Population structure and seasonal intra-burrow movement of *Ornithodoros turicata* (Acari: Argasidae). *J. Med. Entomol.* **26**, 279–283.

Aison, S., Johnson, M.K., and Harger, T.R. (1984) Role of birds in dispersal of itch grass (*Rottboellia exalatata* (L.) L.f.) in the southeastern U.S.A. *Prot. Ecol.* **6**, 307–313.

Amin, O.M. (1970) The circadian rhythm of dropping of engorged larvae and nymphs of the American dog tick *Dermacentor variabilis* Say (Acarina: Ixodidae). *J. Med. Entomol.* **7**, 251–255.

Anderson, B.E., Dawson, J.E., Jones, D.C., and Wilson, K.H. (1991) *Ehrlichia chaffeensis*, a new species associated with human ehrlichiosis. *J. Clin. Microbiol.* **29**, 2838–2842.

Anderson, J.R. (1974) Symposium on reproduction of arthropods of medical and veterinary importance. II. Meeting of the sexes. *J. Med. Entomol.* **11**, 7–19.

Anderson, J.F. (1988) Mammalian and avian reservoirs for Borrelia burgdorferi. *Ann. New York Acad. Sci.* **539**, 180–191.

Anderson, J.F. and Magnarelli, L.A. (1984) Avian and mammalian hosts for spirochete-infected ticks and insects in a focus in Connecticut. *Yale J. Biol. Med* **57**, 627–641.

Anderson, J.F., Johnson, R.C., Magnarelli, L.A., and Hyde, F.W. (1986) Involvement of birds in the epidemiology of the Lyme disease agent *Borrelia burgdorferi*. *Inf. Immun.* **51**, 394–396.

Anderson, J.F., Johnson, R.C., Magnarelli, L.A., Hyde, F.W., and Andreadis, T.G. (1987) New infectious spirochete isolated from short-tailed shrews and white-footed mice. *J. clin. Microbiol.* **25**, 1490–1494.

Anderson, J.F., Magnarelli, L.A., and Stafford, K.C., Jr (1990) Bird-feeding ticks transstadially transmit *Borrelia burgdorferi* that infect Syrian hamsters. *J. Wildl. Dis.* **26**, 1–10.

Aschoff, J. (1981) A survey on biological rhythms. *In:* Aschoff, J. (ed) *Handbook of Neurobiology*. Plenum, New York.

Babenko, L.V. (1967) *Seminar on the Ecology, Biology and Control of Ticks and Mites of Public Health Importance*. WHO/VBC 68.57:9–10.

Bairlein, F. (1985) Avian autumn migration in Algerian Sahara. *J. Ornithol.*, **126**, 348.

Baker, R. (1981) *Mystery of Migration*. Viking Press, New York.

Balashov, Yu.S. (1954) Peculiarities of diurnal rhythm of detachment of engorged female *Ixodes persulcatus* from cattle. *Dokl. Akad. Nauk. U.S.S.R.* **98**, 317–319.

Balashov, Yu.S. (1989) The coevolution of ixodid ticks and terrestrial vertebrates. *Parazitologiya* **33**: 457–468.

Balat, F. (1964) Under what circumstances are wild ducks and geese infested with ticks

and what is the epidemiological importance of such infestation? *Schr.-Reihe Landesst. Natursch. Landsch., Pflege Nordrhein Westfalen* **1**: 115–120 (in German, English translation NAMRU3-T230).

Battaly, G.B., Fish, D., and Dowler, R.C. (1987) The seasonal occurrence of *Ixodes dammini* and *Ixodes dentatus* (Acari: Ixodidae) on birds in a Lyme disease endemic area of southeastern New York State. *J. New York Entomol. Soc.* **95**, 461–468.

Belan, I. and Bull, C.M. (1991) Host detection by four Australian tick species. *J. Parasitol.* **77**, 337–340.

Bellrose, F.C. (1976) *Ducks, Geese and Swans of North America.* Stackpole Books, Pennsylvania.

Berdyev, A. (1989) History of the geographic ranges and migration routes of ticks in the genus *Dermacentor* Koch, 1844 (Ixodidae). *Parazitologiya* **23**, 166–72.

Berezin, V.V. (1971) Investigation of the ecology of arboviruses in river deltas of the Caspian and Azov Sea basins. *Inst. Folio Virus Entsel. Akad. Med. Nauk. S.S.S.R.* (in Russian, English translation NAMRU3-T1160).

Berezin, V.V. (1972) Bird migration in Azov and Caspian Basin River deltas and their possible role in distributing arboviruses. pp. 183–186. *In:* Chereapnov, A.I. (ed) *Transcontinental Associations of Migratory Birds and their Role in the Transmission of Arboviruses.* Nauka Publishers, Novosibirsk (in Russian, English translation NAMRU3-T1558).

Bhat, H.R., Sreenivasan, M.A., Goverhdhan, M.K., Naik, S.V., and Banarjee, J. (1978) Antibodies to Kyasanur forest disease virus in bats in the epizootic-epidemic area and neighbourhood. *Ind. J. Med. Res.* **68**, 387–392.

Blaskovic, D. (1967) The public health importance of tick-borne encephalitis in Europe. *Bull. Wld. Hlth. Org.* **36** (Suppl. 1), 5–13.

Bloemer, S.R. and Zimmerman, R.H. (1988) Ixodid ticks on the coyote and gray fox at Land Between the Lakes, Kentucky–Tennessee, and implications for tick dispersal. *J. Med. Entomol.* **25**, 5–8.

Borisov, V.I., Popov, A.V., and Sadekova, L.K. (1969) Investigation of ectoparasites of migratory birds in Povolzhie, *Vth Symposium on the Study of the Role of Migrating Birds in the Distribution of Arboviruses* (*Abstracts*). Academy of Sciences, Novosibirsk, pp. 100.

Boshell, M.J. (1969) Kyasanur Forest disease: ecologic considerations. *Am. J. Trop. Med. Hyg.* **18**, 67–80.

Boshell, M.-J., Rajagopalan, P.K., Patil, A.P., and Pavri, K.M. (1968) Isolation of Kyasanur Forest disease virus from ixodid ticks: 1961–1964. *Ind. J. Med. Res.* **56**, 541–568.

Bourliere, F. (1975) Mammals, small and large: the ecological implications of size. *In:* Golley, F.B., Petrusewicz, K. and Ryszkowski, L. (eds) *Small Mammals: Their Productivity and Population Dynamics.* University Press, Cambridge.

Brink, P., Svedmyr, A., and von Zeipel, G. (1965) Migrating birds at Ottenby Sweden as carriers of ticks and possible transmitters of tick-borne encephalitis virus. *Oikos* **16**, 88–89.

Burgdorfer, W. and Varma, M.G.R. (1967) Trans-stadial and transovarial development of disease agents in arthropods. *Ann. Rev. Entomol.* **12**, 347–376.

Burgdorfer, W., Barbour, A.G., Hayes, S.F., Benach, J.L., Grunwaldt, E., and Davis, J.P. (1982) Lyme disease– a tick-borne spirochetosis? *Science* **216**, 1317–1319.

Burgdorfer, W., Hayes, S.F., and Benach, J.L. (1988) Development of *Borrelia burgdorferi* in ixodid tick vectors. *Ann. New York Acad. Sci.* **539**, 172–179.

Camicas, J.-L., Wilson, M.L., Cornet, J.-P., Digoutte, J.-P., Calvo, M.-A., Adam, E., and Gonzales, J.-P. Ecology of ticks as potential vectors of Crimean–Congo hemorrhagic fever virus in Senegal: epidemiological implications. *Arch. Virol. Suppl.* **1**, 303–322.

Camin, J.H. (1963). Relations between host-finding behavior and life histories in ectoparasitic acarine. pp. 411–424. *In:* Naegle, J.A. (ed) *Advances in Acarology.* Comstock, Ithaca, NY.

Campbell, A. and MacKay, P.R. (1979) Distribution of the American dog tick, *Dermacentor variabilis* (Say), and its small-mammal hosts in relation to vegetation types in a study area in Nova Scotia. *Can. J. Zool.* **57**, 1950–1959.

Causey, O.R., Kemp, G.E., Madbouly, M.H., and David-West, T.S. (1970) Congo virus from domestic livestock, African hedgehogs, and arthropods in Nigeria. *Am. J. Trop. Med. Hyg.* **19**, 846–850.

Churchill, R.C. (1976) Babesiosis of a horse in Australia. (Letter). *Austral. Vet. J.* **53**, 487.

Clifford, C.M. (1979) *Arctic and Tropical Arboviruses.* Academic Press, New York.

Clifford, C.M. and Hoogstraal, H. (1965) The occurrence of *Ixodes arboricola* Schulze and Schlottke (Ixodoidea: Ixodidae) in Africa on northward migrating birds. *J. Med. Entomol.* **2**, 37–40.

Colbo, M.H. and MacLeod, J. (1976) Ecological studies of ixodid ticks (Acari: Ixodidae) in Zambia. II. Ticks found on small mammals and birds. *Bull. Entomol. Res.* **66**, 489–500.

Daniels, T.J. and Fish, D. (1990) Spatial distribution and dispersal of unfed larval *Ixodes dammini* (Acari: Ixodidae) in Southern New York. *Environ. Entomol.* **19**, 1029–1033.

Davidar, P., Wilson, M., and Ribiero, J.M. (1989) Differential distribution of immature *Ixodes dammini* (Acari: Ixodidae) on rodent hosts. *J. Parasitol.* **75**, 898–904.

Dawson, J.E., Anderson, B.E., Fishbein, D.B., Sanchez, J.L., Goldsmith, C.S., Wilson, K.H., and Duntley, C.W. (1991) Isolation and characterization of an *Ehrlichia* sp. from a patient diagnosed with human ehrlichiosis. *J. Clin. Microbiol.* **29**, 2741–2745.

Diamant, G. (1965) The Bont tick (*Amlyomma hebreum*). Exotic vector of heartwater found in the United States. *Vet. Med./Small An. Clin.* **60**, 847–850.

du Toit, J.T. (1990) Home range-body mass: a field study of African browsing ruminants. *Oecologia* **85**, 301–303.

Eads, R.B., Smith, G.C., and Maupin, G.O. (1982) A CO_2 platform trap for taking adult *Dermacentor andersoni* (Acari: Ixodidae). *Proc. Entomol. Soc. Wash.* **84**, 342–348.

Falco, R.C. (1987) Abundance of the deer tick, *Ixodes dammini* (Acari: Ixodidae), in a Lyme disease endemic area of southern New York State. 183 pp. (Dissertation). Fordham University, NY.

Falco, R.C. and Fish, D. (1988) Prevalence of *Ixodes dammini* near the homes of Lyme disease patients in Westchester County, New York. *Am. J. Epidemiol.* **127**, 826–830.

Filippova, N.A. (1990) The taxonomic aspects of the transmission of the causative agent of Lyme disease. *Parazitologiya* **24**, 257–267.

Filippova, N.A. (1991) A hypothesis for the palaeogenesis of the distribution of the main vectors for Lyme disease. Vol. 1, pp. 109–118. *In:* Dusbábek, F. and Bukva, V. (eds) *Modern Acarology.* Academia, Prague and SPB Academic, The Hague.

Fivaz, B.H. and Petney, T.N. (1989) Lyme disease—a new disease in southern Africa? *J. S. Afr. Vet. Assoc.* **60**, 155–158.

French, J.B., Jr, Schell, W.L., Kazmierczak, J.J., and Davis, J.P. (1992) Changes in the population density and distribution of *Ixodes dammini* (Acari: Ixodidae) in Wisconsin during the 1980's. *J. Med. Entomol.* **29**, 723–728.

Gaidanovich, S.Ya. (1972) Antibodies to Sumakh and Kemerovo viruses in migratory birds. *Mater. Simp. Itogi 6, Simpl Izuch. Virus. Ekol. Svyazan, Ptits., Omsk*, 138–141 (in Russian).

Gonzales, J.P., Camicas, J.L., Cornet, J.P., Faye, O., and Wilson, M.L. (1992) Sexual and transovarian transmission of Crimean–Congo haemorrhagic fever virus in *Hyalomma truncatum* ticks. *Res. Virol.* **143**, 23–28.

Gresikova, M. and Calisher, C.H. (1988) Tick-borne encephalitis. Vol. 4, pp. 177–202. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology.* CRC Press, Boca Raton, FL.

Gresikova, M. and Sekeyova, M. (1990) To the antigenic classification of some viruses from the tick-borne encephalitis complex by monoclonal antibodies. *Acta Virol. (Praha)* **34**, 90–94.

Guerrero, A., Fishbein, D.B., Mesa, E., and Escudero, R. (1991) Human infection by *Ehrlichia canis* in Spain? (Letter). *Med. Clin. (Barc.)* **96**, 236–237.

Gusev, V.M. (1962) Birds as a factor contributing to the expansion of the range limits of certain species of ectoparasites. *Zool. Zh.* **41**, 1061–1066.

Gusev, V.M., Guseva, A.A., Petrosian, E.A., and Eigelis, I.K. (1962) The role of birds in the spread of ticks and fleas. (According to material collected in Azerbaijan SSR). *Zool. Zh.* **41**, 905–912 (in Russian, English translation NAMRU3-T83).

Gusev, V.M., Guseva, A.A., and Resznik, P. (1963) Role of birds in the distribution of fleas (Suctoria) and ticks (Ixodoidea) in Daghestan. *Med. Parazitol. I Parazitarnye Bolezni.* **32**, 738–739.

Gwynne, M.D. and Bell, R.H.V. (1968) Selection of vegetation components by grazing ungulates in the Serengeti National Park. *Nature* **220**, 390–393.

Hallam, A. (1973) *Organisms and Continents Through Time.* Special Papers in Palaeontology No. 12. Palaeontological Association, London.

Halliday, R.B. and Sutherst, R.W. (1990) An Australian record of the American dog tick *Dermacentor variabilis*, and the risk of its establishment outside North America. *Exp. Appl. Acarol.* **8**, 65–70.

Heneberg, N., Heneberg, D., Milosevic, J., and Demitrijevic, V. (1967) Distribution of ticks in the autonomous provinces of Kosovo and Metohija with special regard to *Hyalomma plumbeum plumbeum* Panzer, reservoir and vector of Crimean hemorrhagic fever of man. *Zborn. Vojnomed. Akad. Belograd.* 30–36.

Herman, T.B. and Garvie, M.B. (1985) Mammal–tick interactions and rickettsial epidemiology in Nova Scotia. *Acta Zool. Fenn.* **173**, 201–203.

Hirst, L.F. (1953) *The Conquest of Plague.* Clarendon Press, Oxford.

Holzmann, H., Vorobyova, M.S., Ladyzhenskaya, I.P., Ferenczi, E., Kundi, M., Kunz, C., and Heinz, F.X. (1992) Molecular epidemiology of tick-borne encephalitis virus: cross-protection between European and Far Eastern subtypes. *Vaccine* **10**, 345–349.

Hoogstraal, H. (1961) Migrating birds and their ectoparasites in relation to disease. *E. Afr. Med. J.* **38**, 221–226.

Hoogstraal, H. (1972) Birds as tick hosts and as reservoirs and disseminators of tickborne infectious agents. *Wiad. Parazylt.* **18**, 703–706.

Hoogstraal, H. (1978) Ecology of the Tete serogroup. *Dokl. Simp. Transkont. Svyazi Pereletn. Ptits Rol'v Rasp. Arbovirus.* Novosibirsk, USSR pp. 212–213.

Hoogstraal, H. (1979) The epidemiology of tick-borne Crimean–Congo hemorrhagic fever in Asia, Europe, and Africa. *J. Med. Entomol.* **15**, 307–417.

Hoogstraal, H. (1985) Argasic and Nuttallielid ticks as parasites and vectors. *Adv. Parasitol.* **24**, 135–238.

Hoogstraal, H. and Aeschlimann, A. (1982) Tick host specificity. *Bull. Société Entomol. Suisse* **55**, 5–32.

Hoogstraal, H. and Kim, K.C. (1981) Tick and mammal coevolution, with emphasis on *Haemaphysalis*. pp. 508–568. *In:* Kim, K.C. (ed) *Coevolution.*

Hoogstraal, H., Kaiser, M.N., Traylor, M.A., Gaber, S., Guindy, E. (1961) Ticks (Ixodidae) on birds migrating from Africa to Europe and Asia. *Bull. Wld. Hlth. Org.* **24**, 197–212.

Hoogstraal, H., Kaiser, M.N., Traylor, M.A., Guindy, E., and Gaber, S. (1963). Ticks (Ixodidae) on birds migrating from Europe and Asia to Africa, 1959–61. *Bull. Wld. Hlth. Org* **28**, 235–262.

Hoogstraal, H.H., Traylor, M.A., Gaber, S., Malakatis, G., Guindy, E., and Helmy, I. (1964) Ticks (Ixodidae) on migrating birds in Egypt, spring and fall 1962. *Bull. Wld. Hlth. Org.* **30**, 355–367.

Hoogstraal, H., Kaiser, M.N., Seymour, C., and Gaber, S. (1967) Noteworthy recent tick records from Egypt. 1. *Ixodes arboricola* Schulze and Schlottke infesting resident, migrant and wintering birds in the western coastal desert. *J. Egypt. Pub. Health. Assoc.* **42**, 223–229.

Hoogstraal, H., Boo-Liat Lim, and Anastos, G. (1969) *Haemaphysalis bispinosa* Neumann (Ixodoidea: Ixodidae): Evidence for consideration as an introduced species in the Malay peninsula and Borneo. *J. Parasitol.* **55**, 1075–1077.

Hoogstraal, H., Wassef, H.Y., and Gallagher, M.D. (1985) *Haemaphysalis kutchensis*, an Indian–Pakistani bird and mammal tick, parasitizing a migrant whitethroat in the Sultanate of Oman. *J. Parasitol.* **71**, 129–130.

Iacono-Connors, L. and Schmaljohn, C.S. (1991) Cloning and sequence analysis of the genes encoding the non-structural proteins of Langat virus and comparative analysis with other flaviviruses. *Virology* **188**, 875–888.

Jones, E.N. (1990) Effects of forage availability on home range and population density of *Microtus pennsylvanicus*. *J. Mammal.* **71**, 382–389.

Kahl, K. (1971) Some observations on the occurrence of *Ixodes ricinus* (L) living on migratory birds. *Wiad. Parazyt.* **17**, 417–426.

Kaiser, M.N. and Hoogstraal, H. (1967) Noteworthy recent tick records from Egypt. II. Hosts, distribution and ecology of *Rhipicephalus simus* Koch. *J. Egypt Public Health Assoc.* **42**, 231–242.

Kaier, M.N. and Hoogstraal, H. (1974) Ticks (Ixodoidea) on migrating birds in Cyprus, fall 1967 and spring 1968, and epidemiological considerations. *Bull. Entomol. Res.* **64**, 97–110.

Kaiser, M.N., Hoogstraal, H., and Horner, K.O. (1970) The subgenus Persicargas (Ixodoidea, Argasidae, Argas). 10. A. (P.) streptopelia, new species, a widely distributed parasite of resident and migrating doves in Africa and Cyprus, and epidemiological considerations. *Ann. Entomol. Soc. Am.* **63**, 97–110.

Kaltenrieder, M. (1990) Scototaxis and target perception in the camel tick, *Hyalomma dromedarii*. *Exp. Appl. Acarol.* **9**, 267–278.

Karabatsos, N. (1985) *International Catalogue of Arboviruses. Am. Soc. Trop. Med. Hyg.* San Antonio, TX, 2147 pp.

Keirans, J.E., Oliver, J.H., Jr, and Needham, G.R. (1992) The *Ixodes ricinus/persulcatus* complex defined. (Abstract). *First International Conference on Tick-borne*

Pathogens. Munderloh, U.G. and Kurtti, T.J. (eds). 15–18 September 1992, Saint Paul, Minnesota..

Kharitonova, N.N., Leonov, Yu.A., Dokuchaeva, Yu.I., Barbash, L.A., Sipko, L.L., and Mikhant'ev, A.I. (1978) Ecology of Omsk hemorrhagic fever virus. *Dokl. Simp. Transkont. Svyazi Pereletn. Novosibirsk* 214–217.

Kitron, U., Jones, C.J., and Bouseman, J.K. (1991) Spatial and temporal dispersion of immature *Ixodes dammini* on *Peromyscus leucopus* in northwestern Illinois. *J. Parasitol.* **77**, 945–949.

Kondratenko, V.F. (1976) Importance of ixodid ticks in the transmission and preservation of the causative agent of the Crimean hemorrhagic fever in foci of the infection. *Parazitologiya (Leningrad)* **10**, 297–302 (in Russian, English translation NAMRU3-T1116).

Korch, G.W., Jr (1985) The ecology of the American dog tick, *Dermacentor variabilis* at a site with endemic spotted fever group rickettsia. Ph.D. dissertation, Johns Hopkins University.

Kraft, V.A. (1963) Effects of hydroclimatic factors on the development of epizootics and tularemia outbreaks in Tselinograd Oblast. *Zh. Mikro. Epidem. Immuno.* **5**, 41–48.

Kurbanov, M.M., Berezina, L.K., Zakaryan, V.A., Kiseleva, N.V., and Vatolin, V.P. (1974a) Results of serological investigation of human and domestic animal blood sera with 13 arboviruses in the Karakum canal zone and southeastern Turkmen SSR. *Sb. Tr. Ekol. Virus* **2**, 113–120 (In Russian, English translation NAMRU-3-T778).

Kurbanov, M.M., Gromashevsky, V.L., Berdyev, A., Skvortsova, T.M., Kiseleva, N.V., and Vatolin, V.P. (1974b) Isolation of arboviruses from ticks in the Karakum canal zone. *Sb. Tr. Ekol. Virus* **2**, 109–113 (In Russian, English translation NAMRU-3-T777).

Kurten, B. (1971) *The Age of Mammals*. Columbia University Press, New York.

Lane, R.S., Anderson, J.R., Yaninek, J.S., and Burgdorfer, W. (1985) Diurnal host seeking of adult Pacific Coast ticks, *Dermacentor occidentalis* (Acari: Ixodidae), in relation to vegetational type, meteorological factors, and rickettsial infection rates in California, USA. *J. Med. Entomol.* **22**, 558–571.

Lastavica, C.C., Wilson, M.L., Berardi, V.P., Spielman, A., and Deblinger, R.D. (1989) Rapid emergence of a focal epidemic of Lyme disease in coastal Massachusetts. *New Engl. J. Med.* **320**, 133–137.

Laurance, W.F. and Coan, M.E. (1987) Microhabitat and demographic correlates of tick parasitism in a Northern Great Basin small mammal community. *Am. Mid. Natural.* **118**, 1–9.

Lehman, T. (1993) Ectoparasites: direct impact on host fitness. *Parasitol. Today* **9**, 8–13.

Levine, J.F., Wilson, M.L., and Spielman, A. (1985) Mice as reservoirs of the Lyme disease spirochete. *Am. J. Trop. Med. Hyg.* **34**, 355–360.

Lewis, I.J. (1970) Observations on the dispersal of larvae of the cattle tick *Boophilus microplus* (Can.). *Bull. Entomol. Res.* **59**, 595–604.

Lidicker, W.Z., Jr (1985) An overview of dispersal in non-volant small mammals. *In:* Rankin, M.A. (ed) *Migration: Mechanisms and Adaptive Significance. Contrib. Marine Sci.* **68**, 368–385.

Linzey, A.V. (1989) Response of the white-footed mouse (*Peromyscus leucopus*) to the transition between disturbed and undisturbed habitats. *Can. J. Zool.* **67**, 505–512.

Logan, T.M., Linthicum, K.J., Bailey, C.L., Watts, D.M., Dohm, D.J., and Moulton, J.R.

(1990) Replication of Crimean–Congo hemorrhagic fever virus in four species of ixodid ticks (Acari) infected experimentally. *J. Med. Entomol.* **27**, 537–542.

Lutta, A.S. (1968) *Ixodes triangulicep*s and its distribution in Karelia. *Parazitologiya* **2**, 142–147.

L'vov, D.K. (1970) Significance of chronic and latent infections in the ecology of arboviruses. *Vestn. Akad. Med. Nauk. SSSR* **10**, 63–68.

L'vov, D.K., Karas, F.R., Timofeev, E.M., Tsyrkin, Yu.M., Vargina, S.G., Veselovskaya, O.V., Osipova, N.Z., Grebenyuk, Yu.I., Gromashevski, V.L., Steblyanko, O.N., and Fomina, K.B. (1973) Issyk-Kul virus, a new arbovirus isolated from bats and *Argas* (*Carios*) *vespertilionis* (*Latr.* 1802) *in the Kirghiz S.S.R. Arch. Ges. Virusforsch.* **42**, 207.

MacArthur, R.H. and Wilson, E.O. (1967) *The Theory of Island Biogeography.* Princeton University Press, New Jersey, 203 pp.

Makarova, V.A. and Tarasevich, I.V. (1989) *Questions of Rickettsiology.* Moscow, pp. 75–77.

Marchette, N.J. (1982) *Ecological Relationships and Evolution of the Rickettsiae.* CRC Press, Boca Raton, FL.

Marconi, R.T. and Garon, C.F. (1992) Identification of a third genomic group of *Borrelia burgdorferi* through signature nucleotide analysis and 16s rRNA sequence determination. *J. Gen. Microbiol.* **138**, 533–536.

Mather, T.N. and Spielman, A. (1986) Diurnal detachment of immature deer ticks (*Ixodes dammini*) from nocturnal hosts. *Am. J. Trop. Med. Hyg.* **35**, 182–186.

Matton, P. and van Melckebeke, H. (1989) Spread of heartwater along trek routes in Shaba (south east Zaire). *Vet. Rec.* **125**, 531.

Matuschka, F.R., Richter, D., and Spielman, A. (1991) Differential detachment from resting hosts of replete larval and nymphal *Ixodes* ticks. *J. Parasitol.* **77**, 341–345.

McEnroe, W.D. (1971) The effect of automobile traffic on American dog tick distribution (*Dermacentor variabilis*, Say: Acarina, Ixodidae). *Environ. Pollution* **2**, 135–143.

McCullough, D.R. (1985) Long range movement of large terrestrial mammals. *In:* Rankin, M.A. (ed) *Migration: Mechanisms and Adaptive Significance. Contrib. Marine Sci.* **27**, 444–465.

McKenna, M.C. (1983) Holarctic landmass rearrangement, cosmic events, and Cenozoic terrestrial organisms. *Ann. Missouri Bot. Gard.* **70**, 459–489.

McNab, B.K. (1963) Bioenergetics and the determination of home range size. *Am. Nat.* **97**, 133–140.

Milne, A. (1949) The ecology of the sheep tick, *Ixodes ricinus* I. Host relationships of the tick Part 2. Observations on hill and moorland grazing in northern England. *Parasitol.* **39**, 173–197.

Minshull, J.I. (1982) Drop-off rhythms of engorged *Rhipicephalus appendiculatus* (Acari: Ixodidae). *J. Parasitol.* **68**, 484–489.

Mohr, C.O. (1961) Relation of ectoparasite load to host size and standard range. *J. Parasitol.* **47**, 978–984.

Morrill, J.C., Soliman, A.K., Imam, I.Z., Botros, B.A., Moussa, M.I., and Watts, D.M. (1990) Serological evidence of Crimean–Congo haemorrhagic fever viral infection among camels imported into Egypt. *J. Trop. Med. Hg.* **93**, 201–204.

Need, J.T. and Escamilla, J. (1991) Lyme disease in South America? (Letter). *J. Inf. Dis.* **163**, 681–682.

Nilsson, A. and Lundqvist, L. (1978) Host selection and movements of *Ixodes ricinus* (Acari) larvae on small mammals. *Oikos* **31**, 313–322.

Nuoreteva, P. and Hoogstraal, H. (1963) The incidence of ticks (Ixodoidea: Ixodidae) on migratory birds arriving in Finland during the spring of 1962. *Ann. Med. Exp. Biol. Fenn.* **41**, 457–468.

Nosek, J. and Grulich, I. (1967) The relationship between the tick-borne encephalitis virus and the ticks and mammals of the Tribec Mountain Range. *Bull. Wildl. Hlth. Org.* **36** (Suppl. 1), 31–47.

Okaeme, A.N. (1986) Ectoparasites and gastrointestinal parasites of nomadic cattle infiltrating into Kainji Lake National Park Nigeria. *Int. J. Zoon.* **13**, 40–44.

Oliver, J.H., Jr (1974) Symposium on reproduction of arthropods of medical and veterinary importance. IV. Reproduction in ticks (Ixodoidea). *J. Med. Entomol.* **11**, 26–34.

Oliver, J.H., Jr (1989) Biology and systematics of ticks (Acari: Ixodidae). *Annu. Rev. Ecol. Syst.* **20**, 397–430.

Oliver, J.H., Jr, Owlsley, M.R., Hutcheson, H.J., James, A.M., Chunseng, C., Irby, W.S., Dotson, E.M., and McLain, D.K. (1993) Conspecificity of the ticks *Ixodes scapularis* and *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **30**, 54–63.

Paevskii, V.A. (1973) *Bird Migrations: Ecological and Physiological Factors.* John Wiley & Sons, New York.

Pak, T.P. and Mikhailova, L.I. (1973) *Crimean hemorrhagic fever in Tadzhikistan.* Izd. "Irfon." Dushanbe (in Russian, English translation NAMRU3-T1000), 154 pp.

Pavlovsky, E.N. (1966) *Natural Nidality of Transmissible Diseases.* University of Illinois Press, Urbana, IL.

Persing, D.H., Telford, S.R., III, Spielman, A., and Barthold, S.W. (1990) Detection of *Borrelia burgdorferi* infection in *Ixodes dammini* ticks with the polymerase chain reaction. *J. Clin. Microbiol.* **28**, 566–572.

Petney, T.N., Van Ark, H., and Spickett, A.M. (1991) On sampling tick populations: the problem of overdispersion. *Onderstepoort J. Vet. Res.* **57**, 123–127.

Pielou, E.C. (1979) *Biogeography.* John Wiley and Sons, New York.

Pomerantzev, B.I. (1948) Basic directions of evolution of Ixodoidea. *Parasit. Sborn. Zool. Inst. Acad. Sci. USSR*, **10**, 5–18 (in Russian, English translation NAMRU3-T13).

Purnell, R.E. (1980) Tick-borne disease as a barrier to efficient land use. *Outlook on Agriculture*, **10**, 230–234.

Rajagopalan, P.K., Paul, S.D., and Sreenivasan, M.A. (1969) Isolation of Kyasanur Forest disease virus from the insectivorous bat *Rhinolophus rouxi* and from *Ornithodoros* ticks. *Ind. J. Med. Res.* **57**, 805–808.

Randolph, S.E. (1979) Population regulation in ticks: the role of acquired resistance in natural and unnatural hosts. *Parasitol.* **79**, 141–156.

Rechav, Y. (1970) Effect of age and sex of gerbils on the attraction of immature *Hyalomma excavatum* Koch, 1844 (Ixodoidea: Ixodidae). *J. Parasitol.* **56**, 611–614.

Rechav, Y. (1979) Drop-off rhythms of engorged larvae and nymphs of the bont tick, *Amblyomma hebraeum* (Acari: Ixodidae), and the factors that regulate them. *J. Med. Entomol.* **14**, 667–687.

Rechav, Y., Zeederberg, M.E., and Zeller, D.A. (1987) Dynamics of African tick (Acari: Ixodoidea) populations in a natural Crimean–Congo hermorrhagic fever focus. *J. Med. Entomol.* **24**, 575–583.

Regnery, R.L., Spruill, C.L., and Plikaytis, B.D. (1991) Genotypic identification of rickettsiae and estimation of intraspecies sequence divergence for portions of two rickettsial genes. *J. Bacteriol.* **173**, 1576–1589.

Romer, A.S. (1966) *Vertebrate Paleontology.* University of Chicago Press, Chicago, IL.

Rosa, P.A., Hogan, D., and Schwan, T.G. (1991) Polymerase chain reaction analyses identify two distinct classes of *Borrelia burgdorferi. J. Clin. Microbiol.* **29**, 524–532.

Rothschild, B.M., Turner, K.R., and DeLuca, M.A. (1988) Symmetrical erosive peripheral polyarthritis in the Late Archaic Period of Alabama. *Science* **241**, 1498–1501.

Samoilova, T.I. and Gembitsky, A.S. (1974) Participation of certain song birds in transporting arboviruses along the west coast route (Kurshkaya sand bar, the Baltic Sea). *In:* Votyyakov, V.I. and L'vov, D.K. (eds) *Ecology of Arboviruses Associated with Birds* (in Russian, English translation NAMRU3-T1222).

Sandell, M., Agrell, J., Erlinge, S., and Nelson, J. (1990) Natal dispersal in relation to population density and sex ratio in the field vole, *Microtus agrestis. Oecologia* **83**, 145–149.

Schein, E. (1988) Equine babesiois. *In:* Ristic, M. (ed) *Babesiosis of Domestic Animals and Man.* CRC Press, Boca Raton, FL.

Shepherd, A.J., Swanepoel, R., Cornel, A.J., and Mathee, O. (1989) Experimental studies on the replication and transmission of Crimean–Congo hemorrhagic fever virus in some African tick species. *Am. J. Trop. Med. Hyg.* **40**, 326–331.

Sinclair, A.R.E. (1983) The function of distance movements in vertebrates. pp. 240–258. *In:* Swingland, I.R. and Greenwood, P.J. (eds) *The Ecology of Animal Movements.* Clarendon Press, Oxford.

Smirnova, S.E., Shanmugam, D.Z.H., Nepesova, N.M., Filipenko, P.I., Mamaev, V.I., and Chumakov, M.P. (1974) Isolation of Crimean hermorrhagic fever virus from *Hyalomma asiaticum* ticks collected in the Turkmenian SSR. *Med. Virol. Moskva* **1**, 176–179 (in Russian, English translation NAMRU3-T940).

Smirnova, S.E., Mamaev, V.I., Nepesova, N.M., Filipenko, P.I., Kalyaeva, V., and Chumakov, M.P. (1978) Study of circulation of Crimean hemorrhagic fever virus in Turkmenian SSR. *Zh. Mikrobiol. Moskva* **55**, 92–97 (in Russian, English translation NAMRU3-T1296).

Smittle, B.J., Hill, S.O., and Philips, F.M. (1967) Migration and dispersal patterns of Fe59-labeled lone star ticks. *J. Econ. Entomol.* **60**, 1029–1031.

Sonenshine, D.E. (1973) Ecology of the American dog tick, *Dermacentor variabilis*, in a study area in Virginia. 3. Studies on dispersal of immature ticks by small-mammal hosts. *Ann. Entomol. Soc. Am.* **66**, 44–46.

Sonenshine, D.E. (1975) Influence of host–parasite interactions on the population dynamics of ticks. *Misc. Publ. Entomol. Soc. Am.* **9**, 243–249.

Sonenshine, D.E. and Clifford, C.M, (1973) Contrasting incidence of Rocky Mountain spotted fever in ticks infesting wild birds in eastern U.S. Piedmont and coastal areas, with notes on the ecology of these ticks. *J. Med. Entomol.* **10**, 497–502.

Sonenshine, D.E. and Stout, J. (1968b) Tick burden in relation to spacing and range of hosts in *Dermacentor variabilis. J. Med. Ent.* **5**, 49–52.

Sonenshine, D.E. and Stout, J. (1968a) Use of old-field habitats by the American dog tick, *Dermacentor variabilis. Ann. Entomol. Soc. Am.* **61**, 679–686.

Sonenshine, D.E. and Ziv, M. (1971) Ecological studies on ticks infesting sheep and small mammals in an unimproved semi-desert pasture in Israel. *J. Med. Entomol.* **8**, 132–144.

Sonenshine, D.E., Atwood, E.L., and Lamb, J.T., Jr (1966) The ecology of ticks transmitting Rocky Mountain spotted fever in a study area in Virginia. *Ann. Entomol. Soc. Am.* **59**, 1234–1262.

Spielman, A. and Rossignol, P.A. (1984) Insect vectors. pp. 167–183. *In:* Warren, K.S. and Mahmoud, A.A.F. (eds) *Tropical and Geographic Medicine.* McGraw-Hill, New York.

Spielman, A., Wilson, M.L., Levine, J.F., and Piesman, J. (1985) Ecology of *Ixodes dammini*-borne human babesiosis and Lyme disease. *Annu. Rev. Entomol.* **30**, 439–460.

Stafford, K.C., III (1992) Oviposition and larval dispersal of *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **29**, 129–132.

Steere, A.C., Malawista, S.E., Snydman, D.R., Shope, R.E., Andiman, W.A., Ross, M.R., and Steele, F.M. (1977) Lyme arthritis: an epidemic of oligoarticular arthritis in children and adults in three Connecticut communities. *Arthritis Rheum* **20**, 7–17.

Swanepoel, R., Shepard, A.J., Leman, P.A., Shepard, S.P., and Miller, G.B. (1985) A common-source outbreak of Crimean–Congo haemorrhagic fever on a dairy farm. *S. Afr. Med. J.* **68**, 635–637.

Swihart, R.K., Slade, N.A., and Bergstrom, B.J. (1988) Relating body size to the rate of home use in mammals. *Ecology* **69**, 393–399.

Telford, S.R., III and Spielman, A. (1989) Competence of a rabbit-feeding *Ixodes* (Acari: Ixodidae) as a vector of the Lyme disease spirochete. *J. Med. Entomol.* **26**, 118–121.

Telford, S.R., III, Mather, T.N., Adler, G.H., and Spielman, A. (1990) Short-tailed shrews as reservoirs of the agents of Lyme disease and human babesiosis. *J. Parasitol.* **76**, 681–683.

Telford, S.R., III, Urioste, S.S., and Spielman, A. (1992) Clustering of host-seeking nymphal deer ticks (*Ixodes dammini*) infected by Lyme disease spirochetes (*Borrelia burgdorferi*) *Am. J. Trop. Med. Hyg* **47**, 55–60.

Terpestra, C. (1990) Nairobi sheep disease virus. pp. 495–500. *In:* Dinter, Z. and Morein, B. (eds) *Virus Infections of Ruminants.* Elsevier Science Publishers, Amsterdam.

Thompson, G.B. (1964). Ticks (Ixodoidea) of medical and veterinary interest from birds in the British Isles. *J. Med. Entomol.* **1**, 45–49.

Traub, R. and Jellison, W.L. (1981) *In:* Burgdorfer, W. and Anacker, R.L. (eds) *Rickettsiae and Rickettsial Diseases.* Academic Press, New York.

Turner, B.N., Iverson, S.L., and Severson, K.L. (1983) Seasonal changes in open-field behavior in wild male meadow voles (*Microtus pennsylvanicus*). *Behav. Neurol. Biol.* **39**, 60–77.

Uspensky, I. and Rubina, M. (1992) Host substitution by *Ixodes persulcatus* (Acari: Ixodidae) larvae in the years of deep depression in the abundance of small mammals. *Folia Parasitol. (Praha)* **39**, 171–176.

Venugopal, K., Buckley, A., Reid, H.W., and Gould, E.A. (1992) Nucleotide sequence of the envelope blycoprotin of Negishi virus shows very close homology to louping ill virus. *Virology* **190**, 515–521.

Vesey-Fitzgerald, D.F. (1960) Grazing succession among East African game animals. *J. Mammal.* **41**, 161–172.

Voinov, I.N. (1978) Study of arbovirus infections in Belorussia and other western regions of the USSR. *Akad. Med. Nauk SSSR* **3**, 20–24 (in Russian, English translation NAMRU3-T1361).

Waladde, S.M. and Rice, M.J. (1982) The sensory basis of tick feeding behavior. pp. 71–118. *In:* Oberchain, F.D. and Galun, R. (eds) *Physiology of Ticks.* Pergamon Press, Oxford.

Walker, D.H., Liu, Q.H., Yu, X.J., Li, H., Taylor, C., and Feng, H.M. (1992) Antigenic diversity of *Rickettsia conorii*. *Am. J. Trop. Med. Hyg.* **47**, 78–86.

Wang, J.G., Walker, D.D., Li, H., Lenz, B., and Jerrells, T.R. (1987) Barbash strain spotted fever group rickettsia is a strain of *Rickettsia conorii* and differs from *Rickettsia siberica*. *Acta Virol. (Praha)* **31**, 489–498.

Watson, G.E. (1971) A serological and ectoparasite survey of migratory birds in northeast Africa. Smithsonian Institute Final Report, DoD Contract DA-HC-19-67-C-0008.

Watson, G.E., Shope, R.E., and Kaiser, M.N. (1972) An ectoparasite and virus survey of migratory birds in the eastern Mediterranean. pp. 176–180. *Transcontinental Connections of Migratory Birds and Their Role in the Distribution of Arboviruses.* Nauka Publishers, Novosibirsk.

Watts, D.M., Ksiazek, T.G., Linthicum, K.J. and Hoogstraal, H. (1989) Crimean–Congo hemorrhagic fever. pp. 177–222. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology.* CRC Press, Boca Raton, FL.

Weisbrod, A.R. and Johnson, R.C. (1989) Lyme disease and migrating birds in the Saint Croix River Valley. *Appl. Environ. Microbiol.* **55**, 1921–1924.

Wilkinson, P.R. (1957) The spelling of pasture in cattle tick control. *Austral. J. Agric. Res.* **8**, 414–423.

Wilkinson, P.J. (1988) African swine fever virus. pp. 17–35. *In:* Pensaert, M.B. (ed) *Virus Infections of Porcines.* Elsevier, Amsterdam.

Wilson, J.G., Kinzer, D.R., Sauer, J.R., and Hair, J.A. (1972) Chem-attraction in the lone star tick (Acarina: Ixodidae) I. Response of different developmental stages to carbon dioxide administered via traps. *J. Med. Entomol.* **9**, 245–252.

Wilson, M.L., LeGuenno, B., Guillaud, M., Desoutter, D., Gonzalez, J.P., and Camicas, J.L. (1990a) Distribution of Crimean–Congo hermorrhagic fever viral antibody in Senegal: environmental and vectorial correlates. *Am. J. Trop. Med. Hyg.* **43**, 557–566.

Wilson, M.L., Gonzales, J.-P., LeGuenno, B., Cornet, J.-P., Guillaud, M., Calvo, M.-A., Digoutte, J.-P., and Camicas, J.-L. (1990b) Epidemiology of Crimean–Congo hemorrhagic fever in Senegal: temporal and spatial patterns. *Arch. Virol. Suppl.* **1**, 323–340.

Wilson, M.L., Gonzales, J.P., Cornet, J.-P., and Camicas, J.-L. (1991) Transmission of Crimean Congo haemorrhagic fever virus from experimentally infected sheep to *Hyalomma truncatum* ticks. *Res. Virol.* **142**, 395–404.

Work, T.H. (1958) Russian spring–summer virus in India. Kyasanur Forest Disease. *Progr. Med. Virol.* **1**, 248–277.

Zimmerman, R.H., McWherter, G.R., and Bloemer, S.R. (1987) Role of small mammals in population dynamics and dissemination of *Amblyomma americanum* and *Dermacentor variabilis* (Acari: Ixodidae at Land between the Lakes, Tennessee. *J. Med. Entomol.* **24**, 370–375.

7

Modeling the Ecological Dynamics of Tick-borne Zoonoses

URIEL KITRON AND ALESSANDRO MANNELLI

1. INTRODUCTION

1.1. What is a Model?

A model is a reconstruction of nature for the purpose of research or applications. Models serve as the basic units of theoretical investigations and provide the necessary link between theory and measurement. Successful models are applicable to the purpose of the study for which they were originally constructed but can not be assumed to be relevant to other purposes (Levins, 1968; King and Soskolne, 1988).

A model is a simplified representation of reality. A functional model includes the essential aspects of reality and a set of assumptions necessary to optimize such a representation (Levins, 1968). The transition between reality and its model include the quantitative representation of a conceptual qualitative picture and the interpretation of a theoretical representation in real life context (King and Soskolne, 1988).

An ideal model would maximize generality, realism, and precision. A model which maximizes precision attempts to describe in fine detail parts of a specific biological system. A general model, in contrast, attempts to describe the essential features of a biological scenario which may be applicable to some extent to a large number of specific systems. A model which maximizes realism is concerned with the correct representation of a segment of reality. It is impossible to maximize generality, realism, and precision simultaneously. Consequently, a single best-purpose model does not exist (Levins, 1968).

Models can be used for data collection and information management, for research, for teaching and demonstration, and for control and management of vector species and disease. The choice of an appropriate model depends on the purpose of the investigation, the data available for the construction of the model, and the software and hardware necessary to formulate and run the model. Naturally, different models are appropriate for teaching, demonstration, research, prediction, and management goals. The use of a model for

a different purpose than the one for which it was constructed may provide erroneous or misleading results.

1.2. Model Types

Many types and classifications of models exist. In this chapter we follow the classification suggested by King and Soskolne (1988) who divided epidemiological models into process models and associative models.

Process models, which include descriptive and simulation models, try to represent the mechanisms underlying a phenomenon and to explore a process in depth. They reproduce an event by means of an understanding of the process that leads to the event and through a reconstruction of the system within which the process takes place. Process models can be used to explore a particular part of the system (i.e., tick life cycle or part of it) or to construct comprehensive models which incorporate tick life cycle and disease transmission. Simulation-process models are discussed in detail in Section 3 and provide most of the examples of models of tick-borne diseases.

Associative models examine the association of independent variables, mainly of environmental and demographic nature, with ecological and epidemiological parameters, and are discussed here as statistical models. We further discuss separately expert decision systems and geographic information systems, remote sensing and spatial analysis, which may be grouped together as computerized tools to gather, store, analyze, and represent information on tick-borne zoonoses, and assist in surveillance and management decisions. Because only relatively few models of tick-borne zoonoses have been published, we also discuss models of tick population dynamics and of non-zoonotic tick-borne diseases. This allows us to present additional issues that need to be considered when modeling tick-borne zoonoses.

1.3. Model Variables

The selection of the parameters driving the simulation of a process model, and the choice of the independent variables for a multivariate statistical analysis, is a critical aspect of modeling. The epidemiology and control of vector-borne diseases depend on a variety of environmental factors (biotic and abiotic) as well as parameters related to the human population (demographic, socio-economic, etc.), animal host population(s), and the availability and capabilities of existing surveillance and health-care delivery systems. Proper assessment of the impact of local conditions and changes in these conditions, requires an understanding of the restrictions operating on vectors, hosts, and transmission systems, and consideration of them in the framework of the landscape ecology and epidemiology of the various diseases.

Of course, it is impossible to collect accurate data on all these variables, and even if such data were available, it would be impossible to incorporate all

the data into a single model. It is necessary to decide on the objective of the proposed model, and to identify parameters which are needed to construct and run the model (Sutherst et al., 1978; Floyd, 1987; Maywald, 1987; Sutherst, 1987). For tick-borne zoonoses, various processes have to be identified and described separately before being incorporated into a single model (possibly with several subroutines). These processes include segments of the tick life cycle (developmental phase, host-finding phase, and parasitic phase), transmission of the infection agent and population dynamics of the host(s).

The transmission and maintenance of a disease within a vector–host–parasite system is regulated by the relationships among the components of the system itself, that Young (1986) defined as the host/tick interface, the parasite/host interface, the tick/host interface, and the parasite/host interface. The four aspects of the epidemiology of tick-borne diseases are of course closely related to each other and it is impossible to treat one aspect without involving the others. In the construction of models for tick-borne diseases, these concepts have been approached in different ways according to the characteristics of the particular disease to be modeled and the objectives of the model itself.

Most models for vector-borne diseases have been directed to the transmission of malaria. For mosquitoes and other flying insects, survival time is of the same order of magnitude as the prepatent period in hosts. Thus, a delicate balance exists between vector life span and disease maintenance. Ticks are relatively long lived with a life span of months or even years, and have the ability to transmit infectious agents transovarially and transstadially. Hence, modeling tick life cycles and tick-borne diseases involves a larger number of variables. Moreover, because ticks often have a complex life cycle with several free-living and parasitic stages, and utilize a large variety of hosts, models of tick-borne zoonoses are often limited to certain aspects of the tick and host life cycles and the tick–host–pathogen interactions. An exhaustive review of the epidemiology of tick-borne Crimean–Congo hemorrhagic fever (CCHF) by Hoogstraal (1979) demonstrates the complexity of the transmission system of tick-borne zoonoses.

To prioritize the modeling of a given process, it is necessary to determine the availability of required data and the feasibility of collecting unavailable data, the sensitivity of modeling the process, and the relevance of the process given the objectives of the model.

2. PROCESS MODELS

2.1. Descriptive Models

Descriptive models consist of diagrams and flow charts that represent real-world systems and help visualize complex processes such as tick life cycles or disease transmission. Descriptive models conceptualize relationships within a system, but the relationships among variables are not quantified and

such models do not allow for simulation of a process over time. They can serve as a conceptual framework for the development of simulation models.

Since most descriptive models deal with tick population dynamics, rather than with tick-borne diseases and since they are not considered models by all researchers, only a few examples are mentioned below.

Rechav (1979) and Rechav et al. (1987) developed descriptive models of African ticks. For *Amblyomma hebraeum*, *Rhipicephalus appendiculatus*, and *R. evertsi*, Rechav describes their horizontal and vertical movement, and discusses the relationship of their dispersal patterns to ecological factors and sampling methods. In a natural CCHF focus, Rechav et al. (1987) describe the seasonal activity and host preference of *Hyalomma marginatum*, *H. truncatum*, and *R. evertsi* and relate that information to the epidemiology of CCHF.

Randolph (1975) relates data obtained in the laboratory on the life cycle of *Ixodes trianguliceps* to seasonal feeding activity patterns observed in the field. Her model demonstrates the plasticity of the life cycle of this tick. Through their ability to delay development for periods of up to 9 months, engorgement can occur throughout much of the year, and a large number of ticks can emerge simultaneously.

The European sheep tick (*Ixodes ricinus*) may be the most extensively studied tick species. Lees and Milne (1951) use diagrams and charts to describe the seasonal and diurnal activity pattern of this tick and its dispersal behavior. Gray (1992, 1981, 1982) and Gardiner et al. (1981) study and describe in detail the dynamics of the life cycle of *I. ricinus*. Semter et al. (1971) and Semter and Hair (1973) describe the ecology and behavior of *Amblyomma americanum*. They associate tick abundance, distribution, and daily and seasonal activity patterns with vegetative habitat type. Norvall et al. (1984) provide a descriptive model of aspects of anaplasmosis. They map the incidence of antibodies to *Anaplasma marginale* in Zimbabwe in 1–3-year-old cattle and relate that distribution to the distribution of tick infestations.

Yuval and Spielman (1990) presented a descriptive model of the important processes underlying the life cycle of *Ixodes dammini* (Fig. 7.1), the vector of Lyme disease, on the East Coast of the US. A large amount of information, based on experimental and theoretical data, is synthesized in a diagram which provides insights into the tick's life cycle. The emphasis is on the timing and duration of the principal phases of the life cycle. Host abundance is proposed as a factor which can determine different pathways that cohorts of ticks may follow in the process of development from eggs to adults and the length of the life cycle. Mechanisms underlying pathogen transmission are explored in the same paper. In conditions of high host abundance, a high proportion of larvae feed in fall and contract *Borrelia burgdorferi* from infected hosts. This can result in a high density of infected nymphs the following spring, which are able to pass the infection to young mice which will, in turn, feed new larvae. This descriptive model provided the conceptual framework for a matrix population model by Sandberg et al. (1992).

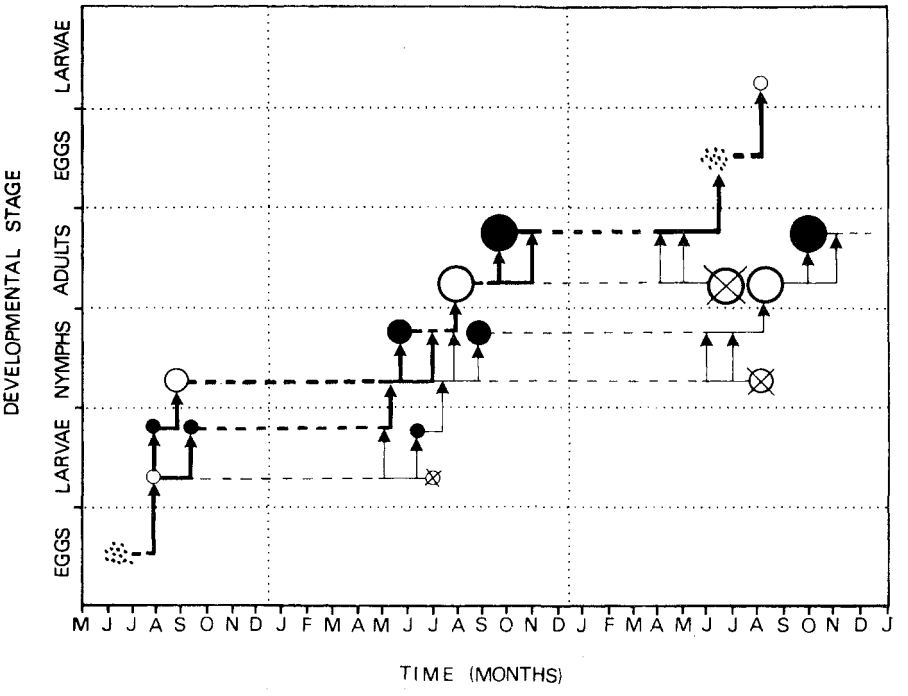


Fig. 7.1. Descriptive model for *Ixodes dammini* population (reproduced by permission of the Entomological Society of America from Yuval and Spielman, 1990). The length of the developmental cycle of cohorts and ticks varies when hosts are abundant (pathways emphasized in bold) and when hosts are scarce (non-emphasized pathways). ○ = unfed; ● = blood-fed; × = dead; —— = active; – – – = inactive.

2.2. Simulation Models

The main subject of this chapter is simulation process models of tick population dynamics and tick-borne diseases, where dynamic relationships among variables are quantified and entered in complex simulations for the purpose of forecasting epidemiological variables. Simulation models represent a dynamic system and incorporate changes over time.

Analytical models, consisting of equations describing the pattern of growth of a population or the basic reproduction rate, have been extensively employed in applied ecology (Conway, 1977; Begon and Mortimer, 1986); the solutions of the equations provide all the information about the system. In simulation models the equations are not necessarily solvable analytically. Rather, the system under study is simulated repeatedly on the computer using different values of variables of interest.

In the simulation process model for tick-borne diseases the system is usually represented by a discrete number of possible states. For ticks these may include life stage, age, and feeding status. Host populations are divided into susceptible,

infected (latent or infectious) and immune (Abbey, 1952; Baily, 1975; Anderson, 1984). The complexity of such systems requires the application of appropriate mathematical techniques such as Leslie matrix models (Leslie, 1945; Caswell, 1989) and dynamic life tables (Haile and Mount, 1987).

2.2.1. The Construction of a Simulation Model

In the construction and development of a simulation model, five principal phases can be recognized (Martin et al., 1987): model formulation, verification, validation, sensitivity analysis, and experimentation.

In the **formulation** of the model, the objectives of the model are stated, and the major features and relationships of the system are synthesized into a logical structure that may be implemented on a computer. The components of the system to be modeled are: **basic elements** (individuals of tick and host populations) which are divided into categories based on attributes that define the **state variable**. Transitions of the basic elements from one state to another (e.g., molting from larvae to nymphs in a tick population model or passing of hosts from the status of susceptible to infected in a disease model), occur according to **rates** (or **parameters**). Values of parameters are a function of complex relationships and interactions among factors (**driving variables**) that are intrinsic to the biology of the elements (intrinsic growth rate, host affinity, etc.) or external (environmental factors, biotic, and abiotic). Individuals move from one state to another at a frequency defined by a **time step**. The time step determines the frequency in which values of the state variable are recalculated (monitored), and thus define the temporal resolution power of the model.

The number of elements that are in each state, at a point in time, define the **structure** of the system. The transition of elements among successive states is the **dynamics** of the system.

In a simple tick population model (Fig. 7.2), individuals are divided on the basis of state (life stage, eggs, larvae, nymphs, and adults). Values for parameters such as fecundity of adult females, hatching rates, and survival, are needed in order to perform a simulation. In a more complex model the state variable is multi-dimensional, so that ticks are classified also according to feeding status, on–off host status, etc. Other parameters such as host finding and feeding success rates are also included. In a vector-borne disease system, important parameters are (among others) vectorial capacity, infectivity rate (of both host and vector), morbidity rate, and immunity development and loss.

The values of driving variables have to be assessed, and modeling the relationships between driving variables and parameters is a key aspect in modeling biological systems. Modelers adopted different approaches and methods (employment of complex mathematical relationships, dependence on theoretical assumptions or available empirical data, emphasis on accuracy, reliability or generalization) based on the purposes of the model. In some models, few (or even only one) aspects of the system are considered and values

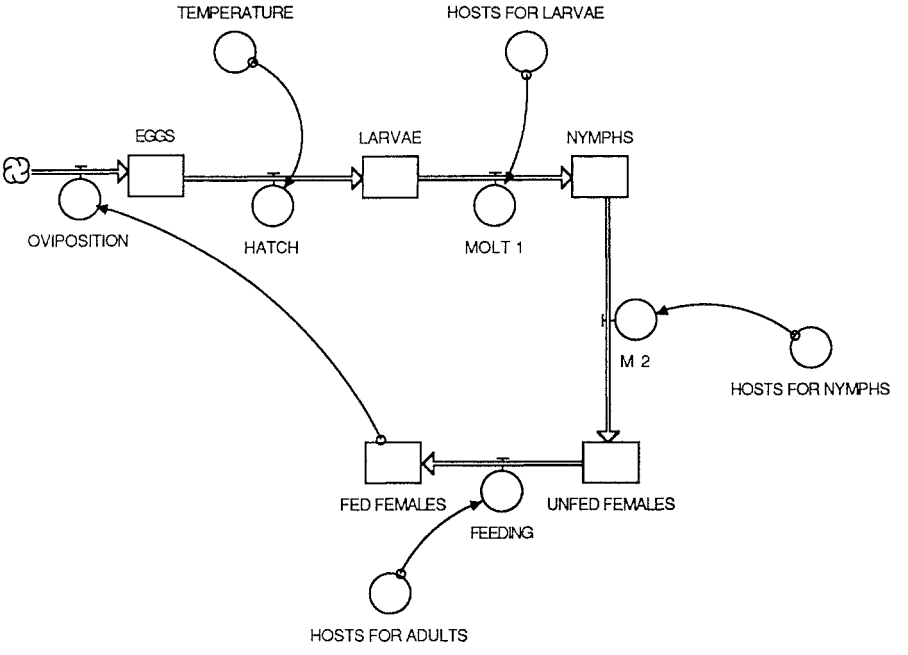


Fig. 7.2. Diagram of a simplified simulation model for a tick life cycle (Stella II, 1990). Rectangular boxes are connected by flow symbols. Transitions of individuals through successive stages occur according to rates which are functions of driving variables. Temperature and densities of hosts are driving variables represented in the diagram. Mortality is not represented. Determination of values for the rates of transition, time step, and initial population structure allow simulation of tick population dynamics.

for some parameters can be entered without modeling the relationships with independent variables. In other cases, a more comprehensive representation of reality is attempted, and particular attention is dedicated to those relationships. In the construction of a simulation model, driving variables are the object of two procedural phases: (1) data collection (determination of the information needed, availability of data etc.); (2) modeling the relationships with the simulation parameters (including the assessment and quantification of the relationships, based on theoretical assumptions, experimental results, or field observations). The validity of the model is mostly based on these phases.

In the phase of **model verification** the general behavior of the model is verified following model implementation. Adequacy of variables and relationships formulated in the previous stage, expressed in equations and functions, is tested. In the case of population models, pattern of growth, as predicted by the model (exponential growth, achievement of population equilibrium) is verified.

Model validation consists of a comparison between simulated results and the observed data. Accuracy of the model is then tested. However,

the availability of experimental and historical data is often a limiting resource.

Sensitivity analysis is the process of running (simulating) the model over a range of values assigned to one or more independent variables. Resulting outputs are then evaluated in order to determine their sensitivity to changes in independent variables. The principal objectives of sensitivity analysis are: to determine the degree of accuracy that is necessary in the evaluation of variables (if changes in a variable result in a great effect on outputs of interest, major research effort has to be made in order to obtain a precise evaluation of the variable); and to determine which phase of the system is more sensitive to control interventions and management strategies.

In model **experimentation** the final application of the model consists of the simulation and comparison of different scenarios, such as management strategies. This can be accomplished through the use of submodels for available management strategies, and combining them in order to obtain maximum efficiency with the minimum management effort.

3. SIMULATION PROCESS MODELS OF TICK POPULATION DYNAMICS

3.1. Matrix Population Models

Matrix population models (Leslie, 1945; Caswell, 1989) integrate population dynamics and population structure and are particularly useful when the life-cycle is most appropriately described in terms of discrete developmental stages. The number of organisms in each defined state or stage of development (structure of the population) is entered in an array of numbers known as the **state vector**. The population structure undergoes a change in time according to parameters which constitute the elements of the **transition matrix**. Multiplication of the state vector at time $t-1$ by the transition matrix gives a projection of the population structure at time t. The simplest application of matrix models is for age-classified organisms, with constant birth and survival rates. Reproductive rates (B_i) are entered in the first row of the matrix, while survival rates (S_i) are entered in the subdiagonal

$$\begin{pmatrix} x_1 \\ x_2 \\ x_3 \\ x_4 \end{pmatrix} (t-1) \; X \begin{pmatrix} B_1 & B_2 & B_3 & B_4 \\ S_1 & 0 & 0 & 0 \\ 0 & S_2 & 0 & 0 \\ 0 & 0 & S_3 & 0 \end{pmatrix} = \begin{pmatrix} x_1 \\ x_2 \\ x_3 \\ x_4 \end{pmatrix} (t) \tag{7.1}$$

The numbers of organisms at time t depend on the numbers of organisms at time $t-1$, where A is the matrix of transition.

$$\mathbf{x}(t) = A \; \mathbf{x} \; (t-1) \tag{7.2}$$

Population dynamic can be simulated and stable age distribution can be reached. The rate of increase per unit of time (R) can be calculated at stable age distribution. R is a characteristic of the transition matrix.

3.1.1. Ixodes dammini

Sandberg et al. (1992) applied a multi-matrix model to the life cycle of the deer tick, *Ixodes dammini*, the vector of Lyme disease in the eastern half of the US. The Leslie single matrix methodology (Leslie, 1945) is modified to represent the non-simultaneous events that characterize the life cycle of *I. dammini* (Yuval and Spielman, 1990). The multi-dimensional state variable is defined by life stage, age, and feeding status. The structure of the population is represented by a vector, and the transitions from one state to another occur according to a chain of 12 transition matrices, one for each month. The time step is therefore fixed in a month. The fecundity of the ticks and the parameters of transition such as feeding, molting to the next stage, hatching to larvae for eggs, and death were obtained from field observations and are modeled according to random tick–host encounters. The multi-matrix modeling method allows ticks of different life stage, and within the same life stage, to undergo those transitions at different times of the year. Year to year variations are not modeled because the structure of each monthly matrix is maintained constant.

The outputs of the simulation are forecasts of seasonal abundance and annual rate of increase of the tick population. The absence of density-dependent constraints yields an exponential population growth. The annual reproductive rate is 0.1155, corresponding to a doubling time of 6 years. The structure of the population stabilizes after 35 years. The seasonal feeding activity of the different tick life stages that is obtained by the simulation, is realistic and demonstrates an internal consistency of the model.

Awerbuch et al. (1992) modified the model by Sandberg et al. (1992) to include year to year variations in the transition parameters. This is obtained by the recalculation of feeding parameters with different values of host abundance. In fact, the authors' objective is to determine the effect in variation in the abundance of white-footed mice (*Peromyscus leucopus*), the principal host for immature stages of *I. dammini* in the northeastern US. Changes in mouse abundance are then simulated based on data collected at Nantucket Island (Massachusetts). The authors conclude that the density of mice critically affects the tick population. Such a tick population may fail to perpetuate unless the abundance of mice rises above a certain threshold.

Kiszewski et al. (1992) hypothesized density-dependent constraints to *I. dammini* population growth based on hyperfututorism (copulation in excess of fertilizing capacity). High tick density increases the probability of mating. Hyperfututoristic males become agametic after two or three effective matings, and then remove or deactivate the sperm previously deposited in female seminal receptacles. The modeling method employed to explore this hypothesis

is based on cellular automata (Wolfram, 1984). Off-host space is represented by a matrix of 1,000 cells where adult ticks, categorized by sex and reproductive status, move through a probabilistic walk and interact with ticks in neighboring cells. Four tick densities (mean number of ticks per cell) are simulated. At low densities, female fertility rises slowly but steadily, but at high densities, when mate finding is facilitated, males quickly exceed their capacity for effective fertilization. Fertility reaches an early peak and then declines rapidly. The model provides a theoretical insight into the process and suggests field studies needed to confirm the hypothesis.

3.1.2. Matrix Tick Population Model for the Analysis of Tick Management Strategies

A matrix population model for the cattle tick *Boophilus microplus* was employed by Sutherst et al. (1979a) as a framework for an analysis of tick-control strategies. The application of acaricide (dipping), pasture spelling, the introduction of resistant cattle breeds, and combinations of these methods, are evaluated in terms of their relative contribution to profit.

In the model, the structure of the tick population is represented by a 12-element vector. Eggs, larvae in pasture, parasitic ticks and adult females on pasture are divided into weekly age classes, resulting in a 1-week time step. The transition matrix includes transition parameters such as egg hatching and survival rates, survival and host-finding rates for larvae, and fecundity rate of adult females. Moreover, a density-dependent mortality of parasitic ticks is included. Density dependence is based on intraspecific competition and its effect was stronger in Zebu-cross cattle than in European breeds (Sutherst et al., 1973, 1979b). The transition parameters are varied for four tick generations to allow for seasonal differences in SE Queensland, Australia.

To test the realism of the model, simulated and observed tick populations on dipped cattle were compared, and the agreement was satisfactory. The simulation of the control methods, in isolation and in combination, results in reductions of the numbers of adult female ticks on cattle and of the losses caused by ticks. The costs and the overall merits of the control strategies are compared. The adoption of resistant cattle with a single dipping or spelling period appears as the best long-term choice.

The model incorporates ecological and economic components and represents a valuable tool in exploring the complex tick life cycle and control strategies. Sensitivity analysis is performed to test the robustness of each control strategy to changes in biological and in economic variables such as initial tick population size and the cost of beef. Moreover, sensitivity analysis points out weaknesses in control and gaps in the available information indicating priorities for research. The model is also effective in considering long-term consequences of tick management policies. The risk of acaricide resistance is considered in the evaluation of dipping strategies and its economical effects are evaluated. The "present value" cost of resistance is calculated by discounting and entered into the "present value" cost of the strategy.

4. LIFE-TABLE MODELS

Ecological life tables were defined by Harcourt (1969) as a "record of a series of sequential measurements that reveal population changes throughout the life cycle of a species in its natural environment." According to this definition, life tables do not simulate a dynamic process but rather reflect a static situation, i.e., empirical results are inserted for the different steps or stages. In a life table for the American dog tick (*Dermacentor variabilis*), Sonenshine (1972) utilizes direct field observations in the same area during a period of several years, combined with data from previous field and laboratory studies. This approach is useful for comparing field studies, establishing their merit, identifying gaps in our knowledge of tick life cycles and the need for further research.

Begon and Mortimer (1986) define "diagrammatic life tables" as a starting point for the construction of simulation models, suitable for modeling the population dynamics of organisms which can be grouped in categories (age, life stage) and which have overlapping generations. This definition is coherent with the dynamic life tables used for tick population models (Haile and Mount, 1987). Begon and Mortimer (1986) describe matrix algebra as a method of expressing dynamic life tables in a much more compact form (cf. Sandberg et al., 1992).

4.1. *Amblyomma americanum* (Lone Star Tick)

The population model of the lone star tick (LST) developed by Haile and Mount (1987) consists of a dynamic life table separated into discrete age classes within each tick stage (Fig. 7.3).

The model is geared to demonstrate the effect of various environmental methods, and suggest future research. The state variable is defined on the basis of life stage and age for each tick. Further division is made between the host-seeking phase, the engorgement phase on a host and the post-feeding phase. The time step is fixed as 1 week: it approximates the shortest development and engorgement time. Calculation of the number of individuals that transfer to the next age or stage during each time step is based on rates of survival, development, fecundity, and host finding. The effects of environmental variables on developmental processes are determined by accumulation of degree-weeks above a threshold temperature. Survival rates for free-living stages are calculated for three types of habitat: bottomland, upland, and meadow.

Density-dependent constraints are included in the model. Simulation starts from the introduction of a number of eggs on a selected week or of a population distribution of overwintering ticks. Weather and day length data, habitat type, host density, host-finding factor, and type of output can be selected. The primary output from the model is a graphic plot of the weekly population of ticks on hosts during each simulation year.

Simulation results are compared with actual population trends for specific areas. Overall agreement is considered good, and availability of actual density

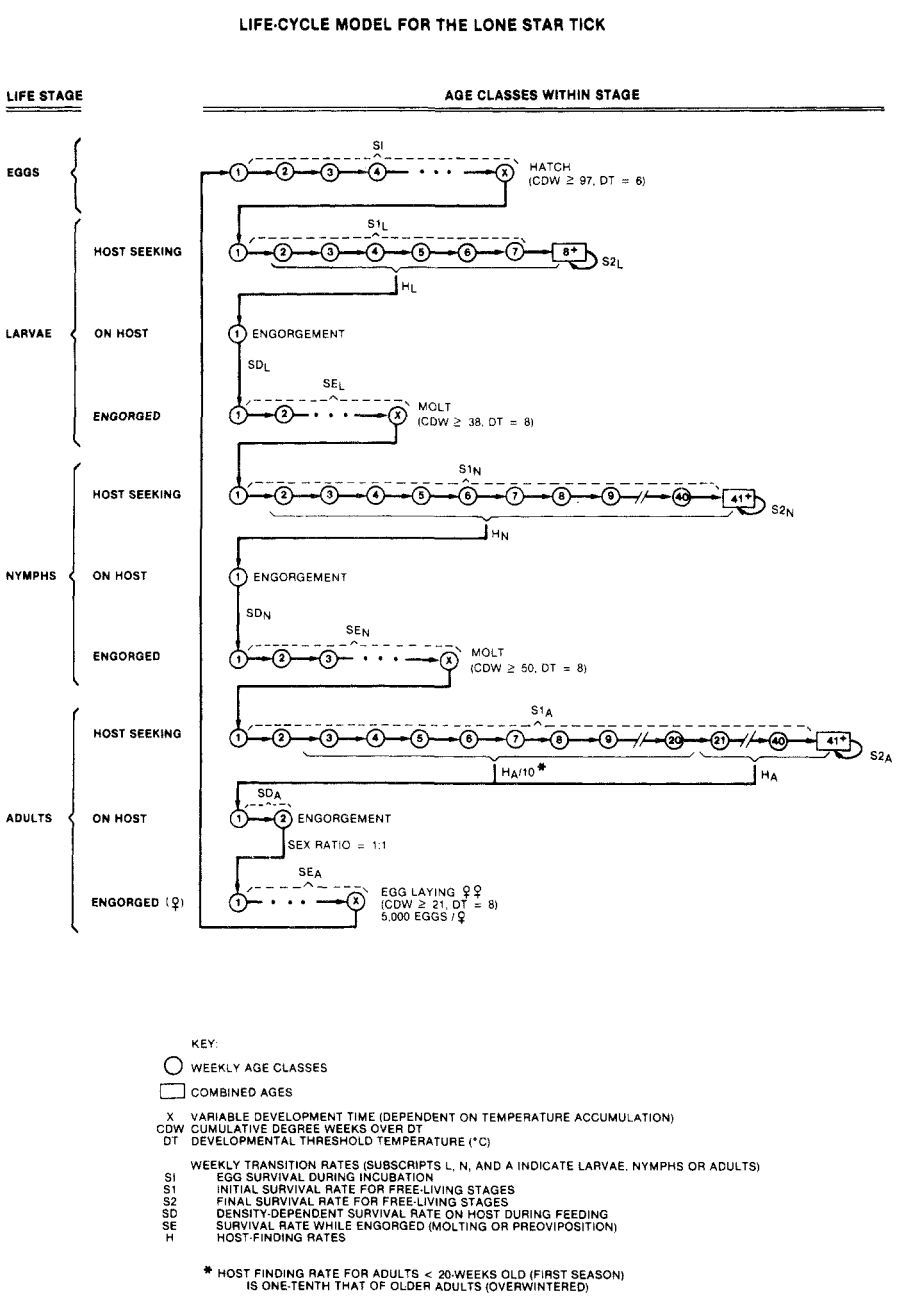


Fig. 7.3. Diagram of the simulation model for *A. americanum* population dynamics. (Reproduced by permission of The Entomological Society of America from Haile and Mount, 1987.)

estimates are considered as limits of the model. Sensitivity analysis is performed for several input levels of day length, weather, habitat, host density, and host finding. Growth rate (R) is sensitive to all variables, while generation time (T) remains almost constant. R and T are calculated at various geographic locations, based on historical temperature and relative humidity data as model inputs. For areas outside the natural range of the LST, population equilibrium density is equal to 0 because the initial input of eggs cannot increase due to an R value < 1, thus validating the model. In a later paper (Mount and Haile, 1987), submodels of management techniques are incorporated into the original model. Submodels are built to test the effectiveness of three management options (considered alone and in various combinations): acaricide application, vegetation management, and host management.

The objective of simulation of management strategies is to determine the minimum level and frequency of each technology or combination of technologies, needed to reduce and maintain the population level of LST below an arbitrary threshold suggested by Mount and Dunn (1983).

The effect of acaricide application on population of *A. americanum* is not highly sensitive to the timing of treatment. It revealed a flexibility of implementation useful in cases in which precise timing is difficult to achieve. Vegetation management techniques can be selectively implemented in optimum tick habitat where densities of *A. americanum* are higher than the mean density of the population.

Implementation of host management techniques alone requires long periods to reduce the density of adults below the threshold level. Each technology may be used at lower levels when integrated with other technologies, in order to obtain satisfactory management of *A. americanum.* Lone star tick simulation (LSTSIM) does not address the problem of the clumped distribution of the tick. Thus, densities of *A. americanum* in optimum habitat could be high, while the overall density is below the threshold level.

4.2. *Dermacentor variabilis* (American Dog Tick)

The simulation model for the American dog tick (ADT) (Mount and Haile, 1989) is a dynamic life-table model with the same general characteristics as the lone star tick model described above (Haile and Mount, 1987). Several aspects of the effects of environmental variables on tick population dynamics are modified and refined. Temperature dependence of female ADT fecundity is modeled by a quadratic equation obtained from a regression analysis. Association between temperature, saturation deficit and survival rate, and association between temperature, day length and host-finding rate, are mathematically and graphically modeled.

Simulation runs require a choice of inputs for geographic location, basic biological data file, and the initial tick population. A selection of inputs relative to environmental variables is made before simulation runs. Geographic and weather data files are available. Initial tick population size is also selected. The

primary output from the simulation is a graphic plot of the weekly population of ticks on hosts during each simulation year. Graphic displays of tick life cycle and of environmental variables are also available. Growth rate per generation, R and generation time T, can be calculated for a given set of input parameters.

Simulation runs are performed using historical average weather data and other environmental variables as inputs for 14 geographic locations in North America. R and T, and equilibrium seasonal activity pattern were calculated. Values of $R > 1$ result for areas inside the known range of *D. variabilis*, while $R < 1$, corresponding to an equilibrium population level = 0 are obtained for areas outside the tick range.

Validation of simulated results through comparison to observed results are made using the yearly average number of free-living adults/ha during peak activity periods (May–July) in three locations. Observed estimates of tick density per ha are obtained from drag-count data. Average deviations are calculated and overall agreement is considered acceptable. Some deviations are explained as variation in actual host populations, maintained constant in the simulation. Potential experiment error in the observations, the limitation of the model, and lack of precise inputs for the simulation run, are also taken into account.

Sensitivity analysis is performed on several input levels of environmental variables. The growth parameters R and T shows different sensitivities to all variables, while T remains almost constant. The effect of input level on R is expressed as the ratio of maximum to minimum value for each variable. Greatest effect is detected for host density, followed by weather, habitat and day length.

Sonenshine (1975), underlined the importance of the knowledge of tick and host population dynamics in order to predict trends in tick-borne diseases and developed a model to evaluate the effects of changes in host abundance on tick population. Cyclical changes in the population of the meadow vole (*Microtus pennsylvanicus*) were particularly important. High vole density resulted in a substantial increase of the yield of fed ADT larvae and nymphs, with a consequent increased risk of tick bites and transmission of tick-borne infections, especially in the habitat utilized by the meadow vole.

4.3. *Ixodes ricinus*

The numerical response of tick populations to increased host density is not always linear. Plowright and Paloheimo (1977) suggest that sparse populations of the tick *Ixodes ricinus* on a high number of sheep hosts may fail to increase due to insufficient mating. The authors calculate the rate of increase for the tick population using arbitrary values for egg production, attachment to host, and tick survival. The incidence of mating is the critical factor for the growth of the population. It is calculated by an analytical model where host finding is assumed to occur randomly and the distribution of ticks on sheep is Poisson. The binomial distribution and its normal approximation are used to determine the probability of tick mating.

The results of the model show that for each initial value of the number of ticks there is an optimum host population size that corresponds to the maximum tick rate of increase. Higher sheep densities result in lower tick population growth. Moreover, when sheep population is low, the tick population rate is insensitive to changes in the size of the tick population, but highly sensitive to changes in the size of the sheep population. Conversely, when the tick population is low, this rate is relatively insensitive to changes in sheep numbers, but sensitive to changes to tick numbers.

The analytical model is incorporated into a tick population model to evaluate the probability of tick extinction as a function of host and tick population sizes. Extinction is less likely when the sheep population is at an optimum value for tick population growth, particularly when the distribution of ticks among sheep is aggregated.

Several variables affecting tick population dynamics are not taken into account in this model which, therefore, is not a predictive one. Rather, it is primarily a general model which effectively synthesizes a complex relationship in an attempt to answer a particular question. It produces the important result that host reduction for tick control may not always be an effective management strategy.

Gardiner and Getinby (1983) developed a weather-based prediction model for the life cycle of *I. ricinus*. The effect of rates on fecundity, development, activity, engorgement, and mortality of each tick life stage is incorporated into a model for predicting occurrence of tick activity depending on varying climatic conditions. They use a sinusoidal curve to relate temperature to development and a gamma curve to describe pattern of egg output. The model predicts greater larval and adult activity in the fall and greater nymphal activity in the spring.

4.4. *Rhipicephalus appendiculatus*

Floyd (1987) and Maywald (1987) developed this model in the context of a wide treatise on environmental factors acting as driving variables on the processes underlying tick life cycles. The driving variables affecting those phases are evaluated based on their importance for the validity of the model and their usefulness for a management approach, the sensitivity of the system to those variables, the availability of relevant data and the priority level for future research.

The tick life cycle is divided into developmental phases, host-finding phase and a parasitic phase. The developmental phases include the processes in the tick life cycle from the drop of engorged female into the pasture to the point when the larvae are ready to attach to a host (Maywald, 1987). For three-host tick species, development of engorged larvae and nymphs is also considered. The host-finding phases include the ecological processes of survival and being picked up by a host (Floyd, 1987). Survival is mainly affected by climatological variables. The host-finding rate is the result of relationships among tick activity

(regulated by extremes of temperatures, dryness, and day length), host activity and stocking rate. Within the parasitic phase, the time spent by the tick on the host, the amount of blood ingested, transmission of pathogens or foreign materials are modeled.

The population model for *Rhipicephalus appendiculatus* is based on the life cycle of a typical three-host tick and it uses a weekly time step. Driving variables are mainly climatological and related to vegetation, host or tick population. Density-dependent constraints are based on host resistance. In order to compare simulated and actual values of seasonal abundance of *R. appendiculatus*, a location with a simple pattern of seasonal tick abundance is selected and simulation is implemented using fitted variables. Peaks of tick abundance obtained by model simulation show a good correspondence with observed patterns, with variations attributed to tick sampling methods and to insufficient information on host resistance. Annual variation in various ecological processes and seasonal abundance is simulated. This model can be expanded to study disease and management, and because of the 1 day time step it is especially suitable for modeling disease transmission.

King et al. (1988) developed a climate-based model for the development of *R. appendiculatus* in East Coast fever zones as a basis for future studies of disease-control programs. In this model, the duration of life-cycle periods which are related to temperature are predicted based on calculation of the development fraction achieved each day. A sinusoidal curve represents the relationship between temperature and time. The predictive technique is used to compare variations in the pattern of the *R. appendiculatus* life cycle in five sites in Kenya, from which accurate climatological data are available. The modal development times are predicted for each life stage in the five sites and compared to observed data. General agreement between predicted and observed results is found. Life-cycle duration is also predicted by the model.

Gettinby et al. (1988) used network representations and computer simulations to investigate the relationship between tick (*R. appendiculatus*) resistance to acaricides and genetic fitness. The model uses a 1-day time step and allows for variations in constants determining tick development and survival, and efficacy of dipping in acaricides. The model provides predictions on the mean number of years before which resistance will appear.

5. PROCESS MODELS OF TICK-BORNE DISEASES

5.1. A General Epidemiological Model

The common characteristics of (the dynamics of) tick-borne diseases are represented in a general model developed by Goldfarb (1986). The human population is categorized according to exposure to tick bite, infection, immunity, and age group. In the flow chart (Fig. 7.4) these state variables are represented by boxes, and transition rates between states are represented by arrows. A proportion X of non-infected human individuals (N) may pass to the state B (bitten). The prevalence of infected ticks (k) determines the probability of

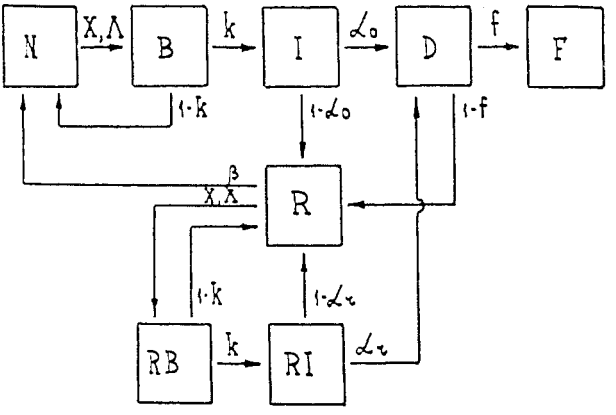


Fig. 7.4. Diagram for a general simulation for tick-borne diseases. (Reproduced by permission of The Entomological Society of America from Goldfarb, 1986.) See text for explanation.

becoming infected (I). The proportion of infected individuals who develop clinical illness is determined by the parameter α. A diseased individual can either die (fatality rate f) or develop immunity and become resistant (R). Immunity is lost at a rate β. An immune individual who is bitten (RB) can return to R with stronger immunity or develop the disease (D). Some of the parameters included in the model (e.g., frequency of tick bite Λ, proportion of infected ticks) can be obtained from field surveys. Parameters that cannot be directly observed are obtained on the basis of theoretical assumptions or through iterative procedures. Number of tick bites per person is assumed to have a Poisson distribution. When this assumption is invalid, direct observation must be carried out.

Infection rate (average number of infected cases per person per unit of time) is then calculated based on tick infection rate and number of bites per person. In order to obtain the incidence rate of the disease it is necessary to know the disease rate (α), which is assigned a value that optimizes both observed incidence rates and values obtained by model simulation.

5.2. Crimean–Congo Hemorrhagic Fever

The procedure described above is illustrated in a paper by Goldfarb et al. (1980). A good agreement between observed and estimated theoretical incidence is attained for Crimean–Congo Hemorrhagic Fever (CCHF) in the Rostov Region (USSR) in 1968. The corresponding value for α is 0.2153. The same value is used to generate accurate forecasts of the disease incidence in subsequent years demonstrating conformity of the model to the real epidemic process.

The age-specific proportion of immune persons, R, is determined from the infection rate. The initial simple model is applicable when R is small (no immune

response to infections or low infection rate), so that patients that are repeatedly infected can be ignored. The simulation explains the lack of a stable immune portion of human population. The application of the model to hyperendemic foci, characterized by high levels of reinfection, requires the calculation of several values for α. In fact, the human population is divided into subsets on the basis of the strength of their immunity. The risk of contracting the disease after infection is calculated for each population subset from an estimate of the rate at which the risk decreases when the immune level increases. The parameter β (immunity loss) is included in the calculation of the proportion of immune persons.

The level of infection rate can also be utilized to distinguish simple and hyperendemic foci and to compare the activity of one focus of disease with another or of the same focus at different time intervals. Epidemic activity of different tick-borne diseases can also be compared.

The simple model is applied to four tick-borne infections (Goldfarb, 1986): tick-borne encephalitis, CCHF, Siberian tick typhus, and Kemerovo tick fever. Estimates of parameters are more reliable in tick-borne encephalitis and CCHF foci and the agreement between observed and estimated age-specific prevalence of immunity confirms the accurate estimates of all values, including the infection rate.

For simulation of a hyperendemic focus of tick-borne encephalitis, a series of parameters α for the subpopulations with increasing immune levels is employed, and the obtained incidence estimate is in agreement with the observed incidence.

5.3. Rocky Mountain Spotted Fever

A model for the transmission of Rocky Mountain spotted fever (RMSF) among its tick vector and mammal hosts and humans was developed by Cooksey et al., 1990 and Haile et al., 1990. This is probably the most complete attempt to construct a comprehensive model for a tick-borne zoonosis.

The model is obtained by incorporating the original ADT model (Mount and Haile, 1989) with simplified submodels for small and medium-sized mammals. Mammals are divided into susceptible, prepatent, rickettsemic, and recovered individuals. Two identical life tables represent infected and susceptible ticks (Fig. 7.5). Transmission of infection is regulated by parameters of biotic and environmental nature. Unavailable parameters are obtained by fitting their value to simulations yielding realistic results. New host infection is regulated by the prevalence of infection in ticks and by a tick infectivity factor which accounts for the virulence of rickettsial strains (proportion of infected and infective ticks that are capable of infection).

The number of infective ticks per host (ITH) is calculated from the infected tick density (ITD), host density (HD), and the tick infectivity factor (TIF):

$$ITH = (ITD/HD) \times TIF \tag{7.3}$$

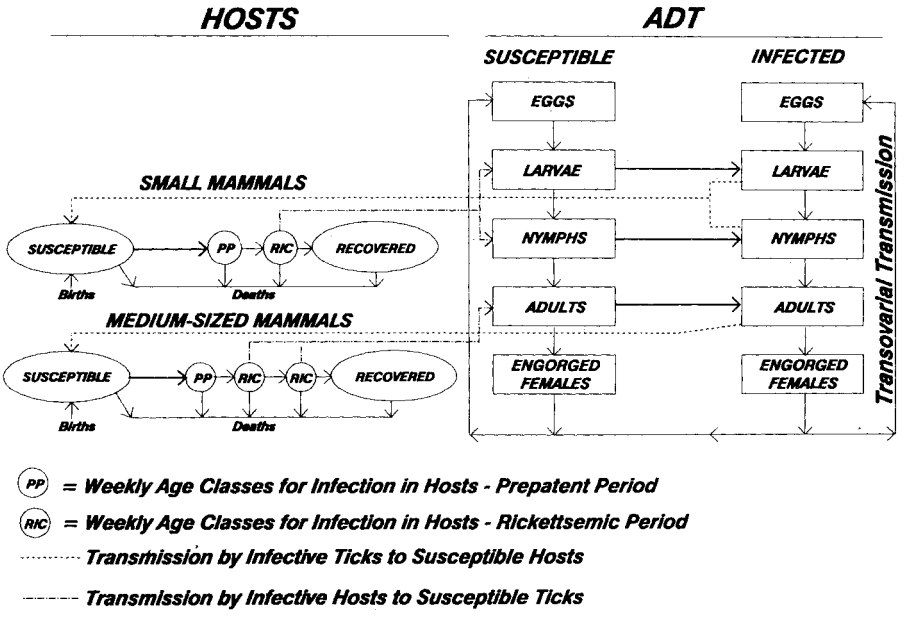


Fig. 7.5. Diagram of the simulation model for the transmission of *Rickettsia rickettsii* between *D. variabilis* and mammal hosts. (Reproduced by permission of The Entomological Society of America from Cooksey et al., 1990.) The models for susceptible and infected tick populations are a simplified scheme of Haile and Mount (1987).

Rickettsial transmission rate depends on the number of infective ticks per host as multiple feedings on a single host result in only one new infection. Monte Carlo simulations are used to distribute various numbers of infective ticks among hosts in order to determine the relationship between intensity of tick infestation and transmission of *Rickettsia rickettsii* to mammal hosts.

Infection of susceptible ticks (the rate of transition of ticks from the state of susceptible to infected) is a function of the proportion of mammals that are rickettsemic and of an infectivity level that accounts for the virulence of rickettsial strain (proportion of rickettsemic hosts that are infective to ticks). A standard infectivity level value of 0.1 (10%) is selected by iterative simulations to fit simulated percentage infections in hosts to observed antibody prevalence data. The model includes both transstadial and transovarial transmission of infection in tick population.

Host finding for humans is modeled as in medium-sized mammals, but at a much lower rate because of lower human density in tick habitat. Human host-finding rate is estimated as a constant using iterative simulation to match the simulated and observed number of cases in Virginia (averaged over a 30-year period). The estimated human host-finding factor of 0.00045 is less than 0.05% of the rate for medium-sized mammals. The total number of cases per year in a state is the sum of weekly simulation results per hectare multiplied by the estimated area of tick habitat.

Simulated human cases are compared with cases observed in nine selected states within the continuous breeding range of the tick. Differences among states are simulated by changing weather and daylength variables. A correlation coefficient (r) of 0.93 is obtained for simulated and observed data suggesting that the model is reasonably valid.

The number of human cases and infection rates of ticks and mammals are sensitive to all variables, both environmental and biotic. The maximum/minimum ratio of human cases indicated that yearly levels of RMSF transmission are most sensitive to changes in weather and host density.

A tick density threshold for the transmission of the disease to humans, corresponding to an equilibrium yearly rate of increase ($R = 1$) is obtained by iterative simulations changing the number of hosts for immature ticks and holding all other variables constant. A transmission threshold is identified at a tick density level of 252 unfed adults per hectare. In an RMSF endemic area, tick-vector maintenance below this level results in a gradual reduction of the number of human cases (Fig. 7.6).

Two different management scenarios are simulated to validate the transmission threshold. Tick population is first reduced below the transmission threshold by reducing small mammals density during years 1–3. In the first scenario (Fig. 7.7a) tick population is maintained at 249 adults/ha with an additional 33% reduction in small mammal density during years 8–30. The number of human cases is reduced to a low level after 3 years and remains at a low rate. In the second scenario (Fig. 7.7b), small mammal density is not

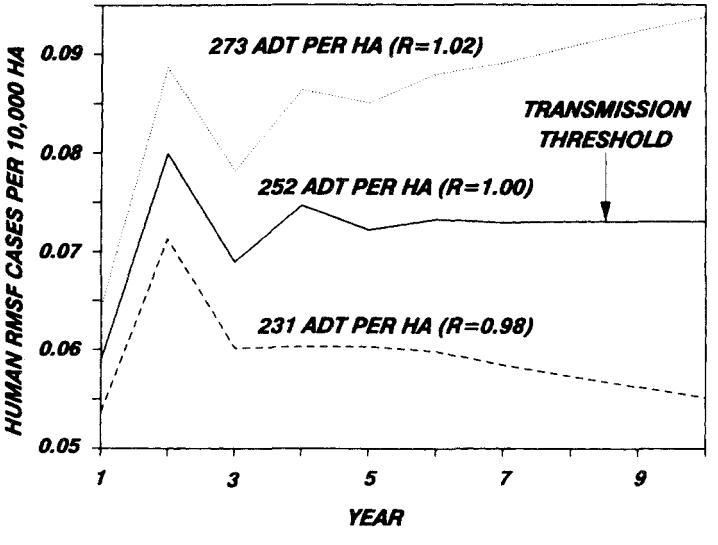


Fig. 7.6. Dynamic simulation of human cases of Rocky Mountain spotted fever with three constant densities of *D. variabilis*. (Reproduced by permission of The Entomological Society of America from Cooksey et al., 1990.) When tick density is below the transmission threshold of 252 unfed adults/ha, human cases decrease.

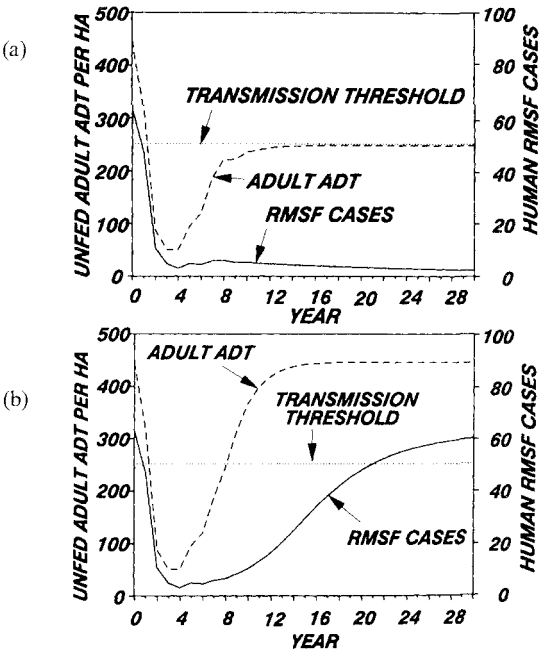


Fig. 7.7. Results of the simulation of tick populations and human cases of Rocky Mountain spotted fever under two different management scenarios. (Reproduced by permission of The Entomological Society of America from Cooksey et al., 1990.) Tick density is below a transmission threshold through a reduction in small mammals (a) and human disease cases decline. With no reduction in small mammals after year 3, the tick population and disease cases undergo a slow recovery (b).

reduced after year 3 and tick population level recovers. The number of human cases increases gradually through a long recovery period of 22 years, including an 8-year recovery period for the vector. This long recovery period provides an explanation for the long-term cyclic nature of RMSF.

This model contains several simplifying assumptions and not all the necessary data are available (e.g., the role of other tick vectors and the population dynamics of host mammals are not included). Nevertheless, this model demonstrates some effects of biotic and environmental variables on RMSF transmission and can be used to evaluate control strategies, and as a research tool for RMSF and other tick-borne diseases.

5.4. Bovine Babesiosis

In the spreadsheet model developed by Smith (1992) for bovine babesiosis, the main driving variable is the *Boophilus* spp. tick burden on hosts. The *Babesia bovis* inoculation rate, h (estimated through the daily probability of infection of

any animal in the herd), is calculated weekly:

$$h = 2(M) \times (s) \tag{7.4}$$

where M is the number of engorged females/animal/day, and s is the sporozoite rate, defined as the proportion of larvae that harbor infective babesial parasites.

The babesial inoculation rate is used to identify a range of risk for disease outbreak defined as "the range of babesial inoculation rates over which the greatest immunological and ecological instability occurs" (Smith, 1983). Within such a range of inoculation rates, most of the cattle receive a primary infection above 9 months of age and develop clinical disease. When the inoculation rate is above the upper limit of the range, 75% of the calves are infected within the first 9 months of life and are able to develop immunity without the disease. Under the lower limit of the range, *Babesia* disappear.

The model is first verified under a steady-state condition (stable tick burden). Excellent agreement is found with the results of a previous life-cycle model (Smith, 1983). The simulation demonstrates that burdens of 2.07 and 5 ticks per host correspond to inoculation rates of 0.0002 and 0.005, the lower and upper limits of the zone of risk. The response of inoculation rates to fluctuating tick burdens is then simulated. Tick counts data from cattle of several breeds in two locations in Brazil and in Paraguay are entered into the model. The results of the simulation confirm the low risk of disease in the herds despite the absence of tick-control practices and of marked seasonal variations in tick counts.

The model proves to be particularly useful in anticipating the effect of low or fluctuating population of ticks caused by tick-control strategies on the risk of babesiosis outbreak. Several levels of tick control are simulated. Greatest enzootic instability (inoculation rate = 0.001) corresponds to 69% tick control in Brazil and to 83% in Paraguay. Model simulations suggest that a decrease in tick burden can result in the eradication of babesiosis without eradicating the tick vector. However, tick control programs must be implemented carefully because the tick burden can fall within the so-called "zone of risk" and can determine high risk of disease outbreak.

The babesiosis model developed by Haile et al. (1992) is combined with a dynamic life table for *Boophilus* spp. ticks that differs from those for *A. americanum* and for *D. variabilis* because it is adapted to a one-host tick life cycle. Larvae, nymphs, and adults are on a single host for an overall time of 3.5 weeks. The model is particularly effective in simulating the effects on tick population dynamics of biotic and environmental variables such as weather, habitat, cattle density, tick density, type of cattle, and geographical location.

In this model, cattle are categorized as infected or susceptible, and according to age class (1–38 weeks and >39 weeks of age). Weekly cohorts of infected animals allow for variations in infectivity based on time elapsed since infection (age of the infection). The relationship between infected tick burden and inoculation rate is defined as the proportion of cattle receiving an infection during each time step and is calculated using Monte Carlo simulations. Variations in host-finding rate are utilized to determine different tick densities

on hosts (standard female ticks per cow per day). Tick-density thresholds are established by iterative model simulations: (1) minimum tick density to maintain *Babesia* spp. transmission (maintenance threshold); (2) minimum density required to inoculate 99.5% of calves by 9 months of age (inoculation threshold). The range of tick densities between these two thresholds corresponds to the zone of high risk of disease outbreak (Smith, 1983).

Simulations in ten locations in the Americas, Africa, and Australia for *Boophilus microplus* and in six locations for *B. annulatus*, with *Bos taurus* × *B. indicus* cattle, strains show that the maintenance threshold is most influenced by the *Babesia* species. The effects of location, strain of cattle, and species of ticks are less evident. The maintenance threshold for *Babesia bovis* ranges from three to eight female ticks per host per day, while it is consistently equal to two for *B. bigemina*.

The inoculation threshold is determined at three locations. For *B. bovis* transmitted by *B. microplus* to *B. taurus*, it varies from 16 ticks in San Juan (Puerto Rico) to 39 ticks in Brisbane, where the season of tick activity is shorter. *B. bigemina* has a generally lower inoculation threshold that varies little from one location to another.

The model confirms the theoretical possibility of the eradication of *Babesia* (cf. Smith, 1983, 1992). It also demonstrated that keeping tick density under the maintenance level to prevent outbreaks of babesiosis is difficult in locations where environmental conditions are favorable to ticks. The introduction of infection to a susceptible herd is also simulated resulting in rapid *Babesia* growth and an epidemic of babesiosis.

The ideal strategy for the control of bovine babesiosis is to maintain tick density above the inoculation threshold, while at the same time preventing losses due to tick infestation. This objective is achievable according to the model because all simulations result in inoculation rates below an economic threshold of 40 standard female ticks per cow per day (Turner and Short, 1972; Sutherst et al., 1979).

5.5. Lyme Disease

Ginsberg's (1988) model of the spread of Lyme disease provides an initial attempt to explore dynamic relationships within the tick vector–vertebrate host–spirochete system which determine the spread of Lyme disease. The prevalence of *Borrelia burgdorferi* infection in the deer tick (*I. dammini*) is calculated from the prevalence of infection in a defined host species, the infectiousness of the host (proportion of susceptible ticks that are infected after feeding) and the proportion of ticks that feed on the host species. Simulations are run using different values of host infectiousness and efficiency of vertical transmission of the infection among hosts (corresponding to a two-way sensitivity analysis). The prevalence of spirochetes in questing nymphs after a 50-year simulation period is higher when both parameters are high.

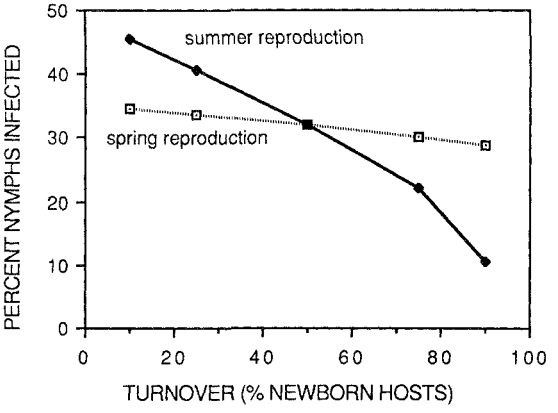


Fig. 7.8. Predicted prevalence of *Borrelia burgdorferi* in nymphal *Ixodes dammini*, as a function of host turnover. (Reproduced by permission of The New York Academy of Science from Ginsberg, 1988.) High summer host reproduction results in a low infection level of nymphs. Prevalence is not sensitive to changes in spring reproduction.

The effect of host turnover (reproduction) is then studied in a dynamical fashion. Simulations are performed with different proportions of newborn hosts. Given a low rate of vertical transmission, a high reproductive rate of ticks in the summer results in a reduction of prevalence of infection in nymphs (Fig. 7.8). This can be explained by the non-simultaneous activity of the immature stages of *Ixodes dammini*. Nymphs are active in spring and early summer, while larval activity peaks in late summer. Hosts which are born in the summer are unlikely to transmit spirochetes to feeding larvae resulting in a low prevalence of infection in nymphs. However, when reproduction occurs mainly in the spring, newborns can be infected by nymphs in spring and in early summer. Larvae that feed on these same hosts in late summer are more likely to become infected nymphs.

Mather et al. (1989) developed a simple mathematical model which incorporated three biological parameters defining the role of an animal reservoir of an arthropod-borne pathogen (specifically *B. burgdorferi*): host density, infectivity to the vector, and degree of host vector contact. The authors assume these parameters to be acting independently.

The infectivity of an individual host to the vector (i_s) is defined by the proportion of the number of larvae that become infected (l_i) out of the total number (l_t) of larvae derived from that host (l_s):

$$i_s = \frac{l_{i_s}}{l_{t_s}} \tag{7.5}$$

The average infectivity (I_s) in a sample of (n) individuals belonging to a species expressed the probability of a tick becoming infected after engorging on

a specific host species (specific infectivity):

$$I_s = \frac{\sum_s i_s}{n_s} \tag{7.6}$$

The expected number of infected nymphs (N_s) produced by individuals of a host species in a given endemic site, is the average number of larval *I. dammini* infesting a host multiplied by the host's specific infectivity (I_s):

$$N_s = L_s I_s \tag{7.7}$$

Including host density (D_s), the reservoir inoculation rate (R) can be calculated relative to all species present:

$$R_s = \frac{N_s D_s}{\sum_s N_s D_s} \tag{7.8}$$

The reservoir potential R expressed the relative contribution made by that species toward infection of the larval tick population. The model is used to compare the relative role of white-footed mice (*P. leucopus*), chipmunks (*Tamias striatus*), and voles (*M. pennsylvanicus*) in the infection of larval *I. dammini* with *B. burgdorferi* in coastal Massachusetts. Specific infectivity is experimentally obtained for each species. Reservoir potential value is calculated incorporating animal densities (as "mean number known alive") and mean larval infestation levels detected in three endemic sites in northeastern US. The prominent role of mice in this geographic location is clearly demonstrated. It is, however, pointed out that there is a possibility of obtaining different results by means of the application of the same model in other ecological communities, characterized by different host species composition (cf. Mannelli et al., 1993).

The relative contribution to the feeding of adults *I. dammini* was calculated in a location in Long Island, New York, by Wilson et al. (1990). The collection of data on host abundance and on number of ticks per host allows the calculation of the forage ratio (Hess et al., 1968). This value, previously employed in mosquito ecology, is the ratio between the proportion of all hosts belonging to that particular species:

$$FR_s = \frac{f_s / \sum f}{N_s / \sum N} \tag{7.9}$$

where f_s is the number of adult ticks collected from species "s" and N_s is the number of individuals belonging to species "s". A forage ratio greater than one indicates "preference" of the tick for the host species. A value lower than one means "avoidance."

White-tailed deer (*Odocoileus virginianus*), raccoons (*Procyon lotor*), opossums (*Didelphis virginiana*), and domestic cats were included in the study by Wilson et al. (1990). The forage ratio for deer, which hosts the vast majority of the ticks, is 3.06, which led to the conclusion by the authors that the abundance

of deer largely determines the abundance of *I. dammini* and the maintenance of Lyme disease.

6. STATISTICAL MODELS

Statistical (associative) models have generally been applied to studies of the population dynamics of ticks and of the epidemiology of non-infectious diseases, and less frequently to epidemiological studies of tick-borne diseases.

In statistical models, the relationships among variables are evaluated probabilistically and according to the strength of associations. Attention is not directed to underlying mechanisms, and phenomena are analyzed in a purely statistical way using multivariate analysis methods.

Applications of associative models to tick-borne diseases include the statistical analysis of the relationships between environmental and human-related variables, and the geographical distribution of disease and disease transmission parameters. Multivariate analysis methods such as stepwise multiple regression, principal component analysis, and discriminant analysis have been applied to the epidemiology of tick-borne disease.

The objectives of multivariate methods are to make predictions on an outcome of interest (tick abundance or incidence of disease) given observed values for a set of independent variables, and to quantify relationships of one or more independent variables to the dependent variable (cf. Kleinbaum et al., 1988). Statistical models are also employed to study the distribution of ticks among host individuals and in the environment.

6.1. Examples of Statistical Models

6.1.1. Dermacentor variabilis

Harlan and Foster (1990) utilized multivariate statistics to associate questing activity of *D. variabilis* in Ohio with micrometeorologic and microenvironmental parameters. Data were collected during the peak period of host-seeking activity of adult ticks. Readings of 15 microenvironmental parameters are individually plotted against estimates of questing activity of adult ticks.

Multivariate procedures used include forward stepwise multiple regression and principal component analysis (PCA). Temperature parameters are important in the multiple regression models, both alone and in interactions with other variates. PCA reduced the number of variables in the system to three. The first component is highly correlated with values of temperature, solar radiation, and vapor pressure deficit, and negatively correlated with relative humidity parameters. Component 2 is positively correlated with wind speed and component 3 with soil moisture.

The above analyses, accompanied by Q-factor analysis and general linear models, demonstrate that ambient temperature is the best general predictor of host-seeking activity of adult ticks, with an apparent upper temperature limit

to questing activity. Solar radiation which co-varied with temperature is not considered a strong predictor.

6.1.2. Colorado Tick Fever

Carey (1979) and Carey et al. (1980) employ PCA and discriminant analysis (DA) in a study of the landscape epidemiology of Colorado tick fever (CTF).

PCA is performed on a condensed data array of 269 sets of habitat variables. The first PC is interpreted as a soil depth gradient, based on the weight of soil related variables. PC_1 and PC_2 (soil moisture gradient) are the major gradients in the habitat variable set and account for two-thirds of the explained variance. Smaller proportions of the variance are explained by ground cover, mammal distribution, and shrub abundance. Soil depth is the most important factor for mammal species distribution, while soil moisture is not a major determinant of any species habitat.

DA is performed in order to classify trap sites based on the presence or absence of mammal species. Two sets of variables are taken into account: habitat, based mostly on soil and vegetation, and ecotype variables which included mammal captures and food items. Discriminating variables for the habitat variable set are compared with those for the ecotype variables. For some mammals, interactions with other species are the determinants of geographical distribution, while other species respond to habitat variables.

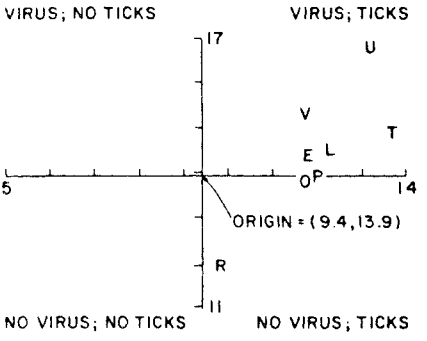
Results of PCA and DA are applied to an analysis of the role of mammalian species as hosts for immature wood tick (*D. andersoni*) and CTF virus. DA is used to classify areas on the basis of tick and virus distribution. Discrimination is performed by means of variables related to soil, vegetation, and the presence of mammalian host species.

The resulting linear discriminating function (DF) for tick and virus is geometrically represented on a plane (Fig. 7.9). The axes separate four quadrants based on the presence of the tick and/or virus. The upper right quadrant represents the area with potential virus calculation in a maintenance cycle, i.e., the CTF nidus. The mean values of the discriminating variables for each mammal's habitat and the overall means of the variables over the study area, are entered into the tick discriminant functions and the virus DF. The resulting DF values are plotted in the graph which represents the functional relationships between the components of the CTF ecosystem.

6.1.3. Rocky Mountain Spotted Fever

Newhouse et al. (1986) evaluate simultaneously the effect of climatic–geographic and human related variables on the occurrence of Rocky Mountain spotted fever.

PCA is performed on ten independent variables whose values were obtained from published records for the 159 counties of Georgia. Based upon scores for



ABSCISSA = ADULT TICK DF = -14.25304 + 1.28404 (SOILD)
+ 0.21876 (% SOIL 2) + 0.24884 (% SHRUB) +
0.18257 (DSLOPE) - 0.07404 (GRASSL)

ORDINATE = VIRUS DF = -16.71534 + 0.30730 (% SOIL 2) +
0.21059 (% SHRUB) + 14.95452 (CHVI-P) +
4.72488 (ROAC-RK) + 0.3041 (NASPEN) +
1.23778 (NJUSC) - 0.0082 (TOPINE)

Fig. 7.9. Geometric representation of the linear discriminating functions for tick and virus in a Colorado tick fever ecosystem. (Reproduced by permission of *The American Journal of Tropical Medicine and Hygiene* from Carey, 1979.) Virus circulation is possible in the upper right quadrant. Letters are habitat coordinates: T = adult *Dermacentor andersoni*; V = Colorado tick fever virus; R = *Spermophilus richardsonii*; L = *S. lateralis*; E = *Eutamias minimus*; P = *Peromyscus maniculatus*; O = mean habitat coordinates. Expressions in equations refer to habitat measurements for soil depth (SOLID, % SOIL 2), shrub coverage (% SHRUB), grass litter (GRASSL), coniferous trees (NTOPINE), slope (DSLOPE), and specific plant species (CHVI-P, ROAC-RK, NASPEN, NJUSC).

two principal components that accounted for 57% of the variability, the counties are divided into four groups by cluster analysis. The incidence of the disease per million people in each of the groups is calculated for the period 1961–1975 as the number of reported cases per total person-year of exposure.

Significant differences in incidence are detected among clusters. From interpretation of principal components (PCs) it is concluded that elevation, class of climax vegetation, and variables related to human activities such as population density, mileage by car and percentage of change in land use are useful predictors for the incidence of the disease.

The results of this study provide a tool for the forecasting of the development of new high-risk areas for RMSF. Moreover, the utility of management strategies, aimed to reduce disease incidence, can be evaluated using this approach. A prospective study could confirm the application of this model to disease prevention.

6.2. Statistical Models for Tick Distribution

Ticks, like many other metazoan parasites, are typically aggregated among hosts resulting in a negative binomial frequency distribution of the number of parasites per host. So fundamental is this departure from randomness, which results in a relatively small number of hosts carrying a large proportion of the parasite population, that Crofton (1971) suggests that "the Negative Binomial distribution is a 'fundamental model' of parasitism in so far as it describes the distribution of parasites among hosts."

More biologically meaningful measures of aggregation are mean crowding, which measures the average number of other ticks per tick on a host or in an area, and patchiness which equals mean crowding over mean density (Lloyd, 1967), and the regression of mean crowding on mean density (Iwao, 1968, 1970). These measures can be used to determine the mechanisms underlying observed aggregated distribution of parasites among hosts (Kitron, 1980).

Aggregated dispersion patterns of *I. dammini* ticks have been analyzed using mean crowding and patchiness (Daniels and Fish, 1990; Kitron et al., 1991, 1992). Daniels and Fish (1990) found that questing *I. dammini* larvae were highly aggregated in August and that their distribution became gradually less aggregated over time. They associated this pattern with gradual dispersal of larvae from oviposition sites. An analysis of changes in the degree of aggregation of larvae on mice (Kitron et al., 1992) indicates that rather than encountering clumps of larvae at a time, mice encounter single larvae in a non-random fashion, or that only single larvae successfully attach to mice following each encounter. This suggests that larvae emerging from one clump of eggs do not infest mice simultaneously in one location. Rather, they either disperse from the oviposition site, or are ready to infest hosts at different times.

7. GEOGRAPHIC AND SPATIAL MODELS

Geographic and spatial models can be used to manage, analyze, and interpret information on distributions of disease, vectors, hosts, and environmental variables, and to generate associations among those distributions. Environmental variables of importance include climate, vegetation, and soil which can be assessed using direct measurements and ground studies, laboratory experiments, and remote sensing data. Geographical information systems (GIS) are particularly equipped to incorporate remote sensing data with ground studies and, with the aid of spatial analysis, offer a way to identify and map the habitat of vector species and potential risk for vector-borne diseases.

7.1. Climate Models

The geographic distribution of tick species and indirectly of tick-borne diseases is limited by climatic factors. Climate is a driving variable in the life cycle of

ticks and the developmental phases of ticks are strongly influenced by meteorological conditions. The suitability of an area for the survival and development of a given species can be assessed with the aid of climatograms. Computerized models have been developed to generate climatic indices which can be used to predict the distribution of tick species (Norval et al., 1992).

Climex is a computer-based system developed by Sutherst and Maywald (1985) which allows for prediction of the potential distribution and relative abundance of an animal species on the basis of biological data and an observed geographic distribution. It has been applied to the distribution of several tick species in Australia, Africa, and globally (Fig. 7.10).

The Climex model uses an ecoclimatic index (EI) which is a measure of the suitability of a given location for the survival of a tick species. This index is calculated as a function of the potential for population growth (growth index) and stress indices which measure the probability of the survival of populations through climatically unfavorable periods. The growth index is a function of a temperature index (TI) and a moisture index (MI). These functions can be made species specific. Stress indices are species-specific functions of dry, wet, cold, or hot conditions which determine the ability of a population to survive unfavorable periods.

Climex input is derived primarily from meteorological data (temperature, relative humidity, etc.) and soil temperature. Parameter values can be approximated using known distributions of tick species with the assistance of laboratory data. Running the model with these values generates indices that can be compared to known distribution and abundance data, and the parameter values can then be interactively adjusted (Fig. 7.10).

7.2. Remote Sensing

Remote sensing (RS) is defined as "the measurement of some property of an object of interest by a sensor that is not in direct physical contact with the object" (Jensen et al., 1989). Most RS systems measure electromagnetic radiation (EMR) that is reflected or reradiated from an object of interest to the remote sensor. Images sensed by satellites have been used to identify habitats of various disease vectors (Hugh-Jones, 1989, 1991). Images vary with regard to resolution and number of wavelength bands. Lower resolution images have a larger swath and are useful for large scale studies of a tick species distribution. Higher resolution images can provide a more detailed map of tick habitats.

Biophysical variables that can be measured using RS include location, elevation, color, chlorophyll absorption characteristics, biomass, temperature, surface texture, and moisture content (Jensen, 1983). Remotely sensed data can be interpreted through calculated indices. A commonly used index is the normalized difference vegetation index (NDVI) based on the red and near infrared wavelength bands and is related to intercepted photosynthetically active radiation. This index can help predict vegetation biomass and has been

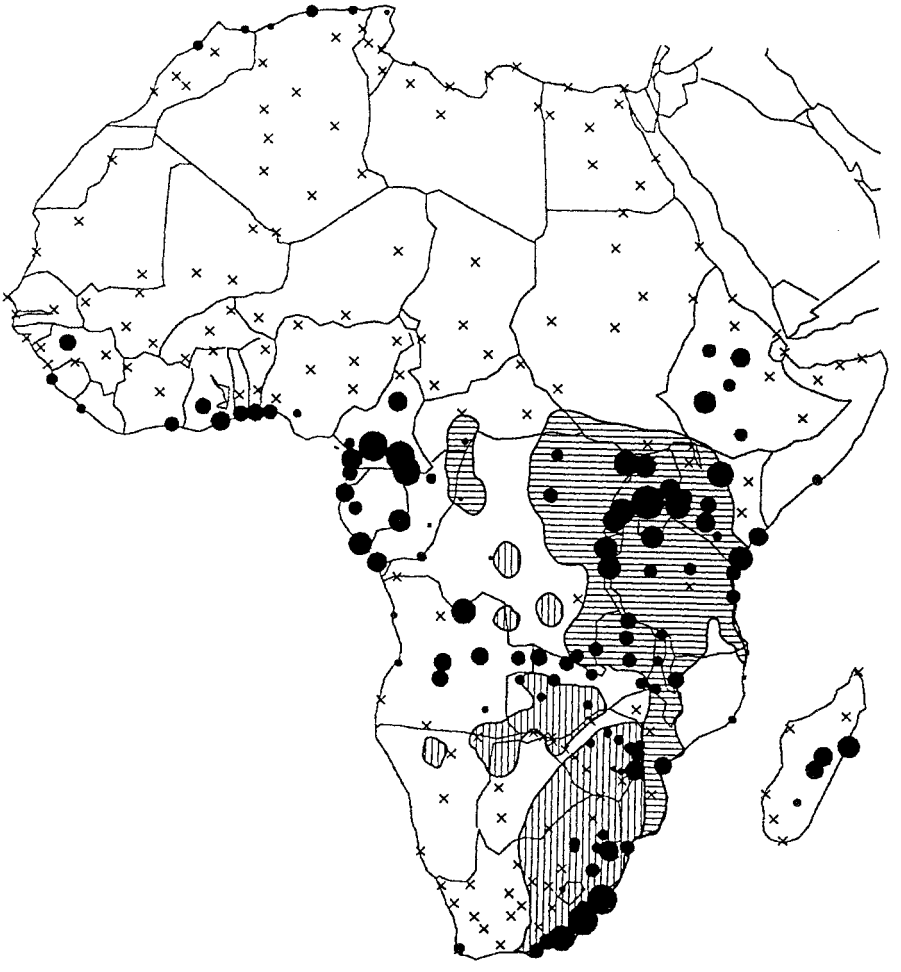


Fig. 7.10. The predicted climatic favorability of different areas of Africa for *Rhipicephalus appendiculatus* and *Amblyomma variegatum*, based on CLIMEX. (Reproduced and modified by permission of the author from Sutherst and Maywald, 1987.) The crosses indicate meteorological stations with climates unsuitable for the tick, while black circles show meteorological stations with suitable climates, with suitability being proportional to the size of the circles. The observed distribution of *R. appendiculatus* alone (shaded area) and with *Theileria parva* (unshaded area) is also shown.

applied to a large-scale study of the distribution of tsetse flies in Africa (Rogers and Randolph, 1991).

The applications of RS to the identification of the habitats of parasites and disease vectors have been summarized recently (Hugh-Jones, 1989, 1991). RS data can assist in mapping parasite, vector, and host habitats, mapping potential vector species distributions, monitoring changes in habitats, and predicting associated changes in vector or host populations, and, finally, in generating risk maps that can be used in control programs.

Remotely sensed data is susceptible to a variety of inaccuracies stemming from the often ill-defined borders between habitats, and errors which may occur in classification. It is always essential to verify (ground-truth) remotely sensed data through studies in the field. The low-resolution studies are susceptible to inaccuracies on the local level and predictions to small areas must be made cautiously.

7.3. Geographic Information Systems (GIS) and Spatial Analysis

A relatively recent tool in mapping and spatial analysis is the geographic information system. A GIS is a computer-based system for automating, manipulating, and displaying mapped information (Burrough, 1986; Chrisman et al., 1989), which includes spatial data in the form of geographic coverages (maps) amd descriptive data in the form of a relational database associated with the mapped features. The four components of a GIS are input, storage, analysis, and output of spatial information (Chrisman et al., 1989).

GIS allows for overlaying of a variety of coverages (e.g., climate, vegetation type, soil pattern, population size) in order to identify factors which may explain the spatial and temporal distribution patterns of vectors and disease. A GIS provides a tool for associating climate and remotely sensed data with ground studies. It is particularly suitable for identifying geographic clusters of disease and for analyzing spatial relationships between disease and risk factors which can be used to construct disease models. For tick-borne zoonoses, spatial information on tick, reservoir host, and human distribution can be associated (overlaid) with information on vegetation (possibly derived through remote sensing) and on soil type in order to help predict the geographic distribution of the disease (Fig. 7.11) either directly or through the incorporation of these relationships in simulation models. Spatial analysis provides methodologies to identify patterns, to test associations in the distributions of multiple factors and to organize complex relationships into meaningful patterns. Statistical analyses of spatial patterns can be applied to the populations of vectors and hosts in order to explain the pattern of distribution of tick-borne zoonoses. Spatial analysis methods include spatial analysis of point patterns (Pielou, 1977). Second-order neighbor analysis (Getis and Franklin, 1987) can be used to associate distribution of ticks and infested hosts with a source of infestation. Measures of spatial autocorrelation, such as Moran's I, can be used to test the degree of similarity of variables in different locations and for the study of static spatial distributions of diseases and environmental factors (Hungerford, 1991). Models which include a spatial component can make use of these methods.

The combination of a GIS and spatial analysis provides a powerful tool to describe and analyze the spatial distribution of disease organisms, vectors of disease and disease cases, and associations among these distributions. Additional information such as climate, type of soil, vegetation cover, demographic conditions, etc. can also be incorporated into a GIS and associated with these distributions. The resulting database can be used for the quantification

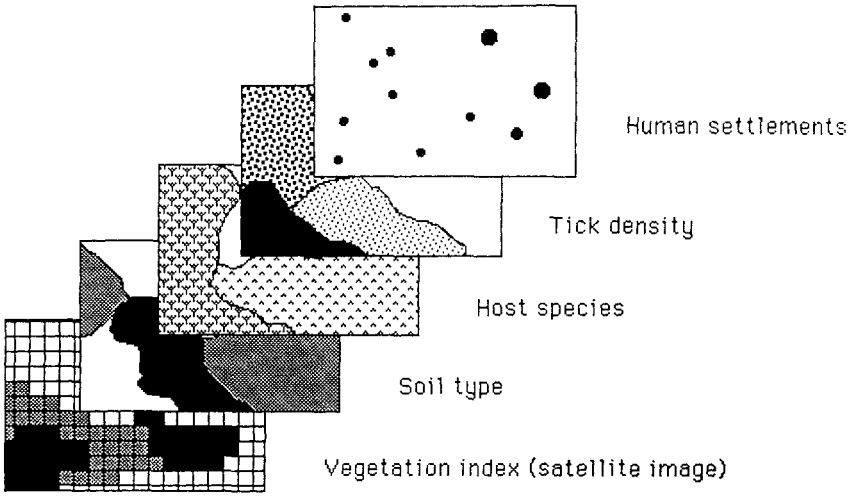


Fig. 7.11. Using the GIS to overlay information which determines the spatial distribution of a tick-borne zoonosis.

of spatial relationships between risk factors and vector or disease distributions, and predicting their spread or maintenance.

Two examples of the application of GIS to disease or tick distributions are described below.

7.3.1. East Coast Fever

Lessard et al. (1990) used a GIS in a study of the epidemiology of East Coast Fever in Africa. They incorporate into the ARC/INFO GIS the distribution of important hosts, vector ticks, and disease presence. Using the Climex model (see above) they assess the distribution of climatic suitability. The objectives of their use of a GIS is to present geographical distributions of African theileriosis and their tick vectors in Africa, to relate the data to selected epidemiological parameters and to define the environmental zones suitable for survival and development of the main tick vector, *Rhipicephalus appendiculatus*. Lessard et al. (1990) and Perry et al. (1990) utilized low-resolution AVHRR satellite data to assess environmental conditions. Vegetation cover and condition are assessed by the NDVI which is based on the difference between the infrared and red reflectance measures. This index is correlated with photosynthesis level. Incorporating these remotely sensed and ground study data, as well as climatic information from the Climex model, they produced a series of maps of the distribution of the mammalian hosts, the tick vectors, the parasite, the disease, the climatic suitability, the heat and dry stress index values, and the NDVI values. This is a large-scale attempt to associate the distribution of a tick species with environmental factors on a continent-wide basis. Due to the unequal

quality of the data throughout the continent and the limitations of the NDVI index, this model is primarily an initial general model and more detailed local studies are still needed. Norval et al. (1992) provide an extensive review of theileriosis models with an in-depth discussion of climate models, GIS, and remote sensing.

7.3.2. Ixodes dammini

Kitron et al. (1991a, 1992) used a GIS to compare the spatial distribution of tick-infested and uninfested white-tailed deer in Illinois, and used second-order neighborhood analysis (Getis and Franklin, 1987) to analyze the spatial distribution of deer around an endemic focus for *I. dammini* and *Borrelia burgdorferi.*

Digitized data on the distribution of deer-harvest locations are overlaid on computerized county maps using an available database on the Illinois GIS. *I. dammini*-infested deer occurrence is associated with the presence of sandy soil, coverage by deciduous forests and with proximity to localities where high tick densities are found. A finer distinction between vegetation and soil types may provide a more informative picture on the specific requirements of this tick species.

Second-order neighborhood analysis is used to examine the proportion of distances between all n points in a spatial database and a given location (usually one of the data points that fall within a specified distance):

$$L(d) = (A \sum k_j/\pi n)^{\frac{1}{2}} \tag{7.10}$$

where $k_{i,j}$ is the summation over all points that are within distance d of point i; A is the area; π and the square root make $L(d)$ linear with respect to d; and $L(d) = d$, when $L(d)$ is produced by a Poisson process.

All deer are clustered around the endemic focus, but this is mostly the result of clustering of infested deer around the endemic focus, indicating that this focus is apparently the only important source of tick infestation in the area (Fig. 7.12).

8. EXPERT DECISION SYSTEMS

Expert decision systems (EDS) are an integral part of artificial intelligence research and applications (Feigenbaum and McCorduck, 1983; Hayes-Roth et al., 1983). Recently, the term advisory systems has been used to indicate that a single expert is often not the sole or major source for constructing the knowledge base and inference engine. The use and applicability of EDS have been detailed by Waterman (1986) and Freznel (1987). Expert systems are computer programs which exhibit high performance in specific problem domains based on large amounts of formally encoded knowledge and the ability to conduct formal reasoning of this knowledge. Originally, expert systems were

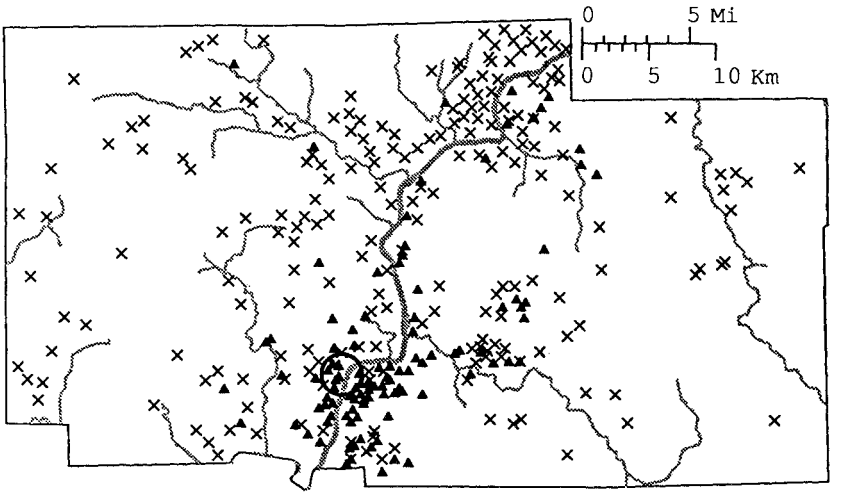


Fig. 7.12. Spatial distribution of deer infested with *Ixodes dammini* (▲) and non-infested deer (×) harvested in northwestern Illinois. (Reproduced by permission of The Entomological Society of America from Kitron et al., 1992.) The circle delineates the hypothesized source of infestation in Ogle county.

designed to do various tasks typically performed by experts: diagnosis, interpretation, design, planning, monitoring, instruction, and prediction. Expert systems consist of a knowledge base (often represented as a set of rules in the form of: IF ⟨condition⟩ THEN ⟨conclusion⟩), an inference mechanism (engine) by which the knowledge base is used to perform given tasks and a user interface through which non-expert users can communicate with the system.

8.1. East Coast Fever

Gettinby and Byrom (1989) constructed a quantitative model using existing knowledge about East Coast Fever (ECF) and applied an expert decision system to store and interpret that knowledge. Based on published findings on the life cycle of *Theileria parva*, they established a set of primary rules as a basis for their model. The model can be used to test the consequences of certain rules or the outcome when rules are in conflict (test rules). When the primary rules and the test rules are not sufficient, secondary rules (based on expert opinion) can be added. A mathematical representation of *T. parva* life cycle is shown in Fig. 7.13. The rule set can be used to calculate parasite growth patterns and effect of parasites on hosts. Information about ECF is expressed in an expert system as a series of rules in which the host status is related to tick challenge. Answers to questions regarding size of tick challenge, proportion of infective ticks, average number of infected acini per tick and number of sporozoites per acinus provide information on the status of a challenged animal. The rules then determine whether an animal dies or recovers.

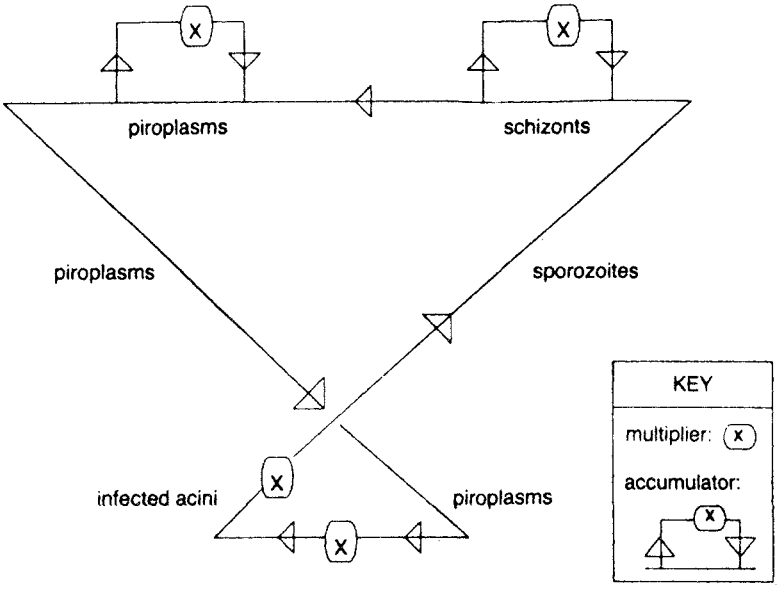


Fig. 7.13. Simplified mathematical representation of the life cycle of the parasite *Theileria parva.* (Reproduced by permission of Elsevier Trend Journals from Gettinby and Byrom, 1989.)

ECFXPERT is an integrated computer model for ECF, its tick vector and its control (Byrom and Gettinby, 1992). The integrated model comprises a tick model, an ECF model, a cattle-dipping model and a drug treatment model. The four simulation models are based on a combination of empirical data and expert information. The model is geared to be site-specific and to be used for undertaking computer experiments on the PC.

The tick model is based on daily time steps to model changes in numbers of the various life stages, using meteorological and tick population data. The disease model uses daily time steps to model changes in numbers of the various categories of host populations (e.g. infected, immune) based on herd age structure and cattle susceptibility as inputs. The intervention models simulate disease control, changes in tick populations and rate of disease in cattle populations.

9. UTILITY OF MODELING TICK-BORNE ZOONOSES

Models provide general conceptual frameworks for understanding the complex dynamic relationships that characterize tick-borne zoonoses, and for discovering patterns common to their transmission systems. Specific cases can be fitted within such conceptual frameworks to highlight the epidemiology of particular diseases and the unique features which determine maintenance and transmission

of infection (cf. Begon and Mortimer, 1986). Thus, models can improve our understanding of essential features of tick-borne zoonoses and help explain epidemiological variables such as incidence, temporal patterns and spatial distribution of the disease.

Several examples of the above concepts can be recognized in the models reviewed in this chapter. The success of models as tools for information management and for determining priorities for research is most readily recognized. Statistical models allow for the recognition of association among variables and the creation of new variables (i.e., principal components) which provide a more succinct summary of available information. Geographic information systems in particular provide a highly effective management tool for spatial information. Expert decision systems can disseminate information to non-experts and decision makers. Simulation models can also direct research efforts to gather necessary data or to improve the quality of existing data.

One of the major goals of models of tick-borne zoonoses is to plan disease control and eradication programs. In applied ecology, models enable one to search for those natural thresholds in dynamic systems which can be exploited when harvesting useful animals and plants, or when attempting to control pests and disease (Conway, 1977). Cooksey et al. (1990) used their simulation model to determine a theoretical tick density threshold for transmission of RMSF. Smith (1992) explored the risk of bovine babesiosis outbreaks associated with tick control, identifying tick-burden levels corresponding to the "zone of risk." Mount and Haile (1987) simulated the effectiveness of tick control technologies, alone or integrated in management strategies, in reducing densities of *A. americanum* below an arbitrary threshold. The model built by Sutherst et al. (1979a) for the control of the tick *B. microplus* includes ecological and economical components and can be considered a true policy model.

In this chapter, we have discussed a variety of model types. Simulation models for tick population dynamics and for tick-borne disease can be incorporated into complex models. Comprehensive simulations help to consider a system in a holistic way, and changes in its components can be assessed in terms of their effects on the entire system.

Simulation and statistical models are typically used separately and applied to different aspects of the ecological dynamics of tick-borne zoonoses. The integration of these model types may provide additional benefits. Thus, the results of the application of statistical models and information management systems to field and experimental data can be integrated in simulation models.

The availability and quality of data, and the understanding of underlying mechanisms determine the reliability and usefulness of model simulations. Insufficient knowledge may lead to the oversimplification of complex systems. On the other hand, excessive complexity deprives models of practical utility (Hedgpeth, 1977).

Overall, the numerical outputs of models should never be followed blindly. Rather, successful models present rapid and inexpensive guidelines for research and management to be validated in real-world situations (cf. Singer, 1984).

10. REFERENCES

Abbey, H. (1952) An examination of the Reed–Frost theory of epidemics. *Human Biol.* **24**, 201–233.

Anderson, R.M. (1985) Strategies for the control of infectious disease agents. pp. 109–141. *In:* Conway, G.R. (ed) *Pest and Pathogens Control: Strategic, Tactical, and Policy Models.* J. Wiley & Sons, Chichester.

Awerbuch, T., Sandberg, S., and Spielman, A. (1992) Critical abundance of hosts perpetuating the tick that transmits the agent of Lyme disease. 40th Annual Meeting of the American Society of Tropical Medicine and Hygiene, Boston, 1–5 December 1992. *Am. J. Trop. Med. Hyg. Suppl.* **45**, 205.

Bailey, N.T.J. (1975) *The Mathematical Theory of Infectious Diseases and Its Applications.* C. Griffin & Co., London.

Begon, M. and Mortimer, M. (1986) *Population Ecology: A Unified Study of Animals and Plants*, 2nd edn. Blackwell Scientific Publications, Oxford.

Burrough, P.A. (1986) *Principles of Geographical Information Systems for Land Resource Assessment.* Clarendon Press, Oxford.

Byrom, W. and Gettinby, G. (1992) Using the computer model ECFXPERT to study ticks and East Coast fever. *Insect Sci. Appl.* **13**, 527–535.

Carey, A.B. (1979) Discriminant analysis: a method of identifying foci of vector-borne diseases. *Am. J. Trop. Med. Hyg.* **28**, 750–755.

Carey, A.B., McLean, R.G., and Maupin, G.O. (1980) The structure of a Colorado tick fever ecosystem. *Ecol. Monogr.* **50**, 131–151.

Caswell, H. (1989) *Matrix Population Models.* Sinauer Assoc. Inc., Sunderland, MA.

Chrisman, N.R., Cowen, D.J., Fisher, P.F., Goodchild, M.F., and Mark, D.M. (1989) Geographic information systems. pp. 776–796. *In:* Gaile, G.L. and Willmot, C.J. (eds) *Geography in America.* Merril, Columbus, OH.

Conway, G.R. (1977) Mathematical models in applied ecology. *Nature* **269**, 291–297.

Cooksey, L.M., Haile, D.G., and Mount, G.A. (1990) Computer simulation of Rocky Mountain spotted fever transmission by the American dog tick (Acari: Ixodidae). *J. Med. Entomol.* **27**, 671–680.

Crofton, H.D. (1971) A quantitative approach to parasitism. *Parasitol.* **62**, 179–193.

Daniels, T.J. and Fish, D. (1990) Spatial distribution and dispersal of unfed larval *Ixodes dammini* (Acari: Ixodidae) in southern New York. *Environ. Entomol.* **19**, 1029–1033.

Feigenbaum, E.A. and McCorduck, P. (1983) *The Fifth Generation.* Addison-Wesley, Reading, MA.

Floyd, R.B. (1987) Modeling tick population. 3. Host-finding phase. pp. 48–51. *In:* Sutherst, R.W. (ed) *Proceedings of an International Workshop of Ticks and Epidemiology of Tick-borne Diseases*, Nyanga, Zimbabwe, 17–21 February 1986.

Frenzel, L.E., Jr (1987) *Understanding Expert Systems.* W. Howard W. Sams & Co., Indianapolis, IN.

Gardiner, W.P. and Gettinby, G. (1983) A weather-based prediction model for the life cycle of the sheep tick, *I. ricinus* L. *Vet. Parasitol.* **13**, 77–84.

Gardiner, W.P., Gettinby, G., and Gray, J.S. (1981) Models based on weather for the development phases of the sheep tick, *I. ricinus* L. *Vet. Parasitol.* **9**, 75–86.

Getis, A. and Franklin, J. (1987) Second-order neighborhood analysis of mapped point patterns. *Ecology* **68**, 473–477.

Gettinby, G. and Byrom, W. (1989) The dynamics of East Coast fever: a modelling perspective for the integration of knowledge. *Parasitol. Today* **5**, 68–73.

Gettinby, G. and Byrom, W. (1992) ECFXPERT: a computer model for the study of East Coast fever. pp. 123–127. *Proceedings of the Eighth Biennial Conference and Brushfire Dynamics Workshop*, Canberra, 25–27 September 1989.

Gettinby, G., Newson, R.M., Calpin, M.M.J., and Paton, G. (1988) A simulation model for the genetic resistance to acaricides in the African brown ear tick, *Rhipicephalus appendiculatus* (Acarina: Ixodidae). *Prevent. Med.* **6**, 183–197.

Ginsberg, H.S. (1988) A model of the spread of Lyme disease in natural populations. *Ann. New York Acad. Sci.* **593**, 379–380.

Goldfarb, L.G. (1986) Epidemiological models of tick-borne infections (Acari: Ixodidae and Argasidae). *J. Med. Entomol.* **23**, 125–131.

Goldfarb, L.G., Chumakov, M.P., Myskin, A.A., Kondratenko, V.F., and Reznikova, O.Yu. (1980) An epidemiological model of crimean hemorrhagic fever. *Am. J. Trop. Med. Hyg.* **29**, 260–264.

Gray, J.S. (1981) The fecundity of *I. ricinus* (Acari: Ixodidae) and the mortality of its developmental stages under field conditions. *Bull. Entomol. Res* **71**, 533–542.

Gray, J.S. (1982) The development and questing activity of *Ixodes ricinus* L. under field conditions. *Bull. Entomol. Res.* **72**, 263–270.

Gray, J.S. (1992) Studies on the activity of *I. ricinus* in relation to the epidemiology of babesiosis in Co. Meath, Ireland. *Br. Vet. J.* **136**, 427–436.

Haile, D.G. and Mount, G.A. (1987) Computer simulation of population dynamics of the lone star tick, *Amblyomma americanum* (Acari: Ixodidae). *J. Med. Entomol.* **24**, 356–369.

Haile, D.G., Mount, G.A., and Cooksey, L.M. (1990) Computer simulation of management strategies for American dog ticks (Acari: Ixodidae) and Rocky Mountain spotted fever. *J. Med. Entomol.* **27**, 686–696.

Haile, D.G., Mount, G.A., and Cooksey, L.M. (1992) Computer simulation of *Babesia bovis* (Babes) and *B. bigemina* (Smith & Kilborne) transmission by *Boophilus* cattle ticks (Acari: Ixodidae). *J. Med. Entomol.* **29**, 246–258.

Harcourt, D.G. (1969) The development and use of life tables in the study of natural insect populations. *Annu. Rev. Entomol.* **14**, 175–196.

Harlan, H.J. and Foster, W.A. (1990) Micrometeorological factors affecting field host-seeking activity of adult *Dermacentor variabilis* (Acari: Ixodidae). *J. Med. Entomol.* **27**, 471–479.

Hayes-Roth, F., Waterman, D.A., and Lenat, D.B. (1983) *Building Expert Systems.* Addison-Wesley, Reading, MA.

Hedgpeth, J.W. (1977) Models and muddles some philosophical observations. *Helgolander Wiss. Meeresunters* **30**, 92–104.

Hess, A.D., Hayes, R.O., and Tempelis, C.H. (1968) The use of the forage ratio technique in mosquito host preference studies. *Mosquito News* **28**, 387–389.

Hoogstraal, H. (1979) The epidemiology of tick-borne Crimean–Congo hemorrhagic fever in Asia, Europe and Africa. *J. Med. Entomol.* **15**, 307–417.

Hugh-Jones, M. (1989) Applications of remote sensing to the identification of habitats of parasites and disease vectors. *Parasitol. Today* **5**, 244–251.

Hungerford, L.L. (1991) Use of spatial statistics to identify and test significance in geographic disease patterns. *Prevent. Vet. Med.* **11**, 237–242.

Iwao, S. (1968) A new regression method for analyzing the aggregation pattern of animal populations. *Res. Pop. Ecol.* **10**, 1–20.

Iwao, S. (1970) Problems of spatial distribution in animal population ecology. Vol. 2, pp. 117–149. *In:* Patil, G. (ed) *Random Counts in Scientific Work.* Pennsylvania University Press, University Park, PE.

Jensen, J.R. (1983) Biophysical remote sensing. *Ann. Assoc. Am. Geogr.* **73**, 111–132.

Jensen, J., Campbell, J., Dozier, J., Estes, J., Hodgson, M., Lo, C.P., Lulla, K., Merchant, J., Smith, R., Stow, D., Strahler, A., and Welch, R. (1989) Remote sensing. pp. 746–775. *In:* Gaile, G.L. and Willmott, C.J. (eds) *Geography in America.* Merril Publishing Co., Columbus, OH.

King, D., Gettinby, G., and Newson, R.M. (1988) A climate-based model for the development of the ixodid tick, *Rhipicephalus appendiculatus* in East Coast fever zones. *Vet. Parasitol.* **29**, 41–51.

King, M.E. and Soskolne, C.L. (1988) Use of modeling in infectious disease epidemiology. *Am. J. Epidemiol.* **128**, 949–961.

Kiszewski, A.E., Awerbuch, T., Sandberg, S. and Spielman, A. (1992) Sexually regulated fertility of ticks—A conceptual model. pp. 328–334. *Proceedings of the First International Conference on Tick-Borne Pathogens at Host–Vector Interface: An Agenda for Research*, Saint Paul, MN, 15–18 September 1992.

Kitron, U. (1980) The pattern of infestation of the beach-hopper amphipod *Orchestoidea corniculata*, by a parasitic mite. *Parasitology* **81**, 235–249.

Kitron, U., Bouseman, J.K., and Jones, C.J. (1991a) Use of ARC/INFO GIS to study the distribution of Lyme disease ticks in an Illinois county. *Prevent. Vet. Med.* **11**, 243–248.

Kitron, U., Jones, C.J., and Bouseman, J.K. (1991b) Spatial and temporal dispersion of immature *Ixodes dammini* (Acari: Ixodidae) on *Peromyscus leucopus* in northwestern Illinois. *J. Parasitol.* **77**, 945–949.

Kitron, U., Jones, C.J., Bouseman, J.K., Nelson, J.A., and Baumgartner D.L. (1992) Spatial analysis of the distribution of *Ixodes dammini* (Acari: Ixodidae) on white tailed deer in Ogle county, Illinois. *J. Med. Entomol.* **29**, 259–266.

Kleinbaum, D.G., Kupper, L.L., and Miller, K.E. (1988) *Applied Regression Analysis and Other Multivariate Methods*, 2nd edn. PWS-Kent Publishing Co., Boston, MA.

Lees, A.D. and Milne, A. (1951) The seasonal and diurnal activities of individual sheep ticks (*Ixodes ricinus* L.). *Parasitol.* **41**, 189–208.

Leslie, P.H. (1945) On the use of matrices in certain population mathematics. *Biometrika* **25**, 183–212.

Lessard, P., L'Eplattenier, R., Norval, R.A.I., Kundert, K., Dolan, T.T., Croze, H., Walker, J.B., Irvin, A.D., and Perry, B.D. (1990) Geographical information systems for studying the epidemiology of cattle diseases caused by *Theileria parva*. *Vet. Rec.* **126**, 255–262.

Levins, R. (1968) *Evolution in Changing Environments.* Princeton University Press, Princeton, NJ.

Lloyd, M. (1967) Mean crowding. *J. Anim. Ecol.* **36**, 1–30.

Mannelli, A., Kitron, U., Jones, C.J., and Slajchert, T.L. (1993) Role of eastern chipmunk as a host for immature *Ixodes dammini* (Acari: Ixodidae) in northwestern Illinois. *J. Med. Entomol.* **30**, 87–93.

Martin, S.W., Meek, A.H., and Willeberg, P. (1987) Veterinary Epidemiology: *Principles and Methods.* Iowa State University Press, Ames, IA.

Mather, T.N., Wilson, M.L., Moore, S.I., Ribeiro, J.M., and Spielman, A. (1989) Comparing the relative potential of rodents as reservoirs of the Lyme disease spirochete (*Borrelia burgdorferi*) *Am. J. Epidemiol.* **130**, 143–150.

Maywald, G.F. (1987) Modelling tick population. 2. Developmental phase. pp. 39–43. *In:* Sutherst, R.W. (ed) *Ticks and Tick-borne Disease. Proceedings of an International Workshop on the Ecology of Ticks and Epidemiology of Tick-borne Diseases*, held at Nyanga, Zimbabwe, 17–21 February 1986.

Mount, G.A. and Dunn, J.E. (1983) Economic threshold for lone star tick (Acari: Ixodidae) in recreational areas based on a relationship between CO_2 and human subject sampling. *J. Econ. Entomol.* **76**, 327–329.

Mount, G.A. and Haile, D.G. (1987) Computer simulation of areawide management strategies for the lone star tick (*Amblyomma americanum* (Acari: Ixodidae). *J. Med. Entomol.* **24**, 523–531.

Mount, G.A. and Haile, D.G. (1989) Computer simulation of population dynamics of the American dog tick (Acari: Ixodidae). *J. Med. Entomol.* **26**, 60–76.

Newhouse, V.F., Keewhan, C., Holman, R.C., Thacker, S.B., D'Angelo, L.J., and Smith, J.D. (1986) Rocky Mountain spotted fever in Georgia, 1961–75: analysis of social and environmental factors affecting occurrence. *Public Health Rep.* **101**, 419–428.

Norval, R.A.I., Fivaz, B.H., Lawrence, J.A., and Brown, A.F. (1984) Epidemiology of tick-borne diseases of cattle in Zimbabwe. II. Anaplasmosis. *Trop. Animal Health Product.* **16**, 63–70.

Norval, R.A.I., Perry, B.D., and Young, A.S. (1992) Modelling of theileriosis and its control. pp. 343–377. *In: The Epidemiology of Theileriosis in Africa.* Academic Press, London.

Perry, B.D., Lessard, P., Norval, R.A.I., Kundert, K., and Kruska, R. (1990) Climate, vegetation and the distribution of *Rhipicephalus appendiculatus* in Africa. *Parasitol. Today* **6**, 100–194.

Pielou, E.C. (1977) *Mathematical Ecology.* John Wiley & Sons, New York.

Plowright, R.C. and Paloheimo, J.E. (1977) A theoretical study of population dynamics in the sheep tick. *Theoret. Pop. Biol.* **12**, 286–297.

Randolph, S.E. (1975) Seasonal dynamics of a host–parasite system: *Ixodes trianguliceps* (Acarina: Ixodidae) and its small mammal hosts. *J. Anim. Ecol.* **44**, 425–449.

Rechav, Y. (1979) Migration and dispersal patterns of three African ticks (Acari: Ixodidae) under field conditions. *J. Med. Entomol.* **16**, 150–163.

Rechav, Y., Zeedeberg, M.E., and Zeller, D.A. (1987) Dynamics of African tick (Acari: Ixodidae) populations in a natural Crimean–Congo hemorrhagic fever focus. *J. Med. Entomol.* **24**, 575–583.

Rogers, D.J. and Randolph, S.E. (1991) Mortality rates and population density of tsetse flies correlated with satellite imagery. *Nature* **351**, 739–741.

Sandberg, S., Awerbuch, T.E., and Spielman, A. (1992) A comprehensive matrix model representing the life cycle of the tick that transmits the agent of Lyme disease. *J. Theor. Biol.* **157**, 203–220.

Semtner, P.J. and Hair, J.A. (1973) The ecology and behavior of the lone star tick (Acari: Ixodidae). IV. The daily seasonal activity patterns of adults in different habitat types. *J. Med. Entomol.* **10**, 337–344.

Semter, P.J., Howell, D.E., and Hair, J.A. (1971) The ecology and behavior of the lone star tick (Acarina: Ixodidae) I. The relationships between vegetative habitat type and tick abundance and distribution in Cherokee Co., Oklahoma. *J. Med. Entomol.* **8**, 329–335.

Singer, B. (1984) Mathematical models of infectious diseases: seeking new tools for planning and evaluating control programs. *In:* Mosley, W.H. and Chen, L.C. (eds). *Child Survival Pop. Devel. Rev.*, Suppl. **10**, 347–365.

Smith, R.D. (1983) Babesia bovis: computer simulation of the relationships between the tick vector, parasite, and bovine host. *Environ. Parasitol.* **56**, 27–40.

Smith, R.D. (1992) Computer simulation of bovine babesiosis using a spreadsheet age–class model with weekly updates. *J. Agric. Entomol.* **8**, 297–308.

Sonenshine, D.E. (1972) Ecology of the American dog tick, *Dermacentor variabilis*, in a study area in Virginia. 1. Studies on population dynamics using radioecological methods. *Ann. Entomol. Soc. Am.* **65**, 1164–1175.

Sonenshine, D.E. (1975) Influence of host–parasite interactions in the population dynamics of ticks. *Misc. Publ. Entomol. Soc. Am.* **9**, 243–249.

Stella II (1990) *User's Guide.* High Performance Systems, Inc., Hanover, NH.

Sutherst, R.W. (1987) Modelling tick population. 4. Parasitic phase. pp. 54–57. *In:* Sutherst, R.W. (ed) *Ticks and Tick-borne Diseases. Proceedings of an International Workshop on the Ecology of Ticks and Epidemiology of Tick-borne Diseases*, Nyanga, Zimbabwe, 17–21 February, 1986.

Sutherst, R.W. and Maywald, G.F. (1985) A computerized system for matching climates in ecology. *Agric. Ecosyst. Environ.* **13**, 281–299.

Sutherst, R.W. and Maywald, G.F. (1987) Modelling tick management 2. The role of Climex. pp. 137–140. *In:* Sutherst, R.W. (ed) *Ticks and Tick-borne Diseases. Proceedings of an International Workshop on the Ecology of Ticks and Epidemiology of Tick-borne Diseases*, Nyanga, Zimbabwe, 17–21 February, 1986.

Sutherst, R.W., Utech, K.B.W., Dallwitz, M.J., and Kerr, J.D. (1973) Intra-specific competition of *Boophilus microplus* (Canestrini) on cattle. *J. Appl. Ecol.* **10**, 855–862.

Sutherst, R.W., Wharton, R.H., and Utech, K.B.W. (1978) *Guide to Studies on Tick Ecology.* CSIRO Australian Division of Entomology, Technical Paper no. 14.

Sutherst, R.W., Norton, G.A., Barlow, N.D., Conway, G.R., Birley, M., and Comins, H.N. (1979a) An analysis of management strategies for cattle tick (*Boophilus microplus*) control in Australia. *J. Appl. Ecol.* **16**, 359–382.

Sutherst, R.W., Utech, K.B.W., Kerr, J.D., and Wharton, R.H. (1979b) Density dependent mortality of the tick, *Boophilus microplus*, on cattle—further observations. *J. Appl. Ecol.* **16**, 397–403.

Turner, H.G. and Short, A.J. (1972) Effect of field infestation of gastrointestinal helminths and of the cattle tick (*Boophilus microplus*) on growth of three breeds of cattle. *J. Agric. Res.* **23**, 177–193.

Waterman, D.A. (1986) *A Guide to Expert Systems.* Addison-Wesley, Reading, MA.

Wilson, M.L., Litwin, T.S., Thomas, A.G., Capkanis, M.C., Maclean, D.C., and Spielman, A. (1990) Host-dependent differences in feeding and reproduction of *Ixodes dammini* (Acari Ixodidae). *J. Med. Entomol.* **27**, 945–954.

Wolfram, S. (1984) Cellular automata as models of complexity. *Nature* **311**, 419–424.

Young, A.S. (1987) Modelling the epidemiology of theileriosis. p. 90. *In:* Sutherst, R.W. (ed) *Ticks and Tick-borne Diseases. Proceedings of an International Workshop on the Ecology of Ticks and Epidemiology of Tick-borne Diseases*, Nyanga, Zimbabwe, 17–21 February, 1986.

Yuval, B. and Spielman, A. (1990) Duration and regulation of the developmental cycle of *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **27**, 196–201.

8

Ecologically Based Strategies for Controlling Ticks

EDWARD T. SCHMIDTMANN

1. ECOLOGICAL OVERVIEW

As obligate ectoparasites, all of the approximately 850 known species of ticks must feed from vertebrate hosts for their growth and development. Underlying this dependency on host blood are tick life-history strategies that reflect ecological, physiologic, and behavioral adaptations to finding and feeding from reptilian, bird, or mammalian hosts. Blood-feeding by ticks is relatively brief, from only 10 to 30 min in argasid species and from 3 to 12 days in ixodid ticks; longer periods of attachment invariably reflect mate finding. Thus, ticks spend only days to weeks attached to a host, but are free living for periods of 1 to many years during which they are exposed to ambient climatic factors. These climatic factors, the collective expression of seasonal temperature, humidity, saturation deficit, and rainfall are important determinants of tick survival. Ticks differ from endoparasites, where the host defines the parasite's environment, but they are also subject to host immune responses during their periods of attachment.

2. ECOLOGICALLY BASED STRATEGIES FOR CONTROLLING TICKS—RATIONALE

Reducing tick population density for more than a short time is a difficult task. Tick populations are intimately integrated into respective plant and animal communities, where their development, activity, and inactivity are keyed to seasonally favorable climatic conditions and host availability. The transient, often cryptic free-living stages are not readily reached by treatments, irrespective of method. The parasitic stages of most three-host ticks utilize free-ranging wildlife as hosts, complicating treatment attempts, while the more accessible

one-host ticks associated with domestic livestock rapidly develop genetic resistance under persistent acaricidal pressure. This chapter reviews progress in developing ecologically based and innovative approaches to reducing tick population density, emphasizing environmental and host-associated factors that can be used to regulate tick populations.

It is notable that both argasid and ixodid ticks typically occupy soil, ground debris and leaf-litter habitats during free-living periods. Ecologically, these habitats have durational stability, defined as H/τ, where H is time that the habitat remains suitable for reproduction and τ is generation time (Southwood, 1977a). The association between ticks and durationally stable habitats is meaningful in the context of the r–K continuum developed from analysis of insect life-histories (Southwood, 1977b), since it indicates that ticks tend to be K-adapted. K-adapted species, in addition to occupying stable habitats, typically have relatively constant population levels, low reproductive rates relative to recruitment of adults, and a low potential for migration; K-adapted species also commonly have complex reproductive tactics, and little harmful impact on their supply of food. These life-history characteristics typify many tick species.

In contrast to K-adapted species, r-adapted organisms are typically found in unstable, short-lived habitats, have high rates of reproduction and increase rapidly, and readily move among suitable habitats, e.g., the house fly, *Musca domestica*. The recognition of K- or r-adapted life-history patterns is useful in that it provides insight into the types of control measures that can be effective for suppressing pest populations (Conway, 1976). This chapter uses the strategies appropriate for controlling K-adapted pests as a template for reviewing the status of ecologically based tick control methods.

3. HABITAT MODIFICATION—PLANTS

It is noteworthy that K-adapted insect species generally fail to adapt to man-made environments. That this condition holds for some tick species is empirically apparent from the advance of modern civilization, where growth of cities and extensive urban sprawl have drastically altered land forms, eliminating many natural habitats. These urban habitats, frequented by ever-increasing densities of humans, represent a near-zero risk for exposure to ticks and tick-borne diseases. On the other hand, ticks invariably occupy extensive geographic ranges that preclude modification in other than relatively localized areas. Accordingly, the escalating involvement of urban populations in outdoor recreation and continuing expansion of residential development into rural settings (e.g. tick-infested ecotypes) represent contemporary biocenoses in which humans and companion animals are exposed to increasing contact with ticks and tick-borne disease agents. Human needs for food and fiber likewise support agricultural production by livestock that forage tick-infested pastures and rangelands throughout the world. These widespread interactions between ticks, their habitats, and wild and domestic

animal hosts represent needs and opportunities for ecologically based tick control methods.

3.1. Mechanical Clearing of Vegetation

Studies in the south-central US have investigated brush removal, mowing, opening of overhead canopy foliage and application of herbicides for suppressing populations of the lone star tick, *Amblyomma americanum*. Lone star ticks are common in wooded and brushy vegetation across extensive areas of the southeastern and south-central US states, where high densities are detrimental to outdoor recreation, wildlife health and beef cattle production (Hair and Howell, 1970; Barnard, 1985). In field studies with plots of varying size, Hair and Howell (1970), Clymer et al. (1970), Hoch et al. (1971b), Mount (1981), and Mayer et al. (1982) demonstrated that clearing and mowing understory vegetation, and opening of overhead canopy foliage reduced tick larval density by 50–85%, nymphs 25–93%, and adults 25–75% (Table 8.1). The considerable variation observed in the effects of vegetative modification on tick life stages reflects differences in the seasonal timing of treatments, length of post-treatment intervals and annual climatic conditions. Using a simulation model of tick population dynamics (Haile and Mount, 1987), Mount and Haile (1987) estimated that modification of vegetation over 100% of a woodlands recreational area continuously for 6 years would reduce lone star tick abundance to less than 300 ticks/ha; in contrast, tick density in similar but unmodified areas remained relatively stable, in keeping with a K-adapted species, at densities of 40,000–50,000 ticks/ha.

3.2. Herbicide Treatment

The application of 2,4-D herbicide to leafy understory vegetation in oak–hickory habitat (Clymer et al., 1970; Hair and Howell, 1970; Hoch et al., 1971a) was somewhat less effective than mechanical clearing in suppressing lone star nymph and adult densities (Table 8.2). Like mechanical clearing, herbicide treatment was initially most effective against the larval stage. Herbicide treatment of woody vegetation with picloram pellets had no effect on adult *D. andersoni* in rock–scrubland vegetation of British Columbia (Wilkinson, 1977), but similar treatment applied annually was very effective against lone star ticks in oak–hickory habitat (Barnard, 1986); larval, nymphal, and adult densities were suppressed 66, 83, and 94%, respectively, over a 4-year period. Herbicide treatment was more economical then mechanical removal of vegetation.

Decreased soil moisture and relative humidity were the most important factors that limited lone star tick survival; higher ground level temperatures due to an absence of vegetative cover also decreased tick survival and egg hatch (Hoch et al., 1971a, 1971b). Nymphs were more susceptible than adults to high

Table 8.1. Effect of vegetative management by mechanical clearing or herbicide treatment on tick density

Treatment (habitat type)	Tick species	Test period	Size plots (number of treatments)	Tick life stage	Effect (% suppression)	Author
Vegetation clearing (oak–hickory woodlot)	*Amblyomma americanum*	4 weeks	1 acre ($n = 3$)	Larva	93%	Hair and Howell (1970)
		2 years	1 acre ($n = 3$)	Larva	72%	
		2 years	1 acre ($n = 3$)	Nymph	53%	
		2 years	1 acre ($n = 3$)	Adult	75%	
Herbicide (oak–hickory woodlot)	*Amblyomma americanum*	4 weeks	1 acre ($n = 3$)	Larva	63%	
		2 years	1 acre ($n = 3$)	Larva	53%	
		2 years	1 acre ($n = 3$)	Nymph	23%	
		2 years	1 acre ($n = 3$)	Adult	12%	
Vegetation clearing (oak–hickory woodlot)	*Amblyomma americanum*	7 months	1 acre ($n = 3$)	Larva Nymph Adult	40–50% all stages	Clymer et al. (1970)
Herbicide (oak–hickory woodlot)	*Amblyomma americanum*	7 months	1 acre ($n = 3$)	Larva Nymph Adult	30–40% all stages	
Vegetation clearing (oak–hickory woodlot)	*Amblyomma americanum*	1–3 years	0.45 ha ($n = 3$)	Larva Nymph Adult	47–85% 29–39% 52–62%	Hoch et al. (1971b)
Herbicide (oak–hickory woodlot)	*Amblomma americanum*	1–3 years	0.45 ha ($n = 1$)	Larva Nymph Adult	33–82% 23–24% 12–39%	Hoch et al. (1971b)
Vegetation management (upland oak–hickory)	*Amblyomma americanum*	2 years	1–20 ha ($n = 5$)	Larva Nymph Adult	84% 93% 77%	Mount (1981)
Herbicide (oak–hickory woodlot)	*Amblyomma americanum*	3 years	0.6 ha ($n = 1$)	Larva Nymph Adult	66% 83% 94%	Barnard (1986)

temperatures, perhaps because nymphs must shelter in close association with soil (Meyer et al., 1982).

These studies emphasize the importance to ticks of plant vegetation in providing favorable microhabitat humidity and moderation of sunlight at the soil surface. Tick susceptibility to desiccation and temperature extremes varies with the species, life stage, sex, age, and physiological condition (Needham and Teel, 1991), but maintenance of water balance is a key factor that determines tick off-host survival. Thus, modifying vegetation will reduce population densities of lone star ticks, as well as other species, especially when these alterations result in decreased humidity and increased air temperature at (or near) the soil surface. However, alteration of vegetation also initiates plant community succession (Wilkinson, 1979). Although subsequent habitat instability might be expected to limit tick population survival, rapid regrowth of vegetation promotes increased host activity or density, along with greater opportunities for tick feeding. Invading hosts, attracted by the rapid re-growth that often follows vegetative management, also introduce ticks, especially ovipositing females, and provide opportunities for re-establishment of a population.

3.3. Burning

Controlled or prescribed burning has been used to destroy ticks, but like mechanical or herbicide treatment, burning entrains ecological processes that facilitate tick population re-establishment. Burning affects ticks directly by exposure to lethal temperatures, and indirectly by removing vegetative cover that protects surviving ticks from deleterious post-burn microclimatic conditions. Warren et al. (1987) divided the effects of burning on arthropod population into "acute" and "chronic" phases. Acute-phase effects occur during combustion and pyrolysis, and in the shock stage that occurs between fire passage and vegetative regrowth; tick mortality in this stage depends upon the season of burning and post-burn climatic conditions. Chronic-phase effects are expressed during the recovery stage that follows vegetative regrowth, but before equilibrium is attained between floral and faunal populations. Tick susceptibility to the effects of burning can be expected to differ among life stages, species, and habitats.

For example, open burning of oak–hickory woodlots in eastern Oklahoma (Hoch et al., 1972), resulted in the suppression of 34.5 and 66.6% of marked adult lone star ticks released in two experimental plots where fire consumed $>70\%$ of the leaf-litter layer and generated temperatures that exceeded 60°C at the leaf-litter and duff interface (Table 8.3). Ticks survived better in the duff and upper portions of soil layers than on the surface of leaf-litter or at the soil–leaf-litter interface. In laboratory trials, exposure of *A. americanum* to *c.* 200°C for 2.5 s (mechanical flaming with a torch) resulted in 100% mortality for all life stages (Barnard, 1986). In follow-up field studies, mechanical flaming eliminated lone star larvae and nymphs, but was less effective against adults

Table 8.2. Effect of vegetative modification by burning on tick density

Type fire (habitat)	Tick species	Test period	Size of plots (number of treatments)	Tick life stage	Effect (% suppression)	Author
Accidental fire (coastal grassland)	*Dermacentor variabilis*	1 year	—	Adult	(90)	Smith et al. (1946)
Annual rangeland burning (pine–palmetto flatland)	*Ixodes scapularis*	12 months	1 mi^2 ($n = 1$)	Adult	(96)[a]	Rogers (1953)
		24 months	1 mi^2 ($n = 1$)	Adult	(75)	
		36 months	1 mi^2 ($n = 1$)	Adult	(29)	
Open fire (oak–hickory woodlot)	*Amblyomma americanum*	1 year	0.4 ha ($n = 1$)	Larva	Apparent reduction	Hoch et al., 19(1972)
		1 year	0.4 ha ($n = 1$)	Nymph	Apparent reduction	
		1 year	0.4 ha ($n = 1$)	Adult	—	
		1 year	0.4 ha ($n = 1$)	Larva	Apparent reduction	
		1 year	0.4 ha ($n = 1$)	Nymph	Great increase	
		1 year	0.4 ha ($n = 1$)	Adult	—	
		Immediate	$\frac{1}{3}$ ha ($n = 2$)	Released adults	(34.5 and 66.6)	
		Immediate	$\frac{1}{3}$ ha ($n = 2$)	Indigenous adults	Minor to moderate suppression	
Prescribed burning	*Amblyomma cavennse*	5 months	—	—	(75)	Oldham (1983)
Mechanical flaming (oak–hickory woodlot)	*Amblyomma americanum*	0–3 months	15.25 m^2 ($n = 3$)	Larva	Strong reduction	Barnard (1986)
		0–6 months	15.25 m^2 ($n = 3$)	Nymph	Strong reduction	

(*continued*)

Table 8.2. (continued)

Type fire (habitat)	Tick species	Test period	*Size of* (number of treatments)	*Tick* life stage	Effect (% suppression)	Author
		0–3 months	15.25 m^2 ($n = 3$)	Adult	No immediate reduction	
		1–3 years	15.25 m^2 ($n = 3$)	Larva	317% increase	
		1–3 years	15.25 m^2 ($n = 3$)	Nymph	182% increase	
		1–3 years	15.25 m^2 ($n = 3$)	Adult	200% increase	
Prescribed burning (deciduous forest)	*Ixoides dammini*	12 months	0.8 ha ($n = 1$)	Adult	(83)	Wilson (1986)
		30 months	0.8 ha ($n = 1$)	Adult	(8.0)	
		4 months	1.3 ha ($n = 1$)	Adult	(88)	
		7 months	0.9 ha ($n = 1$)	Adult	(87)	
		17 months	0.9 ha ($n = 1$)	Adult	(−18)	
		0.5 months	1.2 ha ($n = 1$)	Adult	(71)	
		6 months	1.2 ha ($n = 1$)	Adult	(38)	
Open fire burning (mesquite–mixed grass)	*Amblyomma maculatum*	Immediate	1 ha ($n = 3$)	Adult	(89–100)	Scifres et al. (1988)
(bunchgrass, annual forbs)		Immediate	1 ha ($n = 3$)	Adult	(90–100)	
(mesquite, bristlegrass)		Immediate	1 ha ($n = 3$)	Adult	(100)	
(chapparal–mixed grass)		Immediate	1 ha ($n = 3$)	Adult	(78–90)	
(mesquite–mixed grass)		1 year	1 ha ($n = 3$)	Adult	—	
(bunchgrass, annual forbs)		1 year	1 ha ($n = 3$)	Adult	—	
(mesquite, bristlegrass)		1 year	1 ha ($n = 3$)	Adult	Burned > unburned	
(chapparal–mixed grass)		1 year	1 ha ($n = 3$)	Adult	Burned > unburned	
Open fire burning (oak, beech–maple forest)	*Ixodes scapularis*	2 months	15 ha ($n = 1$)	Nymph	(49)	Mather et al. (1993)

[a]Calculated from authors' data based on mean values for 3 years of study.

Table 8.3. Effects of host-targeted tick control treatments

Method	Host	Tick (life stage)	Plot size (habitat)	Experimental design: Number of treatments	Experimental design: Number of controls	Experimental design: Test period	% infested (ticks/host): Treated plots	% infested (ticks/host): Control plots	Effect on population density[a]	Authors
Bait box	Meadow voles	*Dermacentor variabilis* (larvae and nymphs)	2–300 m traplines	1	1	3 weeks	0 (0)	−(7.6)	—	Sonenshine and Haines (1985)
			2,040 m^2	1	1	6 weeks	23 (0.7)	57.1 (3.8)	—	
	White-footed mice		2,040 m^2 (old field)	1	1	10 weeks	10.3 (0.1)	90 (4.5)	—	
Permethrin-treated cotton	White-footed mice	*Ixodes dammini* and *D. variabilis* (larvae, nymphs)	7 × 7 m and 5 × 10 m grids (Forest)	2	2	4 months	28	99	0	Mather et al. (1987)
Permethrin-treated cotton	White-footed mice	*Ixodes dammini* (larvae, nymphs)	7.4 ha (Forest—residential)	1	1	1st season	3	98	0	Mather et al. (1988)
						2nd season	0	96	10 × reduction	

(*continued*)

Table 8.3. (continued)

Method	Host	Tick (life stage)	Plot size (habitat)	Experimental design: Number of treatments	Experimental design: Number of controls	Experimental design: Test period	% infested (ticks/host): Treated plots	% infested (ticks/host): Control plots	Effect on population density[a]	Authors
Permethrin-treated cotton (Damminix)	White-footed mice	*Ixodes dammini* (larvae or nymphs)	1 ha (woodland)	1	1		—	—	No significant difference	Daniels et al. (1991)
			1 ha (recreational)	1	1	1st season	—	—	No significant difference	
			1.5 ha (residential)	1	1		—	—	Treatment > control area	
			(woodland)	1			33 (15.3)	100 (17.6)	Treatment > control area	
			(recreational)	1		2nd season	67 (12.5)	91 (18.1)	No significant difference	
			(residential)	1			36 (1.5)	80 (4.3)	Treatment > control area	
Permethrin-treated cotton (Damminix)	White-footed mice	*Ixodes dammini* nymphs	7.3 ha (woodland—recreational)	1	1	2 years	0–1 (0–0.1)	74–100 (7.6–15.3)		Deblinger and Rimmer (1991)
				1			<0.1	(7.6–15.3)		
Permethrin-treated cotton (Damminix)	White-footed mice	*Ixodes dammini* (larvae and nymphs)	0.4 ha (residential)	5	5	1st season	35	62.5	No significant difference (nymphs)	Stafford (1991)
						2nd season	9.2	65.6	No significant difference (adults)	
						3rd season	16.3	66.9	No significant difference (nymphs and adults)	Stafford (1992)

[a] Free-living, host-seeking ticks not associated with a host.

that apparently moved into leaf-litter where they were protected. Burning vegetation selectively affects deer ticks, *Ixodes dammini* (=*scapularis*), in differing microhabitats as can be inferred from data on infection rates with a parasitic nematode, adults of which parasitize deer (Mather et al., 1993). This nematode infected 15% of nymphs from the unburned area but no nymphs in the burned area, ostensibly because burning selectively killed ticks that fed as larvae on deer rather than on white-footed mice or other hosts. Larvae fed on white-footed mice would presumably remain sequestered in host nests and survive burning, whereas larvae that fed on deer would have been in leaf-litter and exposed to fire that reduced nymphal density by 49%.

Burning of shrubland vegetation in south Texas (Scifres et al., 1988) resulted in more than 75% mortality of *A. maculatum*, where the maximum temperature in the fire was $\geqslant 330°C$ and $\geqslant 60\%$ of fine fuel was consumed (Table 8.2). Tick densities in each of four vegetation types were reduced immediately post-burning, but no difference was detected between burned and non-burned plots of mesquite–bristle grass and chaparral–mixed grass the following year. Since burning did not alter the botanical composition of vegetative stands, the variation in tick suppression after the first year may have been attributable to reduced mulch cover that otherwise provides refuge for ticks.

Other studies also document the efficacy of acute- and shock-phase burning on tick density, following by rapid increases in tick density during the post-burn recovery stage (Table 8.3). In Florida, annual burning of pine–palmetto flatland (Rogers, 1953) suppressed adult *I. scapularis* for 2 years, but had little effect thereafter; by year 3, tick density was similar to a plot burned 14 years previously. Similarly, open burning of deciduous woodland plots in coastal Massachusetts, which destroyed herbaceous vegetation, grass, and leaves of most woody vegetation but not the duff layer, resulted in as much as 88% of host-seeking adult deer ticks, *I. dammini*, for periods of 6 months to 1 year (Wilson et al., 1984). As with the prior experience, the effect of burning deciduous woodlands was temporary, and after 30 months no difference in adult deer-tick activity was detectable between burned and non-burned areas. Though proof is lacking, the rapid increases in tick density that follow burning may result from the introduction of ticks on wildlife or cattle (Hock et al., 1972; Barnard, 1986; Spickett et al., 1992).

Thus, a consensus of data illustrate that modifying vegetation by mechanical clearing, application of herbicide, or burning can effectively suppress tick density. The effects of such treatments are, however, generally of short duration, as animals attracted to vegetatively modified areas introduce engorged ticks that detach and find suitable microclimatic conditions amidst regrowth vegetation. In rapidly recolonizing modified habitats, tick populations behave like r-adapted species that are capable of moderate migration. This complicates long-term tick control, but its recognition is important to assessing the costs and benefits of habitat modifications. Tick immigration can be minimized or deterred by repeated tick suppression treatments, increasing the area of the management program, or by excluding wildlife hosts (Wilson and Deblinger, 1993). Tick re-introduction on hosts may be avoided by targeting key hosts

with tick control measures, a subject reviewed under Section 4, "Habitat Modification: Hosts."

3.4. Anti-tick Plants

Many tick species not only depend upon the presence of low vegetation for maintenance of water balance, but also use plants as a platform for questing for suitable hosts. An innovative method for disrupting this relationship uses plants that have anti-tick properties, e.g., the tropical legumes, genus *Stylosanthes*. These plants possess sticky glandular trichomes (hairs) that entrap and kill ticks attempting to ascend the stems (Sutherst et al., 1982). Numerous cultivars of *Stylosanthes* have been developed through breeding to improve forage characteristics as well as the density and stickiness of their trichomes, key factors involved in trapping *B. microplus* larvae (Sutherst et al., 1988). Several species may be of use in protecting cattle in tropical pastures (Zimmerman et al., 1984; Sutherst et al., 1988). However, whether *B. microplus* larvae ascend a given plant for questing depends largely upon where female ticks oviposit, as well as the size and density of *Stylosanthes* stems relative to pasture grasses (Wilson and Sutherst, 1990). Sutherst and Wilson (1986) concluded that *Stylosanthes* alone will not control *B. microplus* in Australia, but a high *Stylosanthes* to grass ratio could help to reduce numbers of *B. microplus*, particularly in view of its nutritional benefits to cattle. Other plants, such as the molasses plant, *Melinis minutflora*, and *Gynandropsis gynandra*, also exhibit repellent and acaricidal properties (Thompson et al., 1978; Malonza et al., 1992). Molasses grass is covered with fine hairs that exude an odorous, sticky secretion. *Rhipicephalus appendiculatus* larvae appear to be unable to climb the grass due to its physical properties (Obenchain, 1979). These plants, like *Stylosanthes*, have greatest potential for protecting cattle from ticks in managed grass communities of livestock pastures; they can be expected to have less impact in rangeland habitats with diverse plant communities.

4. HABITAT MODIFICATION—HOSTS

Blood-feeding on vertebrate hosts by ticks is essential for their growth and reproduction. This dependence of ticks on host animals also presents opportunities for intervention with anti-tick measures targeted to host animals. Procedures for reducing tick numbers by modifying their host's habitat or disrupting tick feeding and mating on hosts have considerable promise as innovative approaches to controlling various tick species.

4.1. Host-targeted Control—Advantages

Some advantages of targeting host animals to suppress tick density are obvious, others obscure. Because many ticks, particularly three-host species, feed on

wildlife, anti-tick treatment can be delivered directly to host animals, as opposed to area-wide application. This can be effective and economical, reducing treatment volume and thereby conserving time and money, while also limiting possible non-target effects. Targeting small mammals to control American dog ticks, *Dermacentor variabilis*, for example, required only 0.99 g active ingredient/acre (Sonenshine and Haines, 1985). Host-targeting of anti-tick treatment also improves tracking the fate of a compound or its metabolites, an important consideration in regulatory approval, consumer safety and public acceptance.

Less apparent, the selective targeting of wildlife with tick control technology can be viewed as an ecological finesse that avoids disruption of the animal structure of the community that might occur if a species was removed or reduced in number. Depopulating or eradicating hosts as a means of tick control, for example, will strongly affect the balance of all animals in a community; e.g., depopulation of a rodent species may be detrimental to predators such as red foxes, hawks, and owls. Moreover, host depopulation does not necessarily reduce tick abundance or do so rapidly. As the population of a host species is reduced, ticks attach to other hosts, thus buffering the treatment's effect on tick density. Following deliberate reduction in meadow vole abundance, greater numbers of larval *D. variabilis* attached to white-footed mice (Smith et al., 1946). Elimination of almost all white-tailed deer from Great Island, Massachusetts, resulted in increased numbers of host-seeking adult *I. dammini* on vegetation for 3 years (Wilson et al., 1988). Likewise, a gradual reduction in white-tailed deer density on Crane Island, Massachusetts (Deblinger et al., 1993) led to a 4–6-fold increase in female ticks on female deer for 7 years. Finally, host depopulation must be repeated periodically to counter tick immigration on hosts or host neonate recruitment, which may lead to conflicts with wildlife regulatory agencies, hunters, and animal welfare advocates. In contrast, with selective targeting of key hosts, animals targeted by treatment remain, and continue to pick up ticks which are exposed to treatment and destroyed. Before commencing such treatments, modeling might be done to predict the probable results (see Awerbuch et al., 1992). As an example, Mount and Haile's (1987) lone star tick simulation model predicts that reducing white-tailed deer by 50% will suppress adult *A. americanum* density by 56% over a 5-year period, whereas controlling 50% of adult ticks attached to deer by targeted application of acaricide provides an 86% reduction (Mount, unpublished).

The pioneering study of Kartman (1958) demonstrated that chipmunks, *Eutamias* spp., and golden-mantled ground squirrels, *Citellus lateralis*, hosts of flea species that transmit plague bacteria, could be enticed into bait boxes containing food bait and insecticide. To acquire the food, these rodents had to pass through insecticide dust that contaminated the hair coat, purging both the host and nest of fleas. Sonenshine and Haines (1985) adapted the bait-box method for treating immature *D. variabilis* on small mammals, using insecticide-treated plastic tubes which they baited with various foods. Voles and mice attracted to the containers coated themselves with insecticide impregnated in dust or oil. Trials conducted in an artificial meadow habitat showed that meadow voles entering baited tubes were parasitized by significantly fewer

larvae and nymphs than voles in an untreated habitat. Further, in a follow-up trial, immature *D. variabilis* on small mammals were reduced from 81 to 100% in plots with bait tubes. The bait-tube tick-control concept appears to offer an efficient, ecologically sensitive method for reducing *D. variabilis* populations and countering their re-introduction. Lamentably, this technology has not been accepted for commercial use, even in well-managed high visitor-use areas, such as parks and outdoor recreational areas.

The recent increase in density and distribution of the deer tick, *Ixodes dammini*, in the northeastern, upper midwestern, and mid-Atlantic regions of the US has stimulated renewed interest in host-targeted control. In view of the importance of white-footed mice, *Peromyscus leucopus*, as a primary host for immature deer ticks (Mather and Spielman, 1986), Mather et al. (1987) targeted this host for control. They modified the bait-tube method as a system for delivering cotton treated with acaricide (permethrin) to white-footed mice as nest provisioning material. Immature deer ticks acquired permethrin either in the nest or by contact with the hair coat of mice. Field studies in coastal Massachusetts with cotton-baited tubes set out at 10 m intervals (Mather et al., 1987), resulted in a great reduction of immature *I. dammini* and *D. variabilis* on white-footed mice in treatment sites relative to non-treated areas. Further, 72% of mice from treated areas were tick free, whereas ticks infested nearly all mice in the non-treated sites. In a follow-up study, *I. dammini* nymphs collected from white-footed mice in a forested residential area were reduced approximately ten-fold as compared to a non-treated site (Mather et al., 1988). No reduction in the number of host-seeking nymphs on vegetation, as determined by flagging, was observed the first (same) year of treatment but a 50% reduction in host-seeking nymphs occurred in the treated area the second year.

Development of this promising method (now termed **Damminix**) was important as it represents one of just a few examples where both scientific and commercial interests have been focused on a host-targeted vector control strategy. However, the subsequent application of this host-targeted method for suppressing tick vectors of Lyme disease has had mixed success when applied in different environments by different workers. In Connecticut, Stafford (1991) applied Damminix tubes twice per year at label directions in five deer-tick-infested areas, with five untreated areas as controls. Immature deer ticks on mice in the treated areas were reduced 37.2% and 91.5%, respectively, in 2 successive years. Nevertheless, no significant decrease in either the number of host-seeking nymphs or adult deer ticks was observed in treated areas either year. To assess the effects on host-seeking adults as well as nymphs, the study was continued for a third year (Stafford, 1992). Again, no significant difference was detected in the number of host-seeking nymphs or adult *I. dammini* on leaf-litter or vegetation between treated and untreated areas. Similar results were reported in New York State (Daniels et al., 1991), where Damminix tubes were tested at three sites over 2 years. Since host-seeking nymphs present in the spring and early summer would be derived from larvae active during the previous year, no effect on density of host-seeking nymphs is expected after the first application. In the second year, the density of host-seeking nymphs did

not differ from untreated areas in any of the treatment sites. Fewer larvae were found on mice captured in a residential area but, as in the Connecticut study (Stafford, 1991), without subsequent reduction in host-seeking nymphs. In contrast, Damminix tubes applied in accordance with label instructions in a single 7.3 ha resort area in coastal Massachusetts (Deblinger and Rimmer, 1991) rendered mice free of immature deer ticks and dramatically reduced the number of host-seeking nymphs. Similar results, with tick-free mice and significantly fewer infected nymphs, were obtained in one site but not in another during a trial of permethrin-treated nesting material on Fire Island, New York (Ginsberg, 1992).

Apart from possible differences due to experimental design and plot size, the variable results obtained in these studies very probably reflect ecological factors, such as differing community structures (i.e., insular/coastal versus inland communities that are predictably more diverse), hence the presence of alternative untreated rodent or avian hosts (Daniels et al., 1991), possible differences in the behavior of white-footed mice relative to sex, e.g., male mice carry more ticks than females (Davidar et al., 1989), nest location, provisioning behavior, or other factors. It also is possible that some immature deer ticks contacting treated mice fail to attach and drop off, yet survive and continue host seeking. These studies illustrate the array of ecological and management factors that govern the efficacy of a host-targeted delivery system; they also attest to the necessity of defining and understanding these factors as they affect commercialization of a promising tick-control strategy.

A second host-targeted approach to controlling *I. dammini* reflects the association between adult deer ticks and white-tailed deer. The recent spread and establishment of deer tick populations through deciduous and coniferous forests of northeastern, upper midwestern, and mid-Atlantic regions of the US follows the post-1930s re-establishment of white-tailed deer (Spielman et al., 1985). Several studies also document the importance of white-tailed deer as hosts for adult deer ticks (Piesman et al., 1979; Carey et al., 1980; Schulze et al., 1984), although raccoons, opossums, red foxes, and dogs also host adults (Fish and Dowler, 1989). Wilson et al. (1990) demonstrated that white-tailed deer serve as hosts for 94.6% of a deer tick population. Because large numbers of female deer ticks must feed to maintain the high population densities common to the northeastern US, host animals must be readily accessible during fall and spring months, accommodate tick engorgement, and facilitate the detachment of engorged females in habitats suitable for oviposition, egg hatch, and the acquisition of hosts by offspring larvae. Among the medium- to large-sized mammals common to deciduous forest communities, white-tailed deer best meet these criteria.

White-tailed deer may be treated with a systemic acaricide to control ticks. Miller et al. (1989) showed that ivermectin administered daily as oral doses of 35 and 50 μg/kg body weight provided 100% control of attached adult *A. americanum* and about 90% control of nymphs. Further, a single 50 μg/kg body weight dose gave >90% control of adults and nymphs attached to treated fawns at the time of treatment, and 70% control when deer were challenged with ticks

3 days after treatment. With a single treatment, the engorgement period of females was longer, they were lighter in engorgement weight, and produced fewer eggs than ticks feeding on untreated fawns. Given the 7–10 day engorgement period of female lone star ticks, ingestion of ivermectin to establish lethal titers of 20 p.p.m. in host blood once per week will provide effective treatment.

These data establish the potential for using a host-targeted treatment method to suppress tick populations that are dependent upon feeding from deer, or perhaps other large herbivores. Host-targeted treatments have historically been used to control ectoparasites of cattle. Insecticide-impregnated ear tags in particular, have provided high-level control of the horn fly, *Haematobia irritans*, and also are effective against the Gold Coast tick, *A. maculatum* (Ahrens and Cocket, 1978) and red ear tick, *Rhipicephalus evertsi* (Young et al., 1988). These tick species attach preferentially to the ears of large mammal hosts and are therefore easily treated; grooming by treated animals enhances the spread of active ingredient from such sustained-released devices over large areas of host anatomy (Beadles et al., 1977). A method (Duncan applicator) for topically treating large herbivores with acaricide has been developed and tested in Africa (Duncan and Monks, 1992).

Because populations of most economically important ticks are maintained by wildlife hosts, the topical, ingestible, and injectable acaricide formulations currently available for pets and domestic livestock are of limited value due to the involvement of humans in making treatments. Nevertheless, the innate and learned behavioral capabilities of wildlife species have considerable potential for responding to chemical signals, foods, etc., and these behaviors may be entrained to facilitate delivery of tick control technology. The further development of delivery systems for applying acaricide to wildlife is therefore appropriate, promising, and needed in developing host-targeted tick-control measures. The rodent bait-box and permethrin-treated cotton-ball methods, as well as the recent implementation of an oral rabies vaccine for wildlife (Wandeler, 1990) support this contention.

4.2. Mating Interference

In most ixodid ticks (Metastriata), mating occurs on the host. Disrupting this process provides additional opportunities for tick control. In an ecological context, mating on a host amortizes the costs of mate finding with the risk of host finding; it also increases the probability of successful feeding in mated females.

Of particular interest as a potential method for interfering with tick mating is the production by ticks of pheromones. These substances promote aggregation, attraction, and attachment to suitable hosts, pairing of sexes, copulation, and spermatophore transfer (see Sonenshine et al., 1986; Hamilton, 1992). Not all tick species produce pheromones that evoke these responses, but ticks lack well-developed visual or auditory systems and therefore chemical-based

communication is commonly employed to mediate mating, which in some species may be complex, in character with K-adapted species. The recognition, isolation, and characterization of pheromone compounds that regulate tick mating have intriguing possibilities for manipulating tick behavior to interfere with tick reproduction.

Feeding female ixodid ticks (Metastriata) secrete an attractant sex pheromone, 2,6-dichlorophenol (2,6-dcp), reported in at least five genera and 14 species (Hamilton, 1992). In the American dog tick, 2,6-dcp induces male ticks to detach and signals the location of the pheromone source. Upon locating a pheromone-secreting female, male American dog ticks contact the female and encounter a second sex pheromone, the mounting sex pheromone in the female's cuticular waxes. This contact pheromone stimulates males to search for the female gonopore (Hamilton and Sonenshine, 1988). The mounting sex pheromones of ixodid ticks are non-volatile compounds, largely sterol esters (Hamilton, 1992), that serve to identify conspecific females, gonopore location and spermatophore transfer. *D. variabilis* and *D. andersoni*, however, also employ a third sex pheromone, the species-specific genital sex pheromone (Allan et al., 1988), that mediates mating and is essential for transfer of the spermatophore. Prostriata, including species of the *I. persulcatus* complex, and argasid ticks apparently do not produce either a mounting or genital sex pheromone, but assembly pheromones that facilitate mate pairing have been reported (Graf, 1975; Dusbábek et al., 1991; Hamilton, 1992). The absence of specific sex pheromones in *Ixodes* spp. and argasid ticks may reflect the fact that most species are nidicolous, and therefore isolated physically and ecologically, as well as confined in close proximity to each other in host's nests.

When combined with a pesticide, 2,6-dcp confuses male ticks so that they wander aimlessly in search of females, resulting in increased kill as they collect pesticide from the hair coat of treated animals (Ziv et al., 1981). Microencapsulated 2,6-dcp plus an insecticide (propoxur) proved more effective in reducing mating among surviving ticks than insecticide treatment without pheromone or untreated controls (Sonenshine et al., 1985). By killing mate-seeking males, the treatment disruped mating and egg-laying success.

A further approach to interrupting mating in American dog ticks by confusing and killing males involved use of the mounting sex pheromone in conjunction with molded decoys impregnated with 2,6-dcp and an acaricide (Sonenshine et al., 1992). Males attracted to these decoys were killed when they attached next to, or attempted to mate; no males mated with engorging female ticks and no spermatophores were deposited. Other tick species also were killed using these pheromone-baited decoys, irrespective of variation in the composition of respective mounting sex pheromones.

Ticks also employ other pheromone-mediated mating systems. Most notable is the system of certain *Amblyomma* spp. ticks, where feeding males produce a pheromone that attracts unfed females and induces them to attach near males (Rechay et al., 1977). This pheromone, the "attraction, aggregation and attachment pheromone" (AAAP) (Norval et al., 1992b), is adaptive in that it enables unfed ticks to identify and locate suitable hosts (e.g., animals with

attached and feeding male ticks) (Norval et al., 1989a), brings the sexes together for mating, and further leads ticks to attach in clusters on areas of host anatomy that are least effectively groomed (Norval et al., 1988). The AAAP in *A. variegatum* and *A. hebraeum*, consists of several primary components, especially 2,6-dcp, methyl salicylate and *o*-nitrophenol, that both elicit attraction and induce attachment among species and life stages, along with secondary components, such as benzyl alcohol, 2-methyl propanoic acid and nonanoic acid, that evoke a single response that may be species or stage specific (Norval et al., 1992b).

Yunker et al. (1992) reported that *o*-nitrophenol produced by males and a proven attractant to both species in the field, was only partially attractive to either species in laboratory trials. This apparent paradox was resolved by discovery that unfed male and female *A. hebraeum* are activated by carbon dioxide (CO_2) but, unlike many ticks, not attracted to it. When activated by CO_2, however, both male and female bont ticks are attracted to pre-fed male *A. hebraeum*, *o*-nitrophenol, and feeding males in decreasing order (Norval et al., 1989b). In comparing the response of nymph and adult *A. hebraeum* and *A. variegatum* to CO_2 plus AAAP, Norval et al. (1992a) demonstrated that nymphs of *A. hebraeum* were strongly attracted to CO_2 at distances of 10–15 m, but were more strongly attracted to CO_2 plus AAAP at greater distances. Adult *A. hebraeum* also were not attracted to CO_2 alone, but the combination of CO_2 and AAAP elicited attraction from up to 25 m. The ability of female bont and tropical bont ticks to identify hosts with attached males represents a means by which females not only locate hosts, but also descriminate between hosts, based on the attachment of conspecific males. This ability is of obvious value to females that pair with males and mate on hosts that have large home ranges, such as migratory herbivores (Norval et al., 1989a); it also is advantageous where host density exceeds tick density.

The use of male-produced AAAP pheromone to kill conspecific female ticks was first demonstrated with female *A. maculatum* (Gladney et al., 1974), where females were killed following attraction to feeding males in areas of host anatomy treated with acaricide. Extracts of fed male *A. hebraeum* mixed with acaricide and applied to free-ranging cattle also were shown to be several times more effective in killing ticks than acaricide alone (Rechav and Whitehead, 1978). In further studies, Norval et al. (1991) showed that a pheromone–acaricide mixture effectively attracted nymphs and female *A. variegatum* to treated sites, with the degree and time to kill dependent on the type of acaricide. The potential for using pheromone–acaricide formulations for controlling bont and tropical bont ticks on cattle is based on the understanding that application of pheromone to hosts without male ticks makes them as attractive to unfed females as hosts with males (Norval et al., 1991). Incorporation of the primary components of AAAP and acaricide in molded plastic "bont tick decoys" that were attached to the tails of cattle (Norval et al., 1992b) provided control of adult *A. hebraeum* for at least 2 months. Individual animals are protected because the pheromone–acaricide treatment also kills males, thus precluding the attachment of females. Such treatment also may prevent or reduce tick

transmission of disease microorganisms that require activation by tick feeding before transmission to the host.

Because bont ticks also parasitize wildlife hosts, which compete with cattle as hosts, this promising new technology will be most effective where livestock are isolated from wildlife in paddocks or pastures. Development of this method for use with wildlife, which commonly both sustain tick populations and serve as reservoirs of disease agents, could provide great benefits in both developed and undeveloped countries.

4.3. Anti-tick Vaccine

The repeated feeding of ticks on a host commonly leads to a progressive expression of resistance. This is acquired resistance characterized by a longer duration of tick attachment, reduced engorgement weight, lessened fecundity, impaired viability of eggs, and tick death (Wikel, 1988). Not surprisingly, research efforts have attempted to capitalize on this relationship through developing an anti-tick vaccine that will trigger the host's immune system to interfere with or suppress tick feeding. Achievement of this goal has been elusive, however, and anti-tick vaccines are currently unavailable. On the other hand, current prospects for success using molecular techniques and tick antigens other than salivary gland secretions are promising. To be practical, an anti-tick vaccine will need to be more effective than naturally acquired resistance to ticks, which is commonly incomplete, transitory, and variable among animals; to be succesful for ticks other than one-host species associated with domestic animals, it will require a delivery system for challenging a range of host animals, especially wildlife.

The immune response of animals to tick feeding is complex and variable among species (Allen, 1992). In general, animals respond to tick feeding by activating white blood cells that mediate inflammation and produce antibodies. In naive (previously unexposed) hosts, the response to tick feeding is a sequential process in which components of tick saliva (immunogens or antigens) stimulate host basophils, neutrophils, and eosinophils that infiltrate the host dermis at the feeding site (Wikel, 1988). These cells release vasoactive substances that dilate capillaries, induce edema, and promote inflammation (see Kaufman, 1989). Also, tick salivary immunogens are captured by Langerhans cells in the dermis (Allen et al., 1979) and are presented to B- and T-lymphocytes that produce antibodies and immune memory cells.

In previously exposed (sensitized) hosts, tick feeding evokes a rapid and intense accumulation of basophils, mast cells, and eosinophils in the feeding lesion, resulting in a characteristic "cutaneous basophil hypersensitivity" (Allen, 1973). Degranulation of host basophils releases histamine (Steeves and Allen, 1990) and probably other pharmacologically active compounds that interfere with tick feeding (Paine et al., 1983). With larval *B. microplus*, this immediate-type hypersensitive reaction results in the formation of epidermal vesicules under the mouthparts that induce premature detachment (Kemp and Bourne,

1980), temporary re-attachment and cessation of feeding (Allen and Kemp, 1982), as well as triggering host-grooming responses (Koudstaal et al., 1978).

Because ixodid ticks need to maintain a feeding site for 3–10 days (depending upon life stage) to acquire a blood-meal, tick saliva also contains compounds that prevent blood clotting and wound repair, such as apyrase, as well as anti-histamine and anti-hemostatic compounds (Ribeiro, 1985). In fact, the feeding patterns of ticks may in part be determined by the extent to which tick saliva suppresses host inflammatory responses, thus permitting successful tick feeding (Ribeiro, 1987).

Argasid tick feeding elicits similar, but less pronounced, host immune responses due, presumably to the short feeding period of soft ticks. Not surprisingly, the feeding of argasid larvae, which may attach for up to several days, induce the strongest expression of immunity (Need et al., 1992).

Investigations of tick feeding have greatly improved knowledge of tick–host interaction, including the recognition that the use of salivary gland antigens as a basis for developing an anti-tick vaccine faces formidable barriers, including the ability of ticks to evade host immune responses. Ramachandra and Wikel (1992) reported the presence in tick saliva of cytokines that block the processing of salivary antigens by host T cells. Further evidence of tick ability to counter the host immune response is reflected in the differing salivary gland activity that occurs during feeding on previously challenged hosts (Walker and Fletcher, 1990), as well as during feeding on a single host (McSwain et al., 1992). Tellam et al. (1992) reviewed other reasons why tick salivary gland secretions, though commonly inducing antibody production, are not effective in protecting against tick feeding.

Ecological factors also may contribute to tick evasion of host immune responses. The life span of many tick species exceeds the gestation period or life span of host animals, thus insuring feeding on young, immunologically naive hosts. Argasid ticks, in particular, stand to benefit from this relationship, given their long life span, short feeding interval and the proximity of host neonates in nests. The one-host life cycle of some ixodid tick species probably represents an adaptation to migratory herbivores (Hoogstral, 1976); it also may facilitate avoidance of host immune response, as the duration of attachment and feeding, 20–30 days with *Boophilus microplus*, may permit early season generations to feed from young, non-sensitized or de-sensitized animals, particularly as immunity is expressed most strongly against the larval stage (Roberts, 1968).

A particularly promising approach to developing an anti-tick vaccine is based on the use of tick antigens which, unlike the tick saliva antigens that hosts routinely experience, are associated with tick tissues not directly exposed to hosts. This recent "concealed antigen" approach has focused on tick midgut cells as the source of antigens (Willadsen and Kemp, 1988; Wong and Opdebeek, 1990), with the understanding that midgut cells are exposed to host blood, including any anti-tick antibodies, during tick feeding. Initial studies with crude extracts of ticks inoculated into cattle produced good immunity to subsequent tick infestation, damaging the tick gut such that erythrocytes leaked into the hemolymph (Kemp et al., 1986). This expression of resistance, which

differs strongly from the immediate hypersensitive response associated with natural tick feeding (Willadsen and Kemp, 1988), apparently results from a complexing of the anti-tick antibody with epitopes on tick midgut digestive cells (Kemp et al., 1986) that process host blood by endocytosis. One protective antigen, the midgut membrane protein Bm86 (Willadsen et al., 1989), is active at extremely low concentrations. Nevertheless, 50,000 engorging adult *Boophilus microplus* were required to produce 100 μg of purified antigen (Willadsen et al., 1988). As reviewed by Tellam et al. (1992), the potential for using this or other concealed antigens as a commercial anti-tick vaccine rests on the genetic manipulation of cloned bacteria, fungi, or insect cell cultures to produce a recombinant molecule that will induce anti-tick antibodies that complex with the midgut antigen. This approach is not restricted to *B. microplus* and other researchers are pursuing similar technology with other tick species (see Wikel, 1988; Rechav, 1992).

As dictated by tick life-history patterns, an anti-tick vaccine can be expected to be most useful for controlling one-host ticks, such as *B. microplus*, that complete the entire life cycle on a domestic animal restricted to a defined area of pasturage. Most three-host tick species are maintained by wildlife hosts, which imposes a difficult, but not impossible, problem for vaccine delivery. The few three-host species that utilize the same animal or animals of the same species as hosts for larvae, nymphs, and adults, such as *Rhipicephalus sanguineus* and some *Hyalomma* spp., respectively, should also be amenable to control with an effective vaccine. Economic incentives, such as the estimated 500 million head of cattle affected by *B. microplus* in South America and Australia (Tellam et al., 1992), will ensure continued research to develop an anti-tick vaccine. In addition to molecular studies of antigen and host immune response, the ecological interactions of ticks and key hosts need to be further characterized as a basis for assessing the feasibility of delivering an anti-tick product.

4.4. Host Depopulation or Exclusion

Irrespective of the advantages that host-targeted control methods have over host exclusion or depopulation, removing a key host species will reduce tick-host finding success and therefore, over time, result in decreased tick density. "Pasture spelling" (Wilkinson, 1957) was conceived as a form of host exclusion in which cattle are kept from paddocks long enough to expose larval *Boophilus microplus* to lethal ambient climatic conditions. In field trials, temporary destocking of cattle until numbers of questing larvae decreased dramatically resulted in much lower densities of ticks on cattle during ensuing grazing periods compared to cattle in tick-infested paddocks; it also decreased the number of acaricide dippings needed to maintain satisfactory control. Similarly, exclusion of sheep from open moorland pasture in Wales for 1 year resulted in a virtual disappearance of all life stages of *I. ricinus* (Randolph and Steele, 1985) (Table 8.4). The halving of sheep density had a negligible effect

on nymph host-finding success, a greater effect on the attachment of larvae, but a marginal effect on tick density overall.

Suppression of tick density as observed in the aforementioned studies can be attributed to the fact that both species are common to pasture communities and largely dependent on feeding from domestic livestock for reproductive success. In many other areas, including Africa where several economically important large mammal ticks are long-lived three-host species that utilize multiple wildlife hosts (Young et al., 1988), host exclusion would not only be impractical, it would also be ineffective.

On the other hand, exclusion of white-tailed deer, the primary host for both adult *I. dammini* and *A. americanum* three-host species in North America, has been shown to reduce the density of both tick species. As examples, exclusion of deer by fencing 2.43 ha plots in Kentucky for a 5-year period (Bloemer et al., 1986) resulted in 98% fewer *A. americanum* larvae, 38% fewer nymphs, and 22% fewer adults than plots with deer. Similarly, a 3-year exclusion of white-tailed deer from a larger 66.8 ha campground area, where introduction of larvae and nymphs on medium-sized mammals was less of a factor, eliminated all lone star tick larvae and nymphs, and adults were reduced by about 60% (Bloemer et al., 1990). The importance of cattle as hosts for lone star tick populations was demonstrated in field plots burned or treated with herbicide (Barnard, 1986), where all life stages of *A. americanum* increased in both treated and control plots open to cattle; larvae exhibited a 12-fold increase in population growth rate due to the introduction of blood-fed females on cattle. Simulation modeling (Mount and Haile, 1987) indicates that reducing white-tailed deer density by 90 and 75% will reduce *A. americanum* density below a threshold of 1 tick/ha by years 4 and 7, respectively. Further, the model predicts that a deer density equal to or less than 6.18/100 ha will not support a lone star tick population in oak–hickory habitat.

The resurgence of *I. dammini* populations in North America has also prompted efforts to suppress deer tick density by reducing or eliminating deer. As noted earlier, removal of most deer from Great Island, Massachusetts (Wilson et al., 1984, 1988), had little effect for several years, but ultimately reduced nymphal density by *c.* 60% after 8 years. Depopulation of deer by hunter harvesting of 40% of the population annually from a private reserve in Ipswich, Massachusetts (Deblinger et al., 1993), eventually resulted in a *c.* 50% decrease in deer tick larvae and nymphs on white-footed mice. Notably, when deer become scarce, adult tick density on the remaining animals tends to increase. This may explain why tick density did not decrease during the first 3 years of depopulation. Simulation modeling (Awerbuch et al., 1992), predicts that removal of 40% of deer annually can be expected to have a negligible effect on *I. scapularis/dammini* density. In Ireland, the density of nymphs of *I. ricinus* was significantly lower in a forested area permanently fenced to exclude Sika deer, *Dama dama*, than in the adjacent deer-inhabited area (Gray et al., 1992). Other methods for excluding white-tailed deer with electric fencing and repellents are reviewed in Olkowski (1990) and Wilson and Deblinger (1993).

These studies illustrate on a practical and theoretical basis that stringent

Table 8.4. Effect of host exclusion or depopulation on host tick burdens

Method (habitat)	Degree of exclusion or depopulation	Experimental design: Plot size	Number of treatment sites	Number of control sites	Test periods	Tick species (life stage)	Results: tick burden on hosts or (tick density)	Author(s)
Pasture spelling (tropical grassland)	Total	40 acres	1	1	3–4 months	*Boophilus microplus*		Wilkinson (1975)
						Larva	Very scarce	
						Adult	Scarce	
		2–3 mi^2	1	0	3–4 months	Adult	Scarce	
Host reduction (deciduous forest)	70%	240 ha	1	1	1 year	*Ixodes dammini*		Wilson et al. (1984)
						Larva	Possible minimal decrease	
						Nymph	Apparent decrease	
Pasture spelling (moorland pasture)	100%	1 ha	1	1	1 year	*Ixodes ricinus*		Randolph and Steele (1985)
						Larva	(Virtual disappearance)	
						Nymph	(Virtual disappearance)	
						Adult	(Virtual disappearance)	
Host exclusion (oak–hickory woodlands)	2.43 ha	Total	1	1	3 years	*Amblyomma americanum*		Bloemer et al. (1986)
						Larva	(98% suppression)	
						Nymph	(38% suppression)	
						Adult	(22% suppression)	

(continued)

Table 8.4. (continued)

Method (habitat)	Degree of exclusion or depopulation	Experimental design: Plot size	Number of treatment sites	Number of control sites	Test periods	Tick species (life stage)	Results: tick burden on hosts or (tick density)	Author(s)
Host reduction (deciduous forest)	50% (1–3 years) near total (3–6 years)	240 ha	1	1	6 years	*Ixodes dammini*		Wilson et al. (1988)
						Larva	Increase, then persistent decrease	
						Nymph	Continuous decrease	
						Adult	Increase in questing ticks on vegetation	
Host exclusion (oak–hickory woodlands)	Total	71 ha	1	1	4 years	*Amblyomma americanum*		Bloemer et al. (1990)
						Larva	(64% reduction: all life stages	
						Nymph		
						Adult		
Host exclusion (deciduous forest)	Total		1	1	2 years	*Ixodes ricinus*		Gray et al. (1992)
						Larva	Significant	
						Nymph	Reduction	
						Adult	Each life stage	
Host reduction (deciduous forest)	Gradual (40% per year)		1	0	7 years	*Ixodes dammini*		Deblinger et al. (1993)
						Larva	*c.* 50% reduction	
						Nymph	*c.* 50% reduction	
						Adult	Continuous 4–6 × increase	

exclusion or near absolute depopulation of a host species can reduce tick density where adult tick feeding, hence recruitment of offspring, is dependent on a single or key animal species. Nevertheless, the beneficial effects of such action tend to be slow in developing, and the impact of host exclusion or depopulation on other animal or plant species needs careful consideration.

5. REPRODUCTIVE INTERFERENCE

Pest control strategies based on interfering with reproduction are predictably appropriate for K-adapted species. Three strategies, namely pheromone-mediated inhibition of mating (discussed above), irradiation-induced sterility, and hybrid sterility have been investigated as potential methods for controlling tick populations.

5.1. Radiation-induced Sterility

The most notable method for interfering with the mating of an arthropod is the sterile insect (male) technique (SIT) developed and used with great success in eradicating the primary screwworm, *Cochliomyia hominivorax*, from the US (Knipling, 1979). Application of the SIT for ticks has been researched with several species, but practical control programs have yet to be implemented. Limitations of the SIT for suppressing tick populations are the need for mass rearing of ticks for inundative releases and concern that released ticks may cause further economic loss or transmit disease agents.

Because female *Amblyomma* ticks are attracted to hosts with attached and feeding males, and males mate repeatedly, the SIT method has particular promise as a strategy for reducing the recruitment of *Amblyomma* offspring. Exposure of unfed females to 250 rad of gamma irradiation had no effect on engorgement weight, egg laying, or egg hatch; exposure to 500 to 1,000 rad resulted in production of only some viable eggs and no eggs, respectively (Drummond et al., 1966). Some viable eggs were produced by females mated with males exposed to 500 rad irradiation, whereas mating with males exposed to 2,500 rad prevented egg hatch. Similarly, exposure to 2–8 kilorad (kr) effectively sterilized male *A. americanum*, *A. hebraeum*, and *R. appendiculatus* (cited in Spickett, 1978).

In addition to sterility, factors such as the vigor, longevity, and competitiveness of irradiated ticks also influence the potential for using SIT for control purposes. Spickett et al. (1978) reported that 6 or 8 kr did not affect the attachment of either male or nonirradiated female *A. hebraeum* to hosts, and mating with spermatophore transfer occurred in all test groups. As desired, females mated with irradiated males did not produce viable eggs. Thus a technology base exists for irradiating ixodid ticks to induce sterility, but reservations about the impact of releasing large numbers of potentially damaging ticks has curtailed further study (Spickett, 1978).

Galun and Warburg (1967) established that high (16,000 rad) dose irradiation was necessary to sterilize males of the argasid tick, *Ornithodoros tholozani*. Such exposure also reduced longevity and sexual competitiveness. *O. tholozani* inhabits caves and nests throughout extensive areas of the Middle East and adjacent southern Asia, where it is an important vector of human relapsing fever. Female *O. tholozani* are, however, more sensitive to irradiation than males, as no eggs were laid after exposure to 4 kr, and 1 kr dosage slowed oogenesis considerably (Galun and Warburg, 1967). Irradiated females also retained blood-meals longer than untreated ticks, a condition that apparently resulted from damage to midgut reserve cells that replace digestive cells during blood-meal digestion. Nevertheless, approximately 30% of females were observed to take a blood-meal after irradiation, and therefore the release of irradiated females in nature was viewed as a questionable procedure.

Because the long-lived males of *Argas persicus*, an argasid tick associated with bird nests, only remain sexually competitive with wild males for a short period of time after irradiation (Sternburg et al., 1973), the possibility of using sterile females for control purposes was also investigated for this argasid tick. In support of this method, Ailam and Galun (1967) predicted that "when the total number of available matings of females is greater than that of the males, the rate of extinction of the population increases when the number of sterile females increases." Further, Galun et al. (1972) found that egg laying in female *A. persicus* was strongly affected by low-level irradiated females, presumably due to the slowed rate of blood digestion following treatment. Unfortunately, wild male *A. persicus* produced smaller but more numerous spermatophores in the presence of introduced irradiated females, and this resulted in higher rates of insemination of non-irradiated females. This condition, along with multiple mating by female *A. persicus*, compromises the potential for using sterile females to control this, and possibly other, argasid tick species. Thus, like ixodid ticks, the SIT method has some potential for suppressing argasid tick density, but numerous factors need to be considered and accepted before sterilized ticks are released in nature.

5.2. Hybrid Sterility

A second form of reproductive interference is based upon the fact that similar species of ticks may mate and produce hybrid offspring that are sterile. Moreover, subsequent mating between hybrid males and wild-type females results in the production of non-viable eggs. Accordingly, if enough hybrid matings occur, a decrease in density of the wild-type populations will occur. Interspecific matings resulting in sterile hybrids have been reported for several tick species, including *B. microplus* and *B. annulatus* (Graham et al., 1972), *B. microplus* and *B. decoloratus* (Spickett and Malan, 1978), *A. variegatum* and *A. hebraeum* (Rechav et al., 1982), and at least under laboratory conditions between *Rhipicephalus appendiculatus* and *R. zambeziensis* (Wouters, 1990). The displacement of one tick species with another through hybrid mating would be

beneficial where economic damage, such as transmission of a disease-causing agent, was reduced or eliminated by establishment of the secondary species. For example, *B. annulatus* and *B. decoloratus* are inefficient vectors of *Babesia bigemina*, an important pathogen of cattle, that is normally transmitted by *B. microplus*.

The genesis for pursuing hybrid sterility as a possible means for controlling tick populations followed observations that natural interbreeding occurs between *B. microplus* and *B. annulatus* (Graham and Price, 1966). Cross-mating between these species resulted in the production of 99% sterile hybrid males (Graham et al., 1972), and fertile hybrid females mated (back crossed) to males of the original (parent) species produce sterile males. Hybrid male sterility persists for three generations (Thompson et al., 1981), while sterile males survive as long and mate with as many females as pure strain males (Davey et al., 1983) and hybrid larvae are as long lived as pure-strain larvae. Consequently, Osburn and Knipling (1982) projected that the release of hybrid *Boophilus* ticks in numbers sufficient to exceed substantially those of a natural population would theoretically be feasible as a means of suppressing or eradicating *B. microplus*. This approach was envisioned for use where inundative release of hybrid males would induce sterility in a population of recently established ticks such that the population would be autocidally reduced or eradicated. As discussed by Davey et al. (1993), the success of a sterile hybrid release program also depends upon the response of hybrid ticks to environmental conditions; for example, if differences exist between the oviposition, fecundity, non-parasitic development, and survival of hybrid and pure-strain ticks that favor the pure strain, the sterile-hybrid technology will be of little value.

As evidence of these considerations, assortative mating (e.g., greater mating among conspecific males and females) was observed with mixed populations of *B. microplus* and *B. decoloratus* (Norval and Sutherst, 1986), and non-random mating occurred when *B. microplus* and type 2 *B. microplus* hybrids were released on cattle (Hilburn et al., 1991). In the latter instance, mating was non-random because hybrid males took longer to reach mating maturity and were not as competitive in establishing mating pairs as male pure-strain *B. microplus*. In a theoretical context, simulation modeling indicates that asymmetrical mating interference (satyrism) is reflected in reduced reproductive success and reduced mating competitiveness (Sutherst, 1987; Ribiero, 1988). The greater number of sterile males needed to overcome compromised male competitiveness could therefore have a strong negative impact on the success of a sterile male release program. On the other hand, recent information concerning the oviposition, fecundity, and non-parasitic stage development of *Boophilus* type 2 hybrids (Davey et al., 1993), indicates that the percentage of ovipositing hybrid and pure strain females is similar and uniformly high, and that the number of eggs produced by hybrid females does not differ from pure strain females. Further, little difference was observed in non-parasitic development times between hybrid and pure-strain ticks throughout the year, which indicates no selective advantage that favors either hybrid or wild populations. A disparity in egg viability that favored production of wild-strain larvae was

noted, however, and because the success of a sterile-hybrid release program is contingent upon the hybrid population overwhelming the native tick population, factors such as this need to be identified and counteracted.

6. PARASITOIDS AND PREDATORS (HYMENOPTERA)

Populations of strongly K-adapted species are generally not effectively regulated by parasites and predators, although the impact of natural enemies on less strongly K-adapted species can be considerable. The role of biological agents in regulating tick populations is poorly appreciated, and for the most part poorly investigated. For example, the extensive literature concerning the bionomics and control of *A. americanum* contains only limited studies on biological control agents or their prospects for regulating populations of lone star ticks. Spickett (1987) provides a general review of biologic agents associated with ticks.

The best-known metazoan parasites of ticks are parasitic wasps (parasitoids) of the family Encyrtidae. In keeping with the term "parasitoid," these free-living wasps oviposit only in ticks, both unattached and attached larvae and nymphs. The eggs of tick parasitoids lay dormant through periods of tick inactivity and molting, hatch with blood-feeding, and the developing larvae then consume the internal contents and pupate in the corpse. Females of the six species of tick parasitoids (Cole, 1965) lay multiple eggs per tick and oviposit in more than one tick. The most publicized tick parasitoid, *Hunterellus hookeri*, is indigenous to warmer areas of the world where it is prevalent in nymphs of the Brown dog tick, *Rhipicephalus sanguineus* (Smith and Cole, 1943). This species also apparently exists throughout the Palearctic region, where it has been reported from *Ixodes* and *Haemaphysalis* tick species (Cole, 1965). Larrousse et al. (1928) released *H. hookeri* collected in France on an island off the coast of Massachusetts for controlling American dog ticks, *D. variabilis*. This release apparently had no measurable effect on the abundance of American dog ticks, but it presumably led to the establishment of *H. hookeri* in North America, plausibly in association with *Ixodes scapularis* (=*dammini*). In a subsequent study conducted in the same area, Smith and Cole (1943) recovered one male and four female *H. hookeri* from an *I. scapularis* nymph attached to a meadow mouse, but 284 other *I. scapularis* nymphs and 2,143 nymphal *D. variabilis* were negative. In another trial, an estimated 44,000 adult *H. hookeri* were released in a cranberry bog, along with 55 meadow voles each carrying about 20 parasitized nymphs. A second release consisted of 47,000 wasps released in a beach grass habitat, along with 6,000 female wasps and 32 white-footed mice each carrying from 20 to 25 parasitized ticks. Immature *D. variabilis* density was low in the cranberry bog following release of *H. hookeri*, and none of 24 nymphs collected from rodents were infected. A year later, only 27 *D. variabilis* nymphs were taken on mice, none of which were infected with *H. hookeri*. Theiler (1969) summarized these studies, stating "the results of work done in America on *H. hookeri* were so disappointing that no one has had the courage to try it again."

Many basic questions about the biology of *H. hookeri* and other tick parasitoids remain to be answered. In the Smith and Cole (1943) study, for example, *H. hookeri* was mass reared and released for control of *D. variabilis*, irrespective of the fact that the prey-search behavior of female *H. hookeri* which, as a parasite of *Ixodes* ticks, may be keyed to locating immature ticks on *P. leucopus* that frequent woodland habitats, rather than immature *D. variabilis* on meadow voles in grassland vegetation. Questions about the timing of experimental release relative to the phenology of both parasitoid and host, a key factor requisite to successful use of parasitoids for control of insects (Stinner, 1977), also remain unresolved. In Australia, Sutherst et al. (1978) suggested that *H. hookeri* would not be effective in regulating populations of *B. microplus* as wasp density could not keep pace with increases in the density of this multivoltine tick.

On the other hand, 27% of host-seeking nymphal *I. dammini* on Naushon Island, near the release site of *H. hookeri* by Larousse in 1926, were infected with *H. hookeri* (Mather et al., 1987). This parasitoid population is of further interest in that only nymphs devoid of *B. burgdorferi*, the etiologic agent of Lyme disease, were parasitized by *H. hookeri*, whereas 14–22% of non-parasitized nymphs were infected with *B. burgdorferi*. This relationship was judged to reflect an antagonism, conceivably mediated by the tick "immune" system, between *H. hookeri* and *B. burgdorferi*. The possibility that female *H. hookeri* oviposit exclusively in larvae feeding or fed on non-spirochete donor hosts, such as white-tailed deer, also was considered; nymphs derived from such larvae would not be infected with *B. burgdorferi*, but might harbor *H. hookeri*. Indeed, it was subsequently observed that nymphs derived from larvae that fed previously on white-footed mice were not parasitized by *H. hookeri*, whereas some nymphs from larvae that fed on white-tailed deer, which are not a source of *B. burgdorferi*, were parasitized (Mather, unpublished). This relationship provides useful insight into the host-searching behavior of *H. hookeri*, as well as clarifying the potential for using *H. hookeri* to suppress *I. dammini* populations. Because larval *I. dammini* in the eastern US feed largely on small rodents, particularly white-footed mice, the potential for suppressing *I. dammini* with *H. hookeri* appears to be limited, even where wasp density is high. On the other hand, if a tick parasitoid preferentially parasitized ticks feeding on a host that serves as the reservoir of a disease agent, the prospect for using the parasitoid would differ considerably.

In selecting potential biocontrol agents, Stinner (1977) outlined needs for the following baseline information: (1) an appreciation for the general adaptive features of the predator; (2) an analysis of the predator's searching capacity; and (3) knowledge of the rate of predator increase relative to prey increase. Other components of successful predator establishment and release programs include pre-release conditioning, determination of optimum release times, distribution, and number. It also is noteworthy that predator populations can be selected genetically for increased efficiency of predation. Thus, in the absence of even rudimentary information about tick parasitoids, it is indeed premature to judge the potential for tick parasitoids to regulate tick populations.

6.1. Ants

In contrast to tick-specific wasp parasitoids, ants are generalist predators that prey on a wide variety of terrestrial arthropods, including ticks. In the French West Indies, the tropical fire ant, *Solenopsis geminata*, is reported to feed readily on engorged, but not unengorged, female *A. variegatum* (Barre et al., 1991). The tropical fire ant attacks engorged larvae, nymphs, and adults of *B. microplus* with a stereotypic behavior in which the legs are cut off near the coxae, and then the ant penetrates the tick and removes internal contents that are returned to the mound. While one group of ants disassembles the tick a second group, perhaps summoned to the prey site, buries it by removing soil from under the corpse (Colon-Guaspe, 1985, cited in Barre et al., 1991). Predation by the tropical fire ant is considered a significant mortality factor in *Boophilus microplus* populations (Butler et al., 1979; Colon-Guaspe, 1985).

In the southeastern US, the establishment and spread of the imported fire ant, *S. invicta*, has aided control of several species of harmful arthropods, including the lone star tick. This ant is highly omnivorous, feeds opportunistically upon any plant or animal materials that it encounters (Lofgren 1975), and is therefore a serious pest in itself. In Louisiana, the spread of *S. invicta* through mixed hardwood timberlands once heavily infested with lone star ticks has been accompanied by a "marked reduction, in fact a virtual disappearance, of ticks from the areas" (Burns and Melancon, 1977). Moreover, in a comparison of lone star tick density in areas treated to control fire ants and similar but untreated areas Burns and Melancon (1977) reported consistent and progressive decreases in the number of adults and nymphs responding to CO_2 bait in untreated areas. Likewise, monitoring lone star tick density in several areas before and after fire ant invasion indicated a near-total eradication of ticks over a 2-year period.

Harris and Burns (1972) demonstrated that *S. invicta* is predaceous on eggs and all engorged life stages of *A. americanum*, and Fleetwood et al. (1984) showed that *S. invicta* predation on lone star ticks set out experimentally in open areas and adjacent post-oak thickets was the greatest mortality factor in either habitat. Higher tick mortality occurred in open pasture habitat, and peak predation, 100% in pasture areas and 59.2% in post-oak thickets, occurred in late summer. Thus all evidence indicates that lone star tick populations are strongly affected by the imported fire ant. Predation on engorged females reduces tick progeny, with the level of predation depending upon time of year and habitat.

Other ant species also have been associated with tick predation, as exemplified by the abundance of *Pheidole megacephala*, an introduced species, in areas of "reputed tick (*Boophilus microplus*) scarcity" in Australia (Wilkinson, 1970). This ant also preys on *A. cajennense* in Cuba (Diego et al., 1983). In experimental studies in Kenya, *P. megacephala* was one of several predators feeding on engorged *B. microplus* set out in short and long grass habitats, where predation rates of 6.8 and 7.1% occurred, respectively (Mwangi et al., 1991).

These studies document the presence of hymenopterous parasitoids and ants associated with ticks in widespread habitats around the world. In the

context of biological control programs developed for insect pests, however, only scant information exists concerning their potential for biocontrol of ticks.

7. INTEGRATED PEST (TICK) MANAGEMENT

Integrated pest management (IPM) can be defined in a number of ways, but all definitions include the integration of more technologies to control a pest species relative to an economic-damage or injury-level threshold that is typically density dependent. IPM has been successful as a rational approach to suppressing pest populations while simultaneously encouraging integration of chemical and alternative control methods. IPM is based upon ecologic processes that regulate populations, with strong consideration to the environmental consequences of pest suppression actions.

Two examples of IPM programs for ticks will be considered, one involving *B. microplus* associated with pastured beef cattle in Australia, the second developed for the lone star tick in the US.

7.1. IPM: *Boophilus microplus*

Under tropical pasture conditions in northern Queensland, *B. microplus* complete 7–10 generations annually and attain densities of up to 10,000 ticks per head on European-breed cattle; *B. microplus* infestations cause reduced weight gain, lower feed conversion efficiency, lower milk yield, and cause hide damage along with being an efficient vector of *Babesia bigemina*, the causative agent of redwater fever. Economic losses attributed to *B. microplus* in Australia and South America were estimated at 100 million and one billion dollars annually, respectively (Horn, 1987; Cobon and Willadsen, 1990). The cattle industry in Queensland, Australia has been plagued by genetic resistance in *B. microplus* to acaricides that resulted from an excessive dependence on chemical control (e.g., dipping cattle) (Sutherst, 1981). High resistance to acaricides has forced the development and use of alternative control methods, particularly pasture spelling and use of tick-resistant cross-breed cattle to suppress tick infestation. Acceptance by cattlemen of tick-resistant cattle, principally Zebu (*Bos indicus*) × European (*Bos taurus*) breeds, has reduced the impact of *B. microplus* infestation from one that threatens a national catastrophe to a condition that still causes economic loss, but does not threaten the very existence of the industry. In a practical sense, the use of tick-resistant cattle modifies the economic injury threshold level, thereby reducing dependency on chemical control (dipping) early in the season when tick density on cattle does not exceed costs of control. In turn, this permits culling and further selection for tick resistance within herds, as well as suppression of tick density based on the understanding that acaricide-resistant ticks that survive dipping in late season are unlikely to produce progeny due to low ambient temperatures.

Thus, in limiting acaricide dipping, this IPM program results in a low-risk minimum-management situation that frees capital, labor, and management for more productive farm activities (Sutherst, 1981).

The host resistance employed in regulating the density of *B. microplus* populations is genetically conferred through breeding of European-breed cattle (*Bos taurus*) with Brahma cattle (*Bos indicus*), and varies from breed to breed and among individuals within breeds (Utech et al., 1978). In highly tick-resistant breeds, such as the true-breeding Droughtmaster (beef breed) and Sahiwal (dairy breed) blood lines, up to 50% of European-breed genes are retained to conserve traits of rapid weight gain and high milk production (Utech et al., 1978). In these cattle, host resistance is expressed largely as an immediate hypersensitivity reaction to feeding larvae, where elevated levels of histamine at attachment sites lead to detachment of larvae (Kemp and Bourne, 1980), tick desiccation and death (Agbede and Kemp, 1985). Hypersensitive reactions at the attachment site also induce licking, rubbing and scratching that dislodge or squash ticks; mutual grooming (allogrooming) of ticks attached in inaccessible areas of host anatomy also occurs (see de Castro and Newson, 1993).

7.2. IPM: *Amblyomma americanum*

A second integrated tick management program, developed for the lone star tick, consists of three basic steps: (1) identifying the tick problem; (2), defining the tick problem by censusing the tick population; and (C) managing the tick problem (Barnard et al., 1988). Identifying the tick problem can be based on tick infestations on vegetation or host animals. Defining the tick problem is done by sampling methods appropriate for collecting and identifying the species and stage of tick, and hosts at risk. The decision to manage ticks is influenced by a relationship in which the benefits of control, expressed as reduced tick abundance, are balanced against the costs of control. In human recreational areas, Mount and Dunn (1983) suggested an economic threshold level of 0.65 adult lone star ticks per CO_2 trap. Several types of acaricide formulations and applicators are recommended for area-wide emergency or seasonal control of lone star ticks, including guidelines for use (Barnard et al., 1988).

Computer simulations (Mount and Haile, 1987) indicate that densities of the lone star tick should be reduced to tolerable levels within 2 years by implementing any of the following integrated tick management strategies: (1) acaricide application in weeks 16 and 27 combined with vegetation management at the 50% level in years 1, 2, 5 and 9; (2) acaricide application in weeks 16 and 27 in year 1, and in week 16 of year 2, combined with host management at the 90% level in years 1–3, and vegetation management at the 75% level in years 4–10; and (3) acaricide application in week 1 combined with host management at the 50% level in all years and vegetation management at the 50% level in years 1, 2, 5, and 9.

8. SUMMARY

In a collective context, the aforementioned integrated tick management programs summarize several concepts essential to effective control of tick populations. First, long-term suppression of a tick population cannot be expected with a one-method, one-treatment approach. Tick populations are intimately associated with plant and animal components of respective communities, and therefore a combination of strategies that focus on tick interactions with both vegetation and animal hosts offer the best promise of success. This interpretation is supported by both field studies and simulation-model estimates, both of which indicate that the integration of two control methods provides more effective and more lasting suppression of tick population density than either method alone. Lastly, irrespective of the control strategy or strategies employed, a well-planned and persistent effort, logically involving a host-targeted method, is necessary to counter tick re-introduction and population regrowth.

9. REFERENCES

Agbede, R.I.S. and Kemp, D.H. (1985) Digestion in the cattle-tick *Boophilus microplus*: light microscope study of the gut cells in nymphs and females. *Internat. J. Parasitol.* **15**, 147–157.

Ahrens, E.C. and Cocke, J. (1978) Comparative test with insecticide-impregnated ear tags against the Gulf coast tick. *J. Econ. Entomol.* **71**, 764–767.

Ailam, G. and Galun, R. (1967) Optimal sex ratio for the control of insects by the sterility method. *Ann. Entomol. Soc. Am.* **60**, 41–43.

Allan, S.A., Phillips, J.S., Taylor, D., and Sonenshine, D.E. (1988) Genital sex pheromones of Ixodid ticks: evidence for the role of fatty acids from the anterior reproductive tract in mating of *Dermacentor variabilis* and *Dermacentor andersoni*. *J. Insect. Physiol.* **34**, 315–323.

Allen, J.R. (1973) Tick resistance: basophils in skin reactions of resistant guinea pigs. *Internat. J. Parasitol.* **3**, 195–200.

Allen, J.R. (1992) An overview of progress in characterizing host immunity to ticks. pp. 206–211. *In:* Munderloh, U.G. and Kurtti, T.J. (eds) *First International Conference on Tick-borne Pathogens at the Host–Vector Interface: An Agenda for Research.* University of Minnesota, St. Paul.

Allen, J.R., Khalil, H.M., and Wikel, S.M. (1979) Langerhans cells trap tick salivary gland antigens in tick-resistant guinea pigs. *J. Immunol.* **122**, 563–565.

Allen, J.R. and Kemp, D.H. (1982) Observations on the behavior of *Dermacentor andersoni* larvae infesting normal and tick-resistant guinea pigs. *Parasitol.* **84**, 195–204.

Awerbuch, T., Sandberg, S., Deblinger, R., and Spielman, A. (1992) Effect of simulated deer reduction on the abundance of the vector of Lyme disease. *Am. J. Trop. Med. Hyg.* **47** (Suppl. 4), 214.

Barre, N., Mauleon, H., Garris, G.I., and Kermanec, A. (1991) Predators of the tick *Amblyomma variegatum* (Acari: Ixodidae) in Guadeloupe, French West Indies. *Exp. Appl. Acarol.* **12**, 163–170.

Barnard, D.R. (1985) Injury thresholds and production loss functions from the lone star

tick, *Amblyomma americanum* (Acari: Ixodidae), on pastured, pre-weaner beef cattle, *Bos taurus*. *J. Econ. Entomol.* **78**, 852–855.

Barnard, D.R. (1986) Density perturbation in populations of *Amblyomma americanum* (Acari: Ixodidae) in beef cattle forate areas in response to two regimens of vegetation management. *J. Econ. Entomol.* **79**, 122–127.

Barnard, D.R., Mount, G.A., Koch, H.G., Haile, D.G., and Garris, G.I. (1988) Management of the lone star tick in recreation areas. *U.S. Dept. Agric. Hdbk*, p. 682.

Beadles, M.L., Gingrich, A.R., and Miller, J.A. (1977) Slow-release devices for livestock insect control: cattle body surfaces contacted by five types of devices. *J. Econ. Entomol.* **70**, 72–75.

Bloemer, S.R., Snoddy, E.L., Cooney, J.C., and Fairbanks, K. (1986) Influence of deer exclusion on populations of lone star ticks and American dog ticks (Acari: Ixodidae). *J. Econ. Entomol.* **79**, 679–683.

Bloemer, S.R., Mount, G.A., Morris, A., Zimmerman, R.H., Barnard, D.R., and Snoddy, E.L. (1990) Management of lone star ticks (Acari: Ixodidae) in recreational areas with acaricide applications, vegetation management, and exclusion of white-tailed deer. *J. Med. Entomol.* **27**, 543–550.

Burns, E.C. and Melancon, D.G. (1977) Effect of imported fire ant (Hymenoptera: Formicidae) invasion on lone star tick (Acarina: Ixodidae) populations. *J. Med. Entomol.* **14**, 247–249.

Butler, J.F., Camino, M.L., and Perez, T. (1979) *Boophilus microplus* and the fire ant *Solenopsis feminata*. Vol. 1, pp. 469–472. *In:* Rodriguez, J.G (ed) *Recent Advances in Acarology*. Academic Press, New York.

Carey, A.B., Krinsky, W.L., and Main, A.J. (1980) *Ixodes dammini* (Acari: Ixodidae) and associated ixodid ticks in south-central Connecticut, USA. *J. Med. Entomol.* **17**, 89–99.

Clymer, B.C., Howell, D.E., and Hair, J.A. (1970) Environmental alteration in recreational areas by mechanical and chemical treatment as a means of lone star tick control *J. Econ. Entomol.* **63**, 504–509.

Cobon, G.S. and Willadsen, P. (1990) Vaccines to prevent tick infestations. pp. 901–909. *In:* Woodrow, G.C. and Levine, M.M. (eds) *New Generation Vaccines*. Dekker, New York.

Cole, M.M. (1965) Biological control of ticks by the use of hymenopterous parasites. A review. *WHO/EBL*, 43.65.

Conway, G.R. (1976) Man versus pests. pp. 257–281. *In:* May, R.M. (ed) *Theoretical Ecology: Principles and Applications*. Blackwell, Oxford.

Daniels, T.J., Fish, D., and Falco, R.C. (1991) Evaluation of host-targeted acaricide for reducing risk of Lyme disease in Southern New York State. *J. Med. Entomol.* **28**, 537–543.

Davey, R.B., Osburn, R.L., and Castillo, C. (1983) Longevity and mating behavior in males and parthenogenesis in females in hybridized *Boophilus* ticks (Acari: Ixodidae). *J. Med. Entomol.* **20**, 614–617.

Davey, R.B., Pound, J.M., and Cooksey, L.M. (1993) Comparative reproduction and nonparasitic development of *Boophilus microplus* and hybridized *Boophilus* ticks (Acari: Ixodidae) under natural field conditions in subtropical South Texas. *Exp. Appl. Acarol.* (in press).

Davidar, P., Wilson, M., and Ribeiro, J.M.C. (1989) Differential distribution of immature *Ixodes dammini* (Acari: Ixodidae) on rodent hosts. *J. Parasitol.* **75**, 898–904.

Deblinger, R.D. and Rimmer, D.W. (1991) Efficacy of a permethrin-based acaricide to

reduce the abundance of *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **28**, 708–711.

Deblinger, R.D., Wilson, M.L., Rimmer, D.W., and Spielman, A. (1993) Reduced abundance of immature *Ixodes dammini* (Acari: Ixodidae) following incremental removal of deer. *J. Med. Entomol.* **30**, 144–150.

de Castro, J.J. and Newson, R.M. (1993) Host resistance in cattle tick control. *Parasitol. Today* **9**, 13–14.

Diego, J.G., Villalba, G., Abrey, R., and Castaneiras, A. (1983) *Pheidole megacephala* como de-predator de *Amblyomma cajennense* en Cuba. *Rev. Salud. Anim.* **5**, 437–440.

Drummond, R.O., Medley, J.G., and Graham, O.H. (1966) Engorgement and reproduction of lone star ticks (*Amblyomma americanum* (L.) treated with gamma-radiation. *Internat. J. Rad. Biol.* **10**, 183–188.

Duncan, I.M. and Monks, N. (1992) Tick control on eland (*Taurotragus oryx*) and buffalo (*Syncerus caffer*). *J. South African Vet. Assoc.* **63**, 7–10.

Dusbábek, F., Simek, P., Jegorov, A., and Triska, J. (1991) Identification of xanthine and hypoxanthine as components of assembly pheromone in excreta of argasid ticks. *Exp. Appl. Acarol.* **11**, 307–316.

Fish, D. and Dowler, R. (1989) Host associations of ticks (Acari: Ixodidae) parasitizing medium-sized mammals in a Lyme disease endemic area of southern New York. *J. Med. Entomol.* **26**, 200–209.

Fivaz, B.H., Rencken, I., Cross, R., Terry, S., and Henry, D. (1991). Specificity and effects of host resistance on the African tick *Rhipicephalus zambeziensis* (Acarina: Ixodidae). *Exp. Appl. Acarol.* **11**, 223–232.

Fleetwood, S.C., Teel, P.D., and Thompson, G. (1984) Impact of imported fire ant on lone star tick mortality in open and canopied pasture habitats of East Central Texas. *Southwest Entomol.* **9**, 158–162.

Galun, R., and Warburg, M. (1967) Irradiation effects on respiration and on blood digestion in the tick *Ornithodoros tholozani* and its importance for the sterile male technique. pp. 248–257. In: *IALA Symp. Proc. Isotopes and Radiation in Entomology*. International Atomic Energy Agency, Vienna.

Galun, R., Sternberg, S., and Mango, C. (1972) The use of sterile females for the control of the tick *Argas persicus* (Oken), *Israel J. Entomol.* **7**, 109.

Gladney, W.J., Grabbe, R.R., Ernst, S.E., and Oehler, D.D. (1974) The Gulf coast tick: evidence of a pheromone produced by males. *J. Med. Entomol.* **3**, 303–306.

Ginsberg, H.S. (1992) Ecology and management of ticks and Lyme disease at Fire Island National Seashore and selected eastern natural parks. *National Park Service Sci. Monogr.* NPS/NRSUNJ/NRSM-92/20, 77 pp.

Graf, J.F. (1975) Ecologie et éthologie d'*Ixodes ricinus* L. en Suisse (Ixodoidea: Ixodidae). Cinquième note: mise un évidence d'une phéromone sexuelle chez *Ixodes ricinus*. *Acarologia* **17**, 436–441.

Graham, O.H. and Price, M.A. (1966) Some morphological variations in *Boophilus annulatus* and *B. microplus* (Acarina: Ixodidae) from northern Mexico. *Ann. Entomol. Soc. Am.* **59**, 450–452.

Graham, O.H., Price, M.A., and Tevino, J.L. (1972) Cross-mating experiments with *Boophilus annulatus* and *B. microplus* (Acarina: Ixodidae). *J. Med. Entomol.* **9**, 531–537.

Gray, J.S., Kahl, O., Janetzke, C., and Stein, J. (1992) Studies on the ecology of Lyme disease in a deer forest in County Galway, Ireland. *J. Med. Entomol.* **29**, 915–920.

Haile, D.G. and Mount, G.A. (1987) Computer simulation of population dynamics of the lone star tick, *Amblyomma americanum* (L.) (Acari: Ixodidae). *J. Med. Entomol.* **24**, 356–369.

Hair, J.A. and Howell, D.E. (1970) Lone star ticks: their biology and control in Ozark recreation areas. *Oklahoma Agric. Exp. Stn. Bull.* B-679.

Hamilton, J.G.C. (1992) The role of pheromones in tick biology. *Parasitol. Today* **8**, 130–133.

Hamilton, J.G.C. and Sonenshine, D.E. (1988) Evidence for the occurrence of mounting sex pheromone on body surface of female *Dermacentor variabilis* (Say) (Acari: Ixodidae). *J. Chem. Ecol.* **14**, 401–410.

Harris, W.G. and Burns, E.C. (1972) Predation of the lone star ticks by the imported fire ant. *Environ. Entomol.* **1**, 362–365.

Hilburn, L.R., Davey, R.B., George, J.E., and Pound, J.M. (1991) Non-random mating between *Boophilus microplus* and hybrids of *B. microplus* females and *B. annulatus* males, and its possible effect on sterile male hybrid control releases. *Exp. Appl. Acarol.* **11**, 23–36.

Hoch, A.L., Barker, R.W., and Hair,, J.A. (1971a) Measurement of physical parameters to determine the suitability of modified woodlots as lone star tick habitat *J. Med. Entomol.* **8**, 725–730.

Hoch, A.L., Barker, R.W., and Hair, J.A. (1971b) Further observations on the control of lone star ticks (Acarina: Ixodidae) through integrated control procedures. *J. Med. Entomol.* **8**, 731–734.

Hoch, A.L., Semter, P.J., Barker, R.W., and Hair, J.A. (1972) Preliminary observations on controlled burning for lone star tick (Acarina: Ixodidae) control in woodlots. *J. Med. Entomol.* **9**, 446–451.

Hoogstraal, H. (1976) Acarina (ticks). pp. 89–103. *In:* Gibbs, A.J (ed) *Viruses and Invertebrates.* North-Holland Publishing Co., Amsterdam.

Horn, S.C. (1987) Bovine ectoparasites and their economic inpact in South America. pp. 25–27. *In:* Leaning, W.H.D. and Guerro, J. (eds) *The Economic Impact of Parasitism in Cattle.* Proceedings of the MSD AGVET Symposium XXIII World Veterinary Congress, Montreal, Canada.

Kaufman, W.R. (1989) Tick-host interaction: a synthesis of current concepts. *Parasitol. Today* **5**, 47–56.

Kartman, L. (1958) An insecticide-baitbox method for the control of sylvatic plague vectors. *J. Hyg. Camb.* **56**, 405–465.

Kemp, D.J. and Bourne, A. (1980) *Boophilus microplus*: the effect of histamine on the attachment of cattle-tick larvae—studies in vivo and in vitro. *Parasitol.* **80**, 487–496.

Kemp, D.H., Agbede, R.I.S., Johnston, L.A.Y., and Gough, J.M. (1986) Immunization of cattle against *Boophilus microplus* using extracts derived from adult female ticks: feeding and survival of the parasite on vaccinated cattle. *Internat. J. Parasitol.* **16**, 115–120.

Knipling, E.F. (1979) The basic principles of insect population suppression and management. *Agric. Hdbk* 512. US Printing Office, Washington, D.C.

Larrousse, B., King, A.G., and Wolbach, S.B. (1928) The overwintering in Massachusetts of *Ixodiphagus caucurtei. Science* **67**, 351–353.

Koudstaal, D., Kemp, D.H., and Kerr, J.D. (1978) *Boophilus microplus*: rejection of larvae from British breed cattle. *Parasitol.* **76**, 379–386.

Lofgren, C.S., Banks, W.A., and Glancy, B.M. (1975) Biology and control of imported fire ants. *Annu. Rev. Entomol.* **20**, 1–30.

Malonza, M.M., Dipeolu, O.O., Amoo, A.O., and Hassan, S.M. (1992) Laboratory and field observations on anti-tick properties of the plant *Gynandropsis gynandra* (L.) Brig. *Vet. Parasitol.* **42**, 123–136.

Mather, T.N. and Spielman, A. (1986) Diurnal detachment of immature deer ticks (*Ixodes dammini*) from nocturnal hosts. *Am. J. Trop. Med. Hyg.* **35**, 182–186.

Mather, T.N., Riberio, J.M.C., and Spielman, A. (1987) Lyme disease and Babesiosis: acaricide focused on potentially infected ticks. *Am. J. Trop. Med. Hyg.* **36**, 609–614.

Mather, T.N., Riberio, J.M.C., Moore, S.I., and Spielman, A. (1988) Reducing transmission of Lyme disease spirochetes in a suburban setting. *Ann. New York Acad. Sci* **539**, 402–403.

Mather, T.N., Duffy, D.C., and Campbell, S.R. (1993) An unexpected result from burning vegetation to reduce Lyme disease transmission risk. *J. Med. Entomol.* **30**, 642–645.

McSwain, J.L., Essenberg, R.C., and Sauer, J.R. (1992) Oral secretion elicited by effect of signal transduction pathways in the salivary glands of *Amblyomma americanum* (Acari: Ixodidae). *J. Med. Entomol.* **29**, 41–48.

Meyer, J.A., Lancaster, J.R., and Simco, J.S. (1982) Comparison of habitat modification, animal control, and standard spraying for control of the lone star tick. *J. Econ. Entomol.* **75**, 524–528.

Miller, J.A., Garris, G.I., George, J.E., and Oehler, D.D. (1989) Control of lone star ticks (Acari: Ixodidae) on Spanish goats and white-tailed deer with orally administered invermectin. *J. Econ. Entomol.* **82**, 1650–1656.

Mount, G.A. (1981) Control of the lone star tick in Oklahoma parks through vegetative management. *J. Econ. Entomol.* **74**, 173–175.

Mount, G.A. and Dunn, J.E. (1983) Economic thresholds for lone star ticks (Acarina: Ixodidae) in recreational areas based on a relationship between CO and human subject sampling. *J. Econ. Entomol.* **76**, 327–329.

Mount, G.A. and Haile, D.G. (1987) Computer simulation of area-wide management strategies for the lone star tick, *Amblyomma americanum* (Acari: Ixodidae). *J. Med. Entomol.* **24**, 523–531.

Mwangi, E.N., Newson, R.M., and Kaaya, G.P. (1991) Predation of free-living engorged female *Rhipicephalus appendiculatus*. *Exp. Appl. Acarol.* **12**, 153–162.

Need, J.T., Butler, J.F., and Peck, A.B. (1992) Cellular responses of laboratory mice to feedings of *Ornithodoros talaje* (Acari: Argasidae). *J. Med. Entomol.* **29**, 423–429.

Needham, G.R. and Teel, P.D. (1991) Off-host physiological ecology of Ixodid ticks. *Annu. Rev. Entomol.* **36**, 659–681.

Norval, R.A.I. and Sutherst, R.W. (1986). Assortative mating between *Boophilus decoloratus* and *Boophilus microplus* (Acari: Ixodidae). *J. Med. Entomol.* **23**, 459–460.

Norval, R.A.I., Floyd, R.B., and Kerr, J.D. (1988) Ability of adults of *Amblyomma hebraeum* (Acarina: Ixodidae) to feed repeatedly on sheep and cattle. *Vet. Parasitol.* **29**, 351–355.

Norval, R.A.I., Andrew, H.R., and Yunker, C.E. (1989a). Pheromone-mediation of host selection in bont ticks (*A. hebraeum*) Koch. *Science* **243**, 364–365.

Norval, R.A.I., Butler, J.F., and Yunker, C.E. (1989b) Use of carbon dioxide and natural or synthetic aggregation–attachment pheromone of the bont tick, *Amblyomma hebraeum*, to attract and trap unfed adults in the field. *Exp. Appl. Acarol.* **7**, 171–180.

Norval, R.A.I., Yunker, C.E., Duncan, I.M., and Peter, T. (1991) Pheromone/acaricide mixtures in control of the tick *Amblyomma hebraeum*: effects of acaricides on attraction and attachment. *Exp. Appl. Acarol.* **11**, 233–240.

Norval, R.A.I., Peter, T., and Meltzer, M.I. (1992a) A comparison of the attraction of nymphs and adults of the ticks *Amblyomma hebraeum* and *A. variegatum* to carbon dioxide and the male-produced aggregation–attachment pheromone. *Exp. Appl. Acarol.* **13**, 179–186.

Norval, R.A.I., Sonenshine, D.E., and Peter, T. (1992b) Attraction/aggregation/attachment (AAAP) of the bont ticks, *Amblyomma hebraeum* and *A. variegatum*. pp. 324–327. *In:* Munderloh, U.G. and Kurtti, T.J. (eds) *First International Conference on Tick-borne Pathogens at the Host–*Vector Interface*: An Agenda for Research.* University of Minnesota, St. Paul.

Obenchain, F.D. (1979) Non-acaricide chemicals for the management of Acari of medical and veterinary importance. Vol. 2, pp. 35–43. *In:* Rodriguez, R.D. (ed) *Recent Advances in Acarology.* Academic Press, New York.

Olkowski, W. (1990) Appendix 3. Deterring or excluding deer. *Common Sense Pest Control.* **VI**, 24–25.

Oxburn, R.L. and Knipling, E.F. (1982) The potential use of sterile hybrid *Boophilus* ticks (Acari: Ixodidae) as a supplemental eradication technique. *J. Med. Entomol.* **19**, 637–644.

Paine, S.H., Kemp, D.H., and Allen, J.R. (1983) *In vitro* feeding of *Dermacentor andersoni* (Stiles): effects of histamine and other mediators. *Parasitol.* **86**, 419–428.

Piesman, J., Spielman, A., Etkind, P., Raebush, T.K., II, and Juranek, D.D. (1979) Role of deer in the epizootology of *Babesia microti* in Massachusetts, USA. *J. Med. Entomol.* **15**, 537–540.

Presley, S.M. and Hair, J.A. (1988) Lone star tick (Acari: Ixodidae) management by host manipulation through habitat modification. *J. Med. Entomol.* **25**, 78–81.

Ramachandra, R.N. and Wikel, S.K. (1992) Modulation of host-immune responses by ticks (Acari: Ixodidae): effect of salivary gland extracts on host macrophages and lymphocyte cytokine production. *J. Med. Entomol.* **29**, 818–826.

Randolph, S.E. and Steele, G.M. (1985) An experimental evaluation of conventional control measures against the sheep tick, *Ixodes ricinus* (L.) (Acari: Ixodidae). II. The dynamics of the tick–host interaction. *Bull. Entomol. Res.* **75**, 501–518.

Rechav, Y. (1992) The potential use of tick homogenates for immunization of livestock in less developed countries with special reference to Africa. pp. 219–225. *In:* Munderloh, U.G. and Kurtti, T.J. (eds) *First International Conference on Tick-Borne Pathogens at the Host–Vector Interface: An Agenda for Research.* University of Minnesota, St. Paul.

Rechav, Y. and Whitehead, G.B. (1978) Field trials with pheromone–acaricide mixtures for control of *Amblyomma hebraeum*. *J. Econ. Entomol.* **71**, 149–151.

Rechav, Y., Whitehead, G.B., and Knight, M.M. (1976) Aggregation response of nymphs to pheromones(s) produced by males of the tick *Amblyomma hebraeum* Koch. *Nature* **259**, 563–564.

Rechav, Y., Parolis, H., Whitehead, G.B., and Knight, M.M. (1977) Evidence of an assembly pheromone(s) produced by males of the bont tick, *Amblyomma hebraeum* (Acarina: Ixodidae). *J. Med. Entomol.* **14**, 71–78.

Rechav, Y., Norval, R.A.I., and Oliver, J.H. (1982) Interspecific mating of *Amblyomma hebraeum* and *Amblyomma variegatam* (Acari: Ixodidae). *J. Med. Entomol.* **19**, 139–142.

Ribeiro, J.M.C. (1987) Role of saliva in blood-feeding by arthropods. *Annu. Rev. Entomol.* **32**, 463–478.

Ribeiro, J.M.C. (1988) Can satyrs control pests and vectors? *J. Med. Entomol.* **25**, 431–440.

Ribeiro, J.M.C., Makoul, G., Levine, J., Robinson, D., and Spielman, A. (1985) Antihemostatic, antiinflammatory and immunosuppressive properties of the saliva of a tick, *Ixodes dammini*. *J. Exp. Med.* **161**, 332–344.

Roberts, J.A. (1968) Resistance of cattle to the tick *Boophilus microplus* (Canestrini). 1. Development of ticks on *Bos taurus*. *J. Parasitol.* **54**, 663–666.

Rogers, A.J. (1953) A study of the ixodid ticks of northern Florida including the biology and life history of *Ixodes scapularis* Say (Ixodidae: Acarina). Unpublished Ph.D. dissertation, University of Maryland, College Park.

Schulze, T.L., Lakat, M.F., Bower, G.S., Parkin, W.E., and Shisler, J.K. (1984) *Ixodes dammini* (Acari: Ixodidae) and other ixodid ticks collected from white-tailed deer in New Jersey, USA. I. Geographical distribution and its relation to selected environmental and physical factors. *J. Med. Entomol.* **21**, 741–749.

Scifres, C.J., Oldham, T.W., Teel, P.D., and Drawe, D.L. (1988) Gulf coast tick (*Amblyomma maculatum*) populations and responses to burning of coastal prairie habitats. *Southwest. Natural.* **33**, 55–64.

Smith, C.N. and Cole, M.M. (1943) Studies of parasites of the American dog tick. *J. Econ. Entomol.* **36**, 569–564.

Smith, C.N., Cole, M.M., and Gouck, H.K. (1946) Biology and control of the American dog tick. *Tech. Bull. 905. US Dept Agric.*, Washington, D.C.

Sonenshine, D.E. and Haines, G. (1985) A convenient method for controlling populations of the American dog tick, *Dermacentor variabilis* (Acari: Ixodidae), in the natural environment. *J. Med. Entomol.* **22**, 577–585.

Sonenshine, D.E., Taylor, D., and Corrigan, G. (1985) Studies to evaluate the effectiveness of sex pheromone-impregnated formulations for control of populations of the American dog tick, *Dermacentor variabilis* (Say) (Acari: Ixodidae). *Exp. Appl. Acarol.* **1**, 23–34.

Sonenshine, D.E., Taylor, D., and Carson, K.A. (1986) Chemically mediated behavior in Acari. *J. Chem. Ecol.* **12**, 1091–1108.

Sonenshine, D.E., Hamilton, J.G.C., Phillips, J.S., Ellis, K.P., and Norval, R.A.I. (1992) Innovative techniques for control of tick disease vectors using pheromones. pp. 308–313. *In:* Munderloh, U.G. and Kurtti, T.J. (eds) *First International Conference on Tick-borne Pathogens at the Host–Vector Interface: An Agenda for Research.* University of Minnesota, St. Paul.

Spickett, A.M. (1978) Effects ^{60}Co irradiation on *Amblyomma hebraeum* Koch, 1844 (Acarina: Ixodidae). *Ondst. J. Vet. Res.* **45**, 197–201.

Spickett, A.M. (1987) The natural control of heartwater vectors. *Onderst. J. Res.* **54**, 535–539.

Spickett, A.M. and Malan, J.R. (1978) Genetic incompatibility between *Boophilus decoloratus* (Koch, 1844) and *Boophilus microplus* (Canestrini, 1888) and hybrid sterility of Australian and South African *Boophilus microplus* (Acarina: Ixodidae). *Ondst. J. Vet. Res.* **45**, 149–153.

Spickett, A.M., Horak, I.G., Van Niekerk, A., and Braack, L.E.O. (1992) The effect of veld burning on the seasonal abundance of free-living ixodid ticks as determined by drag sampling. *Ondst. J. Vet. Res.* **59**, 285–292.

Spielman, A., Levine, M.L. and Piesman, J.F. (1985) Ecology of *Ixodes dammini*-borne human babesiosis and Lyme disease. *Annu. Rev. Entomol.* **30**, 439–460.

Southwood, T.R.E. (1977a) Entomology and mankind. *Am. Sci.* **65**, 30–39.

Southwood, T.R.E. (1977b) The relevance of population dynamics theory to pest status. pp. 35–54. *In:* Chenett, J.M. and Sagar, J.M. (eds) *Origins of Pest, Parasite, Disease and Weed Problems.* British Ecological Society Symposium, 18.

Stafford, K.C., III (1991) Effectiveness of host-targeted permethrin in the control of *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **28**, 611–617.

Stafford, K.C., III (1992) Third-year evaluation of host-targeted permethrin for the control of *Ixodes dammini* (Acari: Ixodidae) in Southeastern Connecticut. *J. Med. Entomol.* **29**, 717–720.

Steeves, G.B.T. and Allen, J.R. (1990) Basophils in skin reactions of mast cell deficient mice infested with *Dermacentor variabilis. Internat. J. Parasitol.* **20**, 655.

Sternburg, S., Peleg, B., and Galun, R. (1973) Effect of irradiation on the mating competitiveness of the male tick *Argas persicus* (Oken). *J. Med. Entomol.* **10**, 473–476.

Stinner, R.E. (1977) Efficacy of inundative releases. *Annu. Rev. Entomol.* **22**. 515–531.

Sutherst, R.W. (1981) Is the Australian pest management approach to tick control relevant to Africa? pp. 79–85. *In:* Whitehead, G.B. and Gibson, J.B. (eds) *Proc. Internat. Conf. Tick Biology and Control.* Tick Research Unit, Rhodes University, Grahamstown, South Africa.

Sutherst, R.W. (1987) The dynamics of hybrid zones between tick (Acari) species. *Internat. J. Parasitol.* **17**, 921–926.

Suthertst, R.W. and Wilson, L.J. (1986) Tropical legumes and their ability to immobilize and kill cattle ticks. pp. 185–194. *In:* Juniper, B.E. and Southwood, T.R.E. (eds) *Insects and Plant Surfaces.* Edward Arnold, London.

Sutherst, R.W., Wharton, R.H., and Utech, K.B.W. (1978) *Guides to Studies on Tick Ecology.* Div. Entomol. Tech. Paper no. 14.

Sutherst, R.W., Jones, R.J., and Schnitzerling, H.J. (1982) Tropical legumes of the genus *Stylosanthes* immobilize and kill cattle ticks. *Nature* **295**, 320–321.

Sutherst, R.W., Wilson, L.J., Reid, R., and Kerr, J.D. (1988) A survey of the ability of tropical legumes in the genus *Stylosanthes* to trap larvae of the cattle tick, *Boophilus microplus* (Ixodidae). *Austral. J. Exp. Agric.* **28**, 473–479.

Tellam, R.I., Smith, D., Kemp, D.H., and Willadsen, P. (1992) Vaccination against ticks. pp. 303–331. In: *Animal Parasite Control Utilizing Biotechnology.* CRC Press, Boca Raton, FL.

Theiler, G. (1969) Factors influencing the existence and the distribution of ticks. The ecology and control of ticks in Southern Africa. *Proc. Symp., Rhodes Univ.*, Grahamstown, South Africa.

Thompson, G.D., Osburn, R.L., Davey, R.B., and Price, M.A. (1981) The dynamics of hybrid sterility between *Boophilus annulatus* and *B. microplus* (Acari: Ixodidae) through successive generations. *J. Med. Entomol.* **18**, 413–418.

Thompson, K.C., Roa, E.J., and Romero, N.T. (1978) Anti-tick grasses as the basis for developing practical tropical tick control packages. *Trop. Anim. Hlth Prod.* **10** **179**–182.

Utech, K.B.W., Wharton, R.H., and Kerr, J.D. (1978) Resistance to *Boophilus microplus* (Canestrini) in different breeds of cattle. *Austral. J. Agric. Res.* **29**, 885–895.

Walker, A.R. and Fletcher, J.D. (1990) *Rhipicephalus appendiculatus* feeding on rabbits and cattle: salivary-gland responses to varying host resistance. *Exp. Appl. Acarol.* **8**, 285–290.

Wandeler, A.I. (1990) Oral immunization of wildlife. pp. 488–503. *In:* Baer, G.M. (ed) *The Natural History of Rabies.* CRC Press, Boca Raton, FL.

Warren, S.D., Scifres, C.J., and Teel, P.D. (1987) Response of grassland arthropods to burning: a review. *Agric. Ecosyst. Environ.* **3**, 105–130.

Wikel, S.K. (1988) Immunological control of hematophagous arthropod vectors: utilization of novel antigens. *Vet. Parasitol.* **29**, 235–264.

Wikel, S.K., Ramachandra, R.N., and Bergman, D.K. (1992) Immunological strategies for suppression of vector arthropods. *Bull. Soc. Vector Ecol.* **17**, 10–19.

Wilkinson, P.R. (1957) The spelling of pasture in cattle tick control. *Austral. J. Agric. Res.* **8**, 414–423.

Wilkinson, P.R. (1970) Factors affecting the distribution and abundance of the cattle tick in Australia: observations and hypotheses. *Acarologia* **12**, 492–508.

Wilkinson, P.R. (1977) Effect of herbicidal killing of shrubs on abundance of adult *Dermacentor andersoni* (Acaina: Ixodidae) in British Columbia. *J. Med. Entomol.* **13**, 713–718.

Wilkinson, P.R. (1979) Ecological aspects of pest management of Ixodid ticks. Vol. II, pp. 25–33. *In:* Rodriguez, R.D. (ed) *Recent Advances in Acarology.* Academic Press, New York.

Willadsen, P. and Kemp, D.H. (1988) Vaccination with "concealed" antigens for tick control. *Parasitol. Today* **4**, 196–198.

Willadsen, P., McKenna, R.V., and Riding, G.A. (1988) Isolation from the cattle tick, *Boophilus microplus*, of antigenic material capable of eliciting a protective immunogenic response in the bovine host. *Internat. J. Parasitol.* **18**, 183–189.

Willadsen, P., Riding, G.A., McKenna, R.V., Kemp, D.H., Tellam, R.L., Nielsen, J.M., Lahnstein, J., Cohen, G.S., and Gough, J.M. (1989) Immunologic control of a parasitic arthropod. Identification of a protective antigen from *Boophilus microplus*. *J. Immunol.* **143**, 1346–1351.

Wilson, L.J. and Sutherst, R.W. (1990) Oviposition sites of *Boophilus microplus* (Canestrini) (Acarina: Ixodidae) in stylosanthes and grass pastures. *J. Austral. Entomol. Soc.* **29**, 101–105.

Wilson, M.L. (1986) Reduced abundance of adult *Ixodes dammini* (Acari: Ixodidae) following destruction of vegetation. *J. Econ. Entomol.* **79**, 693–696.

Wilson, M.L. and Deblinger, R.D. (1993) Vector management to reduce the risk of Lyme Disease. pp. 126–156. *In:* Ginsburg, H. (ed) *Ecology and Environmental Management of Lyme Disease.* Rutgers University Press, New Brunswick, NJ.

Wilson, M.L., Levine, J.F., and Spielman, A. (1984) Effect of deer reduction on abundance of the deer tick (*Ixodes dammini*). *Yale J. Biol. Med.* **57**, 697–705.

Wilson, M.L., Telford, S.R., III, Piesman, J., and Spielman, A. (1988) Reduced abundance of immature *Ixodes dammini* (Acari: Ixodidae) following elimination of deer. *J. Med. Entomol.* **25**, 224–228.

Wilson, M.L., Litwin, T.S., Gavin, T.A., Capkanis, M.C., Maclean, D.C., and Spielman, A. (1990) Host-dependent differences in feeding and reproduction of *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **27**, 949–954.

Wong, J.Y.M. and Opdebeeck, J.P. (1990) Larval membrane antigens protect Hereford cattle against infestation with *Boophilus microplus*. *Parasite Immunol.* **12**, 75–83.

Wouters, G. (1990) A hybridization model for *Rhipicephalus appendiculatus* and *R. zambeziensis* (Acarina: Ixodidae) using glucose-6-phosphate-isomerase isoenzymes. *Exp. Appl. Acarol.* **10**. 69–76.

Young, A.S., Groocock, C.M., and Kabiuki, D.P. (1988) Integrated control of ticks and tick-borne diseases of cattle in Africa. *Parasitol.* **96**, 403–432.

Yunker, C.E., Peter, T., Norval, R.A.I., Sonenshine, D.E., Burridge, N.J., and Butler, J.F.

(1992) Olfactory responses of adult *Amblyomma hebraeum* and *A. variegatum* (Acari: Ixodidae) to attractant chemicals in laboratory tests. *Exp. Appl. Acarol.* **13**, 295–301.

Zimmerman, R.H., Garris, G.I., and Beaver, J.S. (1984) Potential of *Stylosanthes* plants as a component in an integrated pest management approach to tick control. *Prev. Vet. Med.* **2**, 579–588.

Ziv, M., Sonenshine, D.E., Silverstein, R.M., West, J.R., and Gingher, K.H. (1981) Use of sex pheromone, 2,6-dichlorophenol, to disrupt mating by American dog tick *Dermacentor variabilis* (Say). *J. Chem. Ecol.* **7**, 829–840.

II

REPRESENTATIVE TICK-BORNE DISEASES

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9

Dynamic Associations of Tick-borne Diseases Affecting Domestic Animal Health

R. ANDREW I. NORVAL AND BRIAN D. PERRY

1. INTRODUCTION

This chapter on the dynamic associations of tick-borne diseases affecting domestic animal health focuses on two African diseases, theileriosis, caused by *Theileria parva*, and heartwater (cowdriosis), caused by *Cowdria ruminantium*. The regulation of vector distribution and the relationship between vector distribution, the occurrence and abundance of wildlife hosts, and the distribution and epidemiology of the diseases are examined. The impact on the epidemiology of theileriosis, of the seasonal occurrence of the vectors, and the effects on the epidemiology of heartwater of an aggregation–attachment pheromone produced by the vectors are discussed.

1.1. Theileriosis

Theileriosis caused by *T. parva* is a protozoan disease of cattle that occurs in eastern, central, and southern Africa. The most important vector is the brown ear tick, *Rhipicephalus appendiculatus*; other field vectors are *R. zambeziensis* and *R. duttonii* (da Graca and Serrano, 1971; Lawrence et al., 1983). The most important wild reservoir host is the Cape buffalo, *Syncerus caffer* (Grootenhuis, 1989; Grootenhuis and Young, 1981; Uilenberg, 1981; Young, 1981); waterbuck *Kobus* spp. have also been implicated as reservoir hosts (Stagg et al., 1983).

The clinical diseases caused by *T. parva* in cattle are known as East Coast fever (ECF), January disease, and corridor or buffalo disease. ECF was first recognized in southern Africa in 1902, following its introduction from eastern Africa (Lawrence, 1992); the disease was subsequently found to be widely distributed in eastern and central Africa (Norval et al., 1992a). Corridor disease, which was first observed by Lawrence (1935) and later described by Neitz (1955), is characteristically transmitted by ticks from buffalo to cattle and occurs in eastern, central, and southern Africa. January disease, the description of which is confined to Zimbabwe, was first recorded by Lawrence (1937) as an

atypical form of ECF in that country, and was confused with Corridor disease for many years. A trinomial system of classification for *T. parva* on the basis of the clinical syndromes observed was suggested by Uilenberg (1976) and Lawrence (1979); *T. parva parva* for parasites causing ECF, *T. parva bovis* for parasites causing January disease and *T. parva lawrencei* for parasites causing Corridor disease. However, this classification system has now been discarded as it is considered to be invalid based on our current understanding of the genetic composition of the parasite (Anon., 1989; Norval et al., 1992a). The three forms of *T. parva* are now considered to be interchangeable behavioral variants within a single species. The form in which *T. parva* occurs in a given environment is probably a function of a combination of factors determined by the mammalian hosts through which infection is passed and the tick populations which serve as vectors (Norval et al., 1991a); in this chapter the dynamic interactions of some of these factors are outlined.

East Coast fever is characteristically a disease causing high morbidity and high case fatality ($> 90\%$) in susceptible adult cattle of all breeds; in endemic situations the disease causes large losses in calves of exotic Taurine breeds and low to negligible losses in calves of indigenous Zebu or Sanga breeds (Norval et al., 1992a). January disease is characterized by a lower morbidity and case fatality than ECF; it causes negligible calf mortality (all breeds) and the incidence in adult cattle, including Taurine breeds, is low ($<3\%$) (Koch et al., 1990; Koch and Foggin, 1991). Corridor disease is extremely severe and causes almost 100% morbidity and case fatality in cattle when it is transmitted by *R. zambeziensis* (as occurs in southern Africa). However, when the vector is *R. appendiculatus* (as occurs mainly in eastern and central Africa) the disease is generally less severe, and a proportion of cattle recover from infection and become carriers (Norval et al., 1991a, 1992a). It is through these carriers that buffalo-derived *T. parva* infections become adapted to cattle, and subsequent infections to susceptible cattle assume the characteristics of ECF (Barnett and Brocklesby, 1966; Young and Purnell, 1973; Maritim et al., 1992).

Research workers in South Africa found, in the early part of the century, that ECF could be controlled through the control of its tick vectors by short-interval dipping of cattle in acaricides. Since then, dipping has become the mainstay of the control of theileriosis in eastern, central, and southern Africa. East Coast fever disappeared from southern Africa by the 1960s and it has been claimed that this was due largely to the success of the dipping program but, as will be described later in the chapter, another explanation based on the ecology of the vector is possible. In recent decades dipping has become an expensive control option that is not compatible with the changing socio-economic and land-use patterns of post-colonial Africa (Norval et al., 1992a). Alternative control strategies are therefore being developed. These include an infection and treatment method of immunization (Cunningham et al., 1974; Brown et al., 1977; Radley, 1978, 1981), which is now being used on a small to moderate scale in several countries, and various integrated control strategies based on immunization, strategic tick control, and the exploitation of endemic stability in indigenous breeds of cattle (Young et al., 1988; Norval et al., 1992a).

1.2. Heartwater

Heartwater is a rickettsial disease that affects cattle, sheep, goats, and a variety of wild ruminant species. The disease occurs throughout most of sub-Saharan Africa and has spread to several islands in the Indian and Atlantic Oceans, and the Caribbean. Heartwater is known to be transmitted by 12 tick species of the genus *Amblyomma* (Bezuidenhout, 1987), of which the most important vectors to livestock are considered to be the tropical bont tick, *A. variagatum*, and the southern African bont tick, *A. hebraeum* (Pentney et al., 1987). Cattle, sheep, and Cape buffalo have been shown in a laboratory study to be long-term carriers of *C. ruminantium* (Andrew and Norval, 1989). A variety of other wild ruminant species, as well as some non-ruminant species, have also been implicated as reservoirs of infection (Oberem and Bezuidenhout, 1987a).

Heartwater was first recognized in South Africa in 1838 (Neitz, 1968), where it was later shown to be transmitted by the tick *A. hebraeum* (Lounsbury, 1900). The disease became an important problem in livestock in South Africa in the latter half of the 19th century, when infection spread to newly established farming areas in the Eastern Cape and elsewhere (Henning, 1956; Provost and Bezuidenhout, 1987). In the remainder of sub-Saharan Africa losses due to heartwater were only recognized in the 20th century when exotic breeds of livestock were introduced, when the *Amblyomma* vectors spread to previously uninfected areas and when intensive dipping disrupted the endemic stability that is likely to have existed previously (Lawrence and Norval, 1979; Camus and Barré, 1982). The disease has now been recorded from all but five of the sub-Saharan African countries in which it can be expected to occur (Camus and Barré, 1982). Heartwater has also been confirmed on the islands of Madagascar, Mauritius, and Réunion in the Indian Ocean, São Tome in the Atlantic Ocean, and Antigua Guadeloupe and Marie Galante in the Caribbean (Provost and Bezuidenhout, 1987). The presence of *C. ruminantium* and *A. variegatum* in the Caribbean is considered to pose an important threat to the American mainland (Barré et al., 1987).

In South Africa and Zimbabwe, intensive tick control has been implemented as a specific control measure for heartwater (Stampa, 1969; Norval and Lawrence, 1979; Bezuidenhout and Bigalke, 1987). Elsewhere in eastern and central Africa the control of heartwater by means of tick control has not been a deliberate policy but may have occurred where intensive dipping has been implemented as a means of control of ECF. In Zimbabwe (Norval and Lawrence, 1979) and to a lesser extent in South Africa (Stampa, 1969) prolonged intensive tick control has resulted in the eradication of *A. hebraeum* from farming areas from which alternate wild hosts for the adult stage have been absent; eradication has not been achieved where alternate hosts have been present. In Tanzania *A. variegatum* was apparently eradicated from parts of the southern highlands by intensive dipping in the 1950s (Tatchell and Easton, 1986).

An infection and treatment method of immunization against heartwater has been available for 50 years (Neitz, 1939, 1940; Weiss et al., 1952; Haig et

al., 1954), but it has only been in South Africa that a "vaccine" (infected sheep blood) has been produced commercially and has been widely used (Oberem and Bezuidenhout, 1987b; Van der Merwe, 1987). Endemic stability for heartwater is thought to be widespread in Africa and as a result the mortality caused by the disease in livestock is usually minimal. In most instances control measures for heartwater are only necessary where endemic stability has been affected by human activities. One unresolved anomaly, however, is that heartwater losses in livestock are usually greater when the disease is associated with *A. hebraeum* than with *A. variegatum* (Norval, 1983a; Asselbergs et al., 1993).

2. VECTOR DISTRIBUTION

2.1. Geographic Information Systems

Information on the distribution of tick species has always relied primarily on tick survey data from the field. Tick surveys have now been conducted in most African countries and large numbers of distribution records exist for the species that commonly parasitize livestock, including the vectors of theileriosis and heartwater. The compilation of these records into distribution maps with subsequent analysis of the relationships between tick distribution and other variables such as climate, vegetation, host and disease distribution, was a problem when mapping was carried out manually. However, progress in the handling and manipulation of African tick distribution data on a continental basis has recently been made by use of geographic information systems (GIS) (Lessard et al., 1990). Geographic information systems are computer-assisted systems for storing, manipulating, analyzing, and displaying spatial data. One use of GISs is to create map overlays which illustrate and quantify geographically the complex relationships among study variables.

2.2. Theileriosis

Lessard et al. (1990) have assembled and plotted distribution records of *R. appendiculatus*, *R. zambeziensis* and *R. duttonii* reported over approximately 40 years (Fig. 9.1). *Rhipicephalus appendiculatus* has been recorded from 15 countries extending from the Central African Republic and southern Sudan southwards through Zaire, Uganda, Kenya, Burundi, Rwanda, Tanzania, Zambia, Malawi, Mozambique, Zimbabwe, and Botswana, into South Africa where it occurs in the Transvaal, Natal, and the coastal areas of the Eastern Cape Province. *Rhipicephalus zambeziensis* replaces *R. appendiculatus* in the hotter, drier areas of central and southern Africa. This species occurs in southern Tanzania, Zambia, Mozambique, Zimbabwe, Botswana, Namibia, and the northern Transvaal in South Africa. *Rhipicephalus duttonii* occurs only in Angola and Zaire, where most records are from the semi-arid zone that stretches along the Atlantic coast.

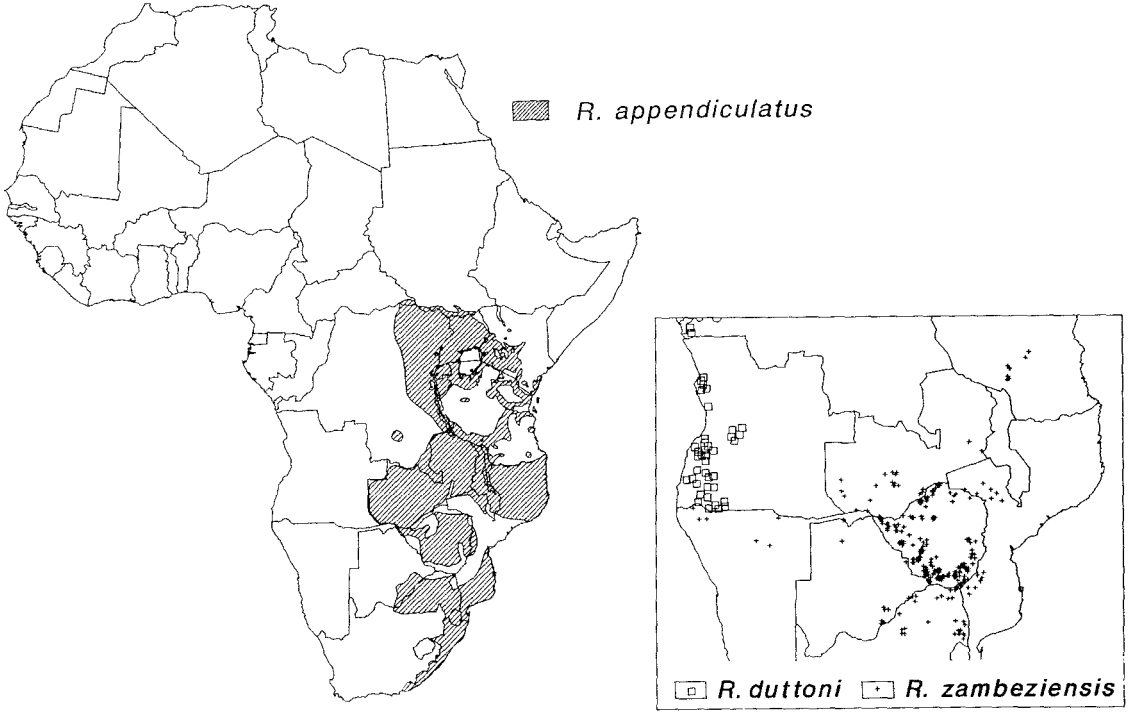

Fig. 9.1. Distribution of *Rhipicephalus appendiculatus* (based on distribution records and expert opinion in the case of Mozambique), *R. zambeziensis* and *R. duttoni* in Africa. Redrawn from data assembled by Lessard et al. (1990).

The distribution of *R. appendiculatus* is by no means continuous, even in those countries in which it occurs most commonly. Its occurrence is influenced by several factors, the most important of which are climate, vegetation and host availability. The climatic requirements of the tick have been defined and quantified and, using the climate matching model CLIMEX, it has been possible to predict the suitability of any particular area for *R. appendiculatus* (Sutherst and Maywald, 1985). CLIMEX calculates an ecoclimatic index (EI), on a scale between 0 and 100, from a growth index moderated by four stress indices (dry, wet, hot, and cold). All the indices are calculated from weekly or monthly maximum and minimum temperatures, rainfall, and evaporation data. Lessard et al. (1990) have estimated EI values for *R. appendiculatus* for the whole of Africa by the interpolation of climatic data from approximately 5,000 meteorological stations scattered over the continent (Fig. 9.2). The resolution of the interpolation is approximately 25 km which gives 43,644 cells covering the African continent. Recently, CLIMEX has been run using a higher resolution interpolated climate database (of 5 min, approximately 8 km) of long-term average monthly data for Zimbabwe, providing a more detailed distribution of EI values in that country (Perry et al., 1991). The distribution of EI values was overlaid with the location of theileriosis outbreaks during the period 1979–1989 (Fig. 9.3).

Perry et al. (1990a) have applied CLIMEX to the interpolating climate database to provide a more precise definition of the potential distribution of *R. appendiculatus* in Africa. Areas of predicted climatic suitability for the tick (Fig. 9.2) extend through most of sub-Saharan Africa; greatest suitability is seen in coastal zones of Cameroon and Equatorial Guinea, the periphery of the Zaire Basin, the Lake Victoria Basin, and the coastal strip of Natal and the Transkei in South Africa. Intermediate and low suitability is seen through much of central and southern Africa. The most unsuitable areas are found in the deserts of the Horn of Africa and southern Africa.

Perry et al. (1990a) noted that there is very close correlation between EI and recorded tick distribution in much of eastern Africa, especially in Kenya, Uganda, Burundi, and Rwanda. There are some exceptions, notably in central Tanzania where, although climate is suitable, *R. appendiculatus* has not been recorded. This is thought to be due to the very low cattle density in the area (Fig. 9.4) as a result of tsetse fly infestation. In central Africa, the distribution of suitable EI values correlates fairly well with known tick distribution. The index may well be accurate in Mozambique, but there are not enough tick collection data to permit evaluation. On the coastal strip of South Africa, the EI is highly correlated with tick distribution. Further inland in South Africa, there is very poor correlation, with no *R. appendiculatus* being recorded in areas where the index values indicate apparent suitability. Most notable among the climatically favorable areas in which *R. appendiculatus* does not occur are large areas of western and central Ethiopia, northern and eastern Zaire, and the coastal strip of western Africa from Cameroon to northern Angola.

The absence of *R. appendiculatus* from the Ethiopian highlands has been discussed by Norval et al. (1991b). It is thought to be the result of the tick not

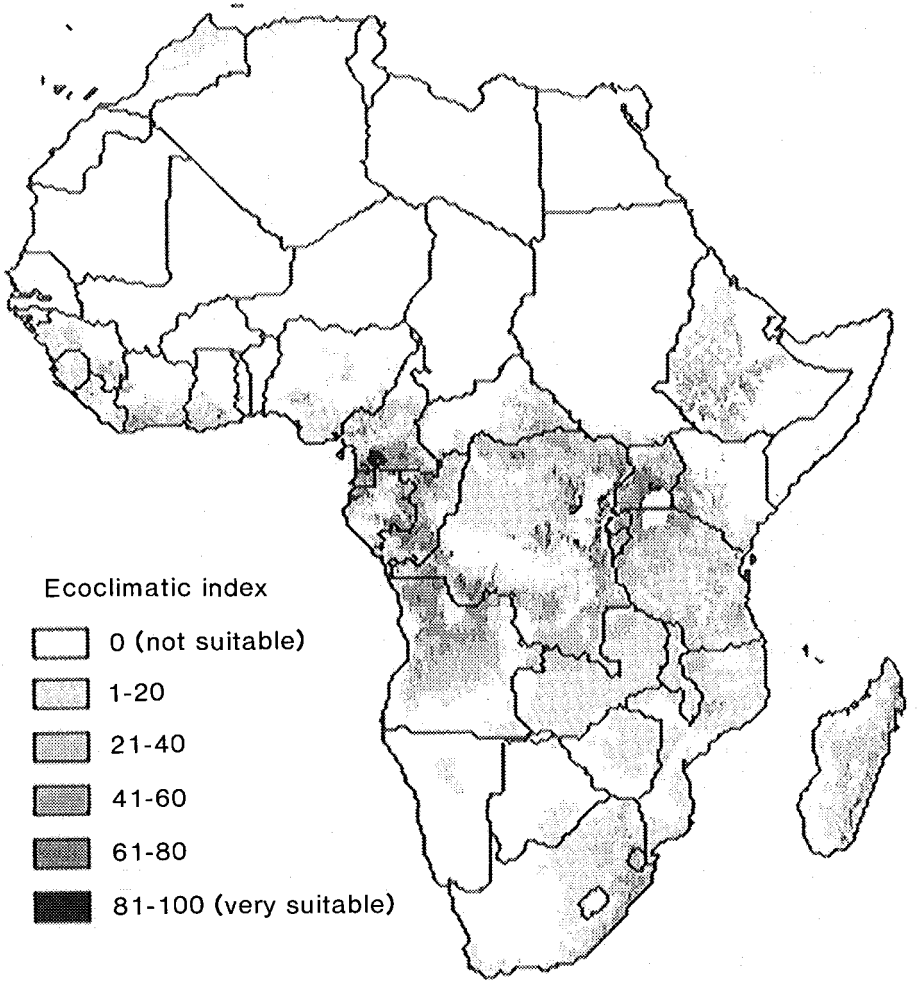

Fig. 9.2. Distribution of ecoclimatic index values of climatic suitability calculated by CLIMEX for *Rhipicephalus appendiculatus* in Africa. Ecoclimatic index values derived from Sutherst and Maywald (1985). Redrawn from Lessard et al. (1990).

having been established in this high rainfall area prior to human settlement, when it was forested and did not support adequate herbivore (host) populations. Deforestation has taken place on a large scale and cattle now occur in abundance. The tick can thus be expected to become established if it is ever introduced. In the past, deserts and large areas affected by trypanosomiasis have formed barriers that have prevented the introduction of *R. appendiculatus* by the movement of infested cattle from neighboring countries.

The absence of *R. appendiculatus* from areas in the west of Africa, predicted by using CLIMEX to be climatically suitable, appears to be due largely to the low densities of cattle (Fig. 9.4) and other herbivore hosts that have existed

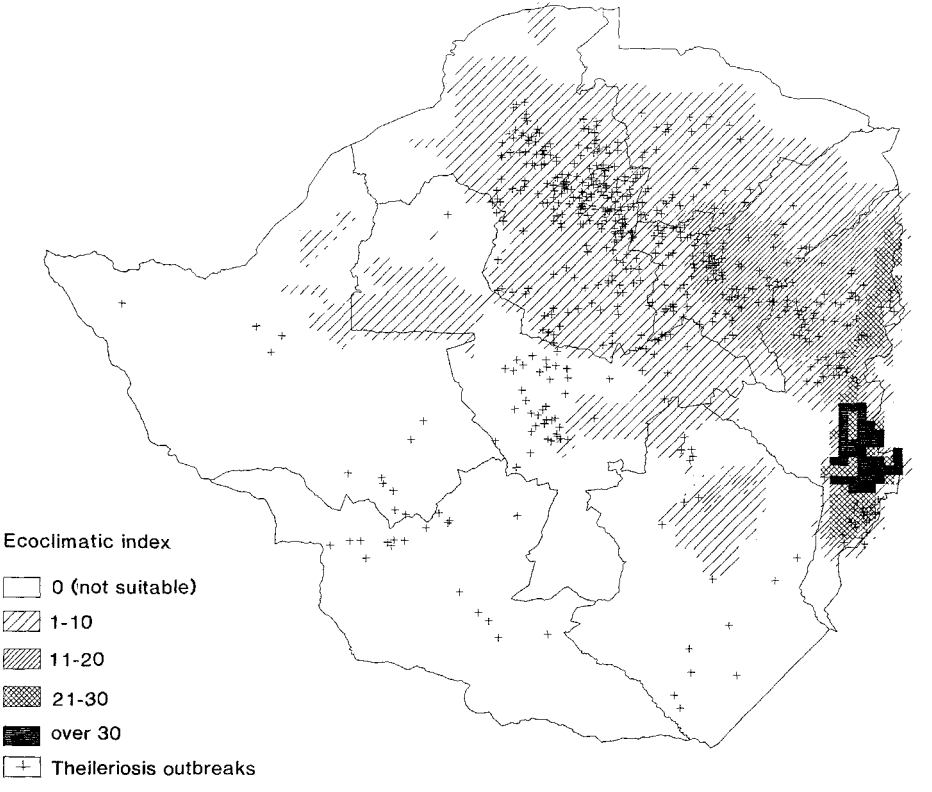

Fig. 9.3. Distribution of ecoclimatic index values of climatic suitability for *Rhipicephalus appendiculatus* in Zimbabwe, derived using the default CLIMEX parameters of Sutherst and Maywald (1985) on an interpolated climate database. The location of theileriosis outbreaks for the period 1979–1989 (Koch, 1990) have been overylayed on the ecoclimatic index distribution. From Perry et al. (1991). With permission of the International Society of Vector Ecology.

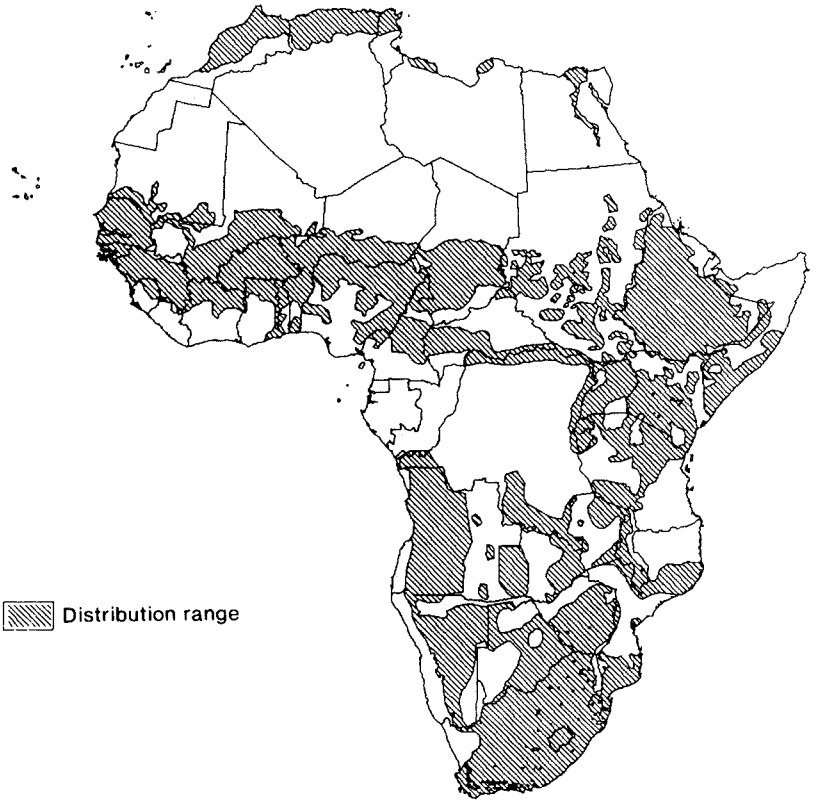

Fig. 9.4. The distribution of cattle in Africa. Redrawn from Lessard et al. (1990).

there in the past. These low host densities have been the result of trypanosomiasis, which has prevented the introduction of cattle, and the presence of large forested areas with little grazing for ground-dwelling herbivores. Deforestation and the introduction of cattle, in association with trypanosomiasis control, are now taking place on a large scale; as a consequence, the risks of the tick spreading to these areas and becoming established are increasing.

The dynamic nature of the distribution of *R. appendiculatus* has been confirmed by observations on the ground. There are records from Tanzania (Yeoman, 1966; Tatchell and Easton, 1986) and Zimbabwe (Norval and Perry, 1990) of extensions of the distribution of the tick into dry areas during years of above average rainfall, and its disappearance when conditions became unfavorable.

The observations from Zimbabwe merit elaboration. *Rhipicephalus appendiculatus* was known to be absent from the hot, dry, southern lowveld in the 1960s. In 1973 the tick was introduced with a herd of sable antelope (*Hippotragus niger*), translocated from a wildlife reserve in a higher rainfall area to a small game park on a cattle ranch. The species became established and spread rapidly, so that by the early 1980s it affected an area of more than one million

hectares. Then, in 1983–1984, the tick disappeared completely from this area. The introduction and spread of the tick occurred during a 7-year wet cycle (above average rainfall for 6 of 7 years) and it disappeared at the end of a 4-year dry cycle (below average rainfall for 3 of 4 years). Analysis of the climatic data using CLIMEX shows that EI values were above 0 in 4 of the 7 years of the wet cycle and remained at 0 throughout the dry cycle. The findings confirm the importance of climate in determining the distribution of *R. appendiculatus* and in causing periodic fluctuations in distribution; they also provide valuable field validation of the CLIMEX model.

At a local level, the occurrence and abundance of *R. appendiculatus* are affected by the amount of vegetation cover (Yeoman, 1967; Norva, 1977), the abundance of suitable ruminant hosts (Norval and Lightfoot, 1982) and acaricide usage (Howell et al., 1981). It should be pointed out that cattle and various wild ungulate species such as buffalo and eland (*Taurotragus oryx*), on which large numbers of all life cycle stages of *R. appendiculatus* can occur, may serve as "amplifier" hosts which increase the abundance of the tick species in certain environments (Norval, 1979a).

Vegetation cover affects microclimate (Minshull and Norval, 1982; Short et al., 1989a, 1989b). Where vegetation cover is reduced by overgrazing and the removal of trees, as has occurred in the communal (peasant) farming areas of Zimbabwe (Fig. 9.5), *R. appendiculatus* tends to disappear. The species become very abundant in the presence of hosts that have a low level of resistance to it (Lightfoot and Norval, 1981). Prolonged intensive acaricide treatment of livestock can cause the localized eradication of *R. appendiculatus*, but the tick can spread if control measures are ever stopped (Norval, 1979b).

2.3. Heartwater

Existing distribution records of *A. variegatum* and *A. hebraeum* have been assembled and plotted by Walker and Olwage (Fig. 9.6). *Amblyomma variegatum* is widely distributed through western Africa south of the Sahel, the horn of Africa, as well as eastern and central Africa. In southeastern Africa, *A. variegatum* is replaced by *A. hebraeum*. The two species have similar climatic requirements and occur in similar habitats but cannot exist sympatrically due to interspecific competition (Rechav et al., 1982; Norval, 1983a; Sutherst, 1987).

Norval et al. (1991c, 1992a) have used the CLIMEX model and the interpolated climate database of Lessard et al. (1990) to identify those areas of Africa that are climatically suitable for *A. variegatum* and *A. hebraeum* (Fig. 9.7). CLIMEX accurately predicts the distribution of *A. variegatum* in western Africa, as far south as Angola, in the Horn of Africa, and in eastern and central Africa, as far south as the Zambezi River valley. The species is absent from areas of predicted high suitability in northern Angola, where it is replaced by *Amblyomma pomposum* (Walker and Olwage, 1987), and from parts of the southern highlands of Tanzania, where it is assumed to have been eradicated

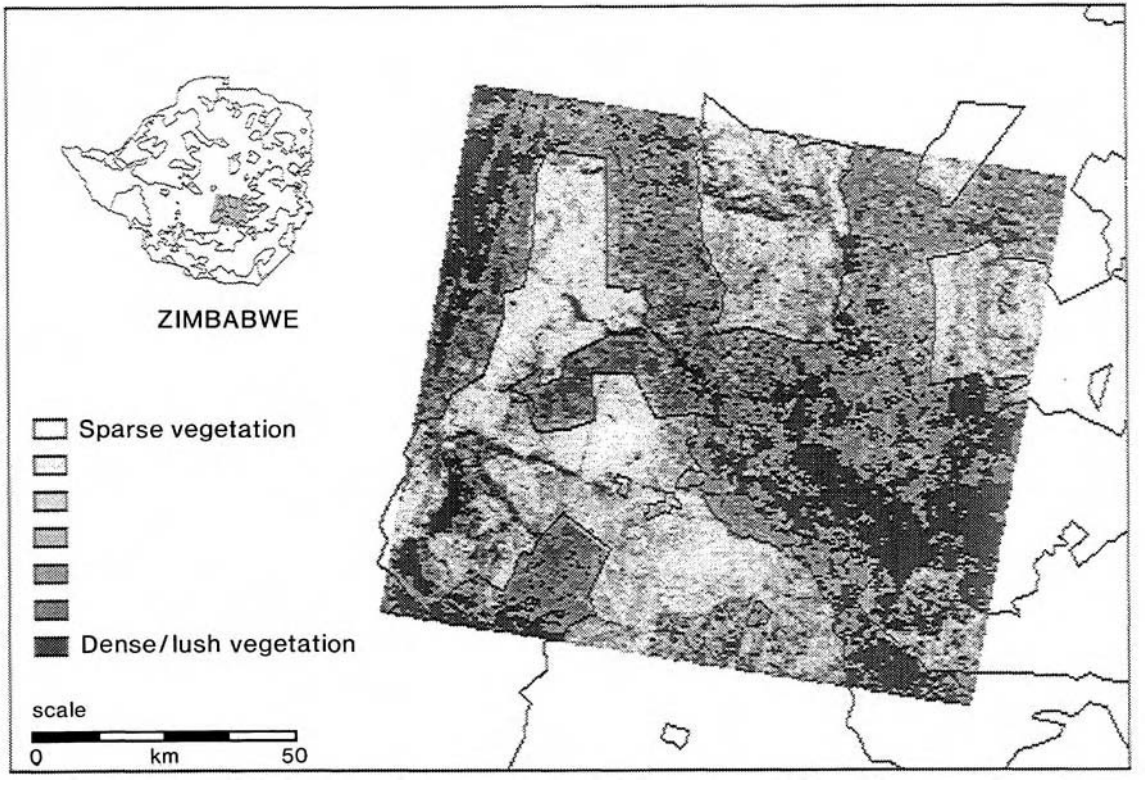

Fig. 9.5. A digital LANDSAT MSS satellite image of the region near Masvingo, Zimbabwe. The black lines represent boundaries between communal lands (overgrazed) and commercial farming areas, overlayed on the LANDSAT image. From Perry et al. (1991).

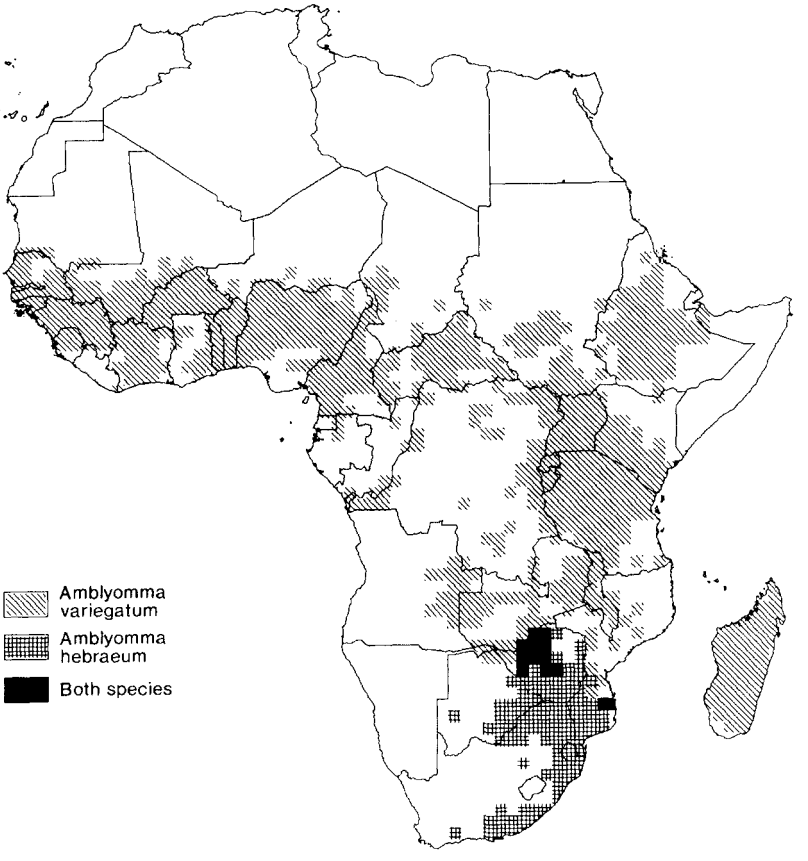

Fig. 9.6. Distribution of *Amblyomma hebraeum* and *A. variegatum* in Africa. Redrawn from Walker and Olwage (1987).

by the intensive use of acaricides (Tatchell and Easton, 1986). In southern Africa *A. hebraeum* occurs in areas of predicted high suitability except on the highveld plateau of northeastern Zimbabwe and in the highveld of the southern Transvaal in South Africa. The absence of the species from the Zimbabwe highveld has been attributed to intensive acaricide usage in an area with few or no alternate wild hosts (Norval et al., 1992b). *Amblyomma hebraeum* is also absent from areas of low to moderate suitability in northern Namibia, northwestern Botswana, and the north of the Cape Province in South Africa.

Amblyomma hebraeum, unlike *R. appendiculatus*, is able to survive in overgrazed habitats such as the Communal Lands of Zimbabwe (Norval, 1983a). Acaricide usage is probably the single most important factor affecting the abundance of this species at a local level (Stampa, 1969; Norval and Lawrence, 1979; Norval et al., 1992b). Cattle and various wild ungulate species such as giraffe (*Giraffa camelopardalis*) and buffalo may serve as amplifier hosts for some African *Amblyomma* species.

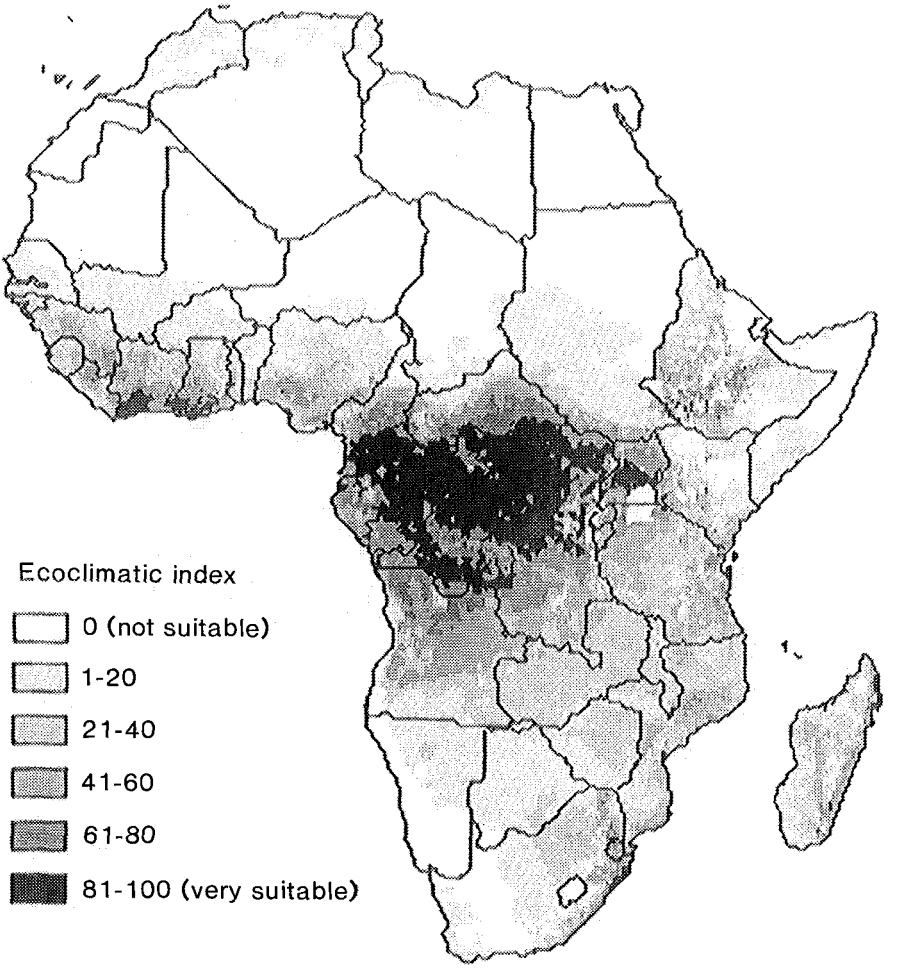

Fig. 9.7. Distribution of ecoclimatic index values of climatic suitability for *Amblyomma variegatum* in Africa, derived using CLIMEX parameter values designated by Norval et al., 1990).

3. DISEASE DISTRIBUTION

3.1. Theileriosis

Lessard et al. (1990) have plotted the known distribution of clinical theileriosis caused by *T. parva* (ECF, Corridor disease, and January disease) in Africa (Fig. 9.8). There is a clear geographical relationship between the occurrence of clinical theileriosis and the distribution of *R. appendiculatus* in eastern Africa (Sudan, Zaire, Uganda, Kenya, Burundi, Rwanda, Tanzania, and Malawi). The relationship is less apparent in central and southern Africa from Zambia southwards, where clinical theileriosis does not occur in all the areas from which *R. appendiculatus* has been recorded, and does occur in some areas from which the tick

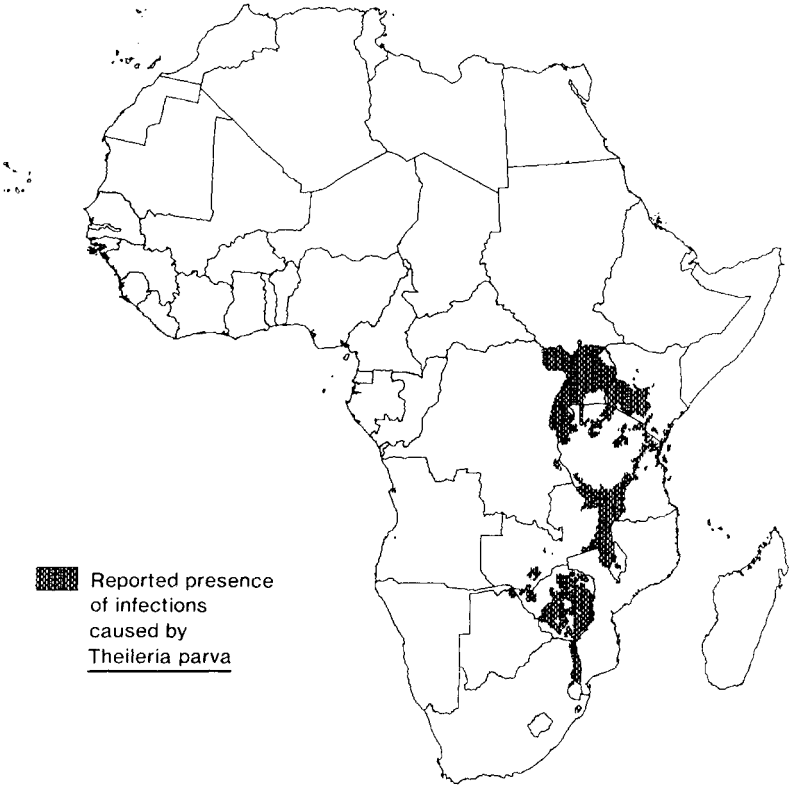

Fig. 9.8. The reported distribution of *Theileria parva* infections in Africa. Redrawn from data assembled by Lessard et al., 1990).

is absent. In Zambia *R. appendiculatus* is very much more widely distributed than clinical theileriosis; in Zimbabwe the distributions of *R. appendiculatus* and clinical theileriosis only overlap in the northeastern and central areas of the country; in Botswana clinical theileriosis is absent despite the presence of *R. appendiculatus* in the southeast; while in South Africa the distributions of *R. appendiculatus* and clinical theileriosis only coincide in a small area in Natal. The relationship between the occurrence of clinical theileriosis and vector distribution in southern Africa is partially clarified when the distribution of *R. zambeziensis* is considered in addition to that of *R. appendiculatus.* It then becomes apparent that the occurrence of the disease in the south and west of Zimbabwe, and in the eastern Transvaal of South Africa is associated with *R. zambeziensis.* However, in other areas in which *R. zambeziensis* occurs, such as Namibia, Botswana, and the western Transvaal, no clinical theileriosis has been recorded. The occurrence of clinical theileriosis in two coastal areas of Angola is clearly linked to the presence of *R. duttonii.*

Hence, while vector distribution is a good predictor of the occurrence of clinical theileriosis in eastern Africa, it is a poor predictor in central and

southern Africa. The apparently anomalous pattern of distribution of clinical theileriosis in central and southern Africa indicates that factors other than the occurrence of vectors *per se* must influence the distribution and epidemiology of *T. parva* in these areas.

3.2. Heartwater

It has not been possible to map the distribution of heartwater because reliable data on the occurrence of the disease are generally unavailable due to the difficulty of diagnosis, which relies on confirmation of the presence of colonies of *C. ruminantium* in the epithelial cells of capillaries in brain smears. Until very recently there have been no serological tests for heartwater, and so it has not been possible to determine the distribution of the disease on the basis of serology.

4. RESERVOIR HOSTS

4.1. Theileriosis

The current distributions of Cape buffalo (Lessard et al., 1990) and waterbuck (Norval et al., 1992a), based on the reports of numerous authors, are shown in Figs 9.9 and 9.10, respectively. In eastern Africa, while there is some overlap between the distributions of both buffalo and waterbuck and clinical theileriosis (Fig. 9.8), neither species is distributed through all the area in which the disease occurs. The occurrence of clinical theileriosis in this region is therefore not directly dependent on the presence of reservoir hosts. In contrast, in southern Africa there does appear to be some association between the distribution of buffalo and that of clinical theileriosis. In South Africa clinical theileriosis in cattle occurs only in areas where buffalo are present; in southeastern Botswana and northern Namibia, where buffalo are absent, there is no clinical theileriosis in cattle despite the presence of vectors. When vector species are considered it is seen that clinical theileriosis in cattle transmitted by *R. zambeziensis* occurs only in the presence of buffalo. In South Africa, *R. appendiculatus* is only a vector of theileriosis when associated with buffalo. However, in Zimbabwe and Zambia, the presence of buffalo is not a prerequisite to *R. appendiculatus* being a vector. Although there is considerable overlap between the distributions of buffalo and waterbuck in southern Africa, where it is impossible to differentiate the roles of the two species as reservoir hosts, the absence of clinical theileriosis from locations where waterbuck alone occurs is an indication that this species is not the primary source of infection.

4.2. Heartwater

The role played by reservoir hosts in the distribution and epidemiology of heartwater is impossible to assess because of the absence of detailed information

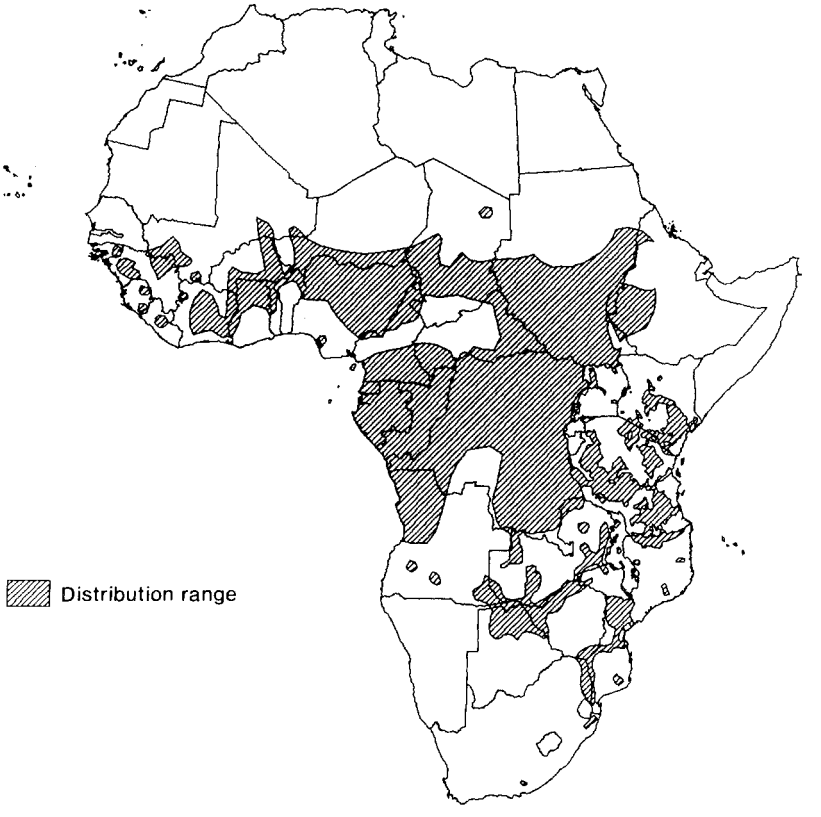

Fig. 9.9. The distribution of Cape buffalo (*Syncerus caffer*) in Africa. Redrawn from data assembled by Lessard et al., 1990).

on the distribution and prevalence of the disease. However, it is known that heartwater can occur in wildlife areas in the absence of domestic ruminants (MacKenzie and Norval, 1980) and that a number of wildlife species can become infected with *C. ruminantium* and may be short- or long-term carriers (Oberem and Bezuidenhout, 1987a; Andrew and Norval, 1989). It is also known that heartwater can persist in livestock in the absence of wildlife (Neitz, 1967).

5. ECOLOGICAL CONSIDERATIONS AFFECTING HOST–VECTOR INTERACTIONS AND DISEASE TRANSMISSION

5.1. Effects of Seasonal Occurrence of *Rhipicephalus appendiculatus* on the Epidemiology of Theileriosis

Rhipicephalus appendiculatus is distributed through areas of Africa which experience a wide range of climatic conditions. In tropical eastern Africa there

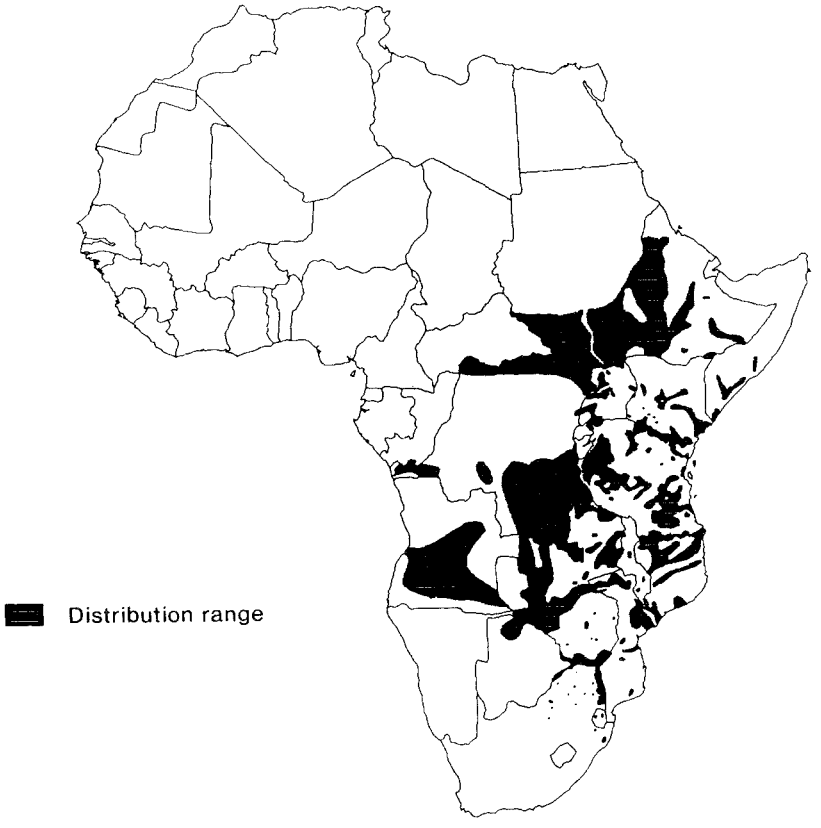

Fig. 9.10. The distribution of waterbuck (*Kobus ellipsiprymnus*) in Africa. From Norval et al., 1992a).

are no clearly defined hot and cold seasons, and day length is almost constant throughout the year. Eastern Africa also experiences no prolonged dry season; rainfall may occur throughout the year, there may be a single short dry season or two short dry seasons. In contrast, in subtropical central and southern Africa, there are well-defined hot and cold seasons, and large changes in day length between seasons. The areas of southern and central Africa in which *R. appendiculatus* occurs mostly experience a single period of rainfall (December–March) and a prolonged dry season which extends through a cold period (June–August) and a hot pre-rainy period (September–November). In the extreme south (Eastern Cape Province of South Africa) the climate is temperate, and there is a short but very cold dry season.

Populations of *R. appendiculatus* have evolved different behavioral strategies in response to these climatic differences. In eastern Africa larvae, nymphs and adults are usually present on hosts throughout the year (Smith, 1969; FAO, 1975; Newson, 1978; Newson and Punyua, 1978; Kaiser et al., 1982; Matthysse and Colbo, 1987) and no diapause occurs in any life cycle stage (Branagan,

1973a, 1973b, 1978). In contrast, in central and southern Africa, *R. appendiculatus* shows a clearly defined pattern of seasonal occurrence (reviews by Short and Norval, 1981a; Norval et al., 1992a), which is regulated by diapause in the unfed adults (Rechav, 1981, 1982; Short and Norval, 1981b; Pegram and Banda, 1990). Diapause is induced by photoperiod (Rechav, 1981). The ticks ascend the vegetation and engage in host seeking when the photophase is at its maximum length, around the start of the wet season, and they descend the vegetation and remain inactive in protected microhabitats at or near the soil surface when the photophase decreases early in the dry season (Short et al., 1989b; Pegram and Banda, 1990). Hence, adults parasitize hosts during the wet season, eggs are laid towards the end of the wet season when microclimatic conditions are most favorable for their survival (Minshull and Norval, 1982; Short et al., 1989a), the desiccation-sensitive larvae (Short et al., 1989b; Pegram and Banda, 1990) are active during the cool humid post-rainy period, nymphs are active in the cool part of the dry season, and the adverse microclimatic conditions of the hot dry season are survived by the diapausing adults which are extremely hardy (Short et al., 1989b).

Diapause is not only an important survival mechanism for *R. appendiculatus* in central and southern Africa, it has a profound effect on the epidemiology of theileriosis. This is because where diapause occurs, the tick passes through only one generation each year, and there is little or no overlap between the activity periods of the nymphs (which pick up infection) and the adults (which transmit infection). The occurrence of the disease is therefore seasonal, being restricted to the period of adult activity and onward transmission is dependent on the infection of nymphs by recovered carrier hosts. Larvae do not appear to become infected by carriers (Norval et al., 1992a). Where there is no diapause *R. appendiculatus* may pass through at least two generations per annum depending on temperatures which determines development rates (King et al., 1988), and all life-cycle stages may occur on hosts simultaneously. In these circumstances disease occurs throughout the year and onward transmission may be from clinically ill animals (with high parasitaemias) as well as from carriers.

As *T. parva* infection rates in adults of *R. appendiculatus* fed as nymphs on clinically ill animals will be higher than those fed on carriers with low parasitaemias (Purnell et al., 1974; Kariuki, 1991), infection rates can be expected to be higher in non-diapausing than in diapausing populations. Higher infection rates can be expected to result in non-diapausing populations being able to transmit clinical theileriosis more frequently than diapausing populations, because the severity of disease caused by *T. parva* infections is dose dependent (Barnett, 1957; Wilde, 1967; Dolan et al., 1984; Fivaz et al., 1989). These factors provide at least a partial explanation as to why ECF occurs in association with non-diapausing populations of *R. appendiculatus* in eastern Africa and why the less severe January disease occurs in association with diapausing populations in Zimbabwe.

Other factors may also contribute to the association of predominantly mild disease (Zimbabwe) or no disease (Botswana, South Africa) with diapausing

populations of *R. appendiculatus*. The infectivity of unfed adults is known to decline with time (Lewis and Fotheringham, 1941; Newson et al., 1984; Young et al., 1987) and, as diapause may last for 3–7 months (Short and Norval, 1981b; Rechav, 1982; Short et al., 1989b), this could contribute to low infection rates. Another contributing factor may be high temperatures which accelerate the loss of infection (Young et al., 1987), as diapausing adults must survive through the hottest time of year before becoming active. Perhaps the most important factor though is the susceptibility of the ticks themselves to infection with *T. parva*. There is now evidence that *R. appendiculatus* populations that originate from eastern Africa tend to become more highly infected than those that originate from southern Africa and, as a consequence, the disease that they transmit is more virulent. For example, in work reported by Irvine et al. (1989), the Boleni stock of *T. parva* from Zimbabwe was used in an immunization experiment. The clinical reactions to the immunizing stock were severe and characteristic of acute ECF when stabilates were made with ticks from Kenya, but were mild when stabilates were prepared in the same way with ticks from Zimbabwe; the apparent cause of this difference may be put down to the degree to which the different tick lines had become infected. One can speculate that resistance to *Theileria* infection, which can reduce longevity in highly infected ticks (Walker et al., 1983), has been selected for in diapausing populations of *R. appendiculatus* in which the unfed adults must survive for longer under field conditions than those in non-diapausing populations.

The December–April pattern of seasonal occurrence of adults of *R. appendiculatus* has been recorded from southern Tanzania (latitude 8°S) southwards through central and southern Africa (Short and Norval, 1981a; Tatchell and Easton, 1986), and it is reasonable to assume that this is the area in which diapause occurs. When considered on a geographical basis, it is seen that the area in which non-diapausing populations of *R. appendiculatus* occur is the area in which there is a close match between the distribution of this vector (Fig. 9.1) and that of clinical theileriosis (Fig. 9.8). In the area in which diapause occurs the relationship between the distribution of *R. appendiculatus* and clinical theileriosis becomes increasingly obscure from north to south until the disease disappears. The changing disease pattern south of latitude 8°C could be related to the increasing importance of diapause as seasonality becomes more pronounced further from the equator or to increasing resistance to infection with *T. parva* in populations of *R. appendiculatus*, or to both of these factors. It is also possible that isolated populations of non-diapausing *R. appendiculatus* occur in some areas, interspersed with diapausing populations.

The importance of diapause in *R. appendiculatus* in the epidemiology of theileriosis caused by *T. parva* became apparent though retrospective studies on the seasonal occurrence and virulence of ECF in southern Africa after its introduction in 1901–1902 (Lawrence, 1991; Norval et al., 1991a). Between 1914 and 1946 outbreaks of ECF in Zimbabwe were most numerous in the months of January/February and May/June/July (Lawrence, 1991). The pattern of seasonal occurrence then changed and the disease became less virulent, assuming the characteristics of what we now know as January disease (Matson,

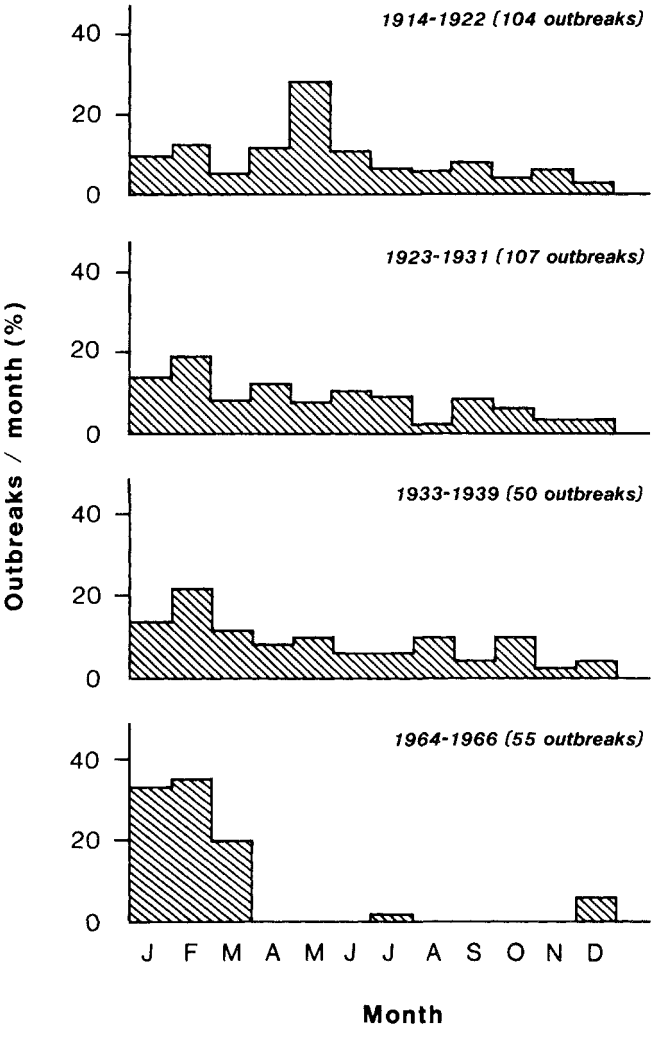

Fig. 9.11. Monthly incidence of new outbreaks of *Theileria parva* infection (East Coast fever and January disease) in Zimbabwe, 1914–1966. From Norval et al. (1991a).

1967; Lawrence and Norval, 1979). At present about 95% of outbreaks occur in January, February, and March, when the adults of *R. appendiculatus* are active (Norval et al., 1985). The change in monthly incidence of theileriosis over the period 1914–1966 is shown in Fig. 9.11. In South Africa, ECF also occurred throughout the year until the last foci of infection were brought under control in the 1940s and early 1950s (Lawrence, 1992); unlike Zimbabwe no cattle-adapted forms of *T. parva* persisted after that time. The changing disease pattern can be explained if one considers that non-diapausing populations of *R. appendiculatus*, in addition to *T. parva*, were introduced to southern Africa

at the beginning of the century (Norval et al., 1991a). This would have been possible because the cattle that introduced the disease were transported from eastern Africa to southern African ports by sea (i.e., in a matter of days), and then inland by rail. The use of ox-drawn transport would then have ensured that the ticks became widely disseminated.

Norval et al. (1991a) used CLIMEX to estimate the climatic suitability of locations in southern Africa for the survival of *R. appendiculatus* in the first half of the century. They found an apparent correlation between climatic conditions and the incidence of ECF, as measured by new outbreaks in Zimbabwe and South Africa (Transvaal). The incidence increased during periods of favorable climatic conditions and decreased during unfavorable periods. It was concluded that vigorous control measures applied during periods when ticks were under severe stress as a result of low rainfall were able to reduce the foci of non-diapausing *R. appendiculatus* and associated ECF in a stepwise fashion, and eventually to eliminate them. The postulated disappearance of introduced eastern African populations of *R. appendiculatus* from southern Africa, in addition to providing an explanation for the change in pattern of seasonal occurrence ECF, also provides a plausible explanation for the concurrent change of behavior of *T. parva* infections in Zimbabwe (i.e., from ECF to January disease) and its disappearance from other parts of the southern African subcontinent. The eradication of ECF from southern Africa can be better explained by the disappearance of particular vector populations that transmitted virulent disease but were not adapted to survive in the region, than by the eradication of *T. parva per se*. This is because of the difficulty of achieving complete control of a vector-borne disease in which there is a well-developed carrier state. Perhaps significantly, none of the other tick-borne diseases of cattle, anaplasmosis, babesiosis and cowdriosis, which have ecologically adapted vectors in southern Africa, have shown any major changes in their distributions in the subcontinent after a half a century or more of intensive tick control (Howell et al., 1981).

5.2. Effects of a Male-produced Aggregation–Attachment Pheromone of *Amblyomma* Ticks on the Occurrence and Epidemiology of Heartwater

Aggregation–attachment pheromones (AAPs) are known to be produced by several ixodid tick species of the genus *Amblyomma*, including *A. hebraeum* and *A. variegatum* (Rechav et al., 1977; Norval and Rechav, 1979). After several days of feeding, males of these species emit a volatile AAP which is used in host location and host selection (Norval et al., 1989a, 1989b; Yunker et al., 1990), it attracts unfed ticks to those areas of the host that are groomed least effectively (Norval et al., 1988a; Norval, 1992), and it brings the sexes together and so facilitates mating (Norval, 1974; Rechav et al., 1977).

Unfed nymphs and adults of *A. variegatum* and *A. hebraeum* do not ascend the vegetation to await passing hosts, as occurs in many other ixodid tick species

such as *R. appendiculatus*. Instead, they seek shelter in favorable microhabitats beneath the debris on the soil surface and only become active in response to specific host stimuli. The stimuli are CO_2, which causes the ticks to emerge on to the soil surface (Norval et al., 1987, 1988b), and the AAP produced by attached males, which provides directional information (Norval et al., 1989a, 1989b, 1992c). The unfed nymphs and adults are able to locate suitable hosts from distances of up to about 25 m in response to CO_2 and AAP (Norval et al., 1992c).

This pheromone-regulated behavior is of considerable importance in the survival of the ticks. It largely eliminates their chances of attaching to unsuitable hosts on which they would be unlikely to survive because of grooming and other factors such as regular treatment with acaricides. For this reason acaricide treatment of cattle appears to have little or no effect on population size if untreated wild hosts, such as giraffe, eland, kudu (*Tragelaphus strepsiceros*) and buffalo, which carry large numbers of adults, share the same pastures as cattle (Norval and Lawrence, 1979; Petney and Horak, 1987). These alternate hosts therefore function as effective reservoirs for the vectors of heartwater, and probably also for *C. ruminantium* itself.

In the absence of alternate hosts *Amblyomma* species are easily controlled using acaricides, because of the long attachment periods of the adults (Norval and Lawrence, 1979; Norval et al., 1992b). Intensive tick control can result in either localized eradication of these tick species or eradication over large areas as has occurred in parts of the highveld of Zimbabwe (Lawrence and Norval, 1979; Norval, 1983a; Norval et al., 1992b). However, the ticks can rapidly re-infest suitable areas if control measures ever fail, as occurred during the pre-independence war in Zimbabwe when *A. hebraeum* became re-established over a large part of the country (Lawrence et al., 1980; Norval, 1983a). The effect of the spread of *Amblyomma* species to uninfested areas is to cause large epizootics of heartwater in domestic livestock (Lawrence et al., 1980). In the long-term the re-infestation of the areas of Africa from which *Amblyomma* species have been eradicated is probably inevitable, due to the costs and organizational problems involved with sustaining intensive tick control programs (Perry et al., 1990b; Norval, 1983b; Norval et al., 1992b). The areas in which alternate hosts are present in addition to livestock will always remain reservoirs of *Amblyomma* species and *C. ruminantium*, from which infection can spread. The AAP which prevents the effective control or eradication of *Amblyomma* ticks over large areas of Africa therefore plays a vital role in the dynamics of heartwater infection.

The AAP is also of importance in the epidemiology of heartwater because it focuses infection in specific hosts (i.e., those with attached males), and this contributes to high infection rates in *Amblyomma* populations (Norval et al., 1990, 1992d). High infection rates in vector populations and a well-developed carrier state, contribute to the endemic stability that occurs for heartwater in much of Africa (Norval et al., 1992d).

The southern African heartwater vector *A. hebraeum* does not have a clearly defined pattern of seasonal occurrence, and larvae, nymphs and adults can be

present on hosts throughout the year. Norval et al. (1992d) attribute this lack of seasonality to the host-finding behavior of the nymphs and adults; the emergence of the ticks for short periods of host-seeking can obviously be fairly independent of weather conditions. The AAP thus allows *A. hebraeum*, at least in part, to overcome the problem of survival through the long dry season of southern Africa. The presence of all stages of the life cycle on hosts throughout the year may also contribute to endemic stability in a part of Africa where this would perhaps not exist if the vectors followed a strictly seasonal pattern of occurrence.

6. DISCUSSION

Progress towards the development of more cost-effective and epidemiologically sound control methods for theileriosis caused by *T. parva* and heartwater is dependent on an understanding of the dynamic associations between the disease organisms, their hosts, the tick vectors, and the environment. In this chapter we have discussed current knowledge of some aspects of the epidemiology of the two diseases, and have shown how methodologies such as GIS and computer models can be used to display, analyze, and interpret some complex interactions that are dynamic in both space and time. Although our understanding of the epidemiology of the diseases is far from complete, sufficient progress has been made to begin to define areas or situations at risk, to predict the outcome of some management interventions and to define the role that immunization may play in the control of the diseases.

A noteworthy example of risk prediction has been the identification, using GIS and the CLIMEX model, of areas of Africa that are climatically suitable for *R. appendiculatus* but are currently uninfested. If *R. appendiculatus* and *T. parva* are ever introduced into these areas, it is reasonable to predict that ECF will become established and will become a major problem. Similarly, it is possible to predict that *Amblyomma* ticks and heartwater will spread into some climatically suitable but presently unaffected areas of Africa, if the currently practiced intensive tick control is relaxed. A more positive finding has been that the ECF problem in southern Africa was probably the result of the introduction of non-diapausing forms of *R. appendiculatus* in association with *T. parva*, rather than simply the introduction of *T. parva*. This means that there is probably no risk of a resurgence of this more virulent form of theileriosis in southern Africa, despite the persistence of *T. parva* in the buffalo population, if *R. appendiculatus* from eastern Africa is prevented from become re-established.

The understanding that *T. parva* is a single taxonomic entity which is expressed in different clinical forms (i.e., ECF, January disease, and Corridor disease) when associated with different mammalian hosts (buffalo or cattle), different tick species (*R. appendiculatus* or *R. zambeziensis*) or different geographic variants within a single tick species (diapausing and non-diapausing populations of *R. appendiculatus*), and that ECF causes greater calf mortality in Taurine than Zebu or Sanga breeds of cattle, allows the effects of various

management or control interventions to be predicted (Norval et al., 1992a). Some examples are as follows. If tick control is relaxed in areas infected with *R. zambeziensis*, clinical theileriosis in cattle presents little risk except if buffalo are present. The risk of Corridor disease will increase in any areas in which theileriosis vectors occur if tick control is relaxed and buffalo are present. Zebu or Sanga cattle can be kept with minimal or no tick control in ECF areas, if endemic stability exists, but in the same circumstances clinical disease will occur in Taurine cattle unless calves are immunized.

With heartwater, the knowledge that infection is widespread in mammalian host and vector populations, that endemic stability can occur with minimal losses, and that the attachment of vectors to hosts is pheromone-mediated also allows the effects of some management and control interventions to be predicted. Some examples are as follows. Losses due to heartwater in cattle, sheep, and goats in endemic areas will remain minimal if no control interventions are undertaken. The risk of losses due to heartwater will increase if tick control is practiced on livestock in the presence of alternate wild hosts, because the AAP will ensure that the wild hosts remain infested with *Amblyomma* ticks and remain sources of infection. The risk of heartwater will increase if intensive tick control is relaxed in areas where the vectors have been eradicated, because such areas are likely to become re-infested. Immunization against heartwater will be required to prevent losses if intensive tick control is practiced in endemic areas and the vectors are suppressed but not eradicated, if the vectors spread to uninfested areas and if susceptible animals are introduced to endemic areas.

Studies on the dynamic associations of tick-borne diseases affecting domestic animal health are being carried out in many areas of the world, but not on the same scale as in Africa where tick-borne diseases of livestock are of considerable importance to both rural communities and the national economies of affected countries. Theileriosis caused by *T. parva* and heartwater have simply been used as examples of how basic epidemiological research can elucidate the causes of disease problems and provide some practical solutions to these problems. The research is continuing, in association with the development of improved diagnostic tests and vaccines, with the ultimate aim of developing disease control systems that are compatible with farming practices in the affected areas, and with the economic constraints that exist.

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8. REFERENCES

Andrew, H.R. and Norval, R.A.I. (1989) The carrier status of sheep, cattle and African buffalo recovered from heartwater. *Vet. Parasitol.* **34**, 261–266.

Anon. (1989) Nomenclature in *Theileria*. pp. 182–186 In: Dolan, T.T. (ed.) *Theileriosis in Eastern, Central and Southern Africa. Proceedings of a Workshop on East Coast Fever Immunization*, Lilongwe, Malawi, 20–22 September 1988. International Laboratory for Research on Animal Diseases, Nairobi.

Asselbergs, M., Jongejan, F., Langa, A., Neves, L., and Afonso, S. (1993) Antibodies to *Cowdria ruminantium* in Mozambican goats and cattle detected by immunofluorescence using endothelial cell culture antigen. *Trop. Anim., Hlth Prod.* **25**, 144–150.

Barnett, S.F. (1957) Theileriosis control. *Bull. Epiz. Dis. Afr.* **5**, 343–357.

Barnett, S.F. and Brockelsby, D. W. (1966) The passage of "Theileria lawrencei (Kenya)" through cattle. *Br. Vet. J.* **122**, 396–409.

Barré, N., Uilenberg, G., Morel, P.C., and Camus, E. (1987) Danger of introducing heartwater onto the American mainland: potential role of indigenous and exotic *Amblyomma* ticks. *Onderst. J. Vet. Res.* **54**, 405–417.

Bezuidenhout, J.D. (1987) Natural transmission of heartwater. *Onderst. J. Vet. Res.* **54**, 349–351.

Bezuidenout. J.D. and Bigalke, R.D. (1987) The control of heartwater by means of tick control. *Onderst. J. Vet. Res.* **54**, 525–528.

Branagan, D. (1973a) Observations on the development and survival of the ixodid tick *Rhipicephalus appendiculatus* (Neumann, 1901) under quasi-natural conditions in Kenya. *Trop. Anim. Hlth Prod.* **5**, 153–165.

Branagan, D. (1973b) The development periods of the ixodid tick *Rhipicephalus appendiculatus* (Neumann) under laboratory conditions. *Bull. Entomol. Res.* **63**, 155–168.

Branagan, D. (1978) Climate and East Coast fever. pp. 126–140. In: Gibson, T.E. (ed.) *Weather and Parasitic Animal Disease*. World Meteorological Organization, Geneva.

Brown, C.G.D., Radley, D.E., Cunningham, M. P., Kirimi, I.M., Morzaria, S.P. and Musoke, A.J. (1977) Immunization against East Coast fever (*Theileria parva* infection of cattle) by infection and treatment: chemoprophylaxis with N-pyrrolidinomethyl tetracycline. *Tropenmed. Parasitol.* **28**, 342–348.

Camus, E. and Barré, N. (1982) *La Cowdriose (heartwater). Revue Générale des Connaissance*. Institut d'Elevage et de Médecine Vétérinaire des pay Tropicaux, Maisons-Alfort, 147 pp.

Cunningham, M.P., Brown, C.G.D., Burridge, M.J., Musoke, A.J., Purnell, R.E., Radley, D.E., and Sempebwa, C. (1974) East Coast fever: titration in cattle of suspensions of *Theileria parva* derived from ticks. *Br. Vet. J.* **13**, 336–345.

da Gracia, H.M. and Serrano, F.M.H. (1971) Contribuicao para o estudo da theileriose cincerina maligna dos bovinos, em Angols. *Acta Vet., Nova Lisboa* **7**, 1–8.

Dolan, T.T., Young, A.S., Losos, G.J., McMillan, I., Minder, C.E., and Soulsby, K. (1984) Dose dependent responses of cattle to *Theileria parva* stabilate infection. *Internat. J. Parasitol.* **14**, 89–95.

FAO (1985) *Kenya: Epizootiological Survey of Tick-borne Cattle Diseases*. Technical report No. 1, AG: DP/KEN/70/522. Food and Agriculture Organization, Rome, 52 pp.

Fivaz, B.H., Norval, R.A.I., and Lawrence, J.A. (1989) Transmission of *Theileria parva bovis* (Boleni strain) to cattle resistant to the brown ear tick *Rhipicephalus appendiculatus* (Neumann). *Trop. Anim. Hlth Prod.* **21**, 129–134.

Grootenhuis, J.G. (1989) The role of wildlife in the epidemiology of cattle theileriosis.

pp. 129–136. In: Dolan, T.T. (ed.) *Theileriosis in Eastern, Central and Southern Africa. Proceedings of a Workshop on East Coast Fever Immunization*, Lilongwe, Malawi, 20–22 September 1988. International Laboratory for Research on Animal Diseases, Nairobi.

Grootenhuis, J.G. and Young, A.S. (1981) The involvement of wildlife in *Theileria* infections of domestic animals in East Africa. In: Irvin, A.D., Cunningham, M.P., and Young, A.S. (eds) *Advances in the Control of Theileriosis; Proceedings of an International Conference*, ILRAD, Nairobi, 9–13 February 1981. Martinus Nijhoff Publishers, The Hague.

Haig, D.A., Alexander, R.A., and Weiss, K.E. (1954) Treatment of heartwater with Terramycin. *J. South Afr. Vet. Med. Assoc.* **25**, 45–48.

Henning, M.W. (1956) *Animal Diseases in South Africa*, 3rd edn. Central News Agency Ltd, Cape Town, 1,239 pp.

Howell, C.J., de Vos, A.J., Bezuidenhout, J.D., Potgieter, F.T., and Barrowman, P.R. (1981) The role of chemical tick eradication in the control or prevention of tick transmitted diseases in cattle. pp. 61–66. In: Whitehead, G.B. and Gibson, J.D. (eds) *Tick Biology and Control; Proceedings of an International Conference*, Grahamstown 27–29 January 1981. Tick Research Unit, Rhodes University, Grahamstown.

Irvin, A.D., Morzaria, S.P., Munatswa, F.C., and Norval, R.A.I. (1989) Immunization of cattle with a *Theileria parva bovis* stock from Zimbabwe protects against challenge with virulent *T. p. parva* and *T. p. lawrencei* stocks from Kenya. *Vet. Parasitol* **32**, 271–278.

Kaiser, M.N., Sutherst, R.W., and Bourne, A.S. (1982) Relationship between ticks and zebu cattle in southern Uganda. *Trop. Anim. Hlth Prod.* **14**, 63–74.

Kariuki, D.P. (1991) Studies on the carrier status of *Theileria parva* in the epidemiology and control of East Coast fever. Ph.D. thesis, University of Salford.

King, D., Gettinby, G., and Newson, R.M. (1988) A climate-based model for the development of the ixodid tick, *Rhipicephalus appendiculatus*, in East Coast fever zones. *Vet. Parasitol.* **29**, 41–51.

Koch, H.T. (1990) Aspects of the epidemiology of January disease (*Theileria parva bovis* infection) in Zimbabwe. Ph.D. thesis, University of Utrecht, 122 pp.

Koch, H.T. and Foggin, C.M. (1991) On the occurrence of January disease (*Theileria parva bovis* infection) in Zimbabwe. pp. 34–46. In: Koch, H.T. (ed) *Aspects of the Epidemiology of January Disease* (Theileria parva bovis, *Infection*) *in Zimbabwe*. Ph.D. thesis, University of Utrecht, Utrecht.

Koch, H.T., Kambeva, L., Norval, R.A.I., Ocama, J.G.R., Masaka, S., Munatswa, F.C., Honhold, N., and Irvin, A.D. (1990) Age resistance to *Theileria parva bovis* infection in calves. *Vet. Parasitol.* **37**, 197–206.

Lawrence, D.A. (1935) *Report of the Director of Veterinary Research for the Year 1934.* Rhodesia Printing and Publishing Co. Ltd, Salisbury, 10 pp.

Lawrence, D.A. (1937) *Report of the Director of Veterinary Research for the Year 1936.* Rhodesia Printing and Publishing Co. Ltd, Salisbury, 12 pp.

Lawrence, J.A. (1979) The differential diagnosis of the bovine theilerias of southern Africa. *J. South Afr. Vet. Med. Assoc.* **50**, 311–313.

Lawrence, J.A. (1991) Retrospective observations on the transmission of East Coast fever in Zimbabwe. *Trop. Anim. Hlth Prod.* **23**, 69–74.

Lawrence, J.A. (1992) History of bovine theileriosis in southern Africa. pp. 2–39. In: Norval, R.A.I., Perry, B.D., and Young, A.S. (eds) In: *The Epidemiology of Theileriosis in Africa.* Academic Press.

Lawrence, J.A. and Norval, R.A.I. (1979) A history of ticks and tick-borne diseases of cattle in Rhodesia. *Rhod. Vet. J.* **10**, 28–40.

Lawrence, J.A., Foggin, C.M., and Norval, R.A.I. (1980). The effects of war on the control of diseases of livestock in Rhodesia (Zimbabwe). *Vet. Rec.* **107**, 82–85.

Lawrence, J.A., Norval, R.A.I., and Uilenberg, G. (1983) *Rhipicephalus zambeziensis* as a vector of bovine Theileriidae. *Trop. Anim. Hlth Prod.* **15**, 39–42.

Lessard, P., L'Eplattinier, R., Norval, R.A.I., Kundert, K., Dolan, T.T., Croze, H., Walker, J.B., Irvin, A.D., and Perry, B.D. (1990) Geographical Information Systems for studying the epidemiology of cattle diseases caused by *Theileria parva*. *Vet. Rec.* **126**, 255–262.

Lewis, E.A. and Fotheringham, W. (1941) The transmission of *Theileria parva* by ticks. *Parasitol.* **33**, 251–277.

Lightfoot, C.J. and Norval, R.A.I. (1981) Tick problems in wildlife in Zimbabwe. 1. The effects of tick parasites on wild ungulates. *South Afr. Wildl. Res.* **11**, 41–45.

Lounsbury, C.P. (1900) Tick heartwater experiments. *Agric. J. Cape Good Hope* **16**, 682–687.

MacKenzie, P.K.I. and Norval, R.A.I. (1980) The transmission of *Cowdria ruminantium* by Amblyomma tholloni. *Vet. Parasitol.* **7**, 265–268.

Maritim, A.C., Young, A.S., Lesan, A.C., Ndungu, S.G., Stagg, D.A., and Ngumi, P.N. (1992) Transformation of *Theileria parva lawrencei* derived from African buffalo (*Syncerus caffer*) by tick passage in cattle and its use in infection and treatment immunization. *Vet. Parasitol.* **43**, 1–14.

Matson, B.A. (1967) Theileriosis in Rhodesia. I. A study of diagnostic specimens over two seasons. *J. South Afr. Vet. Med. Assoc.* **38**, 93–102.

Matthysse, J.G. and Colbo, M.H. (1987) *The Ixodid Ticks of Uganda.* Entomological Society of America, College Park, Maryland, 426 pp.

Minshull, J.I. and Norval, R.A.I. (1982) Factors influencing the spatial distribution of *Rhipicephalus appendiculatus* in Kyle Recreational Park, Zimbabwe. *South Afr. J. Wildl. Res.* **12**, 118–123.

Neitz, W.O. (1939) Die Wirkung von Uleron auf das Herzwasser (*Rickettsia ruminantium*) der Schafe. *Berliner Münchener Tierartzlichen Wochenschrift* **9**, 134.

Neitz, W.O. (1940) Uleron in the treatment of heartwater. *J. South Afr. Vet. Med. Assoc.* **11**, 15.

Neitz, W.O. (1955) Corridor disease: a fatal form of bovine theileriosis encountered in Zululand. *Bull. Epizoot. Dis. Afr.* **3**, 121–123.

Neitz, W.O. (1967) The epidemiological pattern of viral, protophytal and protozoal zoonoses in relation to game preservation in South Africa. *J. South Afr. Vet. Med. Assoc.* **38**, 129–141.

Neitz, W.O. (1968) Heartwater. *Bull. Off. Int. Epizoot.* **70**, 329–336.

Newson, R.M. (1978) The life-cycle of *Rhipicephalus appendiculatus* on the Kenyan coast. pp. 46–50. Wilde, J.K.H. (ed.) *Tick-borne Diseases and their Vectors; Proceedings of an International Conference*, Edinburgh, 27 September–1 October 1976. Centre for Tropical Veterinary Medicine, University of Edinburgh.

Newson, R.M. and Punyua, D.K. (1978) The life cycle of *Rhipicephalus appendiculatus* and associated species in an ecologically marginal situation. pp. 51–55. In: Wilde, J.K (ed.) *Tick-borne Diseases and Their Vectors: Proceedings of an International Conference.* Edinburgh, 27 September–1 October 1976. Centre for Tropical Veterinary Medicine, University of Edinburgh.

Newson, R.M., Chiera, J.W., Young, A.S., Dolan, T.T., Cunningham, M.P., and Radley, D.E. (1984) Survival of *Rhipicephalus appendiculatus* (Acarina: Ixodidae) and

persistence of *Theileria parva* (Apicomplexa: Theileridae) in the field. *Internat. J. Parasitol.* **14**, 483–489.

Norval, R.A.I. (1974) The life cycle of *Amblyomma hebraeum* Koch, 1844 (Acarina: Ixodidae). *J. Entomol. Soc. South Afr.* **37**, 367–367.

Norval, R.A.I. (1974) The life cycle of *Amblyomma hebraeum* Koch, 1985 (Acarina: Ixodidae). *J. Entomol. Soc. South Afr.* **37**, 357–367.

Norval, R.A.I. (1979a) The limiting effect of host availablity for the immature stages on population growth in economically important ixodid ticks. *J. Parasitol.* **65**, 285–287.

Norval, R.A.I. (1979b) Tick infestations and tick-borne diseases in Zimbabwe Rhodesia. *J. South Afr. Vet. Assoc.* **50**, 289–292.

Norval, R.A.I. (1993a) The ticks of Zimbabwe. VII. The genus *Amblyomma*. *Zimb. Vet. J.* **14**, 3–18.

Norval, R.A.I. (1983b) Arguments against intensive dipping. *Zimb. Vet. J.* **14**, 19–25.

Norval, R.A.I. (1992) Host susceptibility to infestation with *Amblyomma hebraeum*. *Insect Sci. Appl.* **13**, 489–494.

Norval, R.A.I. and Lawrence, J.A. (1979) The control of heartwater in Zimbabwe Rhodesia. *Zimb. Rhod. Agric. J.* **76**, 161–165.

Norval, R.A.I. and Lightfoot, C.J. (1982) Tick problems in wildlife in Zimbabwe. Factor infuencing the occurrence and abundance of *Rhipicephalus appendiculatus*. *Zimb. Vet. J.* **13**, 11–20.

Norval, R.A.I. and Perry, B.D. (1990) The introduction, spread and subsequent disappearance of the brown ear-tick *Rhipicephalus appendiculatus*, from the southern lowveld of Zimbabwe. *Exp. Appl. Acarol.* **9**, 103–111.

Norval, R.A.I. and Rechav, Y. (1979) An assembly pheromone and its perception in the tick *Amblyomma variegatum* (Acarina: Ixodidae). *J. Med. Entomol.* **16**, 507–511.

Norval, R.A.I., Fivaz, B.H., Lawrence, J.A., and Brown, A.F. (1985) Epidemiology of tick-borne diseases of cattle in Zimbabwe. III. *Theileria parva* group. *Trop. Anim. Hlth Prod.* **17**, 19–28.

Norval, R.A.I., Yunker, C.E., and Butler, J.F. (1987) Field sampling of unfed adults of *Amblyomma hebraeum* Koch. *Exp. Appl. Acarol.* **3**, 213–217.

Norval, R.A.I., Floyd, R. B., and Kerr, J.D. (1988a) Ability of adults of *Amblyomma hebraeum* (Acarina: Ixodidae) to feed repeatedly on sheep and cattle. *Vet. Parasitol.* **29**, 351–355.

Norval, R.A.I., Yunker, C.E., Gibson, J.D., and Deem, S.L.D. (1988b) Field sampling of unfed nymphs of *Amblyomma hebraeum*. *Exp. Appl. Acarol.* **4**, 173–177.

Norval, R.A.I., Andrew, H.R., and Yunker, C.E. (1089a) Pheromone-mediation of host selection in bont ticks (*Amblyomma hebraeum* Koch). *Science* **243**, 354–365.

Norval, R.A.I., Butler, J.F., and Yunker, C.E. (1989b) Use of carbon dioxide and natural or synthetic aggregation–attachment pheromone of the bont tick, *Amblyomma hebraeum*, to attract and trap unfed adults in the field. *Exp. Appl. Acarol.* **7**, 171–180.

Norval, R.A.I., Andrew, H.R., and Yunker, C.E. (1990) Infection rates with *Cowdria ruminantium* of nymphs and adults of the bont tick (*Amblyomma hebraeum*) collected in the field in Zimbabwe. *Vet. Parasitol.* **36**, 277–283.

Norval, R.A.I., Lawrence, J.A., Young, A.S., Perry, B.D., Dolan, T.T., and Scott. J. (1991a) *Theileria parva*: influence of vector, parasite and host relationships on the epidemiology of theileriosis in southern Africa. *Parasitol.* **102**, 347–356.

Norval, R.A.I., Perry, B.D., Gebreab, F., and Lessard, P. (1991b) East Coast fever: a problem of the future for the horn of Africa? *Prev. Vet. Med.* **10**, 163–172.

Norval, R.A.I., Perry, B.D., Kruska, R., and Kundert, K. (1991c) The use of climate data interpolation in estimating the distribution of *Amblyomma variegatum* in Africa. *Prev. Vet. Med.* **11**, 365–366.

Norval, R.A.I., Perry, B.D., and Young, A.S. (1992a) *The Epidemiology of Theileriosis in Africa.* Academic Press, London, 481 pp.

Norval, R.A.I., Perry, B.D., and Hargreaves, S.K. (1992b) Tick and tick-borne disease control in Zimbabwe: what might the future hold? *Zimb. Vet. J.* **23**, 1–16.

Norval, R.A.I., Peter, T., and Meltzer, M.I. (1992c) A comparison of the attraction of nymphs and adults of the ticks *Amblyomma hebraeum* and *A. variegatum* to carbon dioxide and the male-produced aggregation attachment pheromone. *Exp. Appl. Acarol.* **13**, 179–186.

Norval, R.A.I., Andrew, H.R., and Meltzer, M.I. (1992d) Seasonal occurrence of the bont tick (*Amblyomma hebraeum*) in the southern lowveld of Zimbabwe. *Exp. Appl. Acarol.* **13**, 81–96.

Norval, R.A.I., Andrew, H.R., Yunker, C.E., and Burridge, M.J. (1992d) Biological processes in the epidemiology of heartwater. pp. 71–86. Fivaz, B.H., Petney, T.N., and Horak, I.G. (eds) *Tick Vector Biology. Veterinary and Medical Aspects.* Springer-Verlag, Berlin.

Oberem, P.T. and Bezuidenhout, J.D. (1987a) Heartwater in hosts other than domestic ruminants. *Onderst. J. Vet. Res.* **54**, 271–275.

Oberem, P.T. and Bezuidenhout, J.D. (1987b). The production of heartwater vaccine. *Onderst. J. Vet. Res.* **54**, 485–488.

Pegram, R.G. and Banda, D.S. (1990) Ecology and phenology of cattle ticks in Zambia: development and survival of free-living stages. *Exp. Appl. Acarol.* **8**, 291–301.

Perry, B.D., Lessard, P., Norval, R.A.I., Kundert, K., and Kruska, R. (1990a) Climate, vegetation and the distribution of *Rhipicephalus appendiculatus in Africa. Parasitol. Today* **6**, 100–104.

Perry, B.D., Mukhebi, A.W., Norval, R.A.I., and Barrett, J.C. (1990b) A preliminary assessment of current and alternative tick and tick-borne diseases control strategies in Zimbabwe. Report to the Director of Veterinary Services (Zimbabwe). International Laboratory for Research on Animal Diseases, Nairobi, 41 pp.

Perry, B.D., Norval, R.A.I., Kruska, R.L., Ushewokunze-Obatolu, U., and Booth, T.H. (1991) Predicting the epidemiology of tick-borne diseases of cattle in Zimbabwe using geographic information systems. pp. 214–216. *Proceedings of the 6th International Symposium on Veterinary Epidemiology and Economics*, Ottawa, August 1991.

Petney, T.N. and Horak, I.G. (1987) The effect of dipping on parasitic and free-living populations of *Amblyomma hebraeum* on a farm and on an adjacent nature reserve. *Onderst. J. Vet. Res* **54**, 529–533.

Petney, T.N., Horak, I.G., and Rechav, Y. (1987) The ecology of the African vectors of heartwater, with particular reference to *Amblyomma hebraeum* and *Amblyomma variegatum. Onderst. J. Vet. Res.* **54**, 381–395.

Provost, A. and Bezuidenhout, J.D. (1987) The historical background and global importance of heartwater. *Onderst. J. Vet. Res.* **54**, 165–169.

Purnell, R.E., Ledger, M.A., Omwoyo, P.L., Payne, R.C., and Pierce, M.A. (1974)

Theileria parva: variation in the infection rate of the vector tick, *Rhipicephalus appendiculatus*. *Internat. J. Parasitol.* **4**, 513–517.

Radley, D.E. (1978) *Immunization against Theileriosis by Chemoprophylaxis*. FAO Technical Report A.G., DP/RAF/67/077, FAO, Rome.

Radley, D.E. (1981) Infection and treatment immunization against theileriosis. pp. 227–236. In: Irvin, A.D., Cunningham, M.P., and Young, A.S. (eds) *Advances in the Control of Theileriosis: Proceedings of an Internaional Conference*, ILRAD, Nairobi, 9–13 February 1981. Martinus Nijhoff Publishers, The Hague.

Rechav, Y. (1981) Ecological factors affecting seasonal activity in the brown ear tick *Rhipicephalus appendiculatus*. pp. 187–192. In: Whitehead, G.B. and Gibson, J.D. (eds) *Tick Biology and Control. Proceedings of an International Conference*, Grahamstown, 27–29 January 1981. Tick Research Unit, Rhodes University, Grahamstown.

Rechav,Y. (1982) Dynamics of tick populations (Acari: Ixodoidea) in the Eastern Cape Province of South Africa. *J. Med. Entomol.* **19**, 679–700.

Rechav, Y., Parolis, H., Whitehead, G.B., and Knight, M.M. (1977) Evidence for an assembly pheromone(s) produced by males of the bont tick, *Amblyomma hebraeum* (Acarina: Ixodidae). *J. Med. Entomol.* **14**, 71–78.

Rechav, Y., Norval, R.A.I., and Oliver, J.H. (1982) Interspecific mating of *Amblyomma hebraeum* and *Amblyomma variegatum* (Acari: Ixodidae). *J. Med. Entomol.* **19**, 139–142.

Short, N.J. and Norval, R.A.I. (1981a) Regulation of seasonal occurrence in the tick *Rhipicephalus appendiculatus* Neumann, 1901. *Trop. Anim. Hlth Prod.* **13**, 19–26.

Short, N.J. and Norval, R.A.I. (1981b) The seasonal activity of *Rhipicephalus appendiculatus* Neumann, 1901 (Acari: Ixodidae) in the highveld of Zimbabwe Rhodesia. *J. Parasitol.* **67**, 77–84.

Short, N.J., Floyd, R.B., Norval, R.A.I., and Sutherst, R.W. (1989a) Development rates, fecundity and survival of developmental stages of the ticks *Rhipicephalus appendiculatus*, *Boophilus decoloratus* and *B. microplus* under field conditions in Zimbabwe. *Exp. Appl. Acarol.* **6**, 123–141.

Short, N.J., Floyd, R.B., Norval, R.A.I., and Sutherst, R.W. (1989b) Survival and behavior of unfed stages of the ticks *Rhipicephalus appendiculatus, Boophilus decoloratus* and *B. microplus* under field conditions in Zimbabwe. *Exp. Appl. Acarol.* **6**, 215–236.

Smith, M.W. (1969) Variations in tick species and populations in the Bugisu District of Uganda. Part II. The effects of altitude, climate, vegetation and husbandry on tick species and populations. *Bull. Epiz. Dis. Afr.* **17**, 77–105.

Stagg, D.A., Young, A.S., Leitch, B.L., Grootenhuis, J.G., and Dolan, T.T. (1983) Infection of mammalian cells with *Theileria* species. *Parasitol.* **86**, 243–254.

Stampa, S. (1969) Experiences with heartwater eradication. pp. 79–85. *Proceedings of a Symposium on Tick Biology and Control*. Rhodes University, Grahamstown.

Sutherst, R.W. (1987) The dynamics of hybrid zones between (Acari) tick species. *Internat. J. Parasitol.* **17**, 921–926.

Sutherst, R.W. and Maywald, G.F. (1985) A computerized system for matching climates in ecology. *Agric. Ecosyst. Environ.* **13**, 281–299.

Tatchell, R.J. and Easton, E. (1986) Tick (Acari: Ixodidae) Ecological studies in Tanzania. *Bull. Entomol. Res.* **76**, 229–246.

Uilenberg, G. (1976) Tick-borne livestock diseases and their vectors. 2. Epizootiology of tick-borne diseases. *Wld. Anim. Rev.* **17**, 8–15.

Uilenberg, G. (1981) Theilerial species of domestic animals. pp. 4–37. In: Irvin, A.D., Cunningham, M.P., and Young, A.S. (eds) *Advances in the Control of Theileriosis: Proceedings of an International Conference*, ILRAD, Nairobi, 9–13 February 1981. Martinus Nijhoff Publishers, The Hague.

Van der Merwe, L. (1987) The infection and treatment method of vaccination against heartwater. *Onderst. J. Vet Res.* **54**, 489–491.

Walker, A.R., Latif, A.S., Morzaria, S.P., and Jongejan, F. (1983) Natural infection rate of *Hyalomma anatolicum* with *Theileria* in Sudan. *Res. Vet. Sci.* **35**, 87–90.

Walker, J.B. and Olwage, A. (1987) The tick vectors of *Cowdria ruminantium* (Ixodoidea, Ixodidae, Genus *Amblyomma*) and their distribution. *Onderst. J. Vet. Res.* **54**, 354–379.

Weiss, K.E., Haig, D.A., and Alexander, R.A. (1952) Aureomycin in the treatment of heartwater. *Onderst. J. Vet. Res.* **25**, 41–50.

Wilde, J.K.H. (1967) East Coast fever. *Adv. Vet. Sci.* **11**, 207–259.

Yeomann, G.H. (1966) Field vector studies of epizootic East Coast fever. I. A quantitative relationship between *Rhipicephalus appendiculatus* and the epizooticity of East Coast fever. *Bull. Epizoot. Dis. Afr.* **14**, 5–27.

Yeoman, G.H. (1967) Field vector studies of epizootic East Coast fever. III. Pasture ecoogy in relation to *Rhipicephalus appendiculatus* infection rates on cattle. *Bull Epizoot. Dis. Afr.* **15**, 89–113.

Young, A.S. (1981) The epidemiology of theileriosis in East Africa. pp. 38–55. In: Irvin, A.D., Cunningham, M.D., and Young, A.S. (eds) *Advances in the Control of Theileriosis: Proceedings of an International Conference*, ILRAD, Nairobi, 9–13 February 1981. Martinus Nijhoff Publishers, The Hague.

Young, A.S. and Purnell, R.E. (1973) Transmission of *Theileria lawrencei* (Serengeti) by the ixodid tick *Rhipicephalus appendiculatus*. *Trop. Anim. Hlth Prod.* **5**, 146–152.

Young, A.S., Leitch, B.L., Morzaria, S.P., Irvin, A.D., Omwoyo, P.L., and de Castro, J.J. (1987) Development and survival of *Theileria parva* in *Rhipicephalus appendiculatus* exposed in the Trans-Mara, Kenya. *Parasitol.* **94**, 433–441.

Young, A.S., Groocock, C.M., and Kariuki, D.P. (1988) Integrated control of ticks and tick-borne diseases of cattle in Africa. *Parasitol.* **96**, 493–432.

Yunker, C.E., Andrew, H.R., Norval, R.A.I., and Keirans, J.E. (1990) Interspecific attraction to male-produced pheromones of two species of *Amblyomma* ticks (Acari: Ixodidae). *J. Insect Behav.* **3**, 557–565.

10

Changing Ecology of Rocky Mountain Spotted Fever

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1. INTRODUCTION

Rocky Mountain spotted fever (RMSF) is caused by *Rickettsia rickettsii*, a prototype species of the spotted fever group (SFG) rickettsiae. The disease was not recognized as a clinical entity until 1899 when pioneers began clearing the Bitterroot Valley of northwestern Montana for farms and settlements (Burgdorfer, 1975). Between 1873 and 1910 there were 295 cases of RMSF in Bitterroot Valley of western Montana, 190 of these were fatal (Burgdorfer, 1989). Most RMSF cases occurred in males, as they ventured into the field to hunt, fish, and trap. The development of this RMSF focus and the circumstances surrounding it, i.e., rickettsiae, harbored by ticks, being actively passed to humans establishing their residences in the previously uninhabited lands, have since been repeated in numerous areas. The discovery of the etiologic agent, the tick vector (*Dermacentor andersoni*), and vertebrate reservoirs (rodents) of RMSF in the northwestern Rocky Mountains was a significant scientific achievement in the area of disease ecology.

Today, RMSF continues to be the most important rickettsial disease of humans in terms of morbidity and mortality in the continental United States. RMSF is no longer a disease of adult males, cases are seen with much more frequency in women and children living in suburban areas in the eastern US. RMSF was originally reported from the Rocky Mountain region and, although it has been recognized in 46 states, it has recently become more prevalent in the south Atlantic region. The RMSF distributional changes that occurred during the past several decades are strongly correlated with many man-made ecological changes, and increased levels of inter- and intraspecies interactions. The vector–reservoir system that maintains *R. rickettsii* in nature is multifaceted. Although maintenance of *R. rickettsii* in nature occurs in vector ticks through transovarial and transstadial transmissions for several generations, new lines of ticks may acquire infection from rickettsemic mammalian hosts. Stable transovarial and transstadial transmission of *R. rickettsii* has been observed in laboratory infected *D. andersoni* and *D. variabilis* ticks. However, investigation

of the role of mammalian reservoir hosts has revealed considerable variation in the susceptibility of mammals to develop rickettsemia of sufficient magnitude and duration so as to infect feeding ticks and continue the *R. rickettsii* cycle (Burgdorfer, 1989). Aside from the maintenance and transmission of *R. rickettsii* in nature, which requires intercellular existence within selective target cells, very little is known about *R. rickettsii* and its relationships to other tick endocytobionts. The curious geographical clustering and distribution of RMSF are partially explained by the selective behavior of *R. rickettsii* in its use of a limited number of highly efficient vectors. In addition, transovarial and transstadial passage of *R. rickettsii* within these tick vectors in nature ensures the survival of this pathogen without the complexity inherent to an obligate multi-host reservoir system. In this chapter the ecology of RMSF is viewed in the context of the *R. rickettsii*-selective requirements as an intracellular parasite, and its survival in a dynamic and rapidly changing vector–reservoir system. Although *R. rickettsii*, the etiologic agent of RMSF, has been recognized only in the western hemisphere, RMSF cases are observed in several provinces of Canada, Mexico, Central America (Panama and Costa Rica), and South America (Colombia and Brazil). Nonetheless, the discussion in this chapter is limited to the ecology of RMSF in the United States. For further information on RMSF, as it occurs outside the US, the reader is referred to McDade and Newhouse (1986).

2. GEOGRAPHIC DISTRIBUTION AND INCIDENCE OF ROCKY MOUNTAIN SPOTTED FEVER

The fluctuation of RMSF in the US is rather remarkable if one looks at the incidence of RMSF over the past 90 years and its changing geographic distribution. A major change in RMSF distribution was the preponderance of cases in the western US in the early parts of this century (1900–1945) and the increased prevalence in the eastern states during 1970s through 1980s (McDade and Newhouse, 1986; Burgdorfer, 1989). In addition to the geographic shift, there have been marked fluctuations in the number of reported human cases (Fig. 10.1). While the reported RMSF cases increased by seven-fold in the south Atlantic region during 1980–1984 as compared to 1930–1934, the incidence was decreased almost ten-fold in the Rocky Mountain states in the same period (reviewed in McDade and Newhouse, 1986; Burgdorfer, 1989). During the 1980s, reported RMSF cases followed a declining trend from a high of 1,170 cases in 1981 to approximately 600 cases reported annually in recent years (Anon., 1991). The number and rate of cases have remained stable since 1985 with the overall annual incidence rate ranging from 0.24 to 0.32 per 100,000 population. In the last decade, over 95% of the cases reported to the Centers for Disease Control occur in the southern, southeastern, and eastern states (Anon., 1991). Although North Carolina reported the highest number of the cases ($\bar{X}$ = 179/year), Oklahoma had the highest per capita rate during 7 of the 10 years ($\bar{X}$ = 3.1/100,000).

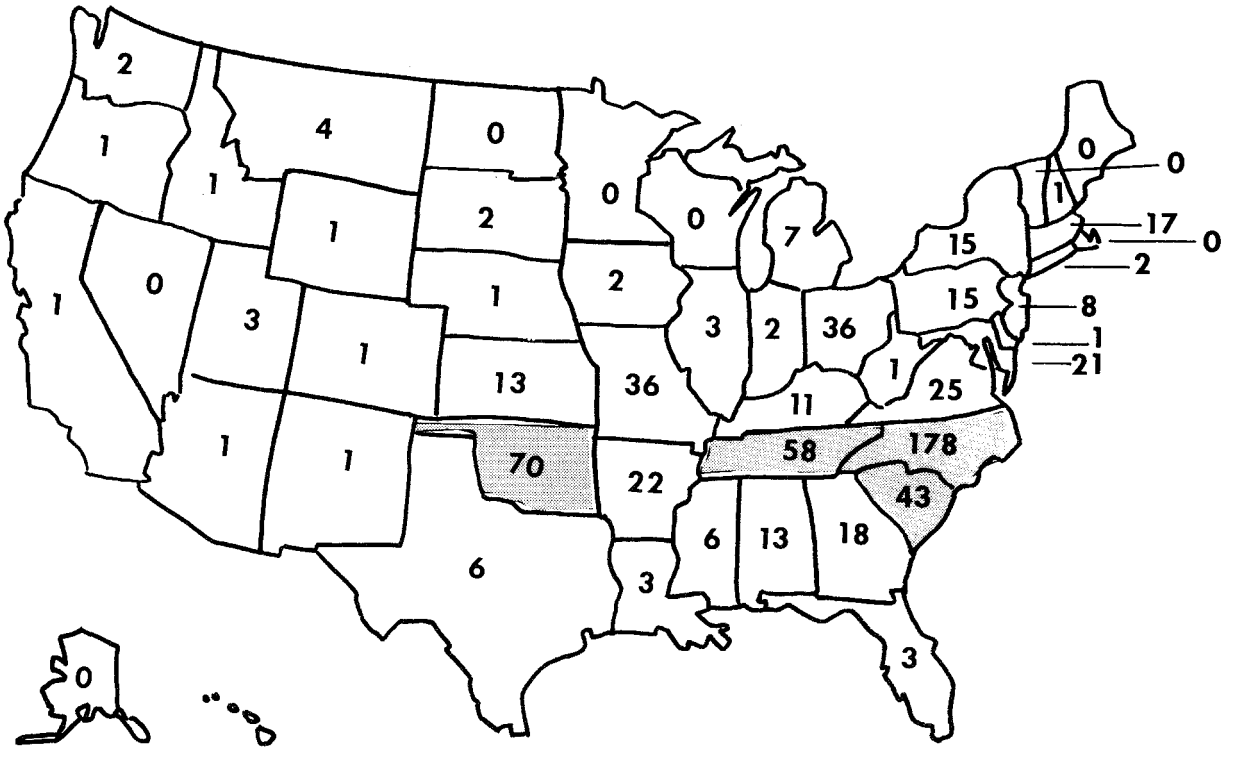

Fig. 10.1. Reported cases of Rocky Mountain spotted fever, by state—United States, 1990 (states that reported ⩾ 1.0 cases/100,000 population are shaded). Data from CDC/MMWR 40, 452, 1991.

During 1989, 224 (37.1%) of 603 reported cases of RMSF were from south Atlantic states (Georgia, Maryland, North Carolina, and Virginia) and 100 (16.6%) from the west south central regions (Anon., 1991). The states with the high incidence rates were Oklahoma (1.9/100,000, 62 cases), North Carolina (1.8/100,000, 118 cases), South Carolina (1.1/100,000, 40 cases), and Missouri (1.0/100,000, 53 cases). Although there has been an overall declining trend in the number of cases in the US, two mid-Atlantic states, New Jersey and Pennsylvania, recently reported large increases in the number of cases (New Jersey: from none in 1988 to 26 cases in 1989; Pennsylvania: from two cases to 23 cases) (Anon., 1991). In 1990, over 50% of the 649 reported cases of RMSF were from the following states: North Carolina (178 cases, 2.7/100,000), Oklahoma (70 cases, 2.2/100,000),Tennessee (58 cases, 1.2/100,000), and South Carolina (43 cases, 1.2/100,000). Despite increased public and primary-care awareness, and improved laboratory recognition of RMSF there is no satisfactory explanation for the observed fluctuations in annual incidence and geographical distribution of RMSF.

3. ECOLOGY OF ROCKY MOUNTAIN SPOTTED FEVER

3.1. The Tick Vectors of *Rickettsia rickettsii*

The spotted fever group includes the etiologic agents of six human diseases (Table 10.1), and 15 other species with low, unknown, or no pathogenicity to humans or certain laboratory animals. *Rickettsia rickettsii*, the etiologic agent of RMSF, is confined to the western hemisphere, and is transmitted to humans during the feeding of infected ixodid ticks. Although many genera and species of ixodid ticks are found naturally infected with rickettsiae, in the US, the wood tick, *D. andersoni*, and the American dog tick, *D. variabilis*, are considered the major vectors of *R. rickettsii* to humans. *D. andersoni* and *D. variabilis* are, respectively, the most common vectors of *R. rickettsii* in the western and eastern US. The lone star tick, *Amblyomma americanum*, the rabbit tick, *Haemaphysalis leporispalustris*, and *D. parumapertus* are also incriminated as vectors of RMSF. Although natural infection with rickettsiae of the SFG has also been observed in the following ticks, *D. occidentalis* and *Ixodes pacificus* from California (Lane et al., 1981), *I. brunneus* from eastern US (Clifford et al., 1969), *I. cookei*, *I. dentatus*, *I. scapularis* and *I. texanus* from Virginia (Burgdorfer, 1975), and *Rhipicephalus sanguineus* from Mississippi (Burgdorfer, 1975), the role of these species in the ecology of RMSF is not well understood. While *D. andersoni* and *D. variabilis* transmit *R. rickettsii* to humans in Canada, *R. sanguineus* and *A. cajennese*, serve as vectors in the western and central parts of Mexico and South America (reviewed in McDade and Newhouse, 1986).

Since the determination of infection in ticks has historically relied heavily on the use of SFG-specific anti-serum in the tick hemolymph test or similar analysis of other tick tissues, and since the etiologic agent of RMSF, *R. rickettsii*, shares many antigenic components with the pathogenic and non-pathogenic members of the SFG rickettsiae (Phillip et al., 1976; Anacker et al., 1987;

Table 10.1. Some epidemiologic features of spotted fever group rickettsiae

Rickettsia species	Disease	Natural cycle		Mode of transmission	Geographical distribution
		Vectors	Host animals		
R. rickettsii	RMSF	*Dermacentor*, *Amblyomma*, *Rhipicephalus*, *Haemaphysalis*	Small mammals, dogs, rabbits, birds	Tick bite	Western hemisphere
R. conori	Boutonneuse fever	*Rhipicephalus*, *Hyalomma*, *Haemaphysalis*, *Dermacentor*	Small mammals, dogs, birds	Tick bite	Middle East, Mediterranean, Africa
R. sibirica	North Asian tick typhus	*Dermacentor*, *Rhipicephalus*, *Haemaphysalis*	Rodents, birds	Tick bite	Siberia, Central Asia, Mongolia
R. australis	Queensland tick typhus	*Ixodes holocyclus*	Rodents, small marsupials	Tick bite	Australia
R. akari	Rickettsial pox	*Liponyssoides sanguineus*	House mice, rats?	Mite bite	North America Russia, Africa
R. japonica	Japanese spotted fever	Tick?	Rodents?	Tick bite?	Japan

Anderson et al., 1990), the results of these widely used tests (Burgdorfer, 1970) should be regarded cautiously. Extensive morphological similarity, at both the electron and light microscopic levels, makes it difficult to differentiate rickettsia, rickettsia-like endocytobionts, and Gram-negative bacteria. Although the hemolymph test in conjunction with fluorescent antibody staining using rabbit or guinea-pig polyclonal antibodies against *R. rickettsii*, distinguishes members of the SFG rickettsiae from rickettsia-like organisms, more elaborate techniques are required to obtain precise identification of rickettsiae in tick (below). However, despite the serological cross-reactivity between the members of the SFG, the development and extensive use of the hemolymph test has been fundamental to tick/ rickettsial surveys in the US and has enabled identification of closely related species of rickettsiae. Compilation of hemolymph SFG infection rates on a state-by-state basis reveals considerable variability. For example, the infection rate for adult *D. variabilis* collected from vegetation and hosts was 2–9% in Connecticut (Magnarelli et al., 1983), 5% in Long Island, NY (Benach et al., 1981), 6% in Kentucky–Tennessee and in Maryland (Azad et al., unpublished), 8.8% in Arkansas, and 10% in Alabama (Burgdorfer, 1975). Similarly, a range of infection rates is often observed for different developmental stages of a given tick within the same area. In a rickettsial/tick survey carried out in Montpelier, Virginia, SFG-infection rates of 2.3, 4.6, and 4.8% for *D. variabilis* larval, nymphal, and adult stages, respectively, were observed (Sonenshine, unpublished; Sonenshine and Stout, 1971; Sonenshine and Clifford, 1973). Rickettsia/tick surveys indicate that *R. rickettsii* is less prevalent in vector ticks than some of the other SFG rickettsiae. The low prevalence of *R. rickettsii* in SFG hemolyph-positive ticks remains puzzling. In many instances where adult ticks were collected in endemic areas with recent reported human cases, very few *R. rickettsii* isolates were obtained. In the US, four described species (*R. rickettsii*, *R. rhipicephali*, *R. montana*, and *R. bellii*) as well as several undescribed species of rickettsiae are commonly found in ixodid ticks (Table 10.2) (Benach et al., 1977; Lovinget al., 1978; Hayes and Burgdorfer, 1979; Philip and Casper, 1981; Philip et al., 1983; Gordon et al., 1984). So far there is no evidence for human infections with *R. rhipicephali*, *R. montana*, and *R. bellii*, nor have any of these species been isolated from patients.

Identification of these rickettsial agents in ticks has been accomplished by isolation of rickettsiae and serologic typing using a microimmunofluorescence assay (microlFA) (Philip et al., 1978). MicrolFA analysis of 106 rickettsial isolates from hemolymph-positive *D. andersoni* from Bitterroot Valley of western Montana identified the presence of 47 *R. rhipicephali*, 41 *R. bellii*, ten *R. rickettsii*, and eight *R. montana* (Philip and Casper, 1981). In a rickettsial/tick survey in Maryland during April through August of 1980 and 1981, we found only two *R. rickettsii* isolates out of 152 hemolymph-positive *D. variabilis* collected from dogs (Table 10.3). Most of the remaining isolates were WB8-2, as yet an undescribed, non-pathogenic, rickettsial species commonly found in *A. americanum* (Burgdorfer et al., 1981). The *R. montana* serotype was predominant in *D. variabilis* in North Carolina where the RMSF incidence is the highest in the US (Anon. 1991). Burgdorfer (1989) reported that, of

Table 10.2. Non-pathogenic spotted fever group rickettsiae.[a]

Rickettsia species	Natural cycle		Geographical distribution
	Vectors	Host animals	
R. montana	*Dermacentor andersoni* and *D. variabilis*	Small mammals	13 states
R. rhipicephali	*Rhipicephalus sanguineus*, *D. andersoni*, *D. variabilis*, *D. occidentalis*	Small mammals	5 states
R. bellii	*Dermacentor andersoni*, *D. variabilis*, *D. occidentalis*, *D. albipictus*, *Haemaphysalis leporispalustris*, *Argas cooleyi*, *Ornithodoros concanensis*	Small mammals	8 states
R. parkeri	*Amblyomma americanum*, *A. maculatum*	Domestic animals	5 states

[a]Excluding five as yet undescribed species of spotted fever group rickettsiae: 364-D, WB-8-2, the East side agent, Tillamook, and the *D. parumapertus* agent.

77 SFG-tick isolates, only one serotyped as *R. rickettsii*, all others were *R. montana*. Infection rates as high as 42% have been reported for WB8-2 in *A. americanum* taken from animals or vegetation in areas surrounding Fayetteville, Arkansas (Burgdorfer et al., 1981). Although *A. americanum* has been found to be an efficient experimental vector for *R. rickettsii*, its role in the transmission of Rocky Mountain spotted fever remains to be elucidated. Despite the occasional occurrence of natural *R. rickettsii* infections in members of the genera *Amblyomma*, *Ixodes* and *Haemaphysalis*, the role of

Table 10.3. Species composition of spotted fever group hemolymph-positive ticks as determined by microimmonofluorescence test

Rickettsial species	*D. andersoni*	*D. variabilis*		
	Montana[a]	Ohio[b]	Long Island[c]	Maryland[d]
Total no. isolates	106	22	100	26
R. rickettsii	10 (9%)	4 (18%)	0 (0%)	2 (8%)
R. rhipicephali	47 (44%)	0 (0%)	0 (0%)	0 (0%)
R. montana	8 (7%)	13 (59%)	100 (100%)	0 (0%)
R. bellii	41 (39%)	4 (18%)	0 (0%)	1 (4%)
Undetermined	—	1 (5%)	—	23 (88%)[e]

[a]Philip and Casper (1981); [b]Gordon et al. (1984); [c]Benach et al. (1977); [d]Azad et al. (unpublished). [e]WB-8-2.

these groups in the ecology of Rocky Mountain spotted fever is not well understood.

3.2. Animal Sources for Infecting Ticks with *Rickettsia rickettsii*

The persistence and maintenance of *R. rickettsii* in nature involves a complicated web of vector–reservoir systems. While infected ticks maintain rickettsiae through transovarial and transstadial transmission, rickettsemic mammalian hosts may serve as sources to infect new lines of uninfected ticks (Koch, 1982; Norment and Burgdorfer, 1984, 1985; Gage et al., 1992). A large variety of mammals and birds serve as hosts for different stages of vector ticks. Since both ticks and rickettsiae are obligate parasites, by the nature of their need they come into contact with the vertebrate hosts. Despite the extensive range of SFG-seropositive vertebrate hosts, relatively few isolates of *R. rickettsii* have been reported. This is largely due to the transient period of rickettsemia, rarely lasting more than 8 days. To date *R. rickettsii* has been isolated from meadow vole (*Microtus pennsylvanicus*), pine vole (*Pitymus pinetorum*), white-footed mouse (*Peromyscus leucopus*), cotton rat (*Sigmodon hispidus*), cottontail rabbit (*Sylbilagus floridanus*), Rocky Mountain cottontail rabbit (*S. nuttallii*), snowshoe hare (*Lepus americanus*), opossum (*Didelphis marsupialis virginiana*), chipmunks (*Eutamias amoenus*), and golden-mantled ground squirrels (*Spermophilus lateralis tescorum*) (McDade and Newhouse, 1986; Burgdorfer, 1989). In contrast to the limited number of *R. rickettsii* isolations from vertebrate hosts, seropositive animals include over 50 species of mammals, particularly lagomorphs and rodents, and several species of ground-frequenting birds (McDade and Newhouse, 1986). Although serological results are indicative of past infection with the rickettsiae, they by no means demonstrate a role for a particular animal in the infection of ticks in a given area. For example, the potential role of the cottontail rabbit as an important reservoir of *R. rickettsii* and the main host for infecting *D. andersoni* ticks was raised because of the remarkable correlation between the occurrence of RMSF cases and the geographic distribution of *S. nuttallii*. However, laboratory studies suggest that, although cottontail rabbits are susceptible to *R. rickettsii*, they do not serve as efficient hosts for infecting ticks (Burgdorfer et al., 1980). A role for cotton rats, *S. hispidus*, in rickettsial maintenance and infection of ticks in the enzootic cycle of RMSF in southern US and Okalahoma was also recently suggested (Gage et al., 1990). Nonetheless, the overall occurrence of human RMSF cases do not seem to depend on the presence of a particular vertebrate reservoir host. Experimental infection of golden-mantled ground squirrels, meadow voles, chipmunks, and snowshoe hares with *R. rickettsii* have shown rickettsemias of sufficient concentrations to infect laboratory-reared ticks (Burgdorfer et al., 1966). Although rickettsemias of 3 to 10 days in duration have been observed in other vertebrates (Bozeman et al., 1967, Burgdorfer et al., 1966, Lundgren et al., 1966), the level of rickettsemia needed to infect feeding ticks was not achieved. According to Burgdorfer et al. (1966), host rickettsemia levels of 10–100 guinea-pig infectious

doses/0.5 ml blood are required for infection of 50% of *D. andersoni*. The role of dogs as a source for infecting ticks also gained notoriety because of the high seropositivity rates of dogs in RMSF endemic areas (Sexton et al., 1976). Although dogs become infected with *R. rickettsii*, and exhibit signs of RMSF illness and rickettsemia, their reservoir potential in the dissemination of *R. rickettsii* to ticks has been dismissed, again owing to the low level and transient nature of the rickettsemias observed in these animals (Norment and Burgdorfer, 1984).

3.3. *Rickettsia rickettsii* Infection in Tick Vectors

Ticks may acquire rickettsiae through transovarial and transstadial transmission, during feeding on the rickettsemic hosts, or via venereal transmission. Passage of rickettsiae into ovarian tissues depends on the severity of infection and a generalized rickettsial dissemination into vector tissues. In efficient tick vectors, SFG rickettsiae produce a generalized infection with rickettsial dissemination into midgut epithelial linings, small intestine, malpighian tubules, hemocytes, salivary glands, and ovarian tissues (Burgdorfer, 1989). Transovarial transmission of *R. rickettsii* plays an important role in the maintenance of this rickettsiosis in nature. *R. rickettsii* infection in field-collected unfed larvae varies from 2% to 15%. In contrast, ticks that acquire *R. rickettsii* yield 30–100% transovarial transmission. Burgdorfer and Brinton (1975) demonstrated that naturally infected *D. andersoni*, *D. variabilis*, and *H. leporispalustris* females passed *R. rickettsii* to 100% of their progeny. These investigators have also shown that *D. andersoni* infected with the Sawtooth strain of *R. rickettsii* maintained 100% transovarial transmission through 12 generations. Transovarial passage nevertheless had an adverse biological effect on tick development and, beginning with the fifth filial generations, increasing numbers of infected ticks died within 1–2 weeks after engorgement (Burgdorfer and Brinton, 1975). In addition, the surviving ticks oviposited smaller numbers of eggs which failed to develop (Burgdorfer and Brinton, 1975).

Acquisition of *R. rickettsii* by ticks feeding on an infected host depends upon the ingestion of a "threshold" number of rickettsiae. In *D. andersoni* the minimum number of rickettsiae required to infect 50% of ticks was estimated to range from 10 to 100 guinea-pig infectious doses/0.5 ml blood (Burgdorfer, 1989). Laboratory experiments with virulent and avirulent strains of *R. rickettsii* revealed that host rickettsemias with sufficient concentrations and duration are required for infecting ticks (Burgdorfer et al., 1966). Susceptible hosts such as meadow voles and chipmunks develop high levels of rickettsemias (100–1,000 guinea-pig infectious dose), sufficient to infect > 50% of feeding ticks. However, under natural conditions, a variety of factors influence the host infection and the outcome of rickettsemias. Very little is known about the behavior of various strains of *R. rickettsii* in the mammalian hosts, their persistence and circulation in the host peripheral blood. Similarly, with the exception of known vectors of RMSF, no information is available for other species of ticks which are

commonly found on small and medium-sized mammals in endemic areas. The overall significance of infected mammalian hosts in rickettsial maintenance and transmission to uninfected ticks is not well understood due to a paucity of field and experimental information.

Venereal transmission of rickettsiae from infected males to female ticks may occur mechanically through spermatozoa or spermiophore fluids (Hayes and Burgdorfer, 1982). The observed absence of rickettsiae in the progeny of venereally infected females (Burgdorfer, 1989) indicates that this mode of rickettsial transmission from infected males to uninfected females is of minor significance.

3.4. Changing Ecology of Rocky Mountain Spotted Fever

In the US, the basis of the dramatic shift of RMSF cases from wooded highland regions of northwestern Rocky Mountains to locations along the eastern Piedmont remains unanswered. Whether this presumed shift of RMSF cases was due to better recognition and prevention of the disease through control measure in the mountain regions, or whether the disease has undergone relocation diffusion is still open to question. The expansion of human populations into previously uninhabited regions has changed the ecology of RMSF by generating harborage sites, and steady food supply for small and medium-sized mammals, such as white-footed mice, rabbits, raccoons, and opossums. Thus, suburban development into the remaining natural habitats has bridged the pre-existing rickettsial cycle and directed a greater flow of rickettsiae into the human habitat where people, mammalian hosts, and vector ticks are brought into close proximity. Infected ticks are brought into human residences by humans, dogs, and cats. Once ticks complete their feeding and drop off within these habitats, rickettsial infections establish a defined cluster. Lack of migratory tendencies of adult *D. variabilis* (Sonenshine et al., 1966) after the completion of feeding allows very limited dispersal of ticks in small areas surrounding the human habitations. Benach et al. (1981) reported that infected ticks became more prevalent in areas after their first appearence. However, the flow of rickettsiae will remain unidirectional since human and dog infections with RMSF will not contribute further to the dissemination of the rickettsiae. Nonetheless, transovarially infected ticks remaining in the surrounding areas will continue to be responsible for the sporadic occurrence of RMSF. The rickettsial flow, from tick to tick through transstadial or transovarial transmission, from tick to rodent host, and from rodent host to tick maintains the cycle in nature. Increased public awareness of RMSF has reduced the rickettsial flow into urban and suburban habitats. This may be one of the reasons for the steady decrease in RMSF cases during the past decade. Small but limited natural habitats in the form of parks and recreational areas within the metropolitan boundaries also have a role in the maintenance of the rickettsial cycle, and occasionally spill over to visiting human populations.

A small number of human RMSF cases have been acquired within the city limits (Durack, 1988).

The cycling of rickettsiae among ticks, and between ticks and mammals has generated interests in lieu of the maintenance of the rickettsial integrity during invertebrate–vertebrate transfer. This view is gaining momentum as the majority of hemolymph-positive ticks, including the principal vector of RMSF, turn out to be infected with the other SFG rickettsiae rather than *R. rickettsii* (Table 10.3). As it has been pointed out by other investigators (see McDade and Newhouse, 1986; Burgdorfer, 1989), the presence of these as yet little known rickettsiae complicate the natural history of *R. rickettsii*. Infection of mammalian hosts with other tick-borne SFG rickettsiae may result in the cross-protection against virulent *R. rickettsii* and consequently reduces the infection rates with this rickettsiae by favoring non-pathogenic organisms. In contrast to *R. rickettsii*, the other SFG rickettsiae exert no adverse effects on the survival and reproductive potential of their vector ticks (Burgdorfer and Brinton, 1975; Hayes and Burgdorfer, 1990), and thereby help to sustain stable transovarial transmission for many tick generations. As a result, infection of ticks with other SFG rickettsiae slowly replaces *R. rickettsii* infection in the tick population. Delineation of these assumptions are just a few exciting challenges for future studies.

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5. REFERENCES

Anacker, R.L., Mann, R.E., and Gonzales, C. (1987) Reactivity of monoclonal antibodies to *Rickettsia rickettsii* with spotted fever and typhus group rickettsiae. *J. Clin. Microbiol.* **25**, 167–171.

Anderson, B.E., McDonald, G.A., Jones, D.C., and Regnery, R.L. (1990) A protective protein antigen of *Rickettsia rickettsii* has tandomly repeated, near-identical sequences. *Infect. Immun.* **58**, 2760–2769.

Anon. (1991) Rocky Mountain spotted fever—United States, 1990. *Morbidity Mortality Weekly Rep.* **40**, 451–453.

Benach, J.L., White, D.J., Burgdorfer, W., Keelan, T., Guirgis, S., and Altieri, R.H. (1977) Changing patterns in incidence of Rocky Mountain spotted fever on Long Island (1971–1976). *Am. J. Epidemiol.* **106**, 380–387.

Benach, J.L., Smith, L.A., and White, D.J. (1981) An example of geographical clustering of *Dermacentor variabilis* adults infected with rickettsiae of the spotted fever group. pp. 611–619. *In:* Burgdorfer, W. and Anacker, R.L. (eds) *Rickettsiae and Rickettsial Diseases.* Academic Press, New York.

Bozeman, F. M., Shirai, A., Humphries, J.W., and Fuller, H. S. (1967) Ecology of Rocky Mountain spotted fever. II. Natural infection of wild mammals and birds in Virginia and Maryland. *Am. J. Trop. Med. Hyg.* **16**, 48–59.

Burgdorfer, W. (1970) Hemolymph test. A technique for detection of rickettsiae in ticks. *Am. J. Trop. Med. Hyg.* **24**, 866–872.

Burgdorfer, W. (1975) A review of Rocky Mountain spotted fever (tick-borne typhus), its agent, and its tick vectors in the United States. *J. Med. Entomol.* **12**, 269–278.

Burgdorfer, W. (1989) Ecological and epidemiological considerations of Rocky Mountain spotted fever and scrub typhus. Vol. 1, pp. 34–47. *In:* Walker, D.H. (ed) *Biology of Rickettsial Diseases.* CRC Press, Inc., Boca Raton, FL.

Burgdorfer, W. and Brinton, L.P. (1975) Mechanisms of transovarial infection of spotted fever rickettsiae in ticks. *Ann. New York Acad. Sci.* **266**, 61–72.

Burgdorfer, W., Friedhoff, K.T., and Lancaster, J.L., Jr (1966) Natural history of tick-borne spotted fever in the U.S.A. Susceptibility of small mammals to virulent *Rickettsia rickettsii. Bull. WHO* **35**, 149–153.

Burgdorfer, W., Cooney, J.C., Mavros, A.J., Jellison, W., and Maser, C. (1980) The role of cottontail rabbits (*Sylvilagus* spp.) in the ecology of *Rickettsia rickettsii* in the United Sates. *Am. J. Trop. Med. Hyg.* **29**, 686–690.

Burgdorfer, W., Hayes, S. F., Thomas, L. A., and Lancaster, J.L., Jr (1981) A new spotted fever rickettsia from the Lone Star tick, *Amblyomma americanum.* pp. 595–602. *In:* Burgdorfer, W. and Anacker, R.L. (eds) *Rickettsiae and Rickettsial Diseases.* Academic Press, New York.

Clifford, C.M., Sonenshine, D.E., Atwood, E.L., Robbins, C.S., and Hughes, L.E. (1969) Tests on ticks from wild birds collected in the eastern United States for rickettsiae and viruses. *Am. J. Trop. Med. Hyg.* **18**, 1057–1061.

Durack, D.T. (1988) Spotted fever comes to town. *New Engl. J. Med.* **318**, 1388–1390.

Gage, K.L., Burgdorfer, W., and Hopla, C.E. (1990) Hispid Cotton rats (*Sigmodon hispidus*) as a source for infecting immature *Dermacentor variabilis* (*Acari*: *Ixodidae*) with *Rickettsia rickettsii. J. Med. Entomol.* **27**, 615–619.

Gage, K.L., Hopla, C.E., and Schwan, T.G. (1992) Cotton rats and other small mammals as hosts for immature *Dermacentor variabilis* (*Acari*: *Ixodidae*) in Central Oklahoma. *J. Med. Entomol.* **29**, 832–842.

Gordon, J.C., Gordon, S.W., Peterson, E., and Philip, R.N. (1984) Epidemiology of Rocky Mountain spotted fever in Ohio, 1981: serologic evaluation of canine and rickettsial isolation from the ticks associated with human case exposure sites. *Am. J. Trop. Med. Hyg.* **33**, 1026–1029.

Hayes, S.F. and Burgdorfer, W. (1979) Ultrastructure of *Rickettsia rhipicephali*, a new member of the spotted fever group rickettsiae in tissues of host vector *Rhipicephalus sanguineus. J. Bacteriol.* **137**, 605–613.

Hayes, S.F. and Burgdorfer, W. (1982) Reactivation of *Rickettsia rickettsii* in *Dermacentor andersoni* ticks: an ultrastructureal analysis. *Infect. Immun.* **37**, 779–785.

Hayes, S.F. and Burgdorfer, W. (1990) Interactions between rickettsial endocytobionts and their tick hosts. pp. 236–251. *In:* Schwemmler, W. and Gassner, G. (eds) *Insect Endocytobiosis: Morphology, Physiology, Genetics, and Evolution.* CRC Press, Inc., Boca Raton, FL.

Koch, H.G. (1982) Seasonal incidence and attachment sites of ticks (*Acari*: *Ixodidae*) on domestic dogs in southeastern Oklahoma and northeastern Arkansas, USA. *J. Med. Entomol.* **19**, 293–298.

Lane, R.S., Emmons, R.W., Dondero, D.V., and Nelson, B.C. (1981) Ecology of tick-borne agents in California. I. Spotted fever group rickettsiae. *Am. J. Trop. Med. Hyg.* **30**, 239–252.

Loving, S.M., Smith, A.B., DiSalvo, A.F., and Burgdorfer, W. (1978) Distribution and prevalence of spotted fever group rickettsiae in ticks from South Carolina, with

an epidemiological survey of persons bitten by infected ticks. *Am. J. Trop. Med. Hyg.* **27**, 1255–1260.

McDade, J.E. and Newhouse, V.F. (1986) Natural history of *Rickettsia rickettsii*. *Ann. Rev. Microbiol.* **40**, 287–309.

Magnarelli, L.A., Anderson, J. F., Phillip, R. N., Burgdorfer, W., and Chapell, W.A. (1983) *Rickettsia*-infected ticks (*Acari*: *Ixodidae*) and seropositive mammals at a focus for Rocky Mountain spotted fever in Connecticut, USA. *J. Med. Entomol.* **20**, 151–156.

Norment, B.R. and Burgdorfer, W. (1984) Susceptibility and reservoir potential of the dog to spotted fever-group rickettsiae. *Am. J. Vet. Res.* **45**, 1707–1710.

Norment, B.R. and Burgdorfer, W. (1985) Susceptibility of small mammals to nonvirulent spotted fever-group rickettsiae. *J. Med. Entomol.* **22**, 200–203.

Philip, R.N. and Casper, E.A. (1981) Serotypes of spotted fever group rickettsiae isolated from *Dermacentor andersoni* (Stiles) ticks in western Montana. *Am. J. Trop. Med. Hyg.* **30**, 230–238.

Philip, R.N. and Casper, E.A., Ormsbee, R.A., Peacock, M.G., and Burgdorfer, W. (1976) Microimmunofluorescence test for the serological study of Rocky Mountain spotted fever and typhus. *J. Clin. Microbiol.* **3**, 51–61.

Philip, R.N., Casper, E.A., Burgdorfer, W., Gerloff, R.K., Hughes, L.E., and Bell, E.J. (1978) Serologic typing of rickettsiae of the spotted fever group by microimmunofluorescence. *J. Immunol.* **121**, 1961–1968.

Philip, R.N., Casper, E.A., Anacker, R. L., Cory, J., Hayes, S.F., Burgdorfer, W. and Yunker, E.C. (1983) *Rickettsia bellii* sp. nov.: a tick-borne rickettsia, widely distributed in the United States, that is distinct from the spotted fever and typhus biogroups. *Internat. J. Syst. Bacteriol.* **33**, 94–106.

Sexton, D.J., Burgdorfer, W., Thomas, L., and Norment, B.R. (1976) Rocky Mountain spotted fever in Mississippii: survey for spotted fever antibodies in dogs and for spotted fever group rickettsiae in dog ticks. *Am. J. Epidemiol.* **103**, 192–197.

Sonenshine, D.E. and Stout, I.J. (1971) Ticks infesting medium-sized mammals in two forest localities in Virginia (*Acarina*: *Ixodidae*). *J. Med. Entomol.* **8**, 217–227.

Sonenshine, D.E. and Cliford, C.M. (1973) Contrasting incidence of Rocky Mountain spotted fever in ticks infesting wild birds in eastern U.S. piedmont and costal areas, with notes on the ecology of these ticks. *J. Med. Entomol.* **10**, 497–502.

Sonenshine, D.E., Atwood, E.L., and Lamb, J.T., Jr. (1966) The ecology of ticks transmitting Rocky Mountain spotted fever in a study area in Virginia. *Ann. Entomol. Soc. Amer.* **59**, 1234–1262.

11

Lyme Disease/Lyme Borreliosis

JOSEPH PIESMAN AND JEREMY S. GRAY

1. HISTORICAL PERSPECTIVE

Ticks have been implicated in the transmission of Lyme disease since 1909, when the Swedish dermatologist Afzelius (1910) first noticed that the pathognomonic skin lesion, erythema migrans, was associated with the bite of the tick *Ixodes ricinus*. A series of early discoveries in Europe (reviewed by Burgdorfer, 1986) suggested that an infectious agent transmitted by *I. ricinus*, most probably a spirochete, caused erythema migrans as well as neurological disease; symptoms resolved with penicillin treatment. This background proved crucial in implicating *I. dammini* as the vector of a "new" pathogen causing a unique cluster of erythema migrans and arthritis in Lyme, Connecticut, and surrounding communities (Steere et al., 1977, 1978; Steere and Malawista, 1979). Studies of Lyme disease ecology became possible when Burgdorfer et al. (1982) isolated the Lyme disease spirochete, subsequently named *Borrelia burgdorferi*, from *I. dammini* ticks. Since 1982, our knowledge of the natural history of Lyme disease has exploded. After a decade of research, it is satisfying to review how much we do know about transmission of *B. burgdorferi* and the biology of its tick vectors. It is more important, however, to identify the gaps in our knowledge of the spirochete, and its relationships with tick vectors and vertebrate reservoirs. These gaps currently impede our efforts to combat transmission of the Lyme disease spirochete. As we integrate our knowledge of Lyme disease ecology, it is important to remember how early observations in Europe led to the discovery of *B. burgdorferi* in North America. While certain aspects of Lyme disease ecology remain constant among different geographic areas, other aspects are more flexible, changing dramatically from area to area. Collating worldwide knowledge of Lyme disease ecology is an essential step in designing reliable strategies for the prevention of Lyme disease.

2. VECTORS IN NORTH AMERICA

The principal vector of *B. burgdorferi* in North America is *I. dammini*. As with all members of the "*I. ricinus* complex", *I. dammini* ticks require moist

microclimates for survival. These moist microclimates can most easily be found where leaf-litter is well established. In island and coastal communities, dense brush dominated by bayberry and shrub oak provide the ideal habitat for *I. dammini* and its primary hosts, white-footed mice and white-tailed deer. In inland locations (e.g., Westchester Co., New York, or the border between Minnesota and Wisconsin) typical mixed deciduous forests, with high canopy, provide the ideal habitat for the Lyme disease spirochete maintenance cycle. An interesting intermediate habitat where *I. dammini* is found is presented by the inland coastal plains of New Jersey. This habitat is characterized by sandy soils resembling coastal areas, but also contains a high forest canopy with a lower level of blueberry bush cover (Schulze et al., 1991a). Thus, the principal Lyme disease vector, *I. dammini*, demonstrates a fair degree of plasticity in the types of habitat it occupies.

There are, however, some interesting limits to the ecological plasticity demonstrated by *I. dammini*. Areas heavily devoted to agriculture, in the mid-western US, which have large areas cleared of forest and only small sanctuaries for deer, do not seem to have large permanent populations of *I. dammini* (Pinger et al., 1991). In northern Illinois, populations of *I. dammini* have become permanently established solely along major rivers and their tributaries (Kitron et al., 1992). Perhaps a critical density of woodlands is necessary for populations of *I. dammini* to become permanently established. The use of remote satellite sensing combined with computer software systems for analyzing ecological information (e.g., geographical information systems) holds promise for defining the course of future dispersal of *I. dammini*. These systems will only prove useful if they are used in a predictive sense, with "ground truthing" to verify predictions.

Many researchers have developed the impression that deciduous leaf-litter is necessary for the maintenance of *I. dammini* populations, and that coniferous forests dominated by pines and spruce do not support large populations of these ticks. In western Pennsylvania, however, hemlock (*Tsuga canadensis*) stands support permanent populations of *I. dammini* and white-footed mice infected with *B. burgdorferi* (Lord et al., 1992). The distribution of *I. dammini* in Canada presents an additional curious situation. The only site in Canada where a well-established population of *I. dammini* is present is Long Point, Ontario (Lindsay et al., 1991), a location which projects into Lake Erie. Interestingly, Presque Isle, Pennsylvania, is an isolated endemic focus of Lyme disease on the direct opposite side of Lake Erie. Perhaps large bodies of water moderate the environment, allowing the establishment of *I. dammini* populations. At present, despite extensive sampling, only isolated individuals of *I. dammini* have been located in Canada outside of Long Point, Ontario. The ecological factors that limit the dispersal of *I. dammini* to the north present an intriguing topic for future research.

Coastal populations of *I. dammini* can be found from Maine to Maryland. Along the Maryland–Virginia border interesting ecological changes have an important bearing on tick populations. The bayberry and shrub-oak-dominated coastal habitat gives way to a traditional southern mixed deciduous–coniferous

forest. While small rodents predominate toward the north, reptilian hosts dominate toward the south. At this interface, *I. dammini*-like immature ticks can be found feeding on small rodents in the state of Maryland, while *I. scapularis*-like immature ticks can be found feeding as immatures on lizards in Virginia. In areas where immature *I. dammini* ticks are feeding on rodents, infection rates of these ticks with *B. burgdorferi* are fairly high. To the south, infection of *I. scapularis* with *B. burgdorferi* is rare. The original description of *I. dammini* as a species distinct from *I. scapularis* was based on the difference in hosts chosen by immature ticks and the differences in morphology, primarily in the nymphal stage (Spielman et al., 1979). Research is in progress to determine whether these two species (*I. dammini* and *I. scapularis*) can interbreed in the laboratory, as well as their degree of genetic relatedness based on gene sequencing and isoenzyme analysis. The degree of gene flow between the *I. dammini* populations to the north and *I. scapularis* to the south will be critical in determining whether these two populations are in fact distinct species. Investigations conducted along the interface of these two populations should prove crucial. Interestingly, populations of *I. scapularis* can be found along the eastern shore of Virginia, behaving in a manner typical of this species, with few rodents being infested by immature ticks and infection with *B. burgdorferi* rare or absent. On the extreme northern edge of the eastern shore of Virginia, however, a population of typical *I. dammini* can be found in the Chinoteague Wildlife Refuge on Assateague Island. This area has a bayberry bush dominated habitat with an abundant small rodent population consisting of *Peromyscus leucopus* and sylvatic house mice (*Mus musculus*). In this isolated Virginia site, *I. dammini* feed abundantly on rodents, and the occurrence of spirochetal infection in these ticks and rodents is not uncommon (Sonenshine, unpublished). North–South transects conducted in areas where the change from typical *I. dammini* habitat to *I. scapularis* habitat should prove revealing.

The ecological conditions that permit *I. pacificus* to survive in western North America are even more variable than those supporting the survival of *I. dammini*. Although the traditional area for *I. pacificus* has long been thought to be solely the Pacific coastal strip, from California to British Columbia, we now know that inland populations of this tick species are fairly common. *I. pacificus* has been found in 54 of the 58 California counties (Clover, unpublished). Moreover, isolated populations of *I. pacificus* have been located in special circumstances in inland locations in the state of Arizona (Olson et al., 1992). These Arizona populations of *I. pacificus* are found in "sky-islands" at elevations $>7{,}000$ feet above sea level, among stands of shrub oak vegetation. These habitats can be found close to xeric habitats; *I. pacificus* adults were flagged from a coffeeberry bush (*Garrya wrightii*) right next to a prickly pear cactus (Ribeiro, unpublished). The tick populations may recede to moist, shady habitats when sufficient drying takes place following the spring snowmelt. Apparently, these ticks are able to find moist microclimates in this xeric environment, perhaps due to leaf-litter provided by shrub-oak vegetation. Questions remain as to how these isolated populations of *I. pacificus* become established and how they are genetically related to the main *I. pacificus* population along the Pacific coast of North America.

The presence of *I. dammini* in highly populated suburban settings in the northeast US is a key factor in explaining the large number of Lyme disease cases reported from the northeastern US in the last decade (Miller et al., 1990). Studies on the distribution of infected ticks on residential properties have added to our understanding of Lyme disease transmission risk. Landscape ecology studies demonstrated that, although *I. dammini* is primarily a woodland species, infected nymphal ticks can be found in ecotonal vegetation, ornamental plantings, and, occasionally, even on maintained lawns (Maupin et al., 1991). Although infected ticks are relatively rare on maintained lawns compared to the woodland population, lawn-inhabiting ticks may be the most likely to come into contact with residents and transmit the Lyme disease spirochete. These considerations will prove important when devising prevention and control strategies (see below).

The manner in which *I. dammini* is dispersing to new locations and thus spreading the Lyme disease epidemic is intriguing. It appears that *I. dammini* is now inhabiting locations where it has not been observed previously. The main problem with these types of observations is that few people have made rigorous efforts to detect *I. dammini* where it has been rare or absent, prior to the establishment of active tick populations. Evidence for the absence of *Ixodes dammini* is usually in the form of casual reports of ectoparasite searches with no reports of *I. dammini*, prior to studies conducted by trained acarologists once *I. dammini* is permanently established. Opportunities to make observations while *I. dammini* is actively dispersing exist, however. For instance, Lyme disease and *I. dammini* appear to be spreading northward from Westchester Co., New York, along the Hudson River Valley (White et al., 1991). The dynamics of this dispersal and subsequent establishments of the Lyme disease cycle will be important to observe, so we can learn to forestall such spread in the future. The manner in which *I. dammini* and Lyme disease disperses appears to be more like an amoeba than a wildfire. Areas close to, or contiguous with, highly disease endemic areas initially become infested with a few adult ticks on deer. Eventually, the *I. dammini* populations build up to the point where nymphal ticks infected with *B. burgdorferi* commonly infest people. Consequently, from the human standpoint, it appears as if an explosive epidemic has occurred in a new location; in fact, the endemic cycle has been establishing itself over an extended period. The dynamics of this dispersal and establishment process are richly deserving of study.

3. RESERVOIRS IN NORTH AMERICA

Vertebrate animals serve two important functions in the maintenance cycle of the Lyme disease spirochete: (1) as reservoirs, maintaining the spirochete in nature; and (2) as hosts, maintaining tick populations. In areas where *I. dammini* is the principal Lyme disease spirochete vector, the white-footed mouse, *Peromyscus leucopus*, clearly plays an important role as a reservoir of *B. burgdorferi* and as a host for immature stages of the tick. A plethora of studies

in coastal New England have added to our knowledge of the contribution of the white-footed mouse in the Lyme disease enzootic cycle (Anderson et al., 1985; Levine et al., 1985; Donahue et al., 1987; Mather et al., 1989a). In coastal New England, the white-footed mouse is clearly the most abundant mammal, and the host most frequently parasitized by both nymphal and larval *I. dammini*; the majority of these mice are infected with *B. burgdorferi* and infectious to ticks. Several control methods have utilized this predominance of *P. leucopus* to develop host-targeted control technologies (see below).

In certain ecologic situations, however, *P. leucopus* may not be the predominant reservoir host. A striking example of an alternative to the white-footed mouse as the predominant reservoir can be found on Monhegan Island, Maine. This island has an abundant white-tailed deer population, and *I. dammini* adults can be collected with ease in various locations on the island. The island lacks *Peromyscus* spp., however; thousands of trap-nights have failed to produce any evidence of white-footed mice on this island (R. Smith, unpublished). But, $>\frac{1}{4}$ of field-collected nymphs and about 40% of adult *I. dammini* collected from vegetation on Monhegan Island are infected with *B. burgdorferi*. The reservoir that infects ticks in this habitat is apparently the Norway rat, *Rattus norvegicus*. These rats are peridomestic on the island during warmer months and can be found up to several hundred meters from the nearest human dwelling. The rats are heavily infested with immature *I. dammini* and infect $>\frac{1}{2}$ of the larvae derived from them. In contrast, larvae derived from cats, birds, muskrats, and deer on the island have not been found to be infected with *B. burgdorferi*. Thus, in at least one instance, definitive proof exists that the Lyme disease enzootic cycle is being maintained in the absence of white-footed mice.

The western US provides additional examples of alternative reservoir hosts. Extensive attempts to culture *B. burgdorferi* from *Peromyscus* spp. in California have met with limited success. In contrast, *B. burgdorferi* has been frequently isolated from *Neotoma fuscipes*, the dusky-footed woodrat, and *Dipodomys californicus*, the California kangaroo rat (Lane and Brown, 1991). Perhaps these hosts become infected by nidicolous vectors, e.g., *I. neotomae*. The endemic vector of the Lyme disease spirochete in the western US, *I. pacificus*, occasionally feed as larvae on infected woodrats or kangaroo rats, thus becoming infected with *B. burgdorferi*, molting into infected nymphs, and transmitting spirochetes to humans. The fact that larval *I. pacificus* only occasionally feeds on these infected rodents, as opposed to refractory reptiles (Lane and Loye, 1989), helps explain the low infection rates. Another factor that reduces the overall infection rates of *I. pacificus* compared with those of *I. dammini* is that *I. pacificus* is far less efficient in acquiring and maintaining *B. burgdorferi* as is *I. dammini*. Using strains of *B. burgdorferi* isolated from *I. pacificus* in California, larval *I. dammini* and *I. pacificus* were placed simultaneously on infected mice. Infection rates in *I. dammini* were around 70%, but infection rates in *I. pacificus* ranged from 10 to 20% (Piesman, unpublished). Thus, even with strains of *B. burgdorferi* isolated from *I. pacificus*, *I. dammini* proved to be a much more efficient vector. The moderate vector competency and infrequent exposure to infected hosts both contribute to low *B. burgdorferi*

infection rates in *I. pacificus*. A similar parallel cycle takes place in the eastern US. Cottontail rabbits can act as reservoirs of *B. burgdorferi* (Anderson et al., 1989; Telford and Spielman, 1989). Although infected nymphal *I. dammini* feed with regularity on cottontail rabbits, and probably infect these hosts, the rabbit tick, *I. dentatus*, is much more prevalent on these hosts. The vector competency of *I. dentatus* is markedly less than that of *I. dammini*, but in at least one instance, *I. dentatus* successfully transmitted infection to rabbits in the laboratory. In areas where *I. dammini* successfully maintains the enzootic cycle of *B. burgdorferi* in *Peromyscus leucopus*, the existence of the parallel rabbit cycle is probably not very important. In areas where the *I. dammini–Peromyscus* cycle is not efficient, however, the parallel rabbit–*I. dentatus* cycle may play an important role in maintaining the pathogen in nature.

In addition to feeding on rabbits, *I. dentatus* feeds on birds as does *I. dammini*. The issue of whether birds can act as reservoirs and dispersal hosts for *B. burgdorferi* is intriguing. One isolate of *B. burgdorferi* is available from birds; this isolate was made from the liver of a veery (*Catharus fuscescens*) (Anderson et al., 1986a). Moreover, ticks derived as larvae from American robins in Connecticut were frequently infected with *B. burgdorferi* (Anderson et al., 1990). On the other hand, grey catbirds (*Dumetella carolinensis*) collected in Massachusetts did not produce infected ticks (Mather et al., 1989b), and in laboratory experiments, readily available commercial birds (chickens) did not produce evidence of infection (Schwan, unpublished). The role of birds as reservoirs and dispersal hosts of *B. burgdorferi* is still open to further investigations.

Inland locations in the eastern US support incredibly diverse populations of small and medium-sized mammals. Medium-sized hosts such as raccoons, opossums, and skunks are heavily parasitized by immature *I. dammini* (Fish and Daniels, 1990) and may contribute to the overall density of infected nymphal *I. dammini* in suburban communities. An animal of particular interest in suburban habitats is the eastern chipmunk, *Tamias striatus*. This host appears to spend much more time in habitats affected by human activities than does the more timid woodland-loving *Peromyscus leucopus* (Frank, unpublished). Chipmunks bring small numbers of infected ticks onto maintained lawns, an area of high human use in suburban communities (Maupin et al., 1991). Chipmunks may also be important hosts in the mid-western US. In the Upper Peninsula of Michigan, chipmunks may be as important as *Peromyscus* in the maintenance of *B. burgdorferi* (Walker, unpublished). Finally, white-tailed deer serve as hosts for large numbers of all three stages of *I. dammini*. If they were to serve as reservoirs for *B. burgdorferi*, the force of transmission (i.e., zoonotic potency) would be greatly increased. These hosts appear to be incompetent reservoirs, since the infection rate in larvae derived from white-tailed deer in Massachusetts was $<1\%$. Researchers have been able to experimentally infect white-tailed deer with *B. burgdorferi* (Stalknecht, unpublished), but the infectivity of these deer is unknown. Perhaps a window of infectivity exists when these unnatural hosts for *B. burgdorferi* are infectious to ticks, albeit for only brief periods.

The comparative roles of individual host species in various locations must be analyzed for an overall picture of the enzootic cycle of *B. burgdorferi* to emerge. One must keep in mind, however, that new records of an infected animal species here or there are probably less important than understanding the major contributions of known reservoirs (e.g., *Peromyscus leucopus*) in areas where *I. dammini* infection rates are high, and human risk is great.

4. VECTORS IN EUROPE

Ixodes ricinus was first associated with the skin condition erythema (chronicum) migrans (EM) in Sweden by Afzelius in 1909 (Afzelius, 1910) and the causative organism, *B. burgdorferi*, was first detected in *I. ricinus* in Switzerland (Burgdorfer et al., 1983). This was swiftly followed by detection of the organism in *I. ricinus* throughout Europe, usually by the use of immunofluorescence. So far, every population of *I. ricinus* ticks examined for the infection seems to have been infected.

I. ricinus has been shown to be an efficient vector of the organism in the laboratory (Burgdorferet al., 1983), and is undoubtedly the most important European vector in nature due to its widespread distribution, its catholic feeding habits and its readiness to bite humans. This tick has received an enormous amount of attention over the years because of its importance as a vector of livestock diseases and tick-borne encephalitis (TBE), a zoonotic viral encephalitis prevalent in central and eastern Europe.

The abiotic factors that determine survival and development of the non-parasitic stages of *I. ricinus* have been clearly defined, the most important being vegetation or litter that is sufficiently thick to maintain a relative humidity of 80–85% at soil level throughout the year (Kahl and Knülle, 1988). This requirement means that through most of the tick's range the main habitat is deciduous woodland. Coniferous forests are less often identified as tick habitat and, when they are, are usually found to contain small numbers of ticks (Smaha, 1979). This is probably due to a relative lack of vegetative litter but also perhaps to poor utilization by important tick hosts, such as deer and woodmice. Tick survival is also possible in open areas of rough vegetation where the rainfall is high, for example, permanent pastures and hill land in parts of the British Isles. This sort of habitat is most often grazed by sheep, and studies in these areas gave rise to the common name of "sheep tick" for *I. ricinus*, although over most of its range it is more accurately described as a deer tick.

The development and seasonal activity of *I. ricinus* vary considerably in different geographical areas and habitats, but usually conform to a basic pattern, with activity mainly occurring in spring and early summer, and another period of activity due to separate cohorts of ticks in late summer and early fall. This subject has been reviewed recently, with special reference to Lyme borreliosis (Gray, 1991).

In eastern Europe, *I. ricinus* is replaced by the closely related species, *I. persulcatus*, which is found in temperature regions across Asia; this species

has also been reported to be an important vector of *B. burgdorferi* (Korenberg et al., 1987). The seasonal activity of this tick has been well studied in relation to TBE, which it also transmits. In general it seems to behave similarly to *I. ricinus*, except that fall activity rarely occurs.

B. burgdorferi has now been detected in several other ixodid species in Europe, especially other members of the genus *Ixodes*, including *I. hexagonus* (Liebisch et al., 1990), *I. trianguliceps*, and *I. acuminatus* (Doby et al., 1990a), *I. canisuga* and *I. frontalis* (Estrada-Pena, unpublished); only certain of these species belong to subgenus *Ixodes*, which includes all of the established vectors of *B. burgdorferi*. Of these, only *I. hexagonus* has been shown to be vector competent, though this study involved the use of artificial feeding of ticks with cultured spirochetes (Gern et al., 1991). Other ixodids found to be carriers, if not vectors, of the infection include *Haemaphysalis punctata* (Marquez and Constans, 1990) and *Dermacentor reticulatus* (Kahl et al., 1992). At least one argasid, *Argas reflexus*, has also been implicated as a vector (Stanek and Simeoni, 1989) and the organism has even been found in some hematophagous insects (Doby et al., 1990a). The presence of the infection in these arthropods does not mean that they are capable of transmitting it, and most species involved rarely, if ever, parasitize humans. It is unlikely, therefore, that their contribution to transmission will detract from the importance of *I. ricinus* as the main vector of Lyme borreliosis in Europe, though some may have a role in maintaining the infection in the vertebrate reservoir.

Considerable antigenic variation seems to occur between isolates, far more than in North America (Barbour et al., 1985), and furthermore it is now thought that *B. burgdorferi* may comprise several different genomic species sharing common epitopes (Wilske et al., 1991). It is quite likely, therefore, that not all organisms detected in ticks and other arthropods all have the same significance for disease, and many may be non-pathogenic. Infection and pathology studies in laboratory animal models may eventually be required to assess the importance of the various European isolates of *B. burgdorferi*.

5. RESERVOIRS IN EUROPE

The identification of the vertebrate species that act as reservoirs for *B. burgdorferi*, and on which ticks become infected, is crucial to understanding the dynamics of the circulation of the organism in the environment, to the accurate assessment of risk in different habitats, and to the design of effective control measures. In Europe many vertebrate species, both wild and domestic, have been found seropositive for the infection. However, this does not mean that these animals necessarily pass the infection on to feeding ticks, but merely that infected ticks have fed on them and that they have responded immunologically to the presence of the organism. Some evidence suggests that *Borrelia* spirochetes can survive in large European vertebrates: organisms resembling *B. burgdorferi* have been observed in blood smears from cattle in the Netherlands, red deer in Austria (Uilenberg et al., 1988), roe deer in Switzerland

(Aeschlimann et al., 1986), and the European fallow deer in America (Lane and Burgdorfer, 1986). Although joint disease associated with seropositivity has been described in sheep (Hovmark et al., 1986), attempts to establish infections artificially in this species have not been successful (Stuen and Fridriksdottir, 1991). So far no attempts have been made to show that ticks can become infected by feeding on any of these animals. This approach has, however, been carried out with the two woodmouse species, *Apodemus sylvaticus* and *A. flavicollis*, and the bank vole, *Clethrionomys glareolus* (Aeschlimann et al., 1986; Anderson et al., 1986b; Humair et al. (1993), and all three species have been shown to be competent reservoirs of *B. burgdorferi*. In some cases the infection persisted and the animals were able to infect ticks for many months. American studies have shown that birds can act as reservoirs (Anderson et al., 1990). Very little attention has been paid to the role of birds as reservoirs of *B. burgdorferi* in Europe. However, Matuschka and Spielman (1992) provided evidence that the European blackbird, *Turdus merula*, which is common and often infested by *I. ricinus*, does not seem to be a common reservoir of *B. burgdorferi* and may even cleanse feeding ticks of their infections.

Rodents are the only species that have been conclusively shown to be competent reservoirs in Europe, which may explain why the typical Lyme borreliosis habitat seems to be woodland. Although woodland is the main habitat of the most important European vector, *I. ricinus*, in some areas this tick is also found in considerable numbers in open habitats, where it feeds predominantly on livestock and where the contribution of rodents to the maintenance of tick populations is likely to be much less than in woodland. Such habitats occur commonly in the eastern regions of the British Isles, but so far they have not been markedly associated with the occurrence of Lyme borreliosis. This may be due in part to the scarcity of rodents in such habitats. Preliminary findings in Ireland suggest that where ticks appear to feed almost exclusively on cattle, infection rates are very low and in some cases, infection may even be absent (Gray et al., unpublished). The probable importance of rodents as reservoirs was also suggested by a recent study in Ireland, which showed that a much lower infection rate of ticks occurred in an area where most ticks apparently fed on fallow deer (*Dama dama*), than in an adjoining area where deer were largely excluded, but which harbored similar numbers of woodmice (*Apodemus sylvaticus*) (Gray et al., 1992). This also suggests that deer may not be significant reservoirs of the infection, and it is interesting to note in this context that a study in America concluded that the white-tailed deer, *Odocoileus virginianus*, is probably not reservoir competent for *B. burgdorferi* (Telford et al., 1988).

6. ROLE OF HOSTS IN *IXODES RICINUS* POPULATION DYNAMICS IN EUROPE

The abundance of ticks in habitats that harbor reservoir hosts of *B. burgdorferi* is obviously of great importance in determining the intensity

of transmission. Identifying factors that determine tick abundance therefore is crucial to an understanding of the epidemiology of the disease and to its control.

In suitable habitats (see Chapter 5), the abundance of tick hosts will determine the abundance of the ticks themselves. For example, in Scotland and northern England, the presence and abundance of sheep are thought to determine the size of the *I. ricinus* populations (MacLeod, 1934; Milne, 1949). However, this open grazing habitat does not support the variety or numbers of wild tick-hosts compared with woodland, which is probably the most important Lyme borreliosis habitat (Jaenson, 1991). Woodlands usually harbor numerous small mammals, which, in addition to being probable reservoirs of *B. burgdorferi*, also feed large number of immature ticks. Detailed studies have been carried out on the densities of both *I. ricinus* (Loew et al., 1963) and *I. persulcatus* (Kolonin et al., 1978) in various woodland and forest habitats in relation to TBE. However, few attempts have been made to analyze the relative roles of small and large hosts in determining tick densities in such habitats. In a recent study, though, tick numbers were compared on either side of a deer fence in an Irish woodland. Ticks were found to be very much more abundant where deer were available as hosts (Gray et al., 1992). This suggests that deer may be of prime importance in determining *I. ricinus* abundance in woodland, which seems to be in agreement with American studies on the related tick species *I. dammini* (Wilson et al., 1990). If this is the case, it is obviously essential to establish whether deer and other large mammal tick-hosts are reservoir hosts of *B. burgdorferi*. As noted previously, deer are not believed to be competent reservoir hosts for *B. burgdorferi*.

7. DEVELOPMENT AND TRANSMISSION OF *BORRELIA BURGDORFERI* IN *I. RICINUS*

I. ricinus is the only European tick species in which the development and transmission of *B. burgdorferi* have been studied. It has been established that the organisms are most readily demonstrated in the mid-gut of unfed ticks, but that during engorgement some spirochetes translocate to the salivary glands and transmission probably takes place through the salivary secretions (Gern et al., 1990). Other possible modes of transmission are via tick feces and by regurgitation. However, the spirochete has never been found in tick rectal sacs and, while regurgitation has not been ruled out, little evidence exists for it as a means of transmission (Burgdorfer and Hayes, 1989; Burgdorfer et al., 1989). In a minority of unfed ticks, the spirochetes are not confined to the mid-gut and, in females with these disseminated infections, transovarial transmission is likely to occur (Burgdorfer et al., 1983). Since a very small proportion of larvae caught on vegetation by flagging seem to be infected (Mehl et al., 1989; Doby et al., 1990b; Miserez et al., 1990), it would appear that the epidemiological significance of transovarial transmission is low. However, these larval infection rates are not particularly low compared with infections such as *Babesia divergens*, the cause of bovine babesiosis, which are known to be maintained by

transovarial transmission (Donnelly and Peirce, 1975). Thus, it is possible that a small part of the tick population in a habitat could serve as a reservoir for *B. burgdorferi.* It is interesting to note that when larvae are infected artificially by the use of capillary tubes, they can transmit the infection very efficiently to white mice (Kurtenbach et al., 1992).

It is generally recognized that, similar to *I. dammini* in North America, the nymphal instar of *I. ricinus* is probably most responsible for transmission to humans because infection rates are higher than in larvae, and nymphs are both more numerous than adults and are more likely to bite humans. It should be noted, however, that in the case of *I. persulcatus* it is apparently the adults that are of significance in transmitting the infection to humans, with the immatures only being important in maintaining *B. burgdorferi* in small mammals, since they rarely bite humans (Ai et al., 1991). Most *I. ricinus* nymphal activity occurs in spring and early summer (Gray, 1991), so that borreliosis cases usually occur in mid-summer. Since the main peak of nymphal activity occurs slightly earlier than that of larvae in most habitats, an efficient circulation of spirochetes between the vertebrate reservoir hosts and ticks can take place. However, as pointed out by Matuschka et al. (1990), few *I. ricinus* nymphs seem to feed on small mammals, the suggested reservoir hosts, which contrasts with the situation in North America involving *I. dammini* and the white-footed mouse, *P. leucopus.* This suggests that the force of infection (zoonotic potency) in Europe may be lower than in America and that, in areas with high *I. ricinus,* important reservoir hosts other than small mammals may present. It is interesting to note that infection rates of adult ticks are usually higher than those of nymphs, which suggests that a proportion of ticks become infected while feeding as nymphs, despite the small numbers that parasitize small mammals, the only known reservoir hosts in Europe. Although indirect evidence suggests that large mammals may not be very important as sources of infection for nymphs, medium-sized mammals and ground-feeding birds may well be involved, as they seem to be in North America (Anderson et al., 1990; Fish and Daniels, 1990).

8. OTHER HARD TICKS AND THEIR SPIROCHETES

Ixodid ticks may contain spirochetes other than *B. burgdorferi.* The only species of spirochete other than *B. burgdorferi* that has been described in ixodid ticks is *Borrelia theileri* (Theiler, 1904; Smith et al., 1985). This spirochete of cattle has been found in a variety of ticks, including *Boophilus* and *Rhipicephalus.* The possibility that *B. theileri* infects deer and subsequently infects the same species of tick being examined for *B. burgdorferi* must be evaluated. Additionally, spirochetes derived from *Amblyomma americanum* are thought by some investigators to be *B. burgdorferi* based on their reaction with antibodies (Schulze et al., 1984) but spirochetes derived from *A. americanum* in Missouri appear to differ from *B. burgdorferi* in their antigenicity, growth characteristics in culture, and the tick species they infect in the laboratory (G. Maupin, unpublished).

Thus, any standard method for detecting *B. burgdorferi* in ticks will have to incorporate the ability to distinguish *B. burgdorferi* from other spirochetes that are capable of infecting ixodid ticks. Tick–spirochete interactions and the variety of ticks in North America found to contain "spirochetes," are discussed further in Chapter 3.

9. PREVENTION AND CONTROL IN NORTH AMERICA

The Lyme disease spirochete enzootic cycle is now well established in large geographic areas in several parts of the US. Because elimination of tick populations over a wide geographic area is not a feasible goal, at present, public health interventions must focus on preventing contact between people and infected ticks. Several methods have been proposed to effect the prevention and control of Lyme disease : (1) area-wide acaricide treatment; (2) host-targeted acaricides; (3) host exclusion/eradication; (4) vegetation management; (5) personal protection; and (6) integrated pest management (IPM). The following is a brief discussion of these methods as they affect Lyme disease. The fundamental principles underlying these methods and their application to ticks generally are discussed in Chapter 9.

9.1. Area-wide Acaricides

Area-wide application of acaricides has been used effectively to kill ticks. The pioneering work in this field involved aerial application of diazinon granules to control the lone star tick, *Amblyomma americanum* (Mount, 1984; Sardelis et al., 1989). Initial attempts to control *I. dammini* focused on adult ticks; carbaryl and diazinon were sprayed into forested habitats via ground-based hydraulic sprayers (Schulze et al., 1987; Schulze et al., 1988). Subsequently, the application of granular preparations of diazinon, carbaryl, and chlorpyriphos were shown to decrease populations of immature *I. dammini* (Schulze et al., 1991b; Stafford, 1991a). Moreover, timely applications of a new synthetic pyrethroid, cyfluthrin, proved quite effective in reducing the questing populations of nymphal *I. dammini* (Solberg et al., 1992; Fish, unpublished). Cyfluthrin is particularly interesting since it can be used at much lower concentrations than the other insecticides tested. Based on bioassay testing in the laboratory, area-wide application of permethrin should be as effective as cyfluthrin in killing immature *I. dammini* (Maupin, unpublished). The queston of where and when acaracides should be sprayed to produce the maximum public health benefit is subject to debate. Maupin et al. (1991) studied the landscape ecology of *I. dammini* on residential properties in Westchester Co., New York. Ticks were most prevalent in wooded habitat, followed by ecotone, onnamental plantings, and maintained lawn. Only a fraction of the total tick population can be found on maintained lawns. However, this is the component of the tick population that is most likely to come into contact with people. Is the application of potentially toxic

acaricides to maintained lawn worthwhile? To a certain degree, this decision will have to be made by individual homeowners but the public health community is obliged to help the homeowner make this difficult decision. Ideally, a thorough evaluation of the landscape ecology and tick abundance should be made on each property before spraying. This is just not feasible, given limitations on staff in local and regional public health organizations. Perhaps commercial pest companies can routinely include an evaluation of entomological risk, before the decision to spray or not to spray is made.

Although area-wide application is cheap, fast, and effective, the well-known problems with environmental damage, toxicity, and potential liability are as great with acaricides as with any pesticide. The knowledge that large-scale pesticide use may become a thing of the past in the next century should stimulate all public health personnel associated with Lyme disease control to look aggressively for alternatives.

9.2. Alternative Acaricides

Relatively non-toxic alternative pesticides have been developed over the last several years. Some of these compounds have achieved common usage in "organic" gardening and other venues. Desiccants, pyrethrin-containing soaps, and fungi have all been tested for their toxicity to *I. dammini*; some of these compounds are almost as effective as standard acaricides in killing *I. dammini* in the laboratory (Allan, unpublished). The problem with the application of desiccants, for instance, is how to apply these compounds in an area that receives high rainfall during the season of nymphal *I. dammini* activity. Nymphal *I. dammini* are mainly active during the season of highest humidity. The tactics of how to best utilize these alternative acaricides will be most interesting. Other alternative approaches include the potential use of parasitic wasps (Mather et al., 1987a), pheromones, and hormones, to attack tick populations. All of these alternative technologies are in the initial stages of investigation.

9.3. Host-targeted Acaricides

Two principal targets exist for the application of acaricides to hosts, in order to break the enzootic cycle of Lyme disease: (1) rodents acting as hosts for immature ticks; and (2) large animals, e.g., white-tailed deer, acting as hosts for adult ticks. Host-targeted acaricides have been used, with good effect, to interrupt transmission of vector-borne disease. For instance, flea populations that transmit plague bacilli to rodents have been controlled with host-targeted acaricides (Barnes et al., 1972; Beard et al., 1992). Moreover, immature *Dermacentor variabilis* were controlled on rodents by luring them to baited treatment stations, containing peanut butter and rolled oats as bait, and a 5% carbaryl-impregnated talc (Sonenshine and Haines, 1985). A clever adaptation of this approach took advantage of the predilection of the white-footed mouse

for cotton as nesting material; Mather et al. (1987b) designed tubes containing cotton balls impregnated with permethrin. In an initial study, this method was quite effective in reducing the population of immature *I. dammini* infesting *Peromyscus* on Naushon Island, Massachusetts. Subsequent studies in Ipswich, Massachusetts, demonstrated a marked reduction of up to 97% in the population of questing *I. dammini* in the year following treatment and a corresponding decrease in the infection rate of nymphal *I. dammini* collection from treated plots (Mather et al., 1988; Deblinger and Rimmer, 1991). Curiously, similar trials with permethrin-treated cotton balls did not result in a decrease in questing nymphal *I. dammini* populations in New York (Daniels et al., 1991) or Connecticut (Stafford, 1991b). The variation in results among different groups and study locations could have been produced by minor differences in study design; on the other hand, these differences could have resulted from true biological differences derived from variation in the importance of hosts other than *Peromyscus leucopus* in different geographic locations. The proper role of permethrin-treated cotton balls in areas where *I. dammini* are the principal vectors of Lyme disease is unclear at present. The possibility that a similar approach can be used in the western US, where *Neotoma* spp., commonly known as packrats, are the primary reservoir is intriguing.

Systemic acaricides hold some prospects for the control of *I. dammini.* Primary attention has focused on invermectin as a possible treatment of rodents aimed at breaking the Lyme disease enzotic cycle. In the laboratory, ivermectin-treated food produced high enough blood levels in mice to prevent full acquisition of a blood-meal and subsequent molting by immature *I. dammini* (Korch, unpublished). The feasibility of getting a sufficient dose of ivermectin in rodent populations in the field has not yet been thoroughly evaluated. The use of orally administered ivermectin to control adult ticks on large animals has been tested. Ivermectin treatment of captive goats and white-tailed deer resulted in dramatically reduced feeding by lone star ticks (Miller et al., 1989). Control by oral administration of ivermectin to free-ranging large animals has not yet been evaluated. The task of safely administering a sufficient dose of ivermectin to natural populations of white-tailed deer in suburban Lyme disease-endemic settings is daunting. Similar logistical problems should be anticipated when trying to deliver topical acaricides, long used as dips or "rub-ons" with cattle, to natural populations of white-tailed deer.

9.4. Host Exclusion/Eradication

Much attention has been focused on the possibility of reducing or eliminating white-tailed deer populations towards the end of controlling *I. dammini.* Without a doubt, the most comprehensive attempt to impact *I. dammini* through deer management took place on Great Island, Massachusetts. Over a 2-year span, deer were virtually eliminated from this island, and the population of deer was kept down to 1–2 animals for several years. A corresponding decrease in the numbers of immature *I. dammini* found on mice on this island was observed,

while populations of *I. dammini* remained high on non-intervention islands (Wilson et al., 1988). Although *I. dammini* can still be found on this island years after deer have been eliminated, epidemiologic follow-up of the human population suggests that *B. burgdorferi* transmission to people has ceased on Great Island (Telford, unpublished). Perhaps deer eradication can lead to the reduction of *I. dammini* populations to a point below a threshold where transmission to people no longer occurs. However, the lesson of Great Island also teaches us that the tick populations are quite resilient and that they can remain in place despite heroic host-management efforts. The alternative to deer eradication is fencing. This has been used quite effectively against lone star ticks (Bloemer et al., 1990). Preliminary data indicate that large enclosed areas in Westchester Co., New York, that have been fenced in for many decades, have much smaller populations of *I. dammini* inside the enclosure as compared to outside (Daniels, unpublished). Fences, however, are expensive to erect and maintain. The minimum area that can be fenced to exclude deer and still receive protection has not been established.

Rodent eradication has been used effectively to interrupt plague epizootics. The possibility of using poisons to control ubiquitous rodents such as *Peromyscus leucopus* in suburban settings, where danger of accidental poisoning of pets and children exists, makes the approach of rodent eradication virtually impossible for Lyme disease control.

9.5. Vegetation Management

Habitat modifications have long been practiced to control ticks. Folklore in areas of New England indicates that native American peoples practiced burning to control tick populations. The first scientific attempt to use habitat modification for the control of *I. dammini* took place on Great Island, Massachusetts (Wilson, 1986); vegetation was destroyed by burning or mowing, and the population of adult *I. dammini* was measured. Both burning and mowing reduced populations of *I. dammini* for up to 1 year, but the populations eventually returned to their original levels. On Shelter Island, New York, an attempt is currently taking place to measure the impact of controlled burns on all three stages of *I. dammini* (Duffy, unpublished). To date, the overall population of nymphs decreased immediately following a spring burn, but the infection rate of ticks in the burnt plots was higher than in the unburnt plots (Mather et al., 1993). Moreover, larval populations in the fall were higher in burnt plots, presumably because the fresh forage attracted deer to the area. The timing and temperature achieved by burning may be key factors in the utility of this method for tick control. In a study on the landscape ecology of Lyme disease in Westchester Co., several measures for habitat modification were made that could result in decreased transmission risk (Maupin et al., 1991), namely, removal of leaf-litter, reducing shade, construction of borders made of xeric substances, and alterations in the planting of certain ornamental plants. The simplest intervention of leaf-litter removal to decrease ground humidity levels

should be tested immediately. Interventions aimed at habitat modification will have to be conducted in close co-operation with homeowners, since substantial financial investments are often made in landscaping in suburban communities. More radical interventions may be possible in recreational areas commonly used by the public.

9.6. Personal Protection

The first line of defense against Lyme disease spirochete transmission is still personal protection. Proper clothing is essential. Long sleeves and long pants with trouser cuffs tucked into boots should be worn when entering tick-infested areas during the peak season (May–June) of nymphal *I. dammini* activity (Piesman et al., 1987a). Tick researchers even tape the trouser–boot interface to reduce tick access. Following this advice, however, is difficult during summer months when temperatures and humidity levels are most uncomfortable. Certainly, application of repellents is a useful adjunct to personal protection measures. The US Army has long taken the lead in testing repellents against a variety of arthropods. In a test comparing permethrin-treated uniforms to those treated with *N,N*-diethyl toluamide (DEET), permethrin treatment was much more effective than DEET against three species of ticks, including *I. dammini* (Evans et al., 1990). One advantage of DEET is that it can be applied directly to skin but seizures following DEET usage in children have been reported. In addition, the protection offered by DEET, when applied to the skin is fairly short lived. Clearly, the feasibility of repellent usage depends on the population at risk. Soldiers wear uniforms and thus are the best candidates for common repellent use. Children in suburban backyards are probably the most difficult group to protect via repellents.

Common sense suggests that, if ticks do manage to attach, they should be removed as soon as possible. There is a real biological urgency in promoting the prompt removal of attached ticks. Both nymphal (Piesman et al., 1987b) and adult (Piesman et al., 1991) *I. dammini* must be attached for 2 days in order to transmit *B. burgdorferi* efficiently. This may be due to both multiplication of the spirochete during the act of tick feeding (Piesman, unpublished) as well as dispersal of the spirochete to the salivary glands of the tick (Ribeiro et al., 1987; Zung et al., 1989). Ticks can be removed with fine forceps, by grasping them as close to the skin as possible, and applying gentle and constant pressure. The main problem with encouraging prompt tick removal as a public health measure is that nymphal *I. dammini* are so small (<2 mm) that most residents of endemic regions do not recognize them as ticks; they associate the word tick with the much larger adult American dog tick. Lyme disease is a great educator, however, and victims of the disease are usually much better informed about the biology of the Lyme disease cycle compared to the general population. The need for public health education on a local level, directed particularly at high-risk neighborhoods in hyperendemic counties is a necessity for Lyme disease prevention.

10. PREVENTION AND CONTROL IN EUROPE

Jaenson et al. (1990) recently considered the control of Lyme disease vectors in Europe in some detail, and considerable attention has been paid to the control of *I. ricinus* in Europe, since these ticks are important livestock parasites and vectors of tick-borne encephalitis virus. Control measures have consisted mainly of the destruction of the tick microhabitat, principally through improvement of agricultural land, and of the application of persistent acaricides to animals, the latest products being efficient pour-on synthetic pyrethroids (Taylor and Elliot, 1987).

These well-established control measures have not so far been perceived as appropriate, however, for Lyme borreliosis, which seems to be mainly associated with woodland that contains a variety of wild hosts, but especially deer and small mammals. Modification of such habitats is impractical and has rarely been considered in Europe for the purpose of tick control, even in relation to the zoonotic infection, tick-borne encephalitis, which also occurs in woodland foci. Applying acaricides to wild hosts is obviously very difficult, and success will depend largely on the development of self-medication systems, such as the provision of treated cotton waste for use as nesting material by small mammals in North America (Mather et al., 1987b). So far, no attempts have been made to control ticks on deer, but the advent of the benzoylphenyl urea derivatives, with their very low toxicity and considerable persistence after ingestion (Hess et al., 1990), offers some promise in this context. The effects of reduction of deer populations have been investigated in North America (Wilson et al., 1988), but not in Europe, where it would probably be much more difficult, since the most common species, the roe deer, *Capreolus capreolus*, is solitary and secretive. Many other approaches have been suggested, including the use of parasitoids and the use of pheomones as baits (Jaenson et al., 1990), but so far no serious attempts have been made in Europe to control Lyme disease through the suppression of tick populations.

The main prophylactic measures currently available are limited. Most have been developed with tick-borne encephalitis in mind and are equally relevant to Lyme disease. They consist of wearing appropriate clothing, applying various repellents such as DEET (Sixl and Stunzner, 1975), synthetic pyrethroids (Rupes et al., 1976) and even perfumes (Novak, 1981) to clothing, and most importantly disseminating information and advice, particularly regarding the presence of infected areas, wearing suitable clothing when in such areas and, lastly, personal inspection and prompt removal of any ticks.

11. INTEGRATED PEST MANAGEMENT

Rather than relying on any one single method to combat Lyme disease, it is clear that we must progress to the integrated pest management (IPM) approach. This type of approach, now being widely used against agricultural pests, should also be used against vectors of human disease. The most

thoroughly conducted IPM campaign against a tick population was conducted in in the Land Between the Lakes region of Kentucky and Tennessee (Bloemer et al., 1990). These researchers used acaricides (chlorpyriphos), vegetation management (mowing and understory removal), and a deer exclosure (fence) to decrease *A. americanum* populations. Not surprisingly, the IPM approach, using all three methods, worked better than any single method. To date, no comprehensive study in areas of high *I. dammini* abundance has tested the IPM approach. Communities must be surveyed to determine what interventions they will accept. A plan acceptable and appropriate for each community will have to be designed and the effectiveness of IPM tested by a demonstrable decrease in Lyme disease transmission. The rationale for the use of IPM and its cost effectiveness as compared to other methods is discussed further in Chapter 9.

For maximum cost-effectiveness, the real risk to the human population in a particular habitat or geographical area must be assessable. Mathematical models could have an important role in such assessments, particularly in relation to geographical information systems, but also in predicting the impact of implementing control measures. However, the first requirement for realistic risk assessment is the systematic accumulation of relevant biological data so that the complex ecology of this disease may be understood and acted upon. We have our work cut out for us.

12. REFERENCES

Aeschlimann, A., Chamot, E., Gignon, E., Jeanneret, J.P., Kesseler, D., and Walther, C. (1986) *B. burgdorferi* in Switzerland. *Zbl. Bakt. Hyg. A* **263**, 450.

Afzelius, A. (1910) Verhandlungen der dermatologischen Gesellschaft zu Stockholm. *Arch. Dermatol. Syph.* **101**, 404.

Ai, C., Qiu, G., Shi, Z., Wu, X., Liu, X., and Zhao, J. (1991) Host associations and seasonal abundance of immature *Ixodes persulcatus* (Acari: Ixodidae) in a Lyme-disease-endemic area in China. *Exp. Appl. Acarol.* **12**, 251.

Anderson, J.F., Johnson, R.C., Magnarelli, L.A., and Hyde, F.W. (1985) Identification of endemic foci of Lyme disease: isolation of *Borrelia burgdorferi* from feral rodents and ticks (*Dermacentor variabilis*). *J. Clin. Microbiol.* **22**, 36.

Anderson, J.F., Johnson, R.C., Magnarelli, L.A., and Hyde, F.W. (1986a) Involvement of birds in the epidemiology of the Lyme disease agent *Borrelia burgdorferi*. *Infect. Immun.* **51**, 394.

Anderson, J.F., Doby, J.M., Coutarmanac'h, A., Hyde, F.W., and Johnson, R.C. (1986b) Antigenically different *Borrelia burgdorferi* strains from *Ixodes ricinus* in Brittany, France. *Med. Mal. Infect.* **16**, 171.

Anderson, J.F., Magnarelli, L.A., LeFebvre, R.B., Andreadis, T.G., McAninch, J.B., Perng, G.C., and Johnson, R.C. (1989) Antigenically variable *Borrelia burgdorferi* isolated from cottontail rabbits and *Ixodes dentatus* in rural and urban areas. *J. Clin. Microbiol.* **27**, 13.

Anderson, J.F., Magnarelli, L.A., and Stafford, K.C., III (1990) Bird-feeding ticks transstadially transmit *Borrelia burgdorferi* that infect Syrian hamsters. *J. Wildl. Dis.* **26**, 1.

Barbour, A.G., Heiland, R.A., and Howe, T.R. (1985) Heterogeneity of major proteins

in Lyme disease borreliae: a molecular analysis of North American and European isolates. *J. Infect. Dis.* **152**, 478.

Barnes, A.M., Ogden, L.J., and Campos, E.G. (1972) Control of the plague vector, *Opisocrostis hirsutus*, by treatment of prairie dog (*Cynomys ludovicianus*) burrows with 2% carbaryl dust. *J. Med. Entomol.* **9**, 330.

Beard, M.L., Rose, S.T., Barnes, A.M., and Montenieri, J.A. (1992) Control of *Oropysylla hirsuta*, a plague vector, by treatment of prairie dog burrows with 0.5% permethrin dust. *J. Med. Entomol.* **29**, 25.

Bloemer, S.R., Mount, G.A., Morris, T.A., Zimmerman, R.H., Barnard, D.R., and Snoddy, E.L. (1990) Management of Lone Star ticks (Acari: Ixodidae) in recreational areas with acaricide applications, vegetative management, and exclusion of white-tailed deer. *J. Med. Entomol.* **27**, 543.

Burgdorfer, W. (1986) Discovery of the Lyme disease spirochete: a historical review. *Zbl. Bakt. Hyg. A* **263**, 7.

Burgdorfer, W. and Hayes, S.F. (1989) Vector–spirochete relationships in louse borne and tick-borne borreliosis with emphasis on Lyme disease. *Adv. Dis. Vector Res.* **6**, 128.

Burgdorfer, W., Barbour, A.G., Hayes, S.F., Benach, J.L., Grunwaldt, E., and Davis, J.P. (1982) Lyme disease—a tick-borne spirochetosis? *Science* **216**, 1317.

Burgdorfer, W., Barbour, A.G., Hayes, S.F., Peter, O., and Aeschlimann, A. (1983) Erythema chronicum migrans—a tick-borne spirochetosis. *Acta Trop.* **40**, 79.

Burgdorfer, W., Hayes, S.F., and Corwin, D. (1989) Pathophysiology of the Lyme disease spirochete, *Borrelia burgdorferi*, in ixodid ticks. *Rev. Inf. Dis.* **2** (Suppl.), S1442.

Daniels, T.J., Fish, D., and Falco, R.C. (1991) Evaluation of host-targeted acaricide for reducing risk of Lyme disease in southern New York state. *J. Med. Entomol.* **28**, 537.

Deblinger, R.D. and Rimmer, D.W. (1991) Efficacy of a permethrin-based acaricide to reduce the abundance of *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **28**, 708.

Doby, J.M., Imbert-Hameurt, C. Jeanne, B., and Chevrier, S. (1989) Infection de *Ixodes ricinus* (Acarina, Ixodidae) par *Borrelia burgdorferi*, agent des spirochétoses à tique (malaie de Lyme et autres formes cliniques) dans l'ouest de la France. I—Resultats globaux de l'examen de 2,320 tiques. *Bull. Soc. Franc. Parasitol.* **7**, 111.

Doby, J.M., Bigaignon, G., Launay, H., Costil, C., and Lorvellec, O. (1990a) Présence de *Borrelia burgdorferi*, agent de spirchétoses à tiques, chez *Ixodes* (*Exopaliger*) *trianguliceps* Birula, 1894 et *Ixodes* (*Ixodes*) *acuminatus* Neumann, 1901 (Acarina Ixodidae) et chez *Ctenophthalamus baeticus arvernus* Jordan, 1931 et *Megabothris turbidus* (Rothchild, 1909) (Insectes Siphonaptera), ectoparasites de micromammifères des forêts dans l'Ouest de la France. *Bull. Soc. Franc. Parasitol.* **8**, 311.

Doby, J.M., Lemblé, C., Bigaignon, G., Kremer, M., Rolland, C. and Lambert, M.C. (1990b) *Borrelia burgdorferi* agent des spirochétoses à tiques (maladie de Lyme et autres formes cliniques) chez *Ixodes ricinus* (Acari: Ixodidae) en Alsace. Recherche chez 2,223 tiques. *Bull. Soc. Franc. Parasitol.* **8**, 339.

Donahue, J.G., Piesman, J., and Spielman, A. (1987) Reservoir competence of white-footed mice for Lyme disease spirochetes. *Am. J. Trop. Med. Hyg.* **36**, 92.

Donnelly, J. and Peirce, M.A. (1975) Experiments on the transmission of *Babesia divergens* to cattle by the tick *Ixodes ricinus*. *Internat. J. Parasitol.* **5**, 363.

Evans, S.R., Korch, G.W. Jr, and Lawson, M.A. (1990) Comparative field evaluation of permethrin and DEET-treated military uniforms for personal protection against ticks (Acari). *J. Med. Entomol.* **27**, 829.

Fish, D. and Daniels, T.J. (1990) The role of medium-sized mammals as reservoirs of *Borrelia burgdorferi* in southern New York. *J. Wildl. Dis.* **26**, 339.

Gern, L., Zhu, Z., and Aeschlimann, A. (1990) Development of *Borrelia burgdorferi* in *Ixodes ricinus* females during blood feeding. *Ann. Parasitol. Hum. Comp.* **65**, 89.

Gern, L., Toutoungi, L.N., Hu, C.M., and Aeschlimann, A. (1991) *Ixodes* (*Pholeoixodes*) *hexagonus*, an efficient vector of *Borrelia burgdorferi* in the laboratory. *Med. Vet. Entomol.* **5**, 431.

Gray, J.S. (1991) The development and seasonal activity of the tick *Ixodes ricinus*, a vector of Lyme borreliosis. *Rev. Med. Vet. Entomol.* **79**, 323.

Gray, J.S., Kahl, O., Janetzki, C., and Stein, J. (1992) Studies on the ecology of Lyme disease in a deer forest in Co. Galway, Ireland. *J. Med. Entomol.* **29**, 915.

Hess, E.A., Bouvard, H.R., Friedel, T., and Hyman, W.B. (1990) CGA 157'419: a novel benzolyphenyl urea for systemic tick control. *VIIth International Congress of Parasitology*, Paris, August 1990, Abstract p. 1098.

Hovmark, A., Asbrink, E., Schwan, O., Hederstedt, B., and Christensson, D. (1986) Antibodies to *Borrelia* spirochaetes in sera from Swedish cattle and sheep. *Acta Vet. Scand.* **27**, 479.

Humair, P.F., Turrian, N., Aeschlimann, A., and Gern, L. (1993) *Borrelia burgdorferi* in a focus of Lyme borreliosis—epizootiologic contribution of small mammals. *Folio Parasitologica* **40**, 65.

Jaenson, J.G.T. (1991) The epidemiology of Lyme borreliosis. *Parasitol Today* **7**, 39.

Jaenson, J.G.T., Fish, D., Ginsberg, H.S., Gray, J.S., Mather, T.N., and Piesman, J. (1990) Methods for control of tick vectors of Lyme borreliosis. *Scand. J. Infect. Dis. Suppl.* **77**, 151.

Kahl, O. and Knülle, W. (1988) Water vapour uptake from subsaturated atmospheres by engorged immature ixodid ticks. *Exp. Appl. Acarol.* **4**, 73.

Kahl, O., Janetzki, C., Gray, J., Stein, S., and Bauch, R.J. (1992) Tick infection rates with *Borrelia: Ixodes ricinus* versus *Haemaphysalis concinna* and *Dermacentor reticulatus* in two locations in eastern Germany. *Med. Vet. Entomol.* **6**, 363.

Kitron, U., Jones, C.J., Bouseman, J.K., Nelson, J.A., and Baumgartner, D.L. (1992) Spatial analysis of the distribution of *Ixodes dammini* (Acari: Ixodidae) on white-tailed deer in Ogle County, Illinois. *J. Med. Entomol.* **29**, 259.

Kolomin, G.V., Kiselev, A.N., and Bolotin, E.L. (1978) Population density of field ticks (Ixodidae) in western Sikhote-Alin. *Parazitogiya* **12**, 148.

Korenberg, E.I., Kryuchechnikov, V.N., Kovalevskii, Yu.V., Shcherbakov, S.V., Kuznetsova, R.I., and Levin, M.L. (1987) The tick *Ixodes persulcatus*—new carrier of *Borrelia burgdorferi*. *Dokl. Akad. Nauk SSSR* **297**, 1268–1270 (in Russian).

Kurtenbach, K., Dizij, A., Kampen, H., Moter, S., Wallich, R., Schaible, U.E., and Simon, M.M. (1992) European wild rodents as reservoir hosts of *Borrelia burgdorferi*: ecological and immunological aspects. *Vth International Conference on Lyme Borreliosis, Arlington, Virginia, USA*, Abstract no. 293.

Lane, R.S. and Brown, R.N. (1991) Wood rats and kangaroo rats: potential reservoirs of the Lyme disease spirochete in California. *J. Med. Entomol.* **28**, 299.

Lane, R.S. and Burgdorfer, W. (1986) Potential role of native and exotic deer and their associated ticks (Acari: Ixodidae) in the ecology of Lyme disease in California, U.S.A. *Zbl. Bakt. Hyg. A* **263**, 55.

Lane, R.S. and Loye, J.E. (1989). Lyme disease in California: interrelationships of *Ixodes pacificus* (Acari: Ixodidae), the western fence lizard (*Sceloporus occidentalis*), and *Borrelia burgdorferi*. *J. Med. Entomol.* **26**, 272.

Levine, J.F., Wilson, M.L., and Spielman, A. (1985) Mice as reservoirs of the Lyme disease spirochete. *Am. J. Trop. Med. Hyg.* **34**, 355.

Liebisch, A., Olbrich, S., Brand, A., Liebisch, G., and Mourettou-Kunitz, M. (1990) Natüliche Infektionen der Zeckenart *Ixodes hexagonus* mit Borrelien (*Borrelia burgdorferi*). *Tierarztliche Umschau* **44**, 809.

Lindsay, L.R., Barker, I.K., Surgeoner, G.A., McEwen, S.A., Elliott, L.A., and Kolar, J. (1991) Apparent incompetence of *Dermacentor variabilis* (Acari: Ixodidae) and fleas (Insecta: Siphonapter) as vectors of *Borrelia burgdorferi* in an *Ixodes dammini* endemic area of Ontario, Canada. *J. Med. Entomol.* **28**, 750.

Loew, J., Radda, A., Pretzman, G., and Groll, E. (1963) Untersuchungen, in einem Naturherd der Fruhsommer-Meningoencephalitis (FSME) Nierderoster-reich. 1. Mitteilung, Okologie und Saisondynamik von *Ixodes ricinus*. *Zentralbl. Bakteriol. Abt. I. Orig.* **190**, 183.

Lord, R.D., Humphreys, J.G., Lord, V.R., McLean, R.G., and Garland, C.L. (1992) *Borrelia burgdorferi* infection and overwintering in white-footed mice (*Peromyscus leucopus*) in hemlock (*Tsuga canadensis*) habitat in Western Pennsylvania. *J. Wildl Dis.* **28**, 364.

MacLeod, J. (1934) The part played by alternative hosts in maintaining the tick population of hill pastures. *J. Anim. Ecol.* **3**, 161.

Marquez, F.J. and Constan, M.C. (1990) Infection d'*Ixodes ricinus* (L. 1758) et *Haemaphysalis punctata* Canestrini et Fanzago, 1877 (Acarina, Ixodidae) per *Borrelia burgdorferi* dans le nord de la Péninsule Iberique (Pays Basque espagñol et Navarre). *Bull. Soc. Franc. Parasitol.* **8**, 323.

Mather, T.N., Piesman, J., and Spielman, A. (1987a) Absence of spirochaetes (*Borrelia burgdorferi*) and piroplasms (*Babesia microti*) in deer ticks (*Ixodes dammini*) parasitized by chalcid wasps (*Hunterellus hookeri*). *Med. Vet. Entomol.* **1**, 3.

Mather, T.N., Ribeiro, J.M., and Spielman, A. (1987b) Lyme disease and babesiosis: acaricide focused on potentially infected ticks. *Am. J. Trop. Med. Hyg.* **36**, 609.

Mather, T.N., Ribeiro, J.M.C., Moore, S.I., and Spielman, A. (1988) Reducing transmission of Lyme disease spirochetes in a suburban setting. *Ann. New York Acad. Sci.* **539**, 402.

Mather, T.N., Wilson, M.L., Moore, S.I., Ribeiro, J.M., and Spielman, A. (1989a) Comparing the relative potential of rodents as reservoirs of the Lyme disease spirochete (*Borrelia burgdorferi*). *Am. J. Epidemiol.* **130**, 143.

Mather, T.N., Telford, S.R., III, MacLachlan, A.B., and Spielman, A. (1989b) Incompetence of catbirds as reservoirs for the Lyme disease spirochete (*Borrelia burgdorferi*). *J. Parasitol.* **75**, 66.

Mather, T.N., Duffy, D.C., and Campbell, S.R. (1993) An unexpected result from burning vegetation to reduce Lyme disease transmission risks. *J. Med. Entomol.* **30**, 642.

Matuschka, F.R. and Spielman, A. (1992) Loss of Lyme disease spirochetes from *Ixodes ricinus* ticks feeding on European blackbirds. *Exp. Parasitol.* **74**, 151.

Matuschka, F.R., Lange, R., Spielman, A., Richter, D., and Fisher, P. (1990) Subadult *Ixodes ricinus* (Acari: Ixodidae) on rodents in Berlin, West Germany. *J. Med. Entomol.* **27**, 385.

Maupin, G.O., Fish, D., Zultowsky, J., Campos, E.G., and Piesman, J. (1991) Landscape ecology of Lyme disease in a residential area of Westchester County, New York. *Am. J. Epidemiol.* **133**, 1105.

Mehl, R., Bjerknes, T., Sandven, P., and Vandvik, B. (1989) *Borrelia burgdorferi* i skogflatt *Ixodes ricinus* i Norge. *Fauna Nov. Ser. B* **36**, 87.

Miller, G.L., Craven, R.B., Bailey, R.E., and Tsai, T.F. (1990) The epidemiology of Lyme disease in the United States, 1987–1988. *Lab. Med.* **21**, 285.

Miller, J.A., Garris, G.I., George, J.E., and Oehler, D.D. (1989) Control of lone star ticks (Acari: Ixodidae) on spanish goats and white-tailed deer with orally administered ivermectin. *J. Econ. Entomol.* **82**, 1650.

Milne, A. (1949) The ecology of the sheep tick, *Ixodes ricinus* L. Host relationships of the tick. Part 2. Observations on hill and moorland grazings in northern England. *Parasitol.* 39, 173.

Miserez, V., Gern, L., and Aeschlimann, A. (1990) *Borrelia burgdorferi* in ticks of the Canton, Tessin, Switzerland. *Parasitologia* **32**, 293.

Mount, G.A. (1984) Aerial application of diazinon granules for area control of the lone star tick (Acari: Ixodidae). *J. Econ. Entomol.* **77**, 1211.

Novak, D. (1981) Laboratory tests with new tick repellents. *Arch. Roum. Path. Exp. Micr.* **40**, 287.

Olson, C.A., Cupp, E.W., Luckhart, S., Ribeiro, J.M.C., and Levy, C. (1992) Occurrence of *Ixodes pacificus* (Parasitiformes: Ixodidae) in Arizona. *J. Med. Entomol.* **29**, 1080.

Piesman, J., Mather, T.N., Dammin, G.J., Telford, S.R., III, Lastavica, C.C., and Spielman, A. (1987a) Seasonal variation of transmission risk of Lyme disease and human babesiosis. *Am. J. Epidemiol.* **126**, 1187.

Piesman, J., Mather, T.N., Sinsky, R.J., and Spielman, A. (1987b) Duration of tick attachment and *Borrelia burgdorferi* transmission. *J. Clin. Microbiol.* **24**. 557.

Piesman, J., Maupin, G.O., Campos, E.G., and Happ, C.M. (1991) Duration of adult female *Ixodes dammini* attachment and transmission of *Borrelia burgdorferi*, with description of a needle aspiration isolation method. *J. Infect. Dis.* **163**, 895.

Pinger, R.R., Holycross, J., Ryder, J., and Mummert, M. (1991) Collections of adult *Ixodes dammini* in Indiana, 1987–1990, and the isolation of *Borrelia burgdorferi*. *J. Med. Entomol.* **28**, 745.

Ribeiro, J.M., Mather, T.N., Piesman, J., and Spielman, A. (1987) Dissemination and salivary delivery of Lyme disease spirochetes in vector ticks (Acari: Ixodidae). *J. Med. Entomol.* **24**, 201.

Rupes, V., Ledvinka, J., and Chmela, J. (1976) The possibility of individual protection of people against *Ixodes ricinus* ticks. *Cesk. Epidem. Mikr. Imm.* **25**, 345 (in Czech).

Sardelis, M.R., Neidhardt, K., Perich, M.J., Milstrey, E.G., Harlan, H.J., and Boobar, L.R. (1989) Reduction of the *Amblyomma americanum* (Acari: Ixodidae) population at Fort A.P. Hill, Virginia, by aerial application of diazinon granules: correlation of percentage control with received dose. *J. Med. Entomol.* **26**, 494.

Schulze, T.L., Bowen, G.S., Bosler, E.M., Lakat, M.F., Parkin, W.E., Altman, R., Ormiston, B.G., and Shisler, J.K. (1984) *Amblyomma americanum*: a potential vector of Lyme disease in New Jersey. *Science* **224**, 601.

Schulze, T.L., McDevitt, W.M., Parkin, W.E., and Shisler, J.K. (1987) Effectiveness of two insecticides in controlling *Ixodes dammini* (Acari: Ixodidae) following an outbreak of Lyme disease in New Jersey. *J. Med. Entomol.* **24**, 420.

Schulze, T.L., Parkin, W.E., and Bosler, E.M. (1988) Vector tick populations and Lyme disease. A summary of control strategies. *Ann. New York Acad. Sci.* **539**, 204.

Schulze, T.L., Taylor, R.C., Taylor, G.C., and Bosler, E.M. (1991a). Lyme disease: a

proposed ecological index to assess areas of risk in the northeastern United States. *Am. J. Public Health* **81**, 714.

Schulze, T.L., Taylor, G.C., Jordan, R.A., Bosler, E.M., and Shisler, J.K. (1991b) Effectiveness of selected granular acaricide formulations in suppressing populations of *Ixodes dammini* (Acari: Ixodidae): short-term control of nymphs and larvae. *J. Med. Entomol.* **28**, 624.

Sixl, W. and Stunzer, D. (1975) Autan als Zeckenabwehrmittel. *Angew. Parasitol.* **16**, 106.

Smaha, J. (1979) The incidence of the tick (*Ixodes ricinus*) in forest phytocenoses with relations to human hygiene. *Lesnictvi.* **25**, 417.

Smith, R.D., Miranpuri, G.S., Adams, J.H., and Ahrens, E.H. (1985) *Borrelia theileri*: isolation from ticks (*Boophilus microplus*) and tick-borne transmission between splenectomized calves. *Am. J. Vet. Res.* **46**, 1396.

Solberg, V.B., Neidhardt, K., Sardelis, M.R., Hoffmann, F.J., Stevenson, R., Boobar, L.R., and Harlan, H.J. (1992) Field evaluation of two formulations of cyfluthrin for control of *Ixodes dammini* and *Amblyomma americanum* (Acari: Ixodidae). *J. Med. Entomol.* **29**, 634.

Sonenshine, D.E. and Haines, G. (1985) A convenient method for controlling populations of the American dog tick, *Dermacentor variabilis* (Acari: Ixodidae) in the natural environment. *J. Med. Entomol.* **22**, 577.

Spielman, A., Clifford, C.M., Piesman, J., and Corwin, M.D. (1979) Human babesiosis on Nantucket Island, USA: description of the vector, *Ixodes* (*Ixodes*) *dammini*. n.sp. (Acarina: Ixodidae). *J. Med. Entomol.* **15**, 218.

Stafford, K.C., III (1991a) Effectiveness of carbaryl applications for the control of *Ixodes dammini* (Acari: Ixodidae) nymphs in an endemic residential area. *J. Med. Entomol.* **28**, 32.

Stafford, K.C., III (1991b) Effectiveness of host-targeted permethrin in the control of *Ixodes dammini* (Acari: Ixodidae). *J. Med. Entomol.* **28**, 611.

Stanek, G. and Simeoni, J. (1989) Are pigeons' ticks transmitters of *Borrelia burgdorferi* in humans? A preliminary report. p. 42 *In:* Stanek, G., Kristoferitsch, W., Pletschette, M., Barbour, A.G. and Flamm, H. (eds) *Lyme Borreliosis II. Zbl. Bakt. Suppl.* 18.

Steere, A.C. and Malawista, S.E. (1979) Cases of Lyme disease in the United States: locations correlated with distribution of *Ixodes dammini*. *Ann. Intern. Med.* **91**, 730.

Steere, A.C., Malawista, S.E., Snydman, D.R., Shope, R.E., Andiman, W.A., Ross, M.R., and Steele, F.M. (1977) Lyme arthritis; an epidemic of oligoarticular arthritis in children and adults in three Connecticut communities. *Arthritis Rheum.* **20**, 7.

Steere, A.C., Broderick, T.F., and Malawista, S.E. (1978) Erythema chronicum migrans and Lyme arthritis: epidemiologic evidence for a tick vector. *Am. J. Epidemiol.* **108**, 312.

Stuen, S. and Fridriksdottir, V. (1991) Experimental inoculation of sheep with *Borrelia burgdorferi*. *Vet. Rec.* **129**, 315.

Taylor, S.M. and Elliot, C.T. (1987) Deltamethrin and flumethrin pour-on formulations for the control of *Ixodes ricinus* on cattle. *Vet. Rec.* **120**, 278.

Telford, S.R., III and Spielman, A. (1989) Enzootic transmission of the agent of Lyme disease in rabbits. *Am. J. Trop. Med. Hyg.* **41**, 482.

Telford, S.R., III, Mather, T.N., Moore, S.I., Wilson, M., and Spielman, A. (1988) Incompetence of deer as reservoirs of the Lyme disease spirochete. *Am. J. Trop. Med. Hyg.* **39**, 105.

Theiler, A. (1904) Spirillosis of cattle. *J. Comp. Pathol. Ther.*, **17**, 47.

Uilenberg, G., Hinaidy, H.K., Perie, N.M., and Feenstraat, T. (1988) *Borrelia* infections of ruminants in Europe. *Vet. Quart.* **10**, 63.

White, D.J., Chang, H.G., Benach, J.L., Bosler, E.M., Meldrum, S.C., Means, R.G., Debbie, J.G., Birkhead, G.S., and Morse, D.L. (1991) The geographic spread and temporal increase of the Lyme disease epidemic [see comments]. *JAMA* **266**, 1230..

Wilske, B., Anderson, J.F., Baranton, G., Barbour, A.G., Hovind, Hougen, K., Johnson, R.C., and Preac-Mursic, V. (1991) Taxonomy of *Borrelia* spp. *Scand. J. Infect. Dis.* Suppl. **77**, 108.

Wilson, M.L. (1986) Reduced abundance of adult *Ixodes dammini* (Acari: Ixodidae) following destruction of vegetation. *J. Econ. Entomol.* **79**, 693.

Wilson, M.L., Telford, S.R., III, Piesman, J., and Spielman, A. (1988) Reduced abundance of immature *Ixodes dammini* (Acari: Ixodidae) following elimination of deer. *J. Med. Entomol.* **25**, 224.

Wilson, M.L., Ducey, A.M., Litwin, T.S., Gavin, T.A., and Spielman, A. (1990) Microgeographic distribution of immature *Ixodes dammini* ticks correlated with that of deer. *Med. Vet. Entomol.* **4**, 151–159.

Zung, J.L., Lewengrub, S., Rudzinska, M.A., Spielman, A., Telford, S.R., and Piesman, J. (1989) Fine structural evidence for the penetration of the Lyme disease spirochete *Borrelia burgdorferi* through the gut and salivary tissues of *Ixodes dammini*. *Can. J. Zool.* **67**, 1737.

12

Tick-borne Encephalitis Subgroup

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1. INTRODUCTION

This chapter aims to compare the ecological dynamics of important members of the tick-borne encephalitis (TBE) subgroup (Table 12.1), a distinct antigenic complex of the virus family Flaviviridae (Murphy et al., 1994). The TBE subgroup is one of the most intensely studied groups of tick-borne pathogens. Indeed, Hoogstraal (1966) commented that "Anyone with a broad general knowledge of research accomplishments ... in the (TBE) complex of viruses ... has an insight into practically every known factor in maintenance and dissemination of disease-causing agents transmissible between lower animal and man".

1.1. Viruses and the Diseases

Many members of the TBE subgroup cause encephalitis in humans. The notable exceptions are Omsk Hemorrhagic Fever (OHF) virus and Kyasanur Forest Disease (KFD) virus, both of which give rise to hemorrhagic disease. Langat, Karshi, and Royal Farm viruses (Table 12.1) are not regarded as important human pathogens and little is known of their ecologies; they are not considered in this chapter. Negishi virus is closely related to Louping Ill virus and is also excluded (Venugopal et al., 1992).

A striking difference among TBE subgroup viruses is their propensity to cause disease in non-human vertebrates. The two TBE viral subtypes and Powassan (POW) virus are significant pathogens only of humans. In contrast, louping ill (LI) virus infection results in severe and often fatal disease in sheep and red grouse, OHF virus infection causes "muskrat disease," and KFD virus is associated with "monkey disease". Such differences in pathogenicity reflect the unique ecological characteristics of each virus.

Tick-borne encephalitis was recognized clinically in the Far East of the former USSR (now CIS) in the early 1930s. Severe cases of encephalitis were observed in humans residing in the area. A special expedition was organized in 1937 by L.A. Zilber to determine the cause of the disease. Virus isolates were

Table 12.1. Members of the tick-borne encephalitis subgroup. Adapted from Calisher and Karabatsos (1988) and Murphy et al. (1994)

Virus	Subtype	Distribution
Tick-borne encephalitis (Russian spring–summer encephalitis)	European	Europe
	Far Eastern	Asia
	Louping ill	Europe (mainly UK and Ireland)
	Langat	Asia
	Omsk hemorrhagic fever	Western Siberia
	Kyasanur Forest disease	India
Powassan		Canada, northern USA, CIS (Far East)
Karshi		CIS (south)
Royal Farm		Afghanistan

obtained from the blood of patients and from *Ixodes persulcatus* ticks. The disease has several names, including Russian Spring–Summer Encephalitis (RSSE), Far Eastern encephalitis, and forest spring encephalitis (Zilber and Soloviev, 1946). In 1948, a similar though less severe form of encephalitis affected humans residing in central Bohemia, Czech Republic. The virus recovered from the blood of a patient and from *I. ricinus* ticks was related to isolates from RSSE cases (Gallia et al., 1949; Hloucal and Gallia, 1949; Rampas and Gallia, 1949). Similar or milder forms of the disease, called biphasic meningoencephalitis, were observed in other central and eastern European countries. TBE is currently endemic over a wide area—covering Europe, northern Asia, and China—which corresponds to the distribution of the ixodid virus vectors. Several thousand human cases are recorded annually, with considerable variation from year to year (WHO, 1986).

Two subtypes of TBE virus can be differentiated by immunodiffusion and antibody absorption tests (Clarke, 1962, 1964), monoclonal antibodies (Guirakhoo et al., 1989), and nucleotide sequence analysis (Mandl et al., 1988, 1989; Pletnev et al., 1990). In general, Far Eastern subtypes of TBE virus (also known as RSSE virus) cause severe disease in humans with a mortality which can be 50% in some outbreaks; disease associated with European subtypes, also known as western TBE virus or Central European encephalitis (CEE) virus, is less severe and mortality is usually under 5% (Grešiková and Calisher, 1988). However, the literature is frequently vague and there are many discrepancies in distinguishing antigenic relationships, clinical forms, and virus ecology.

Louping ill virus is the UK representative of the TBE subgroup. This virus derives its name from the disease of sheep which has been recognized in southern Scotland for at least two centuries (Smith and Varma, 1981). In 1931, the etiological agent was identified as a virus transmitted by the sheep tick, *Ixodes ricinus* (Greig et al., 1931; MacLeod and Gordon, 1932). The disease is found

throughout much of the upland sheep-farming areas of Scotland, northern and southwestern England, Ireland, and Wales. A similar disease has been recorded in Bulgaria, Turkey, Norway, and Spain (Reid, 1988). In this chapter, the ecology of LI virus is limited to published data relative to the UK. The disease in red-grouse populations is confined to Scotland and northern England, and may cause mortality in 80–95% of birds (Reid et al., 1978; Hudson, 1986). LI virus can cause severe encephalitis in humans; however, there are only about 35 recorded cases and they were mainly confined to laboratory workers, veterinary surgeons, farmers, and abattoir workers (Smith and Varma, 1981).

Though closely related to TBE virus, OHF virus appears to be unique with respect to both the clinical features of the disease and epidemiology, and ecology of the virus (Lvov, 1988). The distribution of OHF across the entire forest–steppe landscape zone of western Siberia within the borders of Omsk, Novosibirsk, Kurgan, and Tumen regions, and is adjacent to TBE foci. OHF has the shortest recorded history of all natural-focal diseases of western Siberia. This reflects the unique ecology of the virus, and the introduction and husbandry of muskrats in the 1940s. Indeed, during early studies OHF was known as "muskrat disease" because infections of humans were associated with this rodent (Kharitonova and Leonov, 1985). The disease was first reported in 1941 when physicians recorded sporadic cases in the forest–steppe area of Omsk Oblast. Between 1945 and 1958, nearly 1,500 cases were recorded in the Omsk region but subsequently only sporadic, individual cases have been registered, and these usually occur in autumn and winter among hunters of muskrats.

Elucidation of the ecology of KFD virus has been described as a "dramatic epidemiological detective story" (Hoogstraal, 1966). In March 1957, abnormal mortality in monkeys was reported from the forests of Shimoga District in Karnataka State, India, and a number of human cases of febrile and often fatal illness were reported from the same area. A virus antigenically related to TBE virus was subsequently isolated from humans, and from sick and dead langur (*Presbytis entellus*), and bonnet macaque (*Macaca radiata*) monkeys found near Kyasanur State forest (Work and Trapido, 1957). Subsequently, the virus was isolated from ticks of the genus *Haemaphysalis* occurring in the forest (Banerjee, 1988). About 400–500 human cases are reported annually, the number reflecting the prevalence of active foci in the forest region. The highest recorded incidence was in 1983, with 1,555 cases and 150 deaths (Banjeree, 1988).

Powassan virus is the North American member of the TBE subgroup, although there are several records of POW virus from ticks and mosquitoes collected in the CIS (Karabatsos, 1985). The virus was originally isolated from a pool of *Dermacentor andersoni* collected in 1952 in Colorado (Thomas et al., 1960). POW virus derives its name from the town in Northern Ontario where the first fatal case of the encephalitic disease was recognized (McLean and Donohue, 1959). About 20 cases of the disease have been reported in North America and there was one fatal case recorded in the CIS (Artsob, 1988).

1.2. Evolutionary Relationships

The evolutionary history of viruses within the TBE subgroup is being charted with the help of nucleotide sequence data. Undoubtedly, the course of evolution is strongly inflenced by virus ecology (Beaty et al., 1988; Nuttall et al., 1991). For example, the pronounced differences in the physiology and behaviour of ticks and insects result in different selection pressures on the arboviruses that they transmit. In respect to virus infection, one of the important differences between ticks and insects is that blood digestion in ticks is intracellular whereas it is extracellular in insects. Thus viruses imbibed in a blood-meal enter a completely different environment in the tick gut compared with the insect gut, and hence they are exposed to different selection pressures. Such differences help explain why evolutionary trees based on sequence data clearly show that the tick-borne flaviviruses are set apart from the mosquito-borne flaviviruses such as yellow fever, dengue, and Japanese encephalitis viruses (Mandl et al., 1993; Venugopal et al., 1994).

The complete genomic sequences of the European and Far eastern subtypes of TBE virus (Mandl et al., 1988, 1989; Pletnev et al., 1990), Langat (Mandl et al., 1991; Iacono-Connors and Schmaljohn, 1992), and Powassan (Mandl et al., 1993) viruses have been determined. In addition, the sequences of the structural proteins of LI (Shiu et al., 1991) and KFD (Venugopal et al., 1994) viruses, and the envelope glycoprotein of OHF (Gritsun et al., 1993) and Negishi (Venugopal et al., 1992) viruses have been published. Interestingly, sequence data indicate that OHF and KFD viruses are not close relatives compared to other members of the TBE subgroup despite similarities in the diseases they cause (Venugopal et al., 1994).

2. TRANSMISSION CYCLES

Transmission cycles are determined by the interactions between tick-borne viruses, their vectors and their vertebrate hosts. Such interactions are governed by the biology and population dynamics of the tick and vertebrate hosts, which in turn are strongly influenced by environmental factors (Blaskovic and Nosek, 1972; Hoogstraal, 1973; Nuttall, 1984). These factors combine to determine the basic reproductive rate (R_0) of the virus infection, i.e., the number of new infections that arise from a single current infection (Anderson and May, 1982, 1991; Dietz, 1988; Hasibeder and Dye, 1988).

One or two primary tick vectors, that play a crucial role in maintaining the transmission cycle of tick-borne flaviviruses, can be identified (Table 12.2). Their host preferences determine which vertebrates are natural hosts of the virus. In addition to the primary vector, a few or several tick species may act as secondary vectors. Such vectors are probably not able to circulate the virus without the participation of the primary vector(s), but they can help to maintain the transmission cycle. Environmental changes, including changes in climate, may increase the role of secondary vectors in the virus transmission cycle.

Table 12.2. Primary tick vectors and their preferred hosts

Virus	Tick vector	Vertebrate host
TBE (Far Eastern)	*Ixodes persulcatus*	Mammals, birds
TBE (European)	*Ixodes ricinus*	Mammals, birds
LI	*Ixodes ricinus*	Sheep
OHF	*Dermacentor reticulatus, Ixodes apronophorus*	Rodents
KFD	*Haemaphysalis spinigera, Haemaphysalis turturis*	Cattle, monkeys, porcupines, rats, shrews
POW	*Ixodes cookei, Ixodes persulcatus*	Rodents, carnivores, birds

Similarly, ancillary vectors may act as the primary vector species in certain specific biotopes.

2.1. Enzootic and Epizootic Transmission Cycles

Enzootic transmission cycles of members of the TBE subgroup have many common features. They rely on a relatively stable tick population and a large population of susceptible mammals, generally insectivores and rodents that have a short life span thus providing a renewable source of susceptible hosts. Where differences in the enzootic cycles are apparent, they relate mostly to the peculiar ecology of the natural foci, e.g., the association of LI virus with sheep, and of OHF virus with rodents in lakeside habitats.

The transmission cycles of most TBE complex viruses involve hard ticks of the family Ixodidae. They are three-host ticks, i.e., each parasitic stage (larva, nymph, adult) feeds on a different host (usually a wide range of species) for a period of a few days. In general, immature stages (larvae and nymphs) feed on small mammals and birds, and adults on larger species. Such trophic relationships are a deterministic feature of enzootic transmission cycles. Larger mammals (goats, cows, and sheep) become infected, but levels of viremia may be low; consequently, they are considered as hosts sustaining vector tick populations rather than as hosts of the virus. The notable exception is LI virus infection of sheep.

Nine tick species can be considered as primary vectors in the enzootic cycles of TBE complex viruses; six of these species belong to the genus *Ixodes* (Table 12.2). Experimental studies have demonstrated that ticks belonging to numerous other species are competent vectors; however, virus isolations from such ticks are rare. Hence they are either secondary vectors, or they are not considered natural vectors because their distribution is not sympatric with that of the virus. For example, *Phipicephalus appendiculatus* is a competent vector of LI and TBE viruses under laboratory conditions (Labuda et al., 1993b; Alexander and Neitz, 1933) but this tick species is confined to Africa, a continent in which viruses of the TBE subgroup have not been recorded. About 20 tick species are involved

or potentially involved as secondary vectors of TBE subgroup viruses. In certain ecosystems or biotopes, the vector–host relationships may differ significantly from the typical enzootic cycle. For example, TBE virus (Far Eastern subtype) has been isolated from the host-specific tick, *Ixodes lividus*, which parasitizes birds (sand martins, *Riparia riparia*), and antibodies to TBE virus have been detected in bats that are hosts of *Ixodes vesperilionis* (Hoogstraal, 1966).

Transmission cycles of Far Eastern and European subtypes of TBE viruses involve ixodid ticks and rodents. The most important rodents usually are those that are most abundant within a focus of infection (generally *Apodemus*, *Clethrionomys*, or *Microtus* species). In contrast to TBE viruses, the transmission cycle of LI virus is highly dependent on sheep farming and land management. Besides vector density, factors determining the prevalence of LI virus within enzootic areas are clearly influenced by the access of sheep to pasture during periods of tick activity, and vaccination regimes for sheep. The proportion of grouse that survive epizootics of LI appears to be insufficient to maintain the virus transmission cycle (Reid et al., 1978).

Enzootic and epizootic tick-borne transmission cycles can also be distinguished for OHF virus. The enzootic cycle involves ixodid ticks (*Dermacentor* and *Ixodes* species) and voles, and the epizootic cycle affects muskrats. However, there is confusion in the literature as to the primary transmission cycle of OHF virus. The review of Lvov (1988) emphasizes the role of tick vectors, whereas Kharitonova and Leonov (1985) describe an enzootic cycle that revolves around water-borne transmission. Water contaminated by the urine of infected rodents is considered to be a source of infection of other rodents as well as birds, and even the larvae of mosquitoes that subsequently transmit the virus to birds.

Transmission cycles of KFD virus involve a large variety of ixodid ticks with *Haemaphysalis spinigera* and *H. turturis* as the primary vectors. Forest rats, shrews, and porcupines are considered the most important hosts in the enzootic cycle whereas epizootic cycles affect monkeys (Banerjee, 1988). Alternative enzootic cycles have been reported outside the disease zone. For example, KFD-seropositive bats were captured in the vicinity of Poona (Boshell, 1969).

Powassan virus is transmitted in an enzootic cycle involving ixodid ticks, and rodents and carnivores. Lagomorphs and birds (the latter in the CIS) may also be involved (Artsob, 1988). Although the virus has been isolated from mosquitoes in the Far East, including larvae of *Aedes togoi* (Leonova et al., 1978), no insect vectors of POW virus have been identified in North America and the role of mosquitoes in the enzootic cycle is unclear.

Most TBE subgroup viruses are transmitted vertically from the infected female via the egg to the succeeding generation (see Section 3.3). The significance of vertical transmission in the ecology of these viruses is undetermined. The frequency of vertical transmission generally appears to be indicating that vertical transmission is not important at the population level of virus infections (Rehacek, 1965; Burgdorfer and Varma, 1967). However, results of modeling the relative contribution of transovarial transmission to the maintenance of tick-borne diseases suggests that even a low level of vertical transmission can

be significant (Randolph, 1994). In addition, virus transmission between infected and uninfected larvae feeding together on an uninfected host may amplify the number of infections in the tick population (see Section 3.3).

2.2. Primary Tick Vector Species and their Hosts

Evidence of primary vectors is based mostly on virus isolation from field-collected ticks. Since the expedition of Zilber and colleagues (Zilber and Xoloviev, 1946), repeated field studies have identified *I. persulcatus* as the principal vector of TBE virus (Far Eastern subtype). Similarly, following the first isolation of TBE virus from *I. ricinus* ticks in Czechoslovakia (Rampas and Gallia, 1949), large numbers of isolates have been obtained from *I. ricinus* in various countries of Europe, reflecting the primary role of *I. ricinus* as a vector of the European TBE viral subtype. The epizootiology of LI virus implicates *I. ricinus* as the sole vector of this virus (Reid, 1988). Like most of the vectors of TBE subgroup viruses, *I. ricinus* and *I. persulcatus* are three-host ticks; all parasitic stages except the male engorge (though males may imbibe a small amount of blood).

Ixodes persulcatus is taxonomically closely related to *I. ricinus* which it replaces in the north east of Europe, from the Baltic Sea shore and extending across northern Asia to Japan. The northern border of its distribution extends across the forests of the central taiga. The developmental biology of *I. persulcatus* is very similar to that of *I. ricinus* (Balashov, 1972), though the seasonal activity of *I. persulcatus* may be shorter, lasting only from the end of April to the beginning of June in colder biotopes (Zilber and Xoloviev, 1946).

Ixodes ricinus is a tick of temperate regions in Europe, found within the latitudes 39° and 65° and extending east of the Caspian Sea as far as 60° longitude (Blaškovič, 1967). This species has even been recorded in North Africa (Yousfi-Monod and Aeschlimann, 1986). *Ixodes ricinus* is of considerable importance in both medical and veterinary medicine. The main diseases caused by pathogens transmitted (or exacerbated) by *I. ricinus* are TBE, LI, Lyme disease, tick pyaemia, tick-borne fever, and babesiosis. Detailed studies on the ecology of *I. ricinus* were undertaken in Britain between 1932 and 1955 (reviewed by Arthur, 1962), and were followed by research in Europe (reviewed by Hoogstraal, 1966; Balashov, 1972). Despite these studies, a recent overview of *I. ricinus* concluded that much of the ecology and biology of this tick species remains undetermined (Gray, 1991). Each stage of *I. ricinus* takes approximately 1 year to develop to the next so the life cycle takes 3 years to complete, though it may vary from 2 to 6 years throughout the geographic range. Unfed ticks can quest for several weeks but do not usually survive from one season to the next.

The incidence of TBE virus infection in *I. ricinus* varies from 0.1% or less to about 5%, depending on the geographic location and particular focus (Grešiková and Calisher, 1988). By comparison, active foci of the Far Eastern subtype of TBE virus contain a comparatively high prevalence of infected ticks. For example, in the Krasnojarsk region of Siberia the incidence of infection

varied from 5 to 43%, with a mean of 16% (Cirkin et al., 1968), and it was estimated at 40% in western Siberia (Levkovich et al., 1967). The difference in infection prevalence may compensate for the shorter seasonal activity of *I. persulcatus* and consequent reduced period for active virus transmission. Variations in annual prevalence of infected ticks and in the virus titer in individual ticks have been recorded, but the reasons for these differences are unknown (Korenberg et al., 1992).

There is little published information on the density-dependent mortality factors that regulate tick populations. The population dynamics of *I. ricinus* and *I. persulcatus* are determined by the availability of large mammalian species that support the adult stages (Milne, 1949a, 1949b), although fluctuations in the number of small mammals and game birds have been correlated with the population dynamics of *I. ricinus* (Smith and Varma, 1981). The immature stages have a more catholic host range than the adults, feeding on most warm-blooded animals as well as reptiles. However, nymphs are less successful than larvae in feeding on small mammals (Milne, 1949b). In western Slovakia there were about 20 times more *I. ricinus* larvae than nymphs on small mammals (Labuda et al., 1991); larva to nymph ratios on birds were almost equal (Ernek et al., 1973). The height above ground at which each developmental stage quests for a host is an important determinant of the species infested. Nymphs are probably the most important vector stage in the transmission of TBE virus because they are more numerous than adults and are less host specific. Since transovarial transmission is evidently rare (Reháček, 1962), larvae are probably more important as acquirers (recipients) of the virus than as transmitters (donors) (but see Section 3.3).

Field investigations have revealed an aggregated distribution of TBE virus-infected ticks (Pretzmann et al., 1967). The non-random distribution consists of "elementary foci" which comprise many "microfoci" associated with the feeding or resting places of maintenance hosts (Blaškovič and Nosek, 1972). Several factors may contribute to the clumped distribution of TBE virus-infected ticks. These include the different spatial distributions of questing ticks (clumped for larvae, and random for nymphs and adults) (Randolph and Steele, 1985), the enhanced efficiency of virus transmission between co-feeding ticks (Labuda et al., 1993a), and tick drop-off in the resting sites of small mammals (Belozerov, 1982; Matuschka et al., 1991). Models of vector-borne diseases predict that R_0 of infections that show a clumped distribuion can exceed the rate expected for a homogeneous distribution, while equilibrium disease prevalence may be lowered or raised (Haseibeder and Dye, 1988). This is because, within at least one elementary focus, there are sites that have a higher vector-to-host ratio than in the homogeous situation with the same total numbers of vectors and hosts. Such sites become microfoci or "hot spots" (Rogers, 1988) in which the virus can persist and from which it can spread. An important consequence of an aggregated infection distribution is the increased difficulty of eradicating the disease.

In the natural foci of LI virus in northern England and southwest Scotland, 94–99% of the tick population is supported by sheep (Milne, 1949a, 1959b).

Indeed, the distribution of infected ticks corresponds better with the distribution of sheep-farming areas in the UK than with the distribution of the vector, *I. ricinus* (Reid, 1984). In certain areas of the UK, red deer (*Cervus elaphus*) or hares (*Lepus europaeus*, *L. timidus*) may be important maintenance hosts of the ticks (Smith and Varma, 1981). As for TBE virus (European subtype), comparatively few infected ticks (about 0.1%) have been found in the field (Reid, 1988). If LI virus is maintained solely in a sheep–tick cycle, adult ticks are probably the principal transmitters of the virus to livestock: *I. ricinus* can acquire the infection as a feeding larva and/or nymph, and most adults parasitize susceptible hosts. Furthermore, since nymphs more than larvae seem to prefer livestock (Milne, 1949b), the nymph is more likely to acquire the infection and the adult more likely to transmit the virus. Given the narrow time-window in which sheep are infective for ticks (Reid, 1984), the role of different tick stages in virus transmission dynamics depends on the overlap in both seasonal activity of the different tick instars and their host utilization. The possible role of alternative host species in the transmission cycle of LI virus has been raised (see Section 3.3).

The primary tick vector of OHF virus in forest–steppe regions is *Dermacento reticulatus* and the hosts are voles, particularly the narrow-skulled vole, *Microtus gregalis*. *Ixodes apronophorus* and water voles are the principal vector and vertebrate host, respectively, in grassy marshes of the western Siberian lowland (Lvov, 1988). The prevalence of ticks infected with OHF virus corresponds with the density of ticks in a given focus. During the epidemic period (1945–1949) of OHF in the lake region of Omsk district, the density of *D. reticulatus* was 10 times greater than during the non-epidemic period of 1959 to 1962. In the former period, all cattle in the region were infested, and larvae and nymphs were mainly found on voles, particularly *M. gregalis*; the prevalence of infected ticks was 6%. In contrast, only 0.1–0.9% infected ticks were found during the non-epidemic period (Lvov, 1988).

Most of the few thousand KFD virus isolates have been obtained from *Haemaphysalis* ticks, particularly nymphs of *H. spinigera*. The prevalence of infected *H. spinigera* collected from the forest floor varies from 0.1 to 1% in disease foci (Banerjee, 1988). *Haemaphysalis spinigera* is by far the dominant tick species and it is the main species attracted to man. Larval and nymphal stages frequently infest monkeys, whereas large numbers of adults are found on domestic cattle. The aggregated distribution of infected ticks (*H. spinigera* and *H. turturis*) in enzootic areas has been attributed to the creation of infection foci at sites where nymphs drop off sick or dead monkeys (Boshell, 1969; Sreenivasan et al., 1983). In the forest region, cattle (hosts of adult ticks) are the most important factors in determining the number and density of *H. spinigera* populations; larvae and nymphs of *H. spinigera* and *H. turturis* occur in approximately equal numbers on small mammals and they also feed on birds (Bhat, 1974, 1979, 1985).

In North America, most isolations of POW virus have been from *Ixodes cookei* (Artsob, 1988). This tick is found throughout the East and Midwest, and feeds on mammals of many species, particularly groundhogs (*Marmota monax*).

The greatest number of POW virus isolates in the Primor'ye region of southeast CIS were recorded from *Ixodes persulcatus* (Hoogstraal, 1980).

2.3. Secondary Tick Vector Species

All competent tick species occurring in sufficiently high numbers and having a sympatric distribution with the primary tick vector may become infected, and subsequently transmit a member of the TBE virus complex. However, although experimental studies have shown that numerous tick species are competent vectors, their ecological roles have not been determined. For example, the vector competence of *I. hexagonus* for TBE virus has been demonstrated in the laboratory, including transmission of TBE virus to hedgehogs, the principal host of this tick species (Streissle, 1961; van Tongeren, 1962), and TBE virus has been isolated from field-collected *I. hexagonus* (Křivanec et al., 1988). *Ixodes arboricola*, a bird tick was shown to be a competent vector in the laboratory (Lichard and Kožuch, 1967). Similarly, *Haemaphysalis concinna*, *H. inermis*, and *H. punctata* are competent vectors, and TBE virus has been isolated from field collected specimens (Grešiková, 1972; Grešiková and Calisher, 1988). In the Khabarovsk region (near the Sea of Japan), *Haemaphysalis* rather than *Ixodes* ticks may be the primary vector of the Far Eastern subtype (Hoogstraal, 1966). The relative roles of *Ixodes ricinus* and *I. persulcatus* as primary and/or secondary vectors of the two TBE viral subtypes are undetermined for parts of Europe where the two species are sympatric. In contrast to TBE virus, the epizootiology of LI virus implicates *I. ricinus* as the exclusive vector even though *Dermacentor reticulatus* and *Haemaphysalis punctata* are present in the UK.

The role of secondary vectors in the transmission cycle of OHF virus depends on ecological factors. *Ixodes apronophorus* may comprise 18–68% of tick collections from small mammals in grassy marshes of the western Siberian lowland. During July and August, water voles (the principal host of *I. apronophorus*) migrate from wet areas to meadows in search of food. In meadows, the voles come in contact with the primary vector of OHF virus, *Dermacentor reticulatus*, the larvae and nymphs of which reach peak activity during this period (Lvov, 1988). Thus, *I. apronophorus* may be considered as either a primary or secondary vector. *Dermacentor marginatus* and *Ixodes persulcatus* play secondary roles in the transmission cycle. However, all tick species may play a secondary role in OHF virus transmission if the principal transmission route is via contaminated water.

In the endemic area of KFD, 36 species of ticks have been recorded. Of the 15 species of *Haemaphysalis* present in the area, KFD virus has been isolated from *H. spinigera* and eight other species. According to the frequency of isolations, they are *H. turturis*, *H. kinneari*, *H. wellingtoni*, *H. bispinosa*, *H. minuta*, *H. cuspidata*, *H. intermedia*, and *H. aculeata*. The virus has also been isolated from *I. ceylonensis* and *I. petauristae*, and one species each of *Dermacentor*, *Amblyomma*, *Rhipicephalus*, and *Ornithodoros* (Trapido et al., 1959; Boshell et al., 1968; Rajagopalan et al., 1969). Besides *H. spinigera*,

experimental transmission of KFD virus has been demonstrated with *H. turturis*, *H. minuta* (Singh et al., 1964), *H. kyasanurensis* (Bhat et al., 1975), *H. wellingtoni* (Bhat and Naik, 1978), I. petaursitae (Boshell and Rajagopalan, 1968; Singh *et al.*, 1968b), *I. ceylonensis* (Singh et al., 1968b), *Dermacentor auratus* (Sreenivasan et al., 1979), and *H. cuspidata* (unpublished data cited by Banerjee, 1988). In addition, transstadial persistence and subsequent virus transmission have been demonstrated by four species of argasid tick and one *Haemaphysalis* species not found in the endemic area. KDF virus has been detected in field-collected larvae of *H. spinigera* and *Ixodes* sp. on only two occasions in several years of study, indicating that vertical transmission may occur but at low frequency (Rao, 1963) (see Section 3.3).

Ixodid ticks of a few species have been implicated as alternative vectors of POW virus (Artsob, 1988). The evidence is based on virus isolations from field-collected specimens of *I. marxi* collected in Ontario and associated with red squirrels (*Tamiasciurus hudsonicus*), and from *D. andersoni* and *I. spinipalpus* in western North America. Field data indicate that the primary enzootic tick vectors and vertebrate hosts of POW virus vary according to the biotope and geographical area (Hoogstraal, 1966). Experimental studies have demonstrated virus transmission by *D. andersoni* and *I. pacificus*, although *I. pacificus* appears to be an inefficient vector (see Section 3.3). In the CIS, POW virus has been isolated from *Haemaphysalis longicornis*, *I. persulcatus*, and *D. silvarum* ticks, and also from *Aedes togoi* and *Anopheles hyrcanus* mosquitoes. However, experimental studies failed to demonstrate POW virus replication following inoculation of *Aedes aegypti* and *Culex fatigans* mosquitoes.

2.4. Direct Transmission Mechanisms

A common feature of many members of the TBE virus subgroup is the evidence of direct routes of virus transmission. It is unlikely that such modes of transmission are significant for the maintenance of virus transmission cycles—with the possible exception of OHF virus—though they may be epidemiologically important.

Virus transmission via milk and milk products has been demonstrated for most members of the TBE virus subgroup. In 1951–1952, an outbreak of TBE affecting at least 600 persons was recorded in the Roznava district of Slovakia. Milk of infected goats was incriminated as the probable source of infection (Blaškovič, 1954). Milk-borne transmission to humans is more common in Europe than in the Far East (Korenberg and Kovalevskiy, 1981). Experimental studies have demonstrated TBE virus in the milk of goats, sheep, and cattle for up to 8 days after infection (Grešková and Calisher, 1988). Although LI has not been reported in naturally infected goats, during experimental studies five kids acquired the infection after ingesting infected milk and all developed severe disease (Reid et al., 1984). In monkeys infected experimentally with KFD virus, there was evidence of virus passage in milk (Shah, 1965). Secretion of POW

virus in goat's milk has been demonstrated experimentally (Woodall and Roz, 1977).

Predation of TBE virus-infected small mammals by buzzards (*Buteo buteo*) and goshawks (*Acceipiter gentilis*) led to infection (Ašmera et al., 1962). Reid (1988) suggested that LI virus infection of rodents could arise from predation of infected ticks. Horizontal spread of OHF virus from rodent to rodent, and rodent to human, may occur via the alimentary and respiratory routes. During epizootics in muskrats, the virus was isolated from urine and faeces of sick animals. A direct water-borne route of transmission has been postulated (Kharitonova and Leonov, 1985).

Some viruses are excreted in tick feces, particularly in undigested blood. TBE virus was detected in the faeces of *I. ricinus* (Benda, 1958b), but POW virus was not excreted by *D. andersoni* or *I. pacificus* (Chernesky, 1969). However, there is no evidence of transmission of TBE subgroup viruses to the vertebrate host via contact with contaminated tick fecal material.

3. INTRINSIC FACTORS AFFECTING VIRUS TRANSMISSION

The virus transmission cycle is influenced by several biotic factors that relate to properties of the virus, the tick vector, the vertebrate host, and the interactions thereof. A key element in R_0 (see Section 2) of tick-borne flavivirus infections is the transmissibility of the virus by ticks.

3.1. Vector

Members of the TBE virus subgroup are true arboviruses, relying on biological transmission by the tick vector for survival. The virus is imbibed during feeding, undergoes replication within tick cells, disseminates from the gut to the salivary glands, and is subsequently transmitted in tick saliva when the next tick instar takes a blood-meal (Blaškovič and Reháček, 1962; Balashov, 1972; Nuttall et al., 1994). The ability of a tick to support virus replication and transmit the virus determines the vectorial competence of a particular tick species. Potential restrictions to infection within the tick are the gut infection barrier, gut escape barrier, salivary gland infection barrier, and salivary gland release barrier (Nuttall et al., 1991). Few studies have been undertaken to define these potential barriers to infection by TBE subgroup viruses (Reháček, 1965). Indeed, the wide range of ixodid tick species that can be infected and can transmit TBE subgroup viruses under experimental conditions suggests that most if not all ixodid species are potential vectors of TBE subgroup viruses.

Despite the vector potential of many ixodid ticks for TBE subgroup viruses, only a few primary vectors are apparent. The reasons for this have not been fully defined. Obviously, virus and tick must be sympatric in their distribution. However, this is not the complete story as many competent tick vectors are

found within the geographic range of TBE subgroup viruses but virus transmission cycles (and R_0) depend on comparatively few species. Two parameters are particularly significant: vector efficiency and vertebrate host preferences.

Vector efficiency is the major determinant of vector status. The efficiency of a tick vector is reflected in the infection threshold and the transmission rate. Infection threshold is defined as the lowest amount of virus capable of causing an infection in approximately 1–5% of the vector population (Chamberlain et al., 1954). Thus the lower the infection threshold, the less virus is required to infect the vector and hence the greater the probability of infection of the vector in nature.

Data on the infection thresholds of different tick species for TBE subgroup viruses are limited (Table 12.3). Furthermore, the data are difficult to compare because of differences in the experimental methods employed, the means of expressing the infection threshold, and the tick stage examined. For example, the infection threshold for POW virus was determined by feeding ticks on a rabbit inoculated with a high dose of virus. Direct comparison of *D. andersoni* and *I. pacificus* revealed a 100-fold difference in the susceptibility to POW virus. The infection threshold of *I. cookei*, the principal vector of POW virus, has not been reported. Similarly, the apparent difference in infection thresholds of *I. ricinus* and *I. persulcatus* for TBE virus needs critical examination.

Estimates of infection threshold are affected by the vertebrate host species used in experiments. Initial investigations of the infection threshold of *I. ricinus* for LI virus, in which larvae were fed on laboratory mice, indicated that viraemic titres of 2.0 to greater than 4.0 $\log_{10}$PFU/0.2 ml (2.7 to >4.7 $\log_{10}$PFU/ml) blood were insufficient to establish infection in larval ticks (Beasley et al., 1978). However, using infected chicks, the threshold levels for larvae and nymphs were approximately 4.0 and 3.0 $\log_{10}$PFU/0.2 ml (4.7 and 3.7 $\log_{10}$PFU/ml), respectively (Reid, 1988). This was in agreement with a threshold of 3.2 $\log_{10}$PFU/0.2 ml (3.9 $\log_{10}$PFU/ml) for nymphs fed on viraemic sheep (Swanepoel, 1968, cited by Reid, 1984).

The concept of infection threshold has been challenged by the results of experimental studies demonstrating efficient transmission of Thogoto virus (family, Orthomyxoviridae) between infected and uninfected ticks co-feeding on non-viremic guinea pigs (Jones et al., 1987). In contrast to guinea pigs, hamsters are highly susceptible to Thogoto virus, developing levels of viremia of up to 8.0 $\log_{10}$PFU/ml blood. The 5% infection thresholds of *Rhipicephalus appendiculatus* and *Amblyomma variegatum* determined by feeding nymphs on viremic hamsters were 2.8 and 2.7 $\log_{10}$PFU/ml blood, respectively (Davies et al., 1990). These levels are in sharp contrast to the infection threshold of <1.0 $\log_{10}$PFU/ml blood (i.e., undetectable viremia) determined using guinea pigs (Jones et al., 1990). Experimental studies to elucidate the mechanism of "non-viremic transmission" revealed that a protein, secreted in the saliva of feeding ticks, enhanced virus transmission (Jones et al., 1992a). The saliva-activated transmission (SAT) factor was demonstrated in the salivary glands of competent vectors of the virus but not in a limited number of non-competent vectors (Jones et al., 1992b).

Table 12.3. Threshold of infection

Virus	Tick vector[a]	Vertebrate	Infection threshold (log_{10})[b]		Reference
			Cited	Per ml	
TBE (Far eastern)	*I. persulcatus* (L)	Guinea pig	3.0 LD_{50}/0.03 ml	4.5 LD_{50}	Dumina (1958)
TBE (European)	*I. ricinus* (L)	*Apodemus flavicollis*	2.0 LD_{50}/0.02 ml	3.7 LD_{50}	Radda et al. (1969)
LI	*I. ricinus* (N)	Sheep	2.5 LD_{50}/0.03 ml[c]	4.0 LD_{50}	Swanepoel (1968)
	I. ricinus (N)	Chick	4.0 PFU/0.2 ml	4.7 PFU	Beasley et al. (1978)
	I. ricinus (N)	Chick	3.0 PFU/0.2 ml	3.7 PFU	Beasley et al. (1978)
OHF			? no data		
KFD	*H. spinigera* (L)	Chick	3.0 LD_{50}/0.03 ml	4.5 LD_{50}	Singh and Anderson (1968)
POW	*D. andersoni* (A)	Rabbit	2.5 LD_{50}/ml	2.5 LD_{50}	Chernesky (1969)
	I. pacificus (A)	Rabbit	4.5 LD_{50}/ml	4.5 LD_{50}	Chernesky (1969)

[a] A = adult; N = nymph; L = larva.

[b] Lowest concentration of virus capable of infecting 1–5% of the vector population (Chamberlain et al., 1954). Virus concentration is expressed as 50% mouse lethal dose or number of plaque-forming units in inoculated cell culture, per volume of blood of the infected vertebrate host on which the tick vector fed. The published values and values corrected to per milliliter are shown.

A transmission strategy similar to that observed with Thogoto virus has been reported for TBE virus. The relevance of infection thresholds was questioned in studies of TBE virus infection of *Ixodes persulcatus* that were fed on non-viremic rabbits (Galimov et al., 1989). Similar studies using *I. persulcatus*, *I. ricinus*, *Dermacentor marginatus*, *D. reticulatus*, and *Rhipicephalus appendiculatus*, demonstrated TBE virus transmission between infected and uninfected ticks feeding either in contact with each other, or physically separated on non-viremic guinea pigs (Alekseev and Shunikhin, 1990; Labuda et al., 1993a). "Non-viremic transmission" of TBE virus was shown to be mediated by a factor associated with tick salivary glands (Alekseev et al., 1991; Labuda et al., 1993b). Saliva-activated transmission of TBE virus shows several differences from that of Thogoto virus, probably reflecting different infection strategies of the two viruses in the vertebrate host. Nevertheless, SAT may play an important role in determining vector efficiency of ticks species that transmit TBE subgroup viruses (see Section 3.2).

The latent period between virus acquisition and the time at which the tick becomes infective is known as the extrinsic incubation period (EIP) (Hardy, 1988). During this period the tick is incapable of virus transmission. It is unlikely that EIP is important in the ecology of TBE subgroup viruses because of the relatively long developmental period between tick blood-meals.

Besides the infection threshold (whether apparent or subliminal), the transmission rate is also important in determining vector efficiency. Owing to the long feeding period of tick vectors of TBE subgroup viruses, the true "rate" (i.e., as a function of time) of transmission is probably irrelevant. Of greater significance is the effectiveness of the tick vector in transmitting the virus to the vertebrate host (and thence to other tick vectors). As expected, tick-borne transmission of TBE subgroup viruses appears to be an efficient process when the vertebrate host is susceptible to infection. For example, under experimental conditions, three TBE virus-infected larvae of *I. ricinus* or *I. persulcatus* that fed for 2 days on mice induced encephalitis (Petrishcheva and Levkovitsch, 1949, cited by Blaškovič and Reháček, 1962); one TBE virus-infected female *I. ricinus* was sufficient to transmit the virus to a mouse (Benda, 1958b); and one KFD virus-infected nymph of *Ornithodoros tholozani* that accidentally was allowed to feed on a human transmitted the virus with resulting disease (Bhat and Goverdhan, 1973). Thus it seems likely that one infected tick is sufficient to infect a susceptible host. However, transmission efficiency may vary between tick species and between tick instars, and thus influence the role of a species/stage in the enzootic cycle. Experimental studies demonstrated that nymphs of *I. ricinus* were more efficient than either nymphs or larvae of *H. inermis*, or larvae of *D. reticulatus* (Grešiková and Calisher, 1988). *Haemaphysalis turturis* was shown to be more efficient than either *H. kinneari* or *H. minuta* at maintaining, and subsequently transmitting KFD virus acquired at the larval stage (Singh et al., 1964).

The response of susceptible vertebrate hosts to tick-borne virus transmission depends on their immune status. In nature, infected ticks feed on vertebrate hosts that are immune to the virus and/or develop a resistance response to tick

infestation. Results of experimental studies indicate that the uptake of virus-specific antibodies by infected ticks has no effect on the infection in ticks other than a possible inhibitory effect on vertical transmission (Alexander and Neitz, 1935; Benda, 1958a, Dumina, 1958; Il'enko, 1960; MacLeod, 1962). In a study of tick-host immunity, laboratory animals immunized with uninfected tick salivary glands were protected against infection when exposed to TBE virus-infected ticks, but were susceptible to challenge by syringe inoculation of TBE virus (Votyakov and Mishaeva, 1980; Mishaeva, 1990). However, attempts to confirm this intriguing result were unsuccessful (Labuda, unpublished data). The ability of vertebrate hosts to reject natural tick infestations has not been examined with regard to virus ecology.

In addition to vector efficiency, host selectivity also plays an important role in determining the role of a tick species and instar in the enzootic cycle. The selected vertebrate species may act as maintenance, amplifying, or reservoir hosts of the virus.

3.2. Maintenance and Amplifying Hosts

Certain vertebrate species play a crucial role in maintaining the transmission cycles of TBE subgroup viruses (Table 12.4). In addition, some species may be important in amplifying the number of infected ticks within an infection focus, though their role may be insufficient *per se* to maintain the virus transmission cycle. Such amplifying hosts are highly susceptible to the virus and infection may result in high mortality. In general, however, it is difficult to distinguish between maintenance and amplifying hosts.

The population dynamics of vertebrate hosts are an important factor in determining the number of susceptible maintenance and amplifying hosts that

Table 12.4. Maintenance, amplifying and reservoir hosts

Virus	Maintenance host	Amplifying host	Reservoir host
TBE (Far eastern)	Field mouse		*I. persulcatus*, hedgehog
TBE (European)	Field mouse, mole		*I. ricinus*, hedgehog, dormouse
LI	Sheep	Red grouse	*I. ricinus*
OHF	Water vole, root vole	Muskrat	Water vole, narrow-skulled vole
KFD	Shrew, porcupine	Monkeys, squirrel	*Ixodes* sp.
POW	Woodchuck, grey squirrel		*I. cookei, I. persulcatus*

Apodemus flavicollis, yellow-necked field mouse; *Apodemus sylvaticus*, long-tailed field mouse; *Arvicola terrestris*, water vole; *Erinaceus europaea*, hedgehog; *Funambulus tristriatus*, palm squirrel; *Hystrix indica*, porcupine; *Lagopus scotius*, red grouse; *Macaca radiata*, bonnet macaque monkey; *Marmota monax*, woodchuck; *Microtus gregalis*, narrow-skulled vole; *Microtus oeconomus*, root vole; *Muscardinus avellanarius*, dormouse; *Ondrata zibethica*, muskrat; *Presbytis entellus*, langur monkey; *Sciurus carolinensis*, grey squirrel; *Suncus murinus*, shrew; *Talpa europaea*, mole; *Tamiasciurus hudsonicus*, red squirrel.

contribute to the enzootic or epizootic transmission cycle. Most TBE subgroup viruses utilize rodents as maintenance and amplifying hosts. Such species have high reproductive rates and life spans that are often less than that of the tick vector. Fluctuations in transmission dynamics are correlated with changes in rodent populations. Increases in rodent populations are followed within 1–2 years by increased tick populations and a higher risk of human infections (Kožuch et al., 1990).

At least ten species of rodents have been implicated as maintenace hosts of TBE virus in Central Europe (Cerný, 1976). The most important are probably field mice, *Apodemus flavicollis* and *A. sylvaticus*. They are generally abundant in infection foci and are readily infested with *I. ricinus* ticks (mostly larvae). Bank voles (*Clethrionomys glareolus*) are also considered to be important hosts because of their abundance, although viremia in adult *C. glareolus* was found to be below the infection threshold for ticks (Ernek et al., 1963; Málková et al., 1965). However, a wide range of viremic titres (0.4–5.4 $\log_{10}LD_{50}$/0.03 ml or 1.1–6.0 $\log_{10}LD_{50}$/ml) was detected in adult *C. glareolus* infected with different TBE virus isolates (Chunikhin and Kurenkov, 1979). During a longitudinal study (1981–1986) of natural foci of TBE virus in West Slovakia, the importance of the most abundant rodent species, *C. glareolus* (52.9%) and *A. flavicollis* (22.5%), was demonstrated (Kozuch et al., 1990). The highest prevalence of neutralizing antibody to the virus was found in sera of *A. flavicollis* (18.1%), followed by *C. glareolus* (15.1%). The European mole (*Talpa europaea*) is also an important host of *I. ricinus* and is naturally infected by TBE virus in Czechoslovakia. Grylich (1960) stressed the need to include this mole–parasite relationship in studies of the ecology and epidemiology of TBE.

A problem in assessing the relative roles of different host species in the transmission cycles of arthropod-borne viruses is that studies are based largely on artificial infection of vertebrate hosts by syringe inoculation. A recent investigation mimicked natural conditions of transmission by allowing TBE virus-infected and uninfected *I. ricinus* ticks to feed together on wild vertebrate hosts (field mice, *A. flavicollis* and *A. agrarius*; bank vole; pine vole, *Pitymys subterraneus*; hedgehog, *Erinaceus europaeus*; and pheasant, *Phaseanus colchicus*) (Labuda et al., 1993c). The greatest numbers of infected ticks were obtained from field mice, susceptible hosts that had undetectable or very low levels of viremia. By contrast, fewer infected ticks were obtained from hosts that had significant (bank voles) or substantial (pine voles) levels of viremia. The results suggest that "non-viremic transmission" (see Section 3.1) plays an important role in the ecology of TBE virus.

The role of birds in the ecology of TBE viruses has not been resolved. Birds belonging to more than 100 species have been associated with infections of the Far Eastern subtype, as determined by serology or virus isolations (Naumov et al., 1963; Hoogstraal, 1966). A serological study of the fledglings of female thrushes (*Turdus pilaris*) with high antibody levels of TBE virus (Far Eastern subtype) showed that one third of the progeny had received antibodies to the virus transovarially from the parent (Lvov and Naumov, 1962). In a disease focus in former Czechoslovakia, sera from birds of 15/20 species were positive

and pheasants were identified as important hosts of the European subtype (Aśmera et al., 1962). However, when TBE virus-infected and uninfected *I. ricinus* ticks experimentally were fed together on pheasants, none of the uninfected ticks became infected (Labuda et al., 1993c). TBE virus was recovered from mallards (*Anas platyrhynchos*) following either virus inoculation or exposure to infected *I. ricinus* nymphs (Ernek et al., 1969). Some of these viremic ducks did not develop antibody to TBE virus and, conversely, some produced antibody but had no detectable viremia. Considering the seemingly limited opportunities for tick infestation of ducks, the ecological significance of these results is unclear.

Experimental and epizootiological studies indicate that sheep are the only significantmaintenace host of LI virus (Reid, 1984, 1988). They consistently exhibited viremia above the infection threshold for ticks during a period of 2–3 days after virus inoculation. Sheep develop complete immunity to LI virus and consequently experience only one active infection with LI virus during their lifetime. Considering the turnover in sheep populations per year, the minimum proportion of sheep susceptible in each spring to LI virus infection is 1/5 of the adult flock which, in Scotland, represents about 0.5 million sheep. In contrast to domestic species, only two of eight native mammals developed viremic tirers that exceeded the threshold: in five of 59 field voles (*Microtus agrestris*) the titer exceeded 10^4 PFU/0.2 ml, and one of three roe deer (*Capreolus capreolus*) had a titer of $10^{3.2}$ PFU/0.2 ml on one day only (Reid, 1988). Red grouse were highly susceptible to infection but showed high mortality; they may act as amplifying rather than maintenance hosts of LI virus. Thus, only sheep were considered to be important in virus transmission, based on artificial infection studies. However, attempts to control LI in grouse by immunizing sheep were not entirely successful suggesting that sheep are not the sole hosts involved in maintaining LI virus infections in nature (Hudson, 1992).

In contrast to LI virus, there is good evidence that the ecology of OHF virus provides for an interplay between different vertebrate host species—muskrats and voles—important in maintaining the enzootic cycle (Kharitonova dn Leonov, 1985). The connecting link is the water vole, a species characterized by high numbers, mobility, and ecological adaptability. At different times of the year, water voles occupy lake-marsh or dry valley habitats, thereby bringing them into contact with a broad spectrum of animals susceptible to OHF virus infection. The infection in muskrats is characterized by high mortality rates (*c.* 80%), indicating an amplifying rather than a maintenance role for this host.

KFD virus has been isolated from a variety of small rodents, squirrels, and shrews that are hosts of immature stages of ticks in Kyasanur forest (Banjeree, 1988). Following experimental infection, high titers of circulating virus were detected in squirrels (*Funambulus tristriatus* and *Petaurista philippensis*), the majority of which died. In contrast, virus titers in rodents (*Rattus r. wronghtoni* and *R. blandfordi*) were low and disease was not evident (Webb, 1965; Boshell et al., 1968; Bhat et al., 1979). Thus it was postulated that these species play different roles in the ecology of KFD virus (Hoogstraal, 1966). Monkeys (*Presbytis entellus* and *Macaca radiata*) are highly susceptible to KFD virus

infection and show a high level of mortality (Work, 1958; Webb and Burston, 1966). Like squirrels, they are more likely to be important as amplifying rather than maintenance hosts. The role of birds in the ecology of KFD virus appears primarily as hosts of immature tick stages with the possible exception of ground birds, which may serve as maintenance hosts (Boshell, 1969). High levels of viremia were detected in red spur fowl (*Galloperdix spadicea*) and jungle fowl (*Gallus sonneratii*), and the virus was isolated from a pool of nymphal *H. spinigera* collected from spur fowls (Rodrigues, 1968).

Cattle infected with KFD virus do not develop significant levels of viremia, and show no signs of disease. Before 1970, goats and sheep were not considered important in the endemic area. Since 1970, however, goats have been introduced in the KFD area in substantial numbers. All stages of *H. intermedia* parasitize goats, and the larvae and nymphs also parasitize small mammals.

Special mention must be made of the porcupine (*Hystrix indica*) which is a common host of immature stages of *H. spinigera*, all the stages of *H. turturis* and *H. kyasanurensis*, and immature stages of *Dermacentor auratus*, *Amblyomma integrum*, and *A. javanense*. Porcupines are known to be present in the KFD area in significant numbers and are infected with different stages of ticks in large numbers; they develop high viremic titers for prolonged periods. Porcupines appear to be an ideal host for maintaining the transmission cycle and disseminating virus-infected ticks on the forest floor (Bhat et al., 1976).

The role of seven wild vertebrate species as amplifying hosts of POW virus was tested by experimental inoculation (Kokernot et al., 1969; Timoney, 1971; Zarnke and Yuill, 1981). Opossum (*Didelphis marsupialis*) and striped skunk (*Mephitis mephitis*) developed only trace levels of viremia; in snowshoe hare (*Lepus americanus*), woodchuck (*Marmota monax*), grey squirrel (*Sciurus carolinensis*), grey fox (*Urocyon cinereoargenteus*), and red fox (*Vulpes vulpes*), the viremia was 0.6–2.5 $\log_{10}$/0.03 ml (1.2–4.0 $\log_{10}$/ml). The viremia in woodchuck and grey squirrel was prolonged, lasting 8–11 days, and might compensate for low virus titers (together with the prolonged feeding period of ticks).

As noted by Hoogstraal (1966), serological studies reveal that many vertebrate species are exposed to viruses in the TBE subgroup but virus isolations from most of them are comparatively rare. Are these simply "wasted" infections that do not contribute to R_0, or can they involve maintenace hosts that play a role in "non-viremic transmission"?

Investigations to identify maintenance and reservoir hosts are based on identifying species that develop levels of viremia above the threshold level determined by classical "viremic transmission". The demonstration of efficient virus transmission at subliminal threshold levels (see above and Section 3.1) poses serious questions for this premise. Indeed, the potential number of maintenance hosts is dramatically increased if, in fact, efficient virus transmission occurs when tick vectors feed together on vertebrate hosts having sub-threshold levels of viremia. Prime candidates for the role of non-viremic maintenance host are species that are prevalent within an infection focus and have a high prevalence of antibody to the virus. For example, *Rattus wroughtoni* is the most abundant rodent in the region of KFD and a high proportion of

Table 12.5. Potential "non-viremic" maintenance hosts

Virus	Vertebrate	Comments
TBE (Far Eastern)	Numerous avian species	High antibody prevalence but resistant to experimental infection
TBE (European)	Fieldmice	Demonstrated experimentally[a]
LI	Hare	Host of larvae and nymphs; sub-threshold viremia
OHF	?	
KFD	Rat	Low viremia, virus isolation, high antibody prevalence
POW	Oppossum, striped skunk	Antibody prevalence; sub-threshold viraemia

Clethrionomys glareolus, bank vole; *Didelphis marsupalis*, oppossum; *Lepus timidus*, hare; *Mephitis mephitis*, striped skunk; *Rattus wrongtoni*, white-bellied rat.
[a]Labuda et al. (1993c).

animals are seropositive for KFD virus, although experimental studies demonstrated low or undetectable levels of viremia (Boshell, 1969; Sreenivasan, personal communication). Examples of vertebrate species that are frequently exposed to virus-infected ticks, but are not regarded as virus hosts because they dl not develop significant levels of viremia, are shown in Table 12.5. These should be re-examined as potential hosts of "non-viremic transmission".

3.3. Reservoir Host

The reservoir is defined here as any host (vertebtate or tick) that maintains the virus during conditions that preclude active virus circulation, e.g, winter or monsoon periods. For TBE subgroup viruses, reservoirs can be more readily identified among the tick vector populations than among the vertebrate hosts of the viruses (Table 12.4).

Vertebrate species that act as reservoirs are typically those in which the virus establishes a chronic long-lasting infection. In the circulation of TBE virus, insectivores (shrews, moles, hedgehogs), which have relatively stable populations in contrast to rodents, are believed to be important reservoir hosts (Grulich, 1960; Kožuch et al., 1967). Imperfect homeotherms, such as hedgehogs and dormice (*Muscardinus avellanarius*), may be particularly important. Experimental studies demonstrated that, in infected hedgehogs and dormice, TBE virus (European subtype) persisted during a 29-day period of hibernation, and the animals then developed viremia during the 8-day period post-hibernation (Kožuch et al., 1963). TBE virus also persists and replicates in various bat species during and after hibernation (Nosek et al., 1961). Persistent infection of voles (*Clethrionomys glareolus*) may also allow them to serve as a reservoir. TBE virus was detected in the organs of voles 21 and 28 days after experimental infection (Ernek et al., 1963). Moreover, in field studies, TBE virus was isolated

from the brain or lung and liver of two voles and one yellow-necked mouse (*Apodemus flavicollis*) captured in mid-winter when ticks were not active (Kožuch et al., 1990). None of these animals had antibody to TBE virus. It remains to be determined whether ticks would become infected when feeding on such animals in the spring.

OHF virus has been repeatedly isolated from the kidneys of narrow-skulled voles (Lvov, 1988). Chronic infection of this species may provide a reservoir mechanism for the virus during adverse conditions. Experimental induction of chronic infections in water voles led Kharitonova and Leonov (1985) to postulate that these hosts serve as reservoirs of the virus in which latent infections are activated by ecological changes associated with adverse conditions. Such changes occur in early spring, and late summer and autumn, when water voles experience cooling during their migration from marshy areas to drier habitats, in which the plant food is less nutritious. Hoogstraal (1980) considered that birds nesting in and near water serve as reservoirs of OHF virus.

Vertical transmission from one tick vector generation to the next provides a potential reservoir mechanism for the long-term persistence of viral infections. Although vertical transmission has been demonstrated for all members of the TBE subgroup except LI and POW viruses (Table 12.6), the ecological significance of this transmission mechanism has not been determined (see Section 2.1). Chumakov (1944) (cited by Blaškovič and Reháček, 1962) reported

Table 12.6. Vertical transmission[a]

Virus	Tick	Reference
TBE (Far Eastern)	*Ixodes persulcatus*	Chumakov (1944), Smorodintsev (1958), Il'enko et al. (1970), Kondrashova and Filippovets (1970)
	Ixodes ricinus	Chumakov et al. (1945)
	Dermacentor nuttalli	Chumakov et al. (1945)
	Hyalomma dromedarii	Chumakov et al. (1945)
	Hyalomma asciaticum	Chumakov et al (1945)
	Dermacentor silvarum	Skrynnik and Ryijov (1941)
	Haemaphysalis concinna	Kozlova and Soloviev (1941), Pavlovsky and Soloviev (1963)
	Haemaphysalis japonica	Tatarinova (1961)
TBE (European)	*Ixodes ricinus*	Benda (1958a), Reháček (1962), Il'enko et al. (1970)
	Ixodes hexagonus	Streissle (1960)
LI	*Ixodes ricinus*[b]	Stockman (1918)
OHF	*Dermacentor reticulatus*	Avakian and Lebedev (1955)
KFD	*Ixodes petauristae*	Singh et al. (1968a)
	Argas persicus	Singh et al. (1971)
	Ornithodoros tholozani	Bhat and Goverdhan (1973)
POW	Not demonstrated	

[a] Vertical transmission is the passage of virus from one tick generation to the next and includes transovarial transmission.

[b] Not confirmed in subsequent studies.

that TBE virus persisted in *I. persulcatus* for 26 months through three tick generations.

The virus titer in the female adult tick determines the maternal vertical transmission rate under experimental conditions: in experimental studies, significantly higher levels of maternal transmission occurred when females were exposed to high virus titers prior to oviposition, rather than when females were infected at a preceding instar (Benda, 1958a; Kondrashova and Filippovets, 1970; Korenberg and Pchelkina, 1984). If this relationship applies in nature, higher frequencies of vertical transmission may be expected during epizootics when ticks (particularly adults) feed on vertebrate hosts that have high viremic titers. In this respect, the lack of evidence for vertical transmission of LI virus is curious, given that adults ticks are likely to feed on sheep that have substantial levels of viremia.

In conjunction with vertical transmission, the clumped distribution of questing larval ticks (which increases the likelihood that several larvae attach and feed together on the same host) may be of ecological significance. Experimental studies with KFD virus demonstrated that the prevalence of ticks infected vertically is greater among engorged F_1 larvae and nymphs than in unfed F_1 larvae (*vide infra*). Thus the co-feeding of larvae may amplify the filial infection rate. This possibility was demonstrated by experimental studies involving co-feeding of TBE virus-infected and uninfected larvae (Labuda et al., 1993d). Uninfected larvae were fed on infected hosts and then cohorts of the emergent nymphs were fed on uninfected hosts. Infectious virus was not detected in the larvae (although a small proportion of larvae were infected as determined by use of the polymerase chain reaction) but 79% of fed nymphs from one cohort were infected. The results indicated that TBE virus was transmitted from a small number of infected nymphs (infected as larvae) to uninfected nymphs as they fed together on an uninfected animal.

Even in the absence of vertical transmission, the long-term survival of viruses in their tick vectors can be a significant maintenance mechanism in which the tick acts as a virus reservoir (Reháček, 1965). The European subtype of TBE virus persisted in *I. ricinus* for at least 9 months in experimentally infected starving females maintained at either room temperature or 4°C (Benda, 1958a), and in fed larvae for 102 days maintained under natural overwintering conditions (Reháček, 1960).

The candidate reservoir host of LI virus is the single tick vector species, *I. ricinus*. However, this is difficult to equate with the view that the tick is an inefficient vector of the virus and transovarial transmission of LI virus in *I. ricinus* does not occur (Reid, 1988). Transovarial transmission of OHF virus by its principal vector, *Dermacentor reticulatus*, has been documented, though the rate of vertical transmission is low and is considered to be inefficient as a reservoir mechanism (Lvov, 1988).

Maintenance of KFD virus through the monsoon period poses a problem similar to that of overwintering in temperate zones. Epizootics in monkeys and epidemics affecting humans correspond with the nymphal season of *Haemaphysalis* ticks but only a few infected nymphs were found in the monsoon,

larvae of *Ixodes* species are prevalent and may, together with small mammals (rodents and shrews), maintain the virus. Thus, an important component of the transmission cycle of KFD virus may be the overlap in activity of *Haemaphysalis* and *Ixodes* ticks, and sharing of susceptible hosts (Boshell, 1969). Vertical transmission of KFD virus may also provide a mechanism for maintaining the virus transmission cycle. Although evidence for transovarial transmission by the primary vector *H. spinigera* is inconclusive (Singh et al., 1963; Singh, personal communication cited by Burgdorfer and Varma, 1967), several other species have been shown to transmit the virus transovarially (Table 12.6), and with relatively high rates of maternal transmission and filial infection (Singh et al., 1968a, 1971). Interestingly, in experimental studies, only 5/38 pools of 50 *Argas persicus* larvae showed evidence of filial infection before feeding whereas 13/13 pools of larvae were found to be infected after feeding on uninfected chicks (Singh et al., 1971). The authors concluded that uninfected larvae had become infected as they fed together with infected larvae on chicks that became viremic. However, as circulating virus was detected in only 4/7 chicks, the data suggest to us that "non-viremic transmission" of KFD virus had occurred.

The reservoir hosts of POW virus have not been identified. The virus may overwinter in larval, nymphal, and adult stages of *Ixodes cookei* (Ko, 1972). Timoney (1971) postulated that chronic infection of the kidney of grey squirrels may provide a mechanism for persistence of the virus.

4. EXTRINSIC FACTORS AFFECTING VIRUS TRANSMISSION

Extrinsic factors influencing the virus transmission cycle are essentially properties of the environment in which the virus circulates and is maintained. Ticks spend more than 90% of their life off the host, hence the most important extrinsic factors relate to the stresses experienced by ticks during this period. Body-water homeostasis is one of the key processes determining tick off-host survival (Needham and Teel, 1991). Ticks are well adapted to controlling their water balance, surviving longer than any other arthropod without food or drinking water. Not surprisingly, the habitat in which members of the TBE virus subgroup are found, and the seasonal dynamics of virus transmission, are strongly influenced by the off-host ecology of the tick vectors.

4.1. Habitat

Hoogstraal (1973) distinguished two distinct ecologically determined types of habitat–tick–vertebrate host relationships: "restricted habitats" that involve shelter-seeking ticks adapted to feeding throughout their life cycle on animals inhabiting burrows, trees, caves, etc.; and "generalized habitats" in which the adult ticks feed on wandering hosts that are usually larger and distinct from

the small or medium-sized hosts parasitized by the immature stages. The ecology of TBE subgroup viruses falls into the latter category.

Foci of TBE virus (Far Eastern subtype) are found within the geographic distribution of *I. persulcatus*. They occur mostly in the taiga landscape which consists of mixed broad-leafed forests of the Manchurian type where the humidity is very high, and in coniferous forests with predominately Okhots conifers. Sporadic foci occur in river valleys covered with marshy meadows, and on undulating plains where suitable habitats overgrown with bushes are scattered among cultivated valleys.

Foci of the European TBE viral subtype occur in various natural or man-made habitats within the distribution of the primary vector, *I. ricinus*; they can vary significantly. For example, in former Czechoslovakia numerous foci of TBE virus are found in three geographically distinct regions: hercynian, in Northern Moravia and Bohemia, occurring also in Germany and Austria; carpathian, formed by the Carpathian Mountains and adjacent territories, and covering most of Slovakia; and pannonian, occupying most of the southern part of Slovakia and continuing into Hungary (Nosek et al., 1968).

A mean annual rainfall of about 800 mm and an annual mean temperature of 8°C are characteristics of TBE endemic regions. The non-parasitic phases of *I. ricinus* do not survive in areas where the relative humidity of their microclimate falls below 80% for prolonged periods, or in areas prone to flooding (Gray, 1991). Favorable microclimatic factors for TBE virus circulation are frequently encountered in thermophilic growths within mixed oak/black-locust forests or in swampy uninundated areas of the Danube River basis. These requirements provide an explanation for the occurrence of *I. ricinus* mainly in deciduous woodland containing small mammals and deer, and in meadows and moorland where there is high rainfall, and there are sufficient livestock to support the adult females (Milne, 1949b).

Louping ill virus is mainly restricted to the rough upland grazings and unimproved pastures of the western seaboad of the UK that are largely devoted to sheep farming, although the disease is also found in some areas of northeast Scotland and England. In the most common habitats there is a thick mat of vegetation and the soil remains damp throughout the summer. A comparison of two hill pastures in Ayrshire, Scotland, revealed a 2–5-fold increase in the seroconversion rate of sheep maintained on a pasture that had large areas of swampy ground with thick grass and rushes compared with one that was sloping and well-drained (Smith et al., 1964; Smith and Varma, 1981). The increase of LI virus infections of grouse in northern England has been associated with the spread of bracken, which provides a thick vegetation mat (Hudson and Rands, 1988).

Epidemiological data for the CIS have revealed ecological correlations between zones of TBE and OHF activity (Lvov, 1988). In the southern taiga, more than 90% of TBE cases have been registered and none of OHF. Most cases of OHF have been recorded in the northern and southern forest-steppes where there are comparatively few records of TBE. In the interface zone between the southern taiga and northern forest-steppe, only about 1% of cases of either

disease have been registered, and in part of the southern steppe there have been a few cases of TBE and none of OHF. Typical landscapes associated with OHF virus comprise forest-steppe with numerous swamps and a wide network of lakes covered with dense reed (Kharitonova and Leonov, 1985). This region represents one of the youngest landscape zones in western Siberia, having replaced the southern taiga landscape (Lvov, 1988).

The activity of KFD virus is restricted to a relatively small area in Karnataka State, India. The area, undulated rather than hilly, is situated on the eastern slopes of the Western Ghats Mountains at an elevation of approximately 600–700 m above sea level. Originally, the vegetation was tall rain forest interspersed with deciduous forest and with heavy undergrowth of bushes. The area has been extensively developed to provide numerous clearings for villages. First reports of KFD were limited to within 100 km^2 of Sagar and Sorab taluks (sub-districts) of the Shimoga district. During subsequent years the disease area has expanded significantly. Newly identified foci have been reported, some in regions not contiguous with established foci. Usually the recognition of new foci is associated with deforestation for agricultural development (Banerjee, 1988).

Typical habitats associated with POW virus are both "generalized" and "restricted" (Artsob, 1988). The former are associated with a variety of tick vectors and vertebrate hosts of the virus, including birds in the CIS. In contrast, the den of woodchucks provides a restricted habitat for the transmission cycle involving *Ixodes cookei* (Ko, 1972).

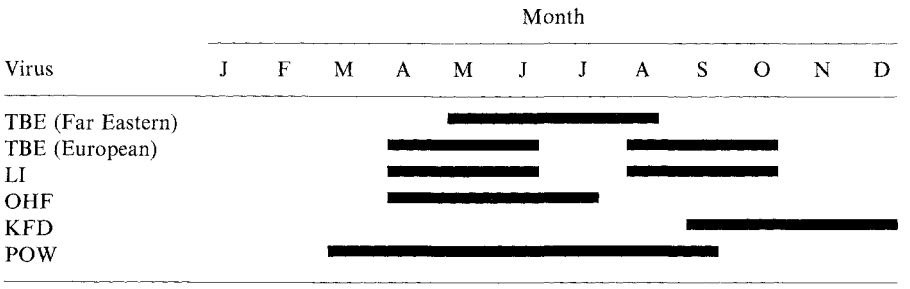
The effect of habitat conditions on virus development in ticks generally has been overlooked. Microclimatic factors were shown to influence the rate of development significantly and also the susceptibility of *I. ricinus* ticks to TBE virus infection. Relative humidity had a direct effect on the infection rate of ticks whereas environmental temperature had no apparent effect (Danielova et al., 1983). The overall results indicated that intrinsic rather than extrinsic factors have a greater influence on TBE virus infection of ticks (Danielova, 1990).

4.2. Seasonality

As in insects, the development of many ticks is synchronized through diapause with seasonal climatic changes (Balashov, 1972; Belozerov, 1982). Thus seasonality plays a major role in virus ecology. Comparison of the seasonal dynamics of TBE subgroup viruses reveals striking differences that are directly related to the activities of the primary tick vectors (Table 12.7).

Diapause, the most important regulator of tick seasonal activity, appears as delayed metamorphosis of engorged larvae and nymphs of *I. ricinus* and *I. persulcatus*, and as interrupted oogenesis in *Dermacentor* females that engorged in summer and autumn. In unfed *Dermacentor* adults, and to a lesser degree in *I. ricinus* nymphs, the so-called behavioral diapause is manifested by decreased questing activity (Balashov, 1972). Whereas morphogenic diapause is accepted, there is some controversy about behavioral diapause (Gray, 1991). Thus, for

Table 12.7. Typical seasonal activity of TBE subgroup viruses



example, it is disputed whether the inactivity of unfed *I. ricinus* is merely a form of quiescence.

The nature of the habitat is very important with regard to seasonality. In exposed areas such as meadows, ticks become activated more quickly, especially in sunshine, and individuals and high humidity, as in dense woodland, ticks may quest for several months (Gray, 1991). Photoperiod, which is dependent on latitude, influences the commencement of development and the timing of activity through diapause mechanisms (Belozerov, 1982). Photoperiod is the dominant entrainment stimulus but temperature has an important secondary influence. Ambient temperature is a good general predictor of tick host-seeking activity (Harlan and Foster, 1990).

The seasonal activity of *I. persulcatus* in Karelia, CIS, commences with adults attacking cattle from late April to July; larvae and nymphs parasitize small mammals throughout June–July (Balashov, 1972). In most parts of its range, *I. ricinus* becomes active and feeds on hosts in spring and early summer, with ticks occurring on vegetation and animals from late March onwards. Depending on environmental conditions, *I. ricinus* shows a bimodal activity pattern with spring and autumn peaks, or a unimodal pattern (Steele and Randolph, 1985; Gray, 1991).

The seasonal incidence of clinical LI closely follows the seasonal pattern of *I. ricinus*. The tick becomes active in the late spring and early summer, followed by a period of inactivity when metamorphosis of engorged larvae and nymphs occurs, and females deposit eggs. Depending on latitude and local climate, *I. ricinus* may become active again in August to October. Commencement of tick activity in the spring was found to correlate with an average day temperature exceeding 7°C (MacLeod, 1939). This threshold temperature is reached in early April in the southwest and in mid-June in the northeast highlands above 300 m. However, it is now accepted that diapause mechanisms rather than environmental temperature govern the seasonal rhythms of tick activity (Belozerov, 1982).

Unlike the *Ixodes* vectors of TBE and LI viruses, the primary vector of OHF virus (*Dermacentor reticulatus*) has a 1-year life cycle. Hibernation and an adult spring wave of parasitism are characteristic of this species.

Spring-engorged females soon oviposit and peak larval parasitism begins in June. Engorged larvae rapidly molt to nymphs, which parasitize rodents at the end of July and begin molting into adults in August. Unfed larvae and nymphs of this species are unable to survive for long periods in nature, whereas the adult stages show exceptional longevity, surviving 3–4 years in the absence of hosts. Consequently, active spring and autumn populations consist of at least three generations. Seasonal activity of the 1-year cycle is ensured by summer inactivity of unfed females and by diapause in engorged females. Thus inactivity and diapause prevent the appearance of immature stages incapable of overwintering (Balashov, 1972).

The seasonal activity of ticks in the KFD area is dictated by the monsoon season, from June to September (Boshell, 1969). The build-up in larval population of *H. spinigera* starts in September, at the end of the rains, remains high through October and November, and starts diminishing in December; there is a small second peak in March. The nymphs appear on the forest floor as well as on captured animals in December and January, and reach their peak population in February and March. The nymphal population declines in April and May, with activity ceasing when the rains begin in mid-June. Adults increase in numbers as the nymphs decrease and reach a peak during the rainy months, but the activity continues up to the dry season in January to February. The activity of *Ixodes* roughly alternates with that of *Haemaphysalis*. *Ixodes petauristae* larvae appear at the beginning of the rains in mid-June and nymphs peak in August–September. Hence *I. petauristae* may provide a reservoir for the enzootic *H. spinigera*–KFD transmission cycle.

The seasonal activity of POW virus varies according to the different ecologies of the virus. Late summer is the time of maximal virus spread in red squirrels in Northern Ontario, but active infection of woodchucks occurs during spring and summer, and of snowshoe hares and marmots in late spring (Artsob, 1988).

As with habitat conditions, there have been few studies on the effect of diapause on virus development in ticks. The effect of diapause of *I. ricinus* ticks on replication of TBE virus was studied by Mishaev and Erofeeva (1979). The virus replicated more intensively in ticks developing without diapause under laboratory conditions. In contrast, comparison of virus replication and persistence in *I. ricinus*-infected larvae maintained under field or laboratory conditions demonstrated higher virus titers, and longer survival in the field-maintained specimens (Kožuch and Nosek, 1985).

5. VIRUS DISSEMINATION

Dissemination is the spread and subsequent establishment of viruses in new locations. As already mentioned, members of the TBE virus subgroup are associated with "generalized habitats" in which large mammals and birds act as disseminators of virus-infected ticks.

5.1. Transportation

Within their known geographical limits, TBE viruses are disseminated by large mammals and birds, either naturally or during the movements of livestock. For example, the driving of sheep from Bosnia to winter pastures in northeastern Croatia was associated with dissemination of TBE virus (Vesenjak-Hirjan and Sooš, 1976). Despite numerous investigations of birds captured during their migrations between Europe, Asia, and Africa, TBE subgroup viruses have not been isolated from either migrating birds or ticks carried by them (Schmidt and Shope, 1971; Balducci et al., 1973).

Reports of LI-like disease and LI virus outside the UK and Ireland—in Norway, Spain, Turkey, and Bulgaria—suggest virus dissemination (Reid, 1988). Possibly the virus has been transported via sheep. This assumes that LI virus evolved from a European subtype of TBE virus, possibly as a result of ecological isolation following the separation of the British Isles from continental Europe. However, genetic studies are needed to determine if the viruses occurring outside the UK and Ireland are indeed LI virus. Sequence analyses to date have shown that Turkish encephalitis virus is a distinct member of the TBE subgroup (Gao et al., 1993a; Whitby et al., 1993) whereas sheep encephalomyelitis in Norway is caused by LI virus (Gao et al., 1993b).

Dissemination of OHF virus by *D. reticulatus* occurs during population explosions of the narrow-skulled vole (Lvov, 1988). The tendency of water voles to migrate from wet habitats to dry habitats for overwintering also acts as a dispersal mechanism for OHF virus. Infections of several avian species, particularly waterbirds such as coot (*Fulica atra*) and gadwall (*Anas strepera*), may provide a mechanism of virus dissemination, although there is no reported evidence to support this.

KFD virus is disseminated comparatively short distances via infected ticks carried by monkeys or birds. This may enable the establishment of secondary infection foci by spread from a primary "hot spot" (Boshell, 1969).

The enzootic cycle of POW virus involving groundhogs and *Ixodes cookei* provides little opportunity for virus dissemination other than in the spring when interburrow activities are highest as males seek out females (Ko, 1972). Resident birds, and many ducks and other birds migrating from China and Southeast Asia into the Primorskiy forests in the far eastern CIS, were reported to have high antibody titers for POW virus (Hoogstraal, 1980). However, there is no epidemiological or epizootiological evidence to indicate that POW virus is disseminated in east Asia by birds.

5.2. Changes in Virus Distribution

The distribution of TBE viruses is well established and, at least in Europe, there is no evidence that the size and location of endemic areas of TBE have changed significantly during the 1980s (WHO, 1986). However, the political

upheavals in Europe and the former USSR will undoubtedly have significant impact on agricultrual practices and land use, movements of livestock and people, and recreational activities. Such changes may well affect the distribution and prevalence of TBE, as illustrated by LI and KFD.

For both LI and KFD viruses, changes in agricultural practices have resulted in increased host availability for the tick vector and a consequent amplification of virus infection at the population level. Land usage in current LI enzootic areas, particularly in Scotland, has changed markedly over the last two centuries. Formerly, LI was scarcely known in the highlands of Scotland. The introduction of extensive sheep farming not only increased the abundance of *I. ricinus* within suitable habitats, but resulted in dissemination of LI virus. It has been suggested that this spread introduced LI virus to the heather (*Caluna vulgaris*) dominated moorland habitat of grouse, and hence that the host–parasite relationship between grouse and LI virus is a comparatively recent one (Hudson, 1986). In contrast, the resistance to LI virus in forest species of birds (*Phaseanus colchicus*, *Tetrao urogalus*, *T. tetrix*) may reflect their exposure to LI or a related virus (Reid, 1984). Thus, while the ancestral origins of LI virus may have been the forest habitat, the current ecology of the virus is dictated by agricultural practices with domestic sheep representing the single essential vertebrate species (Reid, 1988).

The potential for change in the distribution of KFD is of particular concern, given the uncertainties as to the origin and current distribution of the virus. Was the virus imported, or are epizootics (and epidemics) the result of changes in transmission dynamics of an indigenous, enzootic cycle (i.e., an increase in R_0)? Confirmed reports of KFD do not extend back beyond 1956, suggesting that the virus may have been imported. However, examination of thousands of migratory birds, and imported sheep and goats, provided no evidence of transport of relevant tick species (Boshell, 1969). Indeed, epidemics of KFD are commonly associated with reclamation of forest areas for agriculture and human habitation. The evidence suggests that KFD virus circulates in a long-established enzootic cycle, the equilibrium of which is upset by human intrusion. In particular, the introduction of livestock into enzootic areas results in an increase in the population of *H. spinigera* (Boshell, 1969). A direct relationship has been established between the prevalence of KFD virus-infected *H. spinigera* and the number of human cases of the disease (Banerjee and Bhat, 1977).

In addition to political and agricultural changes, the future distribution of TBE subgroup viruses may be affected by changes in global climate. Several models have been developed to describe the relationships between climate and the prevalence and distribution of ticks and tick-borne diseases (Sutherst and Maywald, 1985; Gardiner and Gray, 1986; Gettinby and Byrom, 1989; Randolph, 1994). Obviously, predictions resulting from such models can only be as good as the ecological data on which they are based.

6. CONCLUSIONS

The final section of this chapter is better entitled conundrums rather than conclusions as several aspects of the comparative ecology of TBE subgroup viruses have raised important questions.

This chapter began with an observation by Hoogstraal (1966) that our knowledge of the ecology of TBE viruses provided a fundamental understanding of vector-borne diseases. But nearly 20 years later, Korenberg and Pchelkina (1984) concluded that "Infected and uninfected specimens occur among unfed ticks in all natural TBE foci. They may feed separately or together on vertebrate animals during the period of viremia or in its absence.... These phenomena, which are very important for perception of existence patterns of a virus population, are virtually unstudied".

The ecology of viruses in the TBE subgroup is in many respects an enigma. Enzootic transmission is driven by the ecological dynamics of the vector tick species and yet the infection prevalences in tick vectors are generally low; vector efficiency is considered by several authors to be inefficient, and vertical transmission in ticks occurs at low frequency, if at all, and usually is discounted as a significant ecological factor. Added to these considerations is the observation that parasites are unequally distributed among their hosts (they show a negative binomial distribution), a fact that has not been adequately investigated in the ecology of tick-borne diseases. How then do these viruses survive in nature?

Korenberg (1974) attacked some of the basic dogmas of TBE ecology. The most striking conclusion was that TBE virus infection does not induce life-long immunity. An important consequence of this claim is the R_0 of TBE viruses is not dependent on small mammal populations that have a high turnover, but that larger mammals can be repeatedly infected; hence, the distribution of enzootic foci is independent of the population dynamics of small mammals. This claim needs to be substantiated as it has important implications for TBE virus ecology and for disease control if true.

Notwithstanding the question of immunity to TBE viruses, the observation of Korenberg and Pchelkina (1984) (*vide supra*) may hold the key to the question—how do these viruses survive in nature (i.e, how do they maintain $R_0 < 1$)? Thus, in natural foci, infected and uninfected ticks may feed separately or together on viremic or non-viremic hosts. This complex feature of tick-borne virus ecology has been largely overlooked. Experimental and field-based studies have simplified the inter-relationships between virus–tick–host to a level that may provide answers that are not relevant to the natural ecosystem.

Obviously a tick is a more efficient virus transmitter than a needle and syringe. What is suprising is the usual assumption that experimental results based on syringe inoculation can be directly extrapolated to the field. Recent studies question the validity of this assumption. Comparisons of infection via tick bite with syringe inoculation of TBE virus clearly show that the former method of virus transmission is much more efficient and can proceed without the involvement of an overt viremia (Labuda et al., 1993a, 1993b, 1993c, 1993d).

It seems that tick-borne viruses can survive in nature because their tick vectors are far more efficient than is generally recognized. The challenge now is to define precisely the nature of vector efficiency and, in the light of the results, to re-examine the ecology of TBE subgroup viruses, including the role of virus- and tick-host immunity.

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8. REFERENCES

Alekseev, A.N. and Chunikhin, S.P. (1990) Exchange of the tickborne encephalitis virus between Ixodidae simultaneously feeding on the animals with subthreshold levels of viremia. *Med. Parazitol.* **2**, 48–50.

Alekseev, A.N., Chunikhin, S.P., Rukhkyan, M. Ya., and Stefutkina, L.F. (1991) Possible role of Ixodoidea salivary gland substrate as an adjuvant enhancing the arbovirus transmission. *Med. Parazitol.* **1**, 28–31.

Alexander, R.A. and Neitz, W.O. (1933) The transmission of louping-ill of sheep by ticks (*Rhipicephalus appendiculatus*). *Vet. J.* **89**, 320–323.

Alexander, R.A. and Neitz, W.O. (1935) The transmission of louping ill by ticks (*Rhipicephalus appendiculatus*). *Onderst. J. Vet. Sci. An. Ind.* **6**, 15–33.

Anderson, R.M. and May, R.M. (1982) Population biology of infectious diseases. *Report of the Dahlem Workshop on Population biology of Infectious Disease Agents*, Berlin 1982, March 14–19. Springer Verlag, Berlin, 315 pp.

Anderson, R.M. and May, R.M. (1991) *Infectious Diseases of Humans.* Oxford University Press, Oxford.

Arthur, D.R. (1962) *Ticks and Disease.* Pergamon Press, Oxford, 445 pp.

Artsob, H. (1988) Powassan encephalitis. Vol. IV, pp. 29–49. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology.* CRC Press, Inc., Boca Raton, FL.

Ašmera, J., Šedenka, B., and Nedvídek, J. (1962) Results of parasitological investigations in a natural focus of tick-borne meningo-encephalitis in former Ostravsky territory. *Čsl. Parasitol.* **9**, 5–14 (English translation, NAMRU-3T132).

Avakian, A.A. and Lebedev, A.D. (1955) Natural focalization of haemorrhagic fevers. *J. Microbiol. Moscou* **4**, 20–6 (English translation, NAMRU-3T147).

Balashov, Yu. S. (1972) Bloodsucking ticks (Ixodoidea)—vectors of diseases of man and animals. Nauka, Leningrad, 1968. English translation in *Misc. Publ. Entomol. Soc. Am.* **8**, 161–376.

Balducci, M., Verani, P., Lopes, M.C., and Gregorig. B. (1973) Isolation in Italy of Bahig and Matruh viruses (Tete group) from migratory birds. *Ann. Microbiol. (Inst. Pasteur)* **124B**, 231–237.

Banerjee, J. (1988) Jyasanur forest disease. Vol. III, pp. 93–116. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology.* CRC Press, Inc., Boca Raton, FL.

Banerjee, K. and Khat, H.R. (1977) Correlation between the number of persons suffering from Kyasanur forest disease and the intensity of infection in the tick population. *Ind. J. Med. Res.* **66**, 175–179.

Beasley, S.J., Campbell, J.A., and Reid, H.W. (1978) Threshold problems in infection of *Ixodes ricinus* with the virus of louping ill, pp. 487–500. *In:* Wilde, J.K.H. (ed) *Tick-borne Diseases and Their Vectors'. Proceedings of an International Conference*, Edinburgh, 1976. Edinburgh University Press, Edinburgh.

Beaty, B.J., Trent, D.W., and Roehrig, J.T. (1988) Virus variation and evolution: mechanisms and epidemiological significance. Vol. I, pp. 59–85. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology.* CRC Press, Inc., Boca Raton, FL.

Belozerov, V.N. (1982) Diapause and biological rhythms in ticks. pp. 469–500. *In: Obenchain, F.D. and Galun, R., (eds) Physiology of Ticks.* Current Themes in Tropical Science, Vol. 1. Pergamon Press, Oxford.

Benda, R. (1958a) The common tic, *Ixodes ricinus* L., as a reservoir and vector of tick-borne encephalitis. I. Survival of the virus (strain B3) during the development of the tick under laboratory conditions. *J. Hyg. Epidemiol. Microbiol. Immunol.* **2**, 314–330.

Benda, R. (1958b) The common tick, *Ixodes ricinus* L. as a reservoir and vector of tick-borne encephalitis. II. Experimental transmission of encephalitis to laboratory animals by ticks at various stages of development. *J. Hyg. Epidemiol. Microbiol. Immunol.* **2**, 331–344.

Bhat, H.R. (1974) Infestation of cattle with *Haemaphysalis spinigera* Neumann, 1897 (Acarina: Ixodidae) in Kyasanur Forest disease area, Shimoga district, Karnataka. *Ind. J. Anim. Sci.* **44**, 750–755.

Bhat, H.R. (1979) Life-history of *Haemaphysalis spinigera* Neumann, 1897 (Acarina: Ixodidae). *Ind. J. Anim. Sci.* **49**, 517–522.

Bhat, H.R. (1985) Observation on the biology of *Haemaphysalis spinigera* Neumann, 1897 (Acarina: Ixodidae) under natural conditions of KFD area. *J. Bombay Nat. Hist. Soc.* **82**, 548. (Cited by Banerjee, 1988).

Bhat, H.R. and Naik, S.V. (1978) Transmission of Kyasanur Forest disease virus by *Haemaphsalis wellingtoni*, Nuttall and Wanburton, 1907 (Acarina: Ixodidae). *Ind. J. Med. Res.* **76**, 697–703.

Bhat, H.R., Sreenivasan, M.A., Goverdhan, M.K., and Naik, S.V. (1975) Transmission of Kyasanur Forest disease virus by *Haemaphysalis kyasanurensis* Trapido, Hoogstraal and Rajagopolan, 1964, (Acarina: Ixodidae). *Ind. J. Med. Res.* **63**, 879–887.

Bhat, H.R., Sreenivasan, M.A., Goverdhan, M.K., Naik, S.V., and Banerjee, K. (1976) Susceptibility of *Hystrix indica* Kerr, 1792, indian crested porcupine (Rodentia, Hystricidae) to KFD virus. *Ind. J. Med. Res.* **64**, 1566–1570.

Bhat, H.R., Sreenivasan, M.A., and Naik, S.V. (1979) Susceptibility of common giant squirrel experimental infection with KFD virus. *Ind. J. Med. Res.* **69**, 697–700.

Bhat, U.K.M. and Goverdhan, M.K. (1973) Transmission of Kyasanur forest disease virus by the soft tick, *Ornithodoros crossi. Acta Virol.* **17**, 337–342.

Blaškovič, D. (1954) The epidemic of encephalitis in the Rožňava natural focus of infection. *Slov. Acad. Sci. (Bratislava)*, 265.

Blaškovič, D. (1967) The public health importance of tick-borne encephalitis in Europe. *Bull. WHO* (Suppl. 1) **36**, 5–13.

Blaškovič, D. and Nosek, J. (1972) The ecological approach to the study of tick-borne encephalitis. *Progr. Med. Virol.* **14**, 275–320.

Blaškovič, D. and Rcháček, J. (1962) Ticks as virus vectors in Eastern Europe. pp. 135–157. *In:* Maramorosch, K. (ed) *Biological Transmission of Disease*

Agents. Symposium of the Entomological Society of America, Atlantic City, NY, 28–29 November 1960. Academic Press, New York.

Boshell, J. (1969) Kyasanur Forest disease: ecological considerations. *Am. J. Trop. Med. Hyg.* **18**, 67–80.

Boshell, M.J. and Rajagopalan, P.K. (1968) Preliminary studies on experimental transmission of Kyasanur Forest disease virus by nymphs of *Ixodes petauristae* Warburton, 1933, infected as larvae on *Suncus murinus* and *Ratus blanfordi*. *Ind. J. Med. Res.* **56** (Suppl.), 589–593.

Boshell, M.J., Rajagopalan, P.K., Patil, A.P., and Pavri, K.M. (1968) Isolation of Kyasanur Forest disease virus from Ixodid ticks: 1961–1964. *Ind. J. Med. Res.* **56** (Suppl.), 541–568.

Burgdorfer, W. and Varma, M.G.R. (1967) Trans-stadial and transovarial development of disease agents in arthropods. *Ann. Rev. Entomol.* **12**, 347–376.

Calisher, C.H. and Karabatsos, N. (1988) Arbovirus serogroups: definition and geographic distribution. Vol. I, pp. 19–57. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology*. CRC Press, Inc., Boca Raton, FL.

Cerný, V. (1976) The role of mammals in natural foci of tick-borne encephalitis in Central Europe. *Folia Parasitol.* **22**, 271–273.

Chamberlain, R.W., Sikes, R.K., Nelson, D.B., and Sudia, W.D. (1954) Studies on the North American arthropod-borne encephalitides. Part VI: Quantitative determinations of virus–vector relationships. *Am. J. Hyg.* **60**, 278–285.

Chernesky, M.A. (1969) Powassan virus transmission by ixodid ticks infected after feeding on viraemic rabbits injected intravenously. *Can. J. Microbiol.* **15**, 521–526.

Chumakov, M.P. (1944) Study of ultraviral encephalitides. VI. Transmission of tickborne encephalitis virus to the progeny of ixodid ticks and the question of natural reservoirs of this infection. *Medit. Parazitol.* **13**, 38–41.

Chumakov, M.P., Petrova, S.P., and Sondak, V.A. (1945) Study of ultravirus encephalitis. VII. Artificial adaptation of the virus of tick and Japanese encephalitis to various species of ticks of the family Ixodidae. *Medit. Parazit.* **14**, 18–24 (English translation NAMRU-3 T34).

Chunikhin, S.P. and Kurenkov, V.B. (1979) Viraemia in *Clethrionomys glareolus*–a new ecological marker of tick-borne encephalitis virus. *Acta Virol.* **23**, 257–260.

Cirkin, Ju. M., Krasovskij, M., and Fastovskaja, E.I. (1968) Virophory of *Ixodes persulcatus* ticks in Krasnojarsk region. pp. 204–212. *In:* Paspelovoy-Shtrom, M.V. and Rashinoy, M.G. (eds) *Questions on the Epidemiology of Tick-borne Encephalitis and the Biology of its Natural Focus*. Izd. "Meditsina" (Moscow).

Clarke, D.H. (1962) Antigenic relationships among viruses of the tick-borne encephalitis complex as studied by antibody absorption and agar gel precipitin techniques. pp. 67–75. *In:* Libikova, H. (ed) *Biology of Viruses of the Tick-borne Encephalitis Complex*. Czechoslovak Academy of Science, Prague.

Clarke, D.H. (1964) Further studies on antigenic relationships among the viruses of the group B tick-borne complex. *Bull. WHO* **31**, 45–56.

Danielová, V. (1990) Experimental infection of ticks *Ixodes ricinus* with tick-borne encephalitis virus under different microclimatic conditions. *Folia Parasitol.* **37**, 279–282.

Danielová, V., Daniel, M., Holubová, J., Hájková, Z., Albrecht, V., Marhoul, Z., and Simonová, V. (1983) Influence of microclimatic factors on the development and virus infection rate of ticks *Ixodes ricnius* (L.) under experimental conditions. *Folia Parasitol.* **30**, 153–161.

Davies, C.R., Jones, L.D., and Nuttall, P.A. (1990) A comparative study of the infection thresholds of Thogoto virus in *Rhipicephalus appendiculatus* and *Amblyomma variegatum*. *Am. J. Trop. Med. Hyg.* **43**, 99–103.

Dietz, K. (1988) Density-dependence in parasite transmission dynamics. *Parasitol. Today* **4**, 91–97.

Dumina, A.L. (1958) Experimental study of the extent to which the tick *Ixodes persulcatus* becomes infected with Russian spring–summer encephalitis virus as a result of sucking the blood of immune animals. *Vop. Virusol.* **3**, 156–159.

Ernek, E., Kožuch, O., Lichard, M., Nosek, J., and Albrecht, P. (1963) Experimental infection of *Clethrionomys glareolus* and *Apodemus flavicollis* with tick-borne encephalitis virus. *Acta Virol.* **7**, 434–436.

Ernek, E., Kožuch, O., Nosek, J., and Hudec, K. (1969) The relation between tick-borne encephalitis virus and the wild duck (*Anas platyrhynchos*). I. Acute infection. *Acta Virol.* **13**, 296–302.

Ernek, E., Nosek, J., and Kožuch, O. (1973) The role of birds as tick-hosts. 1 *Int. Arbkolloq. Naturh. Infekt-Krankh. ZentralEurop, Ilmitz and Graz*, Austria, 1973, pp. 112–115.

Galimov, V.R., Galimova, E.Z., Katin, A.A., and Kolchanova, L.P. (1989) The transmission of TBE virus to adult taiga ticks when virusaemia in their hosts is absent. pp. 43–44. *In: Proceedings of the XII All-Union Conference on Natural Focality of Diseases*, Novosibirsk, 10–12 October 1989.

Gallia, F., Rampas, J., and Hollender, L. (1949) Laboratory infection with encephalitis virus. *Cas. Lék. Cesk.* **88**, 224–229.

Gao, G.F., Hussain, M.H., Reid, H.W., and Gould, E.A. (1993a) Classification of a new member of the TBE flavivirus subgroup by its immunological, pathogenetic and molecular characteristics: identification of subgroup-specific pentapeptides. *Virus Res.* (in press).

Gao, G.F., Jiang, W.R., Hussain, M.H., Venugopal, K., Gritsun, T.S., Reid, H.W., and Gould, E.A. (1993b). Sequencing and antigenic studies of a Norwegian virus isolated from encephalomyelitic sheep confirm the existence of louping ill virus outside Great Britain and Ireland. *J. Gen. Virol.* **74**, 109–114.

Gardiner, W.P. and Gray, J.S. (1986) A computer simulation of the effects of specific environmental factors on the development of the sheep tick *Ixodes ricinus* L. *Vet. Parasitol.* **19**, 133–144.

Gettingby, G. and Byrom, W. (1989). The dynamics of East Coast fever: a modelling perspective for the integration of knowledge. *Parasitol. Today* **5**, 68–73.

Gray, J.S. (1991) The development and seasonal activity of the tick *Ixodes ricinus*: a vector of Lyme borreliosis. *Rev. Med. Vet. Entomol.* **79**, 323–333.

Greig, J.R., Brownlee, A., Wilson, D.R., and Gordon, W.S. (1931) The nature of louping-ill. *Vet. Rec.* **11**, 325–333.

Grešiková, M. (1972) Studies on tick-borne arboviruses isolated in Central Europe. *Biol. Works (Bratislava)* **18**, 1–111.

Grešiková, M. and Calisher, C.H. (1988) Tick-borne encephalitis. Vol. IV, pp. 177–202. *In:*Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology*. CRC Press, Inc., Boca Raton, FL.

Gritsun, T.S., Lashkevich, V.A., and Gould, E.A. (1993) Nucleotide and deduced amino acid sequence of the envelope glycoprotein of Omsk haemorrhagic fever virus; comparison with other flaviviruses. *J. Gen Virol.* **74**, 287–291.

Grulich, I. (1960) The European mole (*Talpa europaea* L.) As an important host of the tick (*Ixodes ricinus* L.) in Czechoslovakia. *Zool. Listy* **9**, 171–187.

Guirakhoo, F., Heinz, F.X., and Kunz, C. (1989) Epitope model of tick-borne encephalitis virus envelope glycoprotein E: analysis of structural properties, role of carbohydrate side chain, and conformational changes occurring at acidic pH. *Virology* **169**, 90–99.

Hardy, J.L. (1988) Suceptibility and resistance of vector mosquitoes. Vol. I, pp. 87–126. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology.* CRC Press, Inc., Boca Raton, FL.

Harlan, H.J. and Foster, W.A. (1990) Micrometeorologic factors affecting field host-seeking activity of adult *Dermacentor variabilis* (Acari: Ixodidae). *J. Med. Entomol.* **27**, 471–479.

Hasibeder, G. and Dye, C. (1988) Population dynamics of mosquito-borne disease: persistence in a completely heterogeneous environment. *Theor. Popul. Biol.* **33**, 31–53.

Hloucal, L. and Gallia, F. (1949) An epidemic of neurotropic virus disease in the district of Strakonice in 1948. *Cas. Lék. Cesk.* **88**, 275–276.

Hoogstraal, H. (1966) Ticks in relation to human diseases caused by viruses. *Ann. Rev. Entomol.* **11**, 261–308.

Hoogstraal, H. (1973) Viruses and ticks. Vol. 31, pp. 349–390. *In:* Gibbs, A.J. (ed) *Viruses and Invertebrates.* North-Holland Research Monographs Frontiers of Biology. North-Holland Publishing Company, Amsterdam.

Hoogstraal, H. (1980) Established and emerging concepts regarding tick-associated viruses, and unanswered questions. pp. 49–62. *In:* Vesenmak-Hirjan, J., Porterfied, Vesenjak-Hirjan, J., and Arslanagic, I. E. *Arboviruses in the Mediterranean Countries. Zentrbl. Bakt. Mikro. Hyg. I. Abt. Suppl.* 9. Gustav Fischer Verlag, Stuttgart, New York.

Hudson, P. (1986) *Red Grouse: The Biology and Management of a Wild Gamebird.* Game Conservancy Trust, 250 pp.

Hudson, P. (1992) *Grouse in Space and Time.* Game Conservancy Ltd., Fordingbridge, UK.

Hudson, P.J. and Rands, M.R.W. (1988) *Ecology and Management of Gamebirds.* BSP Professional Books, Blackwell Scientific Publications, Oxford, 263 pp.

Iacono-Connors, L.C. and Schmaljohn, C.S. (1992) Cloning and sequence analysis of the genes encoding the nonstructural proteins of Langat virus and comparative analysis with other flaviviruses. *Virology* **188**, 875–880.

Il'enko, V.I. (1960) The influence on the fate of biphasic meningtoencephalitis virus of feeding the tick vectors on immune animals. *Vop. Virusol.* **5**, 521–5.

Il'enko, V.I., Gorozhankina, T.S., and Smorodintsev, A.A. (1970) Main mechanisms of transovarial transmission of tick-borne encephalitis virus in tick carriers. *Medit. Parazit.* **39**, 263–267.

Jones, L.D., Davies, C.R., Steele, G.M., and Nuttall, P.A. (1987) A novel mode of arbovius transmission involving a nonviremic host. *Science* **237**, 775–777.

Jones, L.D., Davies, C.R., Hodgson, E., Williams, T., Cory, J., and Nuttall, P.A. (1990) Non-viremic transmission of Thogoto virus: vector efficiency of *Rhipicephalus appendiculatus* and *Amblyomma variegatum. Trans. Roy. Soc. Trop. Med. Hyg.* **84**, 846–884.

Jones, L.D., Kaufman, W.R., and Nuttall, P.A. (1992a) Feeding site modification by tick saliva resulting in enhanced virus transmission. *Experientia* **48**, 779–782.

Jones, L.D., Hodgson, E., Williams, T. Higgs, S., and Nuttall, P.A. (1992b) Saliva-activated transmission (SAT) of Thogoto virus: relationship with vector

potential of different haematophagous arthropods. *Med. Vet. Entomol.* **6**, 261–265.

Karabatsos, N. (1985) *International Catalogue of Arboviruses Including Certain Other Viruses of Vertebrates*, 3rd edn. *American Society of Tropical Medicine and Hygiene*, San Antonio, Texas.

Kharitonova, N.N. and Leonov, Yu. A. (1985) *Omsk Haemorrhagic Fever: Ecology of the Agent and Epizootiology*, Chumakov, M.P. (ed in chief). Amerind Publishing Co. Ltd, New Delhi, 230 pp.

Ko, R.C. (1972) Biology of *Ixodes cookei* Packard (Ixodidae) of groundhogs (*Marmota monax* Erxleben). *Can. J. Zool.* **50**, 433–436.

Kokernot, R.H., Radivojevic, B., and Anderson, R.J. (1969) Susceptibility of wild and domesticated mammals to four arboviruses. *Am. J. Vet. Res.* **30**, 2197–2203.

Kondrashova, Z.N. and Filippovets, R.V. (1970) Infection rate of *Ixodes persulcatus* ticks and certain questions of transovarial transmission after dosaged infection with tick-borne encephalitis virus. *Vop. Virusol.* **15**, 703–708 (English translation, NAMRU-3 T1312).

Korenberg, E.I. (1974) Some cntemporary aspects of natural focality and epidemiology of tick-borne encephalitis. *Folia Parasitol.* **23**, 357–366.

Korenberg, E.I. and Kovalevskiy, J.V. (1981) Tick borne encephalitis areas of distribution. *Med. Geog.* **11**, 1–148.

Korenberg, E.I. and Pchelkina, A.A. (1984) Tickborne encephalitis virus titers in engorged adult *Ixodes persulcatus* ticks. *Parazitol.* **18**, 123–127 (English translation, NAMRU-3 T1788).

Korenberg, E.I., Horáková, M., Kovalevsky, J.V., Hubálek, Z., and Karavanov, A.S. (1992) Probability models of the rate of infection with tick-borne encephaltis virus in *Ixodes persulcatus* ticks. *Folia Parasitol.* **39**, 85–92.

Kozlova, A.V. and Soloviev, V.D. (1941) Experimental study of the vector *Haemaphysalis concinna* as vector of the virus of spring–summer encephalitis. *Trudy Voen.-Medit. Akad. Kras. Arm.* **25**, 50–57.

Kožuch, O. and Nosek, J. (1985) Replication of tick-borne encephalitis (TBE) virus in *Ixodes ricinus ticks. Folia Parasitol.* **32**, 373–375.

Kožuch, O., Nosek, J., Ernek, E., Lichard, M., and Albrecht, P. (1963) Peersistence of tick-borne encephalitis virus in hibernating hedgehogs and dormice. *Acta Virol.* **7**, 430–433.

Kožuch, O., Grešíková, M., Nosek, J., Lichard, M., and Sekeyovä, M. (1967) The role of small rodents and hedgehogs in a natural focus of tick-borne encephalitis. *Bull. WHO* **36** (Suppl. 1), 61–66.

Kožuch, O., Labuda, M., Lysy, J., Weismann, P., and Krippel, E. (1990) Longitudinal study of natural foci of Central European encephalitis virus in West Slovakia. *Acta Virol.* **34**, 537–544.

Křivanec, K., Kopecky, J., Tomkova, E., and Grubhoffer, L. (1988) Isolation of tick-borne encephalitis virus from the tick *Ixodes hexagonus. Folia Parasitol.* **35**, 273–276.

Labuda, M., Lysý, M., and Kožuch, O. (1991) On virus–vector relationships in the Central European tick-borne encephalitis foci (West Slovakia). Vol. 2, pp. 29–33. *In:* Dusbábek, F. and Bukva, V. (eds) *Modern Acarology.* Prague and SPB Academic Publishing bv, The Hague, Academia.

Labuda, M., Jones, L.D., Williams, T., Danielova, V., and Nuttall, P.A. (1993a) Efficient transmission of tick-borne encephalitis virus between co-feeding ticks. *J. Med. Entomol.* (in press).

Labuda, M., Jones, L.D., Williams, T., and Nuttall, P.A. (1993b) Enhancement of tick-borne encephalitis virus transmission by tick salivary gland extracts. *Med. Vet. Ent.* (in press).

Labuda, M., Nuttall, P.A., Kozuch, O., Eleckova, E., Zuffová, E., Williams, T., and Sabo, A. (1993c) Non-viraemic transmission of tick-borne encephalitis virus: a mechanism for arbovirus survival in nature. *Experientia* **49**, 802–805.

Labuda, M., Danielova, V., Jones, L.D., and Nuttall, P.A. (1993d) Amplication of tick-borne encephalitis virus infection during co-feeding of ticks. *Med. Vet. Ent.* **7**, 339–342.

Leonova, G.N., Polenova, I.N., Baranov, N.I., Isachkova, L.M., Kruglyak, S.P., Shestakov, V.I., and Somov, G.P. (1978) Arboviruses in Primor'ye region, U.S.S.R. *Sborn. Trud. Inst. Virus imeni D.I. Ivanovsky, Akad. Med. Nauk S.S.S.R.* **3**, 49–52 (English translation, NAMRU-3 T1369).

Levkovich, E.N., Pogodina, V.V., Zasukhina, G.D., and Karpovich, L.G. (1967) Viruses of the tick-borne encephalitis complex. *Akad. Med. Nauk SSSR, Izd. "Meditsina", Leningrad, Otd.*, 245 pp.

Lichard, M. and Kožuch, O. (1967) Persistence of tick-borne encephalitis virus in nymphs and adults of *Ixodes arboricola* and its transmission to white mice. *Acta Virol.* **11**, 480.

Lvov, D.K. (1988) Omsk haemorrhagic fever. Vol. III, pp. 205–216. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology.* CRC Press, Inc., Boca Raton, FL.

Lvov, D.K. and Naumov, R.L. (1962) Transovarian transmission of antibodies to the tick-borne encephalitis virus in *Turdus pilaris* L. *Med. Parazitol.* **31**, 335–338.

MacLeod, J. (1939) The seasonal and annual incidence of the sheep tick, *Ixodes ricinus*, in Britain. *Bull. Entomol. Res.* **30**, 103–118.

MacLeod, J. (1962) Observations on tick-transmission of louping-ill to sheep. *J. Comp. Pathol. Therap.* **72**, 411–419.

MacLeod, J. and Gordon, W.S. (1932) Studies in louping-ill (an encephalomyelitis of sheep). II. Transmission by the sheep tick, *Ixodes ricinus* L. *J. Comp. Path. Therap.* **45**, 240–256.

Málková, D., Smetana, A., Fischer, S., and Marhoul, Z. (1965) Course of infection of *Clethrionomys glareolus* and white mice with tick-borne encephalitis virus freshly isolated from *Ixodes ricinus* ticks. *Acta Virol.* **9**, 367–374.

Mandl, C.W., Heinz, F.X., and Kunz, C. (1988) Sequence of the structural proteins of tick-borne encephalitis virus (Western subtype) and comparative analysis with other flaviviruses. *Virology* **116**, 197–205.

Mandl, C.W., Heinz, F.X., Stöckl, E., and Kunz, C. (1989) Genome sequence of tick-borne encephalitis virus (Western subtype) and comparative analysis of nonstructural proteins with other flaviviruses. *Virology* **173**, 291–301.

Mandl, C.W., Iacono-Connors, L., Wallner, G., Holzmann, H., Kunz, C., and Heinz, F.X. (1991) Sequence of the genes encoding the structual proteins of the low-virulence tick-borne flaiviviruses Langat TP21 and Yelantsev. *Virology* **185**, 891–895.

Mandl, C.W., Holzmann, H., Kunz, C., and Heinz, F.X. (1993) Complete genomic sequence of Powassan virus: evaluation of genetic elements in tick-borne versus mosquito-borne flaviviruses. *Virology* **194**, 173–184.

Matuschka, F.-R., Richter, D., and Spielman, A. (1991) Differential detachment from resting hosts of replete larval and nymphal *Ixodes* ticks. *J. Parasitol.* **77**, 341–345.

McLean, D.M. and Donohue, W.L. (1959) Powassan virus: isolation of virus from a fatal case of encephalitis. *Can. Med. Assoc. J.* **80**, 708–711.

Milne, A. (1949a) The ecology of the sheep tick *Ixodes ricinus* L. Host relationships of the tick. Part I. Review of previous work in Britain. *Parasitol.* **39**, 167–172.

Milne, A. (1949b) The ecology of the sheep tick Ixodes ricinus L. Host relationships of the tick. Part II. Observations on hill and moorland grazing in northern England. *Parasitol.* **39**, 173–197.

Mishaeva, N.P. (1990) Protection of vertebrates from experimental tick-borne encephalitis in active and passive immunization against *Ixodes* antigens. *Vop. Virusol.* 93–98.

Mishaeva, N.P. and Erofeeva, N.I. (1979) The effect of diapause of the tick *Ixodes ricinus* (Ixodidae) upon the multiplication of the tick-borne encephalitis virus in them. *Parazitol.* **13**, 218–222.

Murphy, F.A., Fauquet, C.M., Bishop, D.H.L., Ghabrial, S.A., Jarvis, A.W., Martelli, G.P., Mayo, M.A., and Summers, M.D. (1994) Virus Taxonomy. The Classification and Nomenclature of Viruses. Sixth Report of the International Committee on Taxonomy of Viruses. *Archives of Virology*, Springer-Verlag, Wien (in press).

Naumov, R.L., Levkovich, E.N., and Rzhakhova, O.E. (1963) The part played by birds in circulation of tick-borne encephaltis virus. *Med. Parazitol.* **32**, 18–29 (English translation, MANRU-3 T141).

Needham, G.R. and Teel, P.D. (1991) Off-host physiological ecology of ixodid ticks. *Annu. Rev. Entomol.* **36**, 659–681.

Nosek, J., Grešiková, M., and Reháček, J. (1961) Persistence of tick-borne encephalitis virus in hibernating bats. *Acta Virol.* **5**, 112–116.

Nosek, J., Kožuch, O., and Radda, A. (1968) Untersuchungen über die Ökologie des virus der zentraleuropäischen Enzephalitis im Nordmähren. *Zbl. Bakt., Mikrobiol., Hyg., Abt. I. Org.* **208**, 81–87.

Nuttall, P.A. (1984) Transmission of viruses to wild life by ticks. pp. 135–159. *In:* Mayo, M.A. and Harrap, K.A. (eds) *Vectors in Virus Biology*. Academic Press, New York.

Nuttall, P.A., Jones, L.D., and Davies, C.R. (1991) The role of arthropod vectors in arbovirus evolution. *Adv. Dis. Vector Res.* **8**, 15–45.

Nuttall, P.A., Jones, L.D., Labuda, M., and Kaufman, W.R. (1994) Adaptations of arboviruses to ticks. *J. Med. Entomal.* **31**, 1–9.

Pavlovsky, E.N. and Soloviev, V.D. (1963) Circulation of the virus of spring–summer encephalitis virus in the organism of the tick vector, *Haemaphysalis concinna.* pp. 197–204. *In: Papers on Experimental Parasitology by E.N. Pavlovsky*, Akad. Nauk USSR (English translaton, NAMRU-3 T115).

Petrishcheva, P.A. and Levkovitsch, E.N. (1949) On the spontaneous virus-carrier state of the *Ixodes persulcatus* and *Ixodes ricinus* ticks in new focus of tick-borne encephalitis. *Vop. Kraj. Obshch. Eksp. Parasitol.* **4**, 42.

Pletnev, A.G., Yamshchikov, V.F., and Blinov, V.M. (1990) Nucleotide sequence of the genome and complete amino acid sequence of the polyprotein of tick-born encephalitis virus. *Virology* **174**, 250–263.

Pretzmann, G., Radda, A., and Loew, J. (1967) Die Verteilung virustragender Zechen in Naturherden der Frühsommer-Meningoencephalitis. *Zbl. Bakt. I. Orig.* **203**, 30–46.

Radda, A., Hofman, H., and Pretzmann, G. (1969) Threshold of viramia in *Apodemus*

flavicollis for infection of *Ixodes ricinus* with tick-borne encephalitis virus. *Acta Virol.* **13**, 74–77.

Rajagopalan, P.K. and Anderson, C.R. (1970) Trans-monsoonal persistence of Kyasanur Forest disease virus in *Haemaphysalis* nymphs infected in nature. *Indian J. Med. Res.* **58**, 1184–1187.

Rajagopalan, P. K., Paul, S.D., and Sreenivasan, M.A. (1969) Isolation of Kyasanur Forest disease virus from the insectivorous bat, *Rhinolophus rouxi* and from *Ornithodorus* tick. *Indian J. Med. Res.* **57**, 805–808.

Rampas, J. and Gallia, F. (1949) Isolation of encephalitis virus from *Ixodes ricinus* ticks. *Cas. Lék. Cesk.* **88**, 1179–1180.

Randolph, S.E. (1994) The relative contributions of transovarial and transstadial transmission to the maintenance of tick-borne diseases. *In:* Axford, J.S. and Rees, D.H.E. (eds) *Lyme Borreliosis, NATO Series in Life Sciences* (in press).

Randolph, S.E. and Steele, G.M. (1985) An experimental evaluation of conventional control measures against the sheep tick, *Ixodes ricinus* (L.) (Acari: Ixodidae). II. The dynamics of the tick-host interaction. *Bull. Entomol. Res.* **75**, 501–518.

Rao, T.R. (1963) Potentiality of transovarial transmission of Kyasanur forest disease virus by ticks. *Anais Microbiol.* **11**, 95–98 (cited by Burgdorfer and Varma, 1967).

Reháček, J. (1960) Experimental hibernations of the tick-borne encephalitis virus in engorged larvae of the tick *Ixodes ricinus* L. *Acta Virol.* **4**, 106–109.

Reháček, F. (1962) Transovarial transmission of tick-borne encephalitis virus by ticks. *Acta Virol.* **6**, 220–226.

Reháček, J. (1965) Development of animal viruses and rickettsiae in ticks and mites. *Ann. Rev. Entomol.* **10**, 1–24.

Reid, H. W. (1984) Epidemiology of louping-ill. pp. 161–178. *In:* Mayo, M.A. and Harrap, K.A. (eds) *Vectors in Virus Biology*. Academic Press, London.

Reid, H.W. (1988) Louping-ill. Vol. III, pp. 117–135. *In:* Monath, T.P. (ed) *The Arboviruses: Epidemiology and Ecology*. CRC Press, Inc., Boca Raton, FL.

Reid, H.W., Duncan, J.S., Phillips, J.D.P., Moss, R., and Watson, A. (1978) Studies on louping-ill virus (Flavivirus group) in wild red grouse (*Lagopus lagopus scoticus*). *J. Hyg. Camb.* **81**, 321–330.

Reid, H. W., Buxton, D., Pow, I., and Finlayson, J. (1984) Transmission of louping-ill virus in goat milk. *Vet. Rec.* **114**, 163–165.

Rodrigues, F.M. (1968) A study on the susceptibility of wild birds of Shimoga district, Mysore State, to infection with Kyasanur Forest disease virus, with a view to determining their role in the epidemiology of the disease. Ph.D. thesis, University of Poona (cited by Banerjee, 1988).

Rogers, D.J. (1988) The dynamics of vector-transmitted diseases in human communities. *Phil. Trans. Roy. Soc. Lond. B* **321**, 513–539.

Schmidt, J.R. and Shope, R.E. (1971) Kemerovo virus from a migrating common redstart of Eurasia. *Acta Virol.* **15**, 112.

Shah, K.V. (1965) Experimental infection of lactating monkeys with Kyasanur forest disease virus. *Zcta Virol.* **9**, 71–75.

Shiu, S.Y.W., Ayres, M.D., and Gould, E.A. (1991) Genomic sequence of the structural proteins of Louping ill virus: comparative analysis with tick-borne encephalitis virus. *Virology* **180**, 411–415.

Singh, K.R.P. and Anderson, C.R. (1968) Relation of *Haemaphysalis spinigera* larval infection rates and host viraemia levels of Kyasanur forest disease virus. *Indian J. Med. Res.* **56**, 137–141.

Singh, K.R.P., Pavri, K., and Anderson, C.R. (1963) Experimental transovarial transmission of Kyasanur forest disease virus in *Haemaphysalis spinigera*. *Nature* **199**, 513.

Singh, K.R.P., Pavri, K.M., and Anderson, C.R. (1964) Transmission of Kyasanur forest disease virus by *Haemaphysalis turturis*, *Haemaphysalis papuana kinneari* and *Haemaphysalis minuta*. *Indian J. Med. Res.* **52**, 566–573.

Singh, K.R.P., Goverdhan, M.K., and Bhat, H.R. (1968a) Transovarial transmission of Kyasanur forest disease virus by *Ixodes petauristae*. *Indian J. Med. Res.* **56**, 628–632.

Singh, K.R.P., Goverdhan, M.K., and Rao, T.R. (1968b) Experimental transmission of Kyasanur Forest disease virus to small mammals by *Ixodes petauristae*, *I. ceylonensis* and *Haemaphysalis spinigera*. *Indian J. Med. Res.* **56** (Suppl.) 594–609.

Singh, K.R.P., Goverdhan, M.K., and Bhat, U.K.M. (1971) Transmission of Kyasanur fever virus by soft tick, *Argas persicus* (Ixodoidea: Argasidae). *Indian J. Med. Res.* **59**, 213–218.

Skrynnik, A.N. and Ryzhov, N.V. (1941) Experimental investigation of the ticks *Dermacentor silvarum* Ol. as the vectors of spring–summer encephalitis. *Trudy Voenno-Medit. Akad. Kras. Armii* **25**, 41–49.

Smith, C.E.G. and Varma, M.G.R. (1981) Louping Ill. Vol. 1, pp. 191–200. Steele, J.H. (ed in chief) *CRC Handbook Series in Zoonoses, Section B*, Beran, G.W. (ed): *Viral Zoonoses*, CRC Press, Inc., Boca Raton, FL.

Smith, C.E.G., McMahon, D.A., O'Reilly, K.J., Wilson, A.L., and Robertson, J.M. (1964) The epidemiology of louping ill in Ayrshire: the first year of studies in sheep. *J. Hyg. Camb.* **62**, 53–68.

Smorodintsev, A.A. (1958) Tick-borne spring–summer encephalitis. *Publ. Hlth Mono.* **50**, 26–33.

Sreenivasan, M.A., Bhat, H.R., and Naik, S.V. (1979) Experimental transmission of Kyasanur Forest disease virus by *Dermacentor auratus* Supino. *Indian J. Med. Res.* **69**, 701–707.

Sreenivasan, M.A., Rajagopalan, P.K., and Bhat, H.R. (1983) Spatial distribution of infected *Haemaphysalis* nymphs in the epizootic localities of Kyansanur forest disease. *Indian J. Med. Res.* **78**, 531–536.

Steele, G.M. and Randolph, S.E. (1985) An experimental evaluation of conventional control measures against the sheep tick, *Ixodes ricinus* (L.) (Acari: Ixodidae). I. A unimodal seasonal activity pattern. *Bull. Entomol. Res.* **75**, 489–499.

Stockman, S. (1918) Louping-ill. *J. Comp. Path. Therap.* **31**, 137–193.

Striessle, G. (1960) Die Zecke *Ixodes hexagonus* als Reservoir für das Virus des Frühsommer-Meningoencephalitis. *7 Tagung Österr. Ges. Mikrobiol. Hgy., Pörtschach.* [cited by Blaškovič, D. and Reháček, J. (1967)].

Streissle, G. (1961) Versuche zur Ubertragung des Virus der Frühsommer-meningoencephalitis durch Lederzacken. *Zbl. Bakt. I. Abt. Orig.* **182**, 159–169.

Sutherst, R.W. and Maywald, G.F. (1985) A computerised system for matching climates in ecology. *Agricult. Ecosyst. Environ.* **13**, 281–299.

Swanepoel, R. (1968). Quantitative studies on the virus of louping-ill in sheep and ticks. Oh.D. thesis, University of Edinburgh [cited by Reid (1984)].

Tatarinova, L.G. (1961) The experimental establishment of the role of ticks *Haemaphysalis japonica dougalasi* N. as vectors of the virus of tick encephalitis. *Dokl. Akad. Nauk SSSR* **140**, 510–512.

Thomas, L.A., Kennedy, R.C., and Eklund, C.M. (1960) Isolation of a virus closely

related to Powassan virus from *Dermacentor andersoni* collected along north Cache la Poudre River, Colo. *Proc. Soc. Exp. Biol. Med.* **104**, 355–359.

Timoney, P. (1971) Powassan virus infection in the grey squirrel. *Acta Virol.* **15**, 429.

Tongeren, H.A.E., van (1962) Central European encephalitis—epidemiology and vectors. *Proc. 6th Int. Congr. Trop. Med. Malar.*, Lisbon, 5–13 September 1958, Vol. 5, pp. 174–179. (Anais Inst. Med. Trop., Lisbon, **16**, Suppl. 9.)

Trapido, H., Rajagopalan, P.K., Work, T.H., and Verma, M.E.R. (1959) Kyasanur Forest disease. Part VIII. Isolation of Kyasanur Forest disease virus from naturally infected ticks of the genus *Haemaphysalis*. *Indian J. Med. Res.* **47**, 133–138.

Venugopal, K., Buckley, A., Reid, H.W., and Gould, E.A. (1992) Nucleotide sequence of the envelope glycoprotein of Negishi virus shows very close homology to louping ill virus. *J. Gen Virol.* **190**, 515–521.

Venugopal, K., Gritsun, T., Lashkevich, V.A., and Gould, E.A. (1994) Analysis of the structural gene sequence shows Kyasanur Forest Disease virus as a distinct member in the tick-borne encephalitis virus serocomplex. *J. Gen. Virol.* **75**, 227–232.

Vesenjak-Hirjan, J. and Šooš, E. (1976) The role of migrating sheep in dissemination of tick-borne encephalitis virus. pp. 167–173. *In:* Vesenjak-Hirjan, J. (ed) *Tick-borne Encephalitis in Croatia* (*Yugoslavia*). RAD Jugoslavenske Akademije-Znanosti i Umjetnosti, Zagreb, Book 372.

Votyakov, V.I. and Mishaeva, N.P. (1980) Investigation of a possibility of protecting vertebrates against transmissive infection with tick-borne encephaltis virus. *Vop. Virusol.* **2**, 170–172.

Webb, H.E. (1965) Kyasanur Forest disese virus in three species of rodents. *Trans. Roy. Soc. Trop. Med. Hyg.* **59**, 205–211.

Webb, H.E. and Burston, J. (1966) Clinical and pathological observations with special reference to the nervous system in *Macaca radiata* infected with Kyasanur Forest disease virus. *Trans. Roy. Soc. Trop. Med. Hyg.* **60**, 325–331.

WHO (1986) *Tick-borne Encephalitis and Haemorrhagic Fever with Renal Syndrome.* Report on a WHO Meeting, Baden, 3–5 October, 1983. EURO Reports and Studies, 104.

Whitby, J.E., Whitby, S.N., Jennings, A.D., Stephenson, J.R., and Barrett, A.D.T. (1993) Nucleotide sequence of the envelope protein of a Turkish isolate of tick-borne encephalitis (TBE) virus is distinct from other viruses of the TBE complex. *J. Gen. Virol.* **74**, 921–924.

Woodall, J.P. and Roz, A. (1977) Experimental milk-borne transmission of Powassan virus in the goat. *Am. J. Trop. Med. Hyg.* **26**, 190–192.

Work, T.H. (1958) Russian spring–summer virus in India. Kyasanur Forest disese. *Prog. Med. Virol.* **1**, 248–279.

Work, T.H. and Trapido, H. (1957) Kyasanur Forest disease. Summary of preliminary report of investigations of the Virus Research Centre on an epidemic disease affecting forest villagers and wild monkeys of Shimoga District, Mysore. *Indian J. Med. Sci.* **11**, 340–341.

Yousfi-uonod, R. and Aeschlimann, A. (1986) Recherches sur les ticques (Acarina: Ixodidae), parasites de bovides dan l'ouest Algerien. A. Inventaire systematique et dynamique salsonniere. *Ann. Parasitol. Hum. Comp.* **61**, 341–358.

Zarnke, R.L. and Yuill, T.M. (1981) Powassan virus infection in snowshow hare (*Lepus americanus*). J. Wildl. Dis. **17**, 303–310.

Zilber, L.A. and Xoloviev, V.D. (1946) Far Eastern tick-borne spring–summer (spring) encephalities. *Am. Rev. Soviet. Med.* (Special Suppl.) 80 pp.

13

Ecology of Crimean–Congo Hemorrhagic Fever

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1. INTRODUCTION: THE DISEASE

1.1. Discovery

Literature from as far back as the 12th century describes a hemorrhagic disease, now thought to be Crimean–Congo Hemorrhagic Fever (CCHF), from Tadzhikistan, Central Asia, which was transmitted by an arthropod that parasitized blackbirds (Hoogstraal, 1979). The arthropod described may well have been one or more species of *Hyalomma* tick larvae which frequently are found on blackbirds. In Central Asia, various hemorrhagic diseases known by such names as "cases of a peculiar gastro-intestinal hemorrhage," "acute infectious capillarotoxicosis," "acute infectious hemorrhagic disease," and "Uzbezekistan hemorrhagic fever" were known for centuries to produce a disease similar to CCHF (Chumakov et al., 1976). The first detailed clinical description of CCHF (as Crimean hemorrhagic fever) was made during an epidemic in 1944–1945 in Crimea (Chumakov, 1945, 1946, 1971, 1974). An etiological agent, presumably a virus, was isolated from the blood of patients and from *H. margitanum* (as *H. plumbeum*) ticks. The disease produced an illness characterized by fever, headache, nausea, vomiting, backache, joint ache, and photophobia.

Chumakov et al. (1968) first isolated CCHF virus in brains of suckling mice and rats that died after intracerebral inoculation of acute-phase patient sera and suspensions of *H. margitanum* (as *H. plumbeum*) ticks. The virus contained RNA, could be passed through a 220 μm Millipore (R) filter, and was sensitive to elevated temperatures. The virus was not pathogenic for adult mice, rats, guinea pigs, hamsters, rabbits, and monkeys. In 1967, the virus isolated in Zaire (as Congo virus; Simpson et al., 1967) was found to be antigenically indistinguishable from the virus isolated in Europe and Asia (Casals, 1969; Chumakov et al., 1969).

1.2. History of Epidemics

The history of CCHF outbreaks was reviewed in detail by Hoogstraal (1979) and Watts et al. (1989). The following is a summary of these outbreaks with recent observations. After the initial descriptions from the Crimean Oblast outbreaks, and sporadic cases were detected in Bulgaria (1953–1965, 1968–1973) and in Eurasia in Astrakhan Oblast (1953–1963), Rostov Oblast (1963–1969), Uzbekistan SSR (1944), Kazahkstan SSR (1948–1968), and Tadzhikistan SSR (1943–1970). A similar disease was first observed in Africa in 1956 and described (as Congo virus) from a febrile patient in Zaire (Simpson et al., 1967). Subsequent clinical descriptions in Agrica from Uganda (1958–1977), Mauritania (1983, 1987), Burkina Faso (1983), Republic of South Africa (1981–1986), Tanzania (1986), and Namibia (1986); and in the Middle East from Pakistan (1976), United Arab Emirates (1979–1980), and Iraq (1979–1980) were similar to those reported from Europe and Asia. Evidence of recent CCHF activity in Africa was reported in 1985 in Madagascar (isolated in ticks; Mathiot et al., 1988) and 1990 in Senegal (antibody in domestic animals; Wilson et al., 1990).

1.3. Importance

Although CCHF virus is known to infect many species of vertebrates, humans (and suckling mice and rats) are the only hosts of CCHF virus that appear to develop serious and often fatal disease. Signs of disease in humans occur 2–9 days after infection (Swanepoel et al., 1987). Human infection can occur by: (1) bite from an infected tick; (2) exposure through skin abrasions from a viremic vertebrate host; and (3) inhalation of aerosols containing virus.

In many cases, there is a lack of definitive mortality and morbidity data due to the absence of systematic surveillance. The ratio of inapparent to apparent infections has been estimated to be 5:1 (Goldfarb et al., 1980). In the initial outbreak in the Crimean Oblast in 1944, the cast fatality rate in hospitalized military troops was about 10% (Chumakov, 1974). Between 1953 and 1969 in Astrakhan Oblast, 17% of the 104 cases were fatal (Watts et al., 1989). In Bulgaria from 1953 to 1965, the overall mortality rate in people with overt clinical symptoms was about 17%, with an average annual morbidity rate of 71% (Donchev et al., 1967). Mortality rates in nosocomial infections in hospitals were much higher (40.5%) than the rates observed in people exposed in natural foci. In Eurasia, morbidity has historically been sporadic. Epidemics are often temporally and spatially distinct, and the disease only gradually spreads from the original focus. Little or no disease activity is observed immediately after major epidemics. Although widely endemic in Africa, CCHF cases are reported only sporadically, and may be commonly overlooked or misdiagnosed. There is no reliable way to predict morbidity rates in Eurasia or Africa in the absence of regular and reliable seroepidemiological surveys.

1.4. Virus

Little is known about the structure, genetics, and replication strategies of CCHF virus. The morphology of the virus has been studied in newborn mice and pig embryo kidney cell cultures (Murphy et al., 1968, 1973; Jelinkova et al., 1975; Korolev et al., 1976). As described by Clerx and Bishop (1981) and Clerx et al. (1981), CCHF virus is characterized by having two glycoproteins (G_1, G_2) on the surface of virions and three nucleocapsid [L (4.9×10^6 Da), M ($1.5–1.9 \times 10^6$ Da), and S ($0.6–0.7 \times 10^6$ Da)] RNA species.

Blackburn et al. (1987) and Shepherd et al. (1988) described new techniques, an enzyme-linked immunosorbent assay (ELISA) and reversed passive hemagglutination, to detect CCHF antigen. Several new techniques for use in CCHF serological studies were described by Saluzzo and LeGuenno (1987) and Saluzzo et al. (1988).

The paucity of data concerning many aspects of the molecular and natural biology of CCHF virus is partly due to: (1) the limitations inherent in its use in the laboratory to Biological Safety Level 4 facilities (except strain 10200; Causey et al., 1970); (2) the lack of an adequate laboratory animal model; and (3) its inability to produce cytopathogenic effect (CPA) or plaques in many cell lines.

1.4.1. Relationships to Other Viruses

CCHF virus is the type species of the genus *Nairovirus* (family Bunyaviridae) (Casals and Tignor, 1980). The other 32 related nairoviruses are categorized into six serological groups, including: Crimean–Congo hemorrhagic fever group (CCHF, Hazara, Khasan), Dera Ghazi Khan group (Abu Hammad, Abu Mina, Dera Ghazi Khan, Kao Shuan, Pathum Thani, Pretoria), Hughes group (Hughes, Punta Salinas, Soldado, Zirqa, Farallon, Fraser Point, Great Saltee, Puffin island, Raza, Sapphire), Nairobi sheep disease group (Dugbe, Nairobi sheep disease, Ganjam), Qalyub group (Qalyub, Bandia, Omo), and Sakhalin group (Sakhalin, Avalon, Clo Mor, Paramushir, Taggert, Tillamook) (Karabatsos, 1985). RNA probes have been used to detect nucleotide sequence homology among the members of the Nairobi sheep disease (NSD) and the CCHF serogroups. The sequence relationships showed that the NSD and CCHF serogroups are more closely related to each other than to the other serogroups in the genus (Marriott et al., 1990).

1.4.2. Vertebrate Hosts

Like other zoonotic agents, CCHF virus appears to produce little or no disease in its natural hosts. A complete description of vertebrates from which either CCHF virus has been isolated or antibody to CCHF virus detected in nature was made by Hoogstraal (1979) and Watts et al. (1989). Antibody to CCHF virus has been detected from only one reptile (Horsfield tortoise; Pak and Mikhailova, 1973), and two birds [fowl (Semashko et al., 1975); magpie

(Zarubinsky et al., 1975)] sera. Sera from several species of wild mammals (including baboons, gazelles, hedgehogs, bats, hares, rodents, and carnivores) have been found to have antibodies to CCHF virus. Seroepidemiological studies have also detected antibodies to CCHF virus in domestic cattle, horses, donkeys, sheep, goats, and pigs in Eurasia and Africa. CCHF virus has been isolated in nature from humans, and both domestic and wild vertebrates including: cattle (Woodwall et al., 1965), goats (Causey et al., 1970), sheep (Yu-Chen et al., 1985), hares (Chumakov, 1974), hedgehogs (Kemp et al., 1974), and a multimammate mouse (Saluzzo et al., 1985). Although the number of species of vertebrates implicated in the natural history and ecology of CCHF virus in both Eurasia and Africa is exceptionally varied and extensive, the precise role of each still needs to be determined. A further discussion of field and laboratory studies as they relate to transmission and maintenance of CCHF virus is found in the Section 2.3 on transmission and maintenance cycles.

1.4.3. Arthropod Vectors

CCHF virus isolations have been made from 31 species of ticks and one species of biting midge collected in nature (Table 13.1). Virus isolations from ticks have been made from two species in the family Argasidae and from seven genera of the family Ixodidae. The one virus isolation sample from *Culicoides* spp. (Diptera: Ceratopogonidae) in Nigeria, collected by light traps near a cattle shed, may have contained undigested blood (Causey et al., 1970). In Africa, CCHF virus has been isolated from 13 tick species in nine different countries, including isolations from *Amblyomma variegatum* in four countries, and *Hyalomma impeltatum*, *H. truncatum* (Fig. 13.1), and *H. rufipes* in three countries. In Eurasia and Asia, CCHF virus has been isolated from 17 species of ticks, including five *Hyalomma* and five *Rhipicephalus* spp., in China and the republics of the former USSR. CCHF virus has been isolated from seven species (three *Rhipicephalus* spp. and one each *Hyalomma*, *Dermacentor*, *Boophilus*, and *Ixodes* spp.) in four countries in Europe. In the middle East, CCHF virus has been isolated from five species (two *Hyalomma*, one each *Ambloyomma*, *Boophilus*, and *Ornithodorus* spp.) in three countries.

Several tick species from which CCHF virus has been isolated in nature and other related species have been tested in the laboratory for their ability to maintain and transmit the virus. These studies are described in Section 2.3.

1.4.4. Strain Variation

Most early studies suggested that there are very few significant differences among strains of CCHF virus, in spite of the extremely wide geographic distribution of the virus (Tignor et al., 1980; Casals, 1969; Chumakov et al., 1969). However, recent detailed analysis by genetic sequencing and protein analysis of six geographically dispersed (Senegal to China) strains suggests more differences in strains than previously believed (Lofts et al., USAMRIID, Fort Detrick, MD, personal communication).

Table 13.1. Tick species from which CCHF virus has been isolated, associated vertebrate hosts and locations of isolations

Species	Vertebrate hosts		Location CCHF virus isolated
	Larvae and nymphs	Adult	
Multiple host			
Family Argasidae			
Argas persicus (Oken) *Ornithodoros lahorenis* Neumann[a]	Wild and domestic birds and mammals	Wild and domestic birds and mammals	Uzbekistan, Iran
Three-host			
Family Ixodidae			
Amblyomma variegatum (Fabricius)	Small mammals and birds	Cattle	Uganda, CAR[b], Afghanistan, Senegal, Nigeria
Dermacentor daghestanicus Olenev	Hedgehogs, rodents, hares	All domestic mammals, deer, and humans	Kazakhstan
D. marginatus (Sulzer)	Insectivores, rodents, hares and small carnivores	Livestock, and other domestic and wild herbivores	Bulgaria, Moldavia, Uzbekistan
Haemaphysalis punctata Canestrini and Fanzago	Wild birds and hares	All domestic mammals, and large wild mammals	Moldavia, Ukraine, Russia
H. asiaticum Schulze	Hedgehogs, hares, rodents, and carnivores	Camels and other domestic herbivores, pigs, and gazelles	Azerbaijan, Uzebekistan, Kazakhstan, Tadzhikistan
H. dromedarii Koch	Hares and other small mammals	Camels, cattle, sheep and goats	Azerbaijan, Uzbetkistan, Kazakhstan, Tadzhikistan
H. impeltatum Schulze and Schlottke	Gerbils, jerboas, jirds, hedgehogs, hares, lizards, and birds	Domestic herbivores, antelope, dog, wild pig, camel and cattle	Ethiopia, Nigeria, Senegal
H. impressum Koch	Hedgehogs and rodents	Cattle, horses, camel, sheep and dog	Senegal
H. nitidum Schulze	Hares	Cattle, horses, goats, bushpig, Cape buffalo, Defassa waterbuck and Roan antelope	CAR[b]

Species	Vertebrate hosts: Larvae and nymphs	Vertebrate hosts: Adult	Location CCHF virus isolated
H. truncatum Koch	Hares, birds, rodents and domestic animals	Cattle, goats, sheep, horses, camels, pigs, dogs, and numerous large wild mammals, tortoises and birds	Nigeria, RSA[c], Senegal
Ixodes ricinus Latrielle	Small mammals and birds (larvae) and large mammals and birds (nymphs)	Large mammals	Bulgaria, Hungary, Yugoslavia, Moldavia, Ukraine
Rhipicephalus appendiculatus Neumann	Hares and cattle	Cattle	Uganda
R. e. evertsi Neumann	Hares and large herbivores	Large herbivores	RSA[c]
R. pulchellus Gerstacker	Small and large wild and domestic herbivores	Large herbivores	Kenya
R. pumilio Schulze	Hedgehogs, hares and birds	Hares, hedgehogs, and large rodents, domestic and wild mammals	Turkmenia, Uzbekistan
R. rossicus Yakimov and Kohl-Yakimova	Hedgehogs, rodents, and hares	Wild and domestic mammals from the size of hares to camels	Russia, Armenia
R. sanguineus Latrielle	Insectivores and rodents	Wild and domestic ungulates and carnivores	Bulgaria, Ukraine, Turkmenia
R. turanicus Pomerantsev and Matikashvili	Small mammals	Large mammals	Kirgizia
H. excavatum Koch	Small mammals	Small mammals	Nigeria
Two-host			
Hyalomma anatolicum Koch	Domestic mammals	Domestic mammals	Nigeria, Pakistan, Armenia, Turkmenia, Uzbetkistan, Tadzhikistan
H. detritum Schulze	Cattle and horses	Cattle and horses	Azerbaijan, Uzbekistan, Kazakhstan, Tadzhikistan
H. m. marginatum Koch	Hedgehogs, hares, and birds	Cattle, goats, sheep, horses, and camels	Bulgaria, Yugoslavia, Afghanistan, Russia, Armenia, Azerbaijan, Ukrainia, Kalmyk

(*continued*)

Table 13.1. (continued)

Species	Vertebrate hosts: Larvae and nymphs	Vertebrate hosts: Adult	Location CCHF virus isolated
H. m. rifupes Koch	Birds and hares	Cattle	RSA[c], Mauritania, Nigeria, Senegal
H. turanicum Pomerantsev	Birds and hares	Cattle	Uzbekistan, Kirgizia, Tadzhikistan
R. Bursa Canestrini	Wild and domestic mammals	Wild and domestic mammals	Bulgaria, Greece, Armenia, Azerbaijan, Turkmenia
One-host			
Boophilus annulatus (Say)	Cattle	Cattle	Armenia, CAR[b], Bulgaria, Uzbekistan
B. decoloratus (Koch)	Cattle	Cattle	Nigeria, Senegal
B. microplus (Canestrini)	Cattle	Cattle	Pakistan, Madagascar
B. geigyi Aeschlimann and Morel	Hartebeest	Hartebeest	Senegal

[a] *O. lahorensis* is a two-host tick.
[b] CAR = Central African Republic.
[c] RSA = Republic of South Africa.

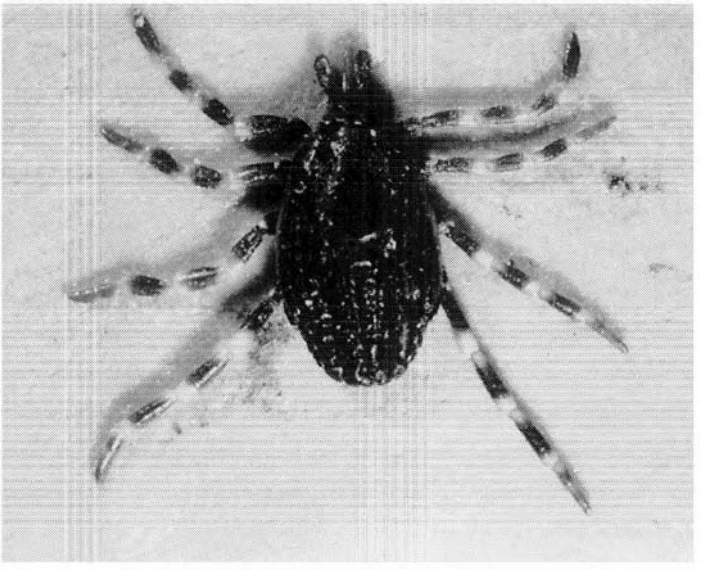

Fig. 13.1. Dorsal view of *Hyalomma truncatum* female..

1.5. Associations with Humans and Animals

Humans are the only documented naturally infected host of CCHF virus. Severe disease and mortality from CCHF infection have been observed in countries in Africa (Van Eeden et al., 1985; Swanepoel et al., 1987), Europe (Donchev et al., 1967), the Middle East (Burney et al., 1980), and Asia (Mikhailov, 1946). In endemic areas, the most common routes of CCHF virus transmission to humans are infection from a bite by an infected tick or possibly by crushing engorged ticks. The typical course of infection resulting from exposure to infected ticks has been reviewed by Hoogstraal (1979) and includes incubation, pre-hemorrhage, hemorrhage, and convalescence periods. In South Africa, the incubation period for CCHF patients previously exposed to ticks was documented to be 2–7 days (Swanepoel et al., 1987). The pre-hemorrhage period, lasting 1–7 days, is characterized by a very sudden onset of symptoms: starting with fever then chills; headache; rheumatic, lumbar and epigastric pains, nausea; vomiting, diarrhea; and loss of appetite. The hemorrhagic period occurs 3–6 days after onset of illness, lasts 1–10 days, and may end in death. Hemorrhages, from petechiae to large hematomas, appear on the mucous membranes and skin, especially on the upper body. Hemorrhage is seen in all organs and tissues, and is manifested externally by bleeding from mouth, gums, nose, conjunctivae, and ears. Convalescence starts suddenly 9–20 days after onset of illness and is characterized by prolonged asthenia, labile pulse, and occasionally temporary loss of hair. Associated problems may persist for more than a year. Disease course can vary from mild to severe.

The disease course in laboratory or medical personnel, infected by contact with virus-contaminated materials from CCHF patients, is generally characterized by severe clinical symptoms and high mortality. Family members caring for sick relatives and health-care workers historically have not taken the necessary measures to prevent CCHF transmission. From 1953 to 1968, 24% of the CCHF patients observed in the Stavropol region, Russia, were people who had cared for CCHF-infected patients. In Tadzhikistan (1943–1972), mortality rates in CCHF patients were 25.2% and 50.0% for tick-transmitted and patient-transmitted cases, respectively (Pak and Mikhailova, 1973). As recently as 1985, nosocomial infections were documented in South Africa (Swanepoel et al., 1987). Nosocomial cases represented 26% ($n = 31$) of the patients seen in 1981–1986. Six cases involved contact with patient blood and two cases involved contact with fomites. Mortality rates were 25% in nosocomial-infected patients and 33.3% ($n = 3$) in cases with confirmed infection from ticks.

Abattoire workers and people associated in some way with infected wild and domestic animals (sheep shearing, other husbandry practices) are also at risk of CCHF infection. Transmission can occur either from contact with a virus-infected animal or tick tissues and/or by the reattachment of dislodged infected ticks. These types of cases have been documented in Eurasia (Stolbov et al., 1965) and Africa (Shepherd et al., 1985; Swanepoel et al., 1985a, 1985b). In South Africa 32% ($n = 31$) of the CCHF cases were caused by exposure to blood of cattle, sheep, or an ostrich, with a mortality rate of 10% ($n = 10$).

2. EPIDEMIOLOGY

2.1. Geographic and Seasonal Distribution

The known distribution of CCHF virus covers the greatest geographic range of any tick-borne virus (Fig. 13.2). Virus isolation and antibody data provide evidence that enzootic foci exist in Europe and Asia (from Portugal to Western China and India), and in Africa (from Senegal to Egypt, Kenya, South Africa, and Madagascar). Of the more than 90 countries found within the geographic range of CCHF, there is recent evidence of CCHF viral activity in only about 50% of these countries (Watts et al., 1989). The geographic distribution of CCHF, as presently known, is related closely to sporadic outbreaks of human disease. Where human outbreaks have not been recognized, little or no information exists concerning CCHF maintenance or transmission. Presumably CCHF virus circulates between ticks and human or non-human vertebrates in nearly all of the countries in which the tick and vertebrate fauna and ecological conditions are similar to those in countries with known CCHF viral activity.

In the northern distribution of CCHF, in Eurasia, virus transmission to humans normally occurs from April to November, with very few cases occuring in the winter months. The time of CCHF viral activity coincides with an increase in the tick population density and adult tick activity (Hoogstraal, 1979). In the temperate and tropical extremes where CCHF occurs, the reported cases are

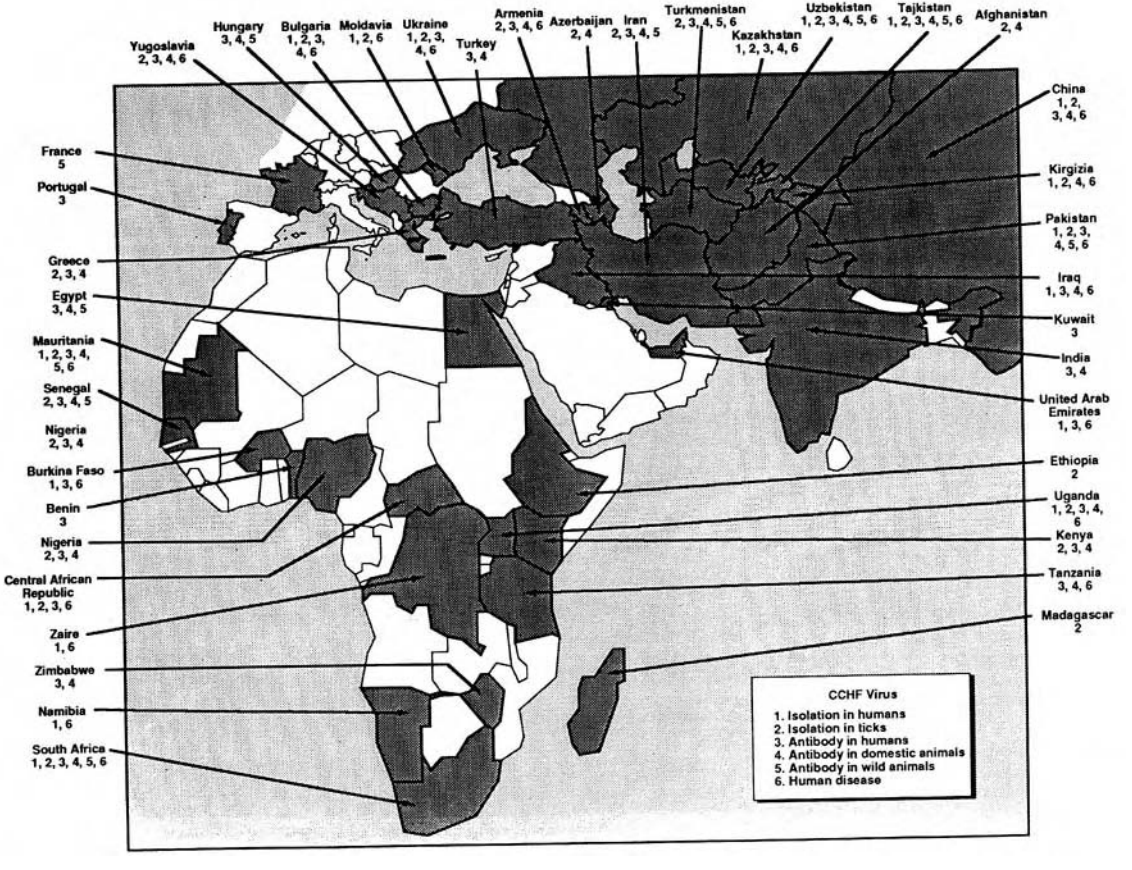

Fig. 13.2. The geographic distribution of CCHF viral isolates and antibody and human disease.

sporadic, and the time of transmission with respect to seasonality is not always clear. Cases of CCHF occurred in Iraq during 1979–1980, in September through November and April through June (Casals, 1981). In the southern-most range of CCHF, more human cases occurred in South Africa in the spring and summer, which coincided with the time of peak activity of the *Hyalomma* adults (Rechav, 1986). In tropical Africa, potential vector ticks have seasonal population peaks corresponding to occurrence of rainfall. However, human disease is far too sporadic to predict peak transmission.

2.2. Risk Factors

People (in endemic foci) susceptible to tick bite, particularly from *Hyalomma* spp., and persons who come in contact with CCHF patients are at the highest risk of infection. People who work outdoors, particularly those who work with domestic animals, are at increased risk of tick bites or of crushing infected ticks. In an enzootic area in rural northern Senegal, Chapman et al. (1991) found that male risk factors included herding, sleeping outside during seasonal animal migrations, bite by *H. truncatum*, tick bite during the cool dry season, and contact with sick animals. Those people butchering animals, caring for CCHF patients, and laboratory workers handling virus material are at risk of contacting CCHF virus. Excluding nosocomial infections, in which age and sex do not appear to be factors in CCHF-virus susceptibility, there is an unequal distribution between males and females based upon their differential exposure to CCHF virus. More males than females, 10–49 years old, became infected in South Africa (Gear, 1982; Gear et al., 1982; Swanepoel et al., 1983, 1985a, 1985b, 1987; Van Eeden et al., 1985).

The risk of nosocomial infection in health-care workers is great, especially during the hemorrhagic period of disease. In Pakistan in 1976, 83% ($n = 6$) of exposed health-care and 46% ($n = 26$) of all potentially exposed persons, including family members, contracted CCHF (Burney et al., 1980).

CCHF infection in laboratory workers was documented in Africa in Uganda (Simpson et al., 1967; Kalunda et al., 1983), Zaire, and the Central Africa Republic (Robin, 1977). Risk associated with contact with infected animals has been well documented throughout the geographic range of CCHF. Possible aerosol transmission is suspected but not confirmed.

Although CCHF virus has been isolated from numerous species of ticks, *Hyalomma* spp. are considered the primary vectors in all CCHF enzootic areas except an environmentally altered focus in Moldavia. Risk areas are those where one or more species of *Hyalomma* occurs. In Rostov Oblast, Russia, the risk of human infection is directly proportional to the rate of attachment of *H. marginatum* on humans (Goldfarb et al., 1980). The distribution of CCHF virus in Eurasia and Africa coincide precisely with the distribution of *Hyalomma* ticks (Hoogstraal, 1956). There appears to be no risk in areas outside the known distribution of *Hyalomma* spp. Within the limits of the zoogeography of *Hyalomma* ticks, specimens are found clustered within specific ecological zones.

It is within these ecological zones that risk of CCHF infection is high. This topic is discussed further in Section 3.1 on environmental conditions.

2.3. Transmission and Maintenance Cycles

2.3.1. Field Studies: Vectors

As early as 1944, *Hyalomma* spp. were implicated in the ecology of CCHF, based upon a relationship between clinical CCHF cases and tick bite. Later, the association was noted among the distribution, population density, and activity of *Hyalomma* ticks and the incidence of CCHF disease. Since 1967, when the development of laboratory techniques led to the isolation and identification of CCHF virus, the virus has been found in 31 different species of ticks collected in Africa, Europe, the Middle East, Asia, and Eurasia (Table 13.1). The virus has been isolated from 12 *Hyalomma*, eight *Rhipicephalus*, four *Boophilus*, and two *Dermacentor*; and one each *Argas*, *Ornithodoros*, *Haemaphysalis*, *Amblyomma*, and *Ixodes* species. CCHF virus has been isolated from approximately 33% of the countries within the geographic range of the virus (Fig. 13.2). By country, CCHF virus has been isolated most often in countries of Eurasia (73%), followed by Europe and the Middle East (37%), with the least number being isolated from countries in Africa (20%). The paucity of isolations from Africa may be an artifact related to the lack of reported human cases in most of the continent. The virus has been isolated from *Hyalomma*, *Rhipicephalus*, and *Boophilus* spp. in Africa, Europe, and Eurasia. However, virus isolations from *Dermacentor* and *Ixodes* have been made from Europe and Eurasia only. Although CCHF viral activity has been found in a wide diversity of ecological habitats in three faunal regions (Palearctic, Oriental, Ethiopian) and in a wide variety of tick species with different life cycles, isolations from *Hyalomma* spp. appear to be the common factor in all areas of viral activity (Fig. 13.3). Of the 18 non-*Hyalomma* spp. from which CCHF virus has been isolated, 17 are known to feed in the same habitats and on the same hosts as the 12 species of *Hyalomma* spp. incriminated in CCHF transmission.

Of the 31 species from which CCHF virus has been isolated, there are 16 three-host, nine two-host, four one-host, and two multi-host species. The role that a tick's life cycle can play in virus transmission is discussed in Section 3.3.5 on life cycle.

Field studies have demonstrated that long-term survival of CCHF virus is possible through transstadial or transovarial transmission of the virus in ticks. Chumakov (1965, 1972) reported finding CCHF virus in unfed *H. marginatum* nymphs and females in the Crimea (Ukraine), and in the Rostov and Astrakhan Oblasts (Russia) in the spring. Isolation of CCHF virus has also been made from unfed *H. anatolicum* adults in Tadzakistan in the spring (Pak et al., 1974, 1975). Because these ticks were unfed, they must have acquired the virus transovarially and then passed the virus transstadially.

Transovarial transmission has been documented in field studies in two tick

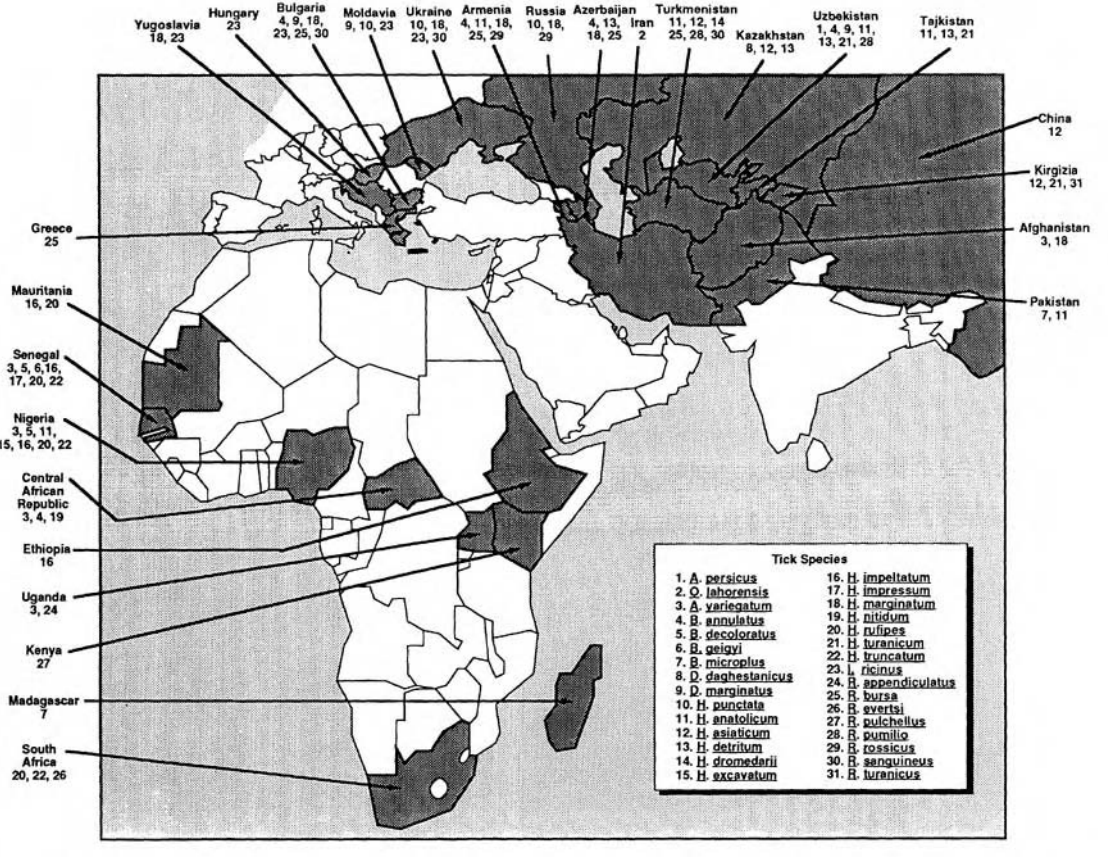

Fig. 13.3. The geographic distribution of isolations of CCHF virus by tick species.

species in Eurasia by isolating CCHF virus in eggs of field-collected *D. marginatus* and *H. marginatum* (Kondratenko et al., 1970). Wilson et al. (personal communication) found CCHF viral antigen in the progeny of *H. truncatum* and *H. impeltatum* adults collected in Senegal. Evidence of transoviral transmission based upon detecting viral antigen only in progeny should be viewed with caution. Logan et al. (1993) demonstrated in laboratory studies that CCHF viral antigen can be detected in the progeny of infected *H. truncatum* in the absence of live virus. Pak (1975) concluded that transoviral transmission of CCHF virus is not important in the epidemiology of the virus in Tadzhikistan after he failed to find the virus in the progeny of field-collected *H. anatolicum* and *H. detritum.*

No CCHF virus was isolated from 25,000 (six species) mosquitoes and sentinel animals from an enzootic foci in Astrakhan in 1967–1969 (Chumakov et al., 1972). CCHF virus was isolated from 1/377 pools of *Culicoides* collected in Nigeria (Causey et al., 1970); however, these specimens may have contained recently ingested vertebrate blood and are not considered to be evidence of infection.

Because of the technological reasons described in Section 1.4, studies on CCHF virus ecology are difficult to conduct. Efforts to maintain surveillance of CCHF infection in ticks have continued to decline in recent years in all geographic areas, and few laboratories in Africa and some regions of Eurasia have the capability of isolating the virus. This trend is expected to continue, owing to a decline in interest and funding of infectious diseases in developing countries, and also to a dramatic decrease in personnel trained to conduct field studies and co-ordinate laboratory studies. The wide range of tick species from which CCHF virus has been isolated, the diversity of life cycles in these ticks, the the wide range of ecological habitats for these ticks, and the uncertain involvement of various vertebrates have contributed to a poor understanding of the complex transmission and maintenance cycles. A hypothesized life cycle for CCHF virus in one-, two-, and three-host tick species is shown in Fig. 13.4.

2.3.2. Field Studies: Vertebrates

Transstadial and transoviral and transmission of CCHF virus are important mechanisms in the survival of the virus between seasons; however, the maintenance of enzootic CCHF virus circulation and the survival of the tick vector depends upon the association between ticks and vertebrates. The precise role of vertebrates in CCHF virus ecology is not well understood. Humans, domestic, and wild animals can become infected but only humans develop overt disease. Human to human transmission has been documented; however, it is not known whether ticks can become infected while feeding on viremic humans. Anti-CCHF antibodies are found in human populations sampled in various countries in Europe, Eurasia, and the Middle East, with the highest antibody rates (29%) reported from animal breeders and persons in contact with CCHF patients in Iraq (Fig. 13.2). In Africa, antibody to CCHF virus in humans occurs in Nigeria (10%) and the Republic of South Africa (up to 7%) (Watts et al.,

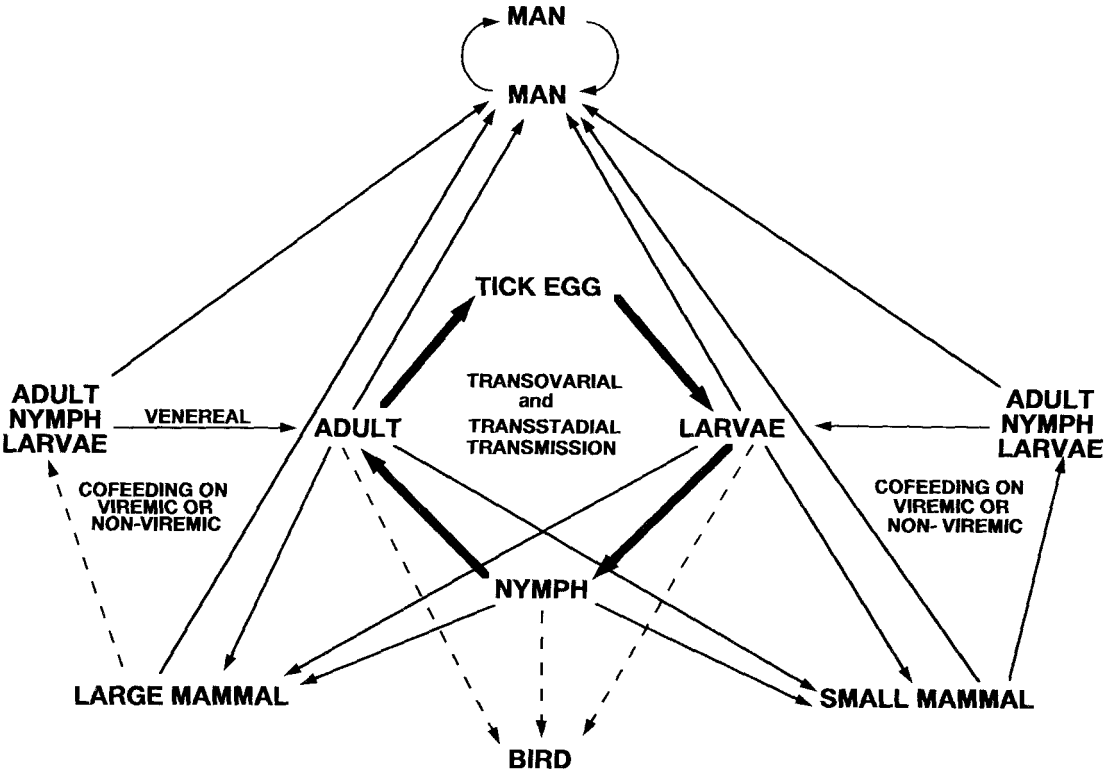

Fig. 13.4. Hypothesized maintenance and transmission cycle of CCHF virus involving *Hyalomma truncatum* and associated vertebrate hosts. ——— = Virus transmission cycle demonstrated by field and laboratory observations; – – – – – = unconfirmed or unknown portions of the cycle.

1989). Recently, Chapman et al. (1991) found that 13.1% of 283 persons living in rural northern Senegal had IgG antibody to CCHF virus.

CCHF virus has been isolated from a few domestic and wild animal species (see Section 1.4.2 on vertebrate hosts). Field data are primarily restricted to serosurveys in areas where disease is thought to be enzootic. These data have been reviewed in detail by Watts et al. (1989).

The high infection rates found in domestic animals in many countries within the range of CCHF virus suggest that these animals are, at least, an important part of the ecology of the tick vectors as a source of blood-meals. It is not known whether domestic animals are an important source of CCHF infection in ticks. A high prevalence of antibody to CCHF virus has been found in cattle in South Africa and Zimbabwe (Swanepoel et al., 1987). In Senegal, Wilson et al. (1990) found that the prevalence of IgG antibody to CCHF virus was the highest in sheep sampled in the northern, arid Sahelian zone (75.7%), and decreased to 0% in the moister Sudano-Guinean and Guinean zones of southern Senegal. *Hyalomma* ticks predominated in the ecological areas where antibody prevalence was highest. Laboratory studies demonstrate that ticks can become infected while feeding on viremic domestic animals (see Section 2.3.3). Morrill et al. (1990) did not find antibodies to CCHF virus in 400 sheep and 200 cattle in Egypt; however, they did detect antibodies in 26% of the camels imported from Kenya and 12% of those imported from Sudan. In South Africa, Shepherd et al. (1987b) found antibodies to CCHF virus in 6% ($n = 1{,}978$) of domestic dogs.

Small mammals appear to have the greatest potential to contribute to maintenance and transmission of CCHF virus in Europe, Eurasia, and the Middle East; however, as discussed below, large herbivores in Africa may also play an important role in regions where they are numerous. Viremia has been demonstrated in several small mammal species, including hares in the republics of the former USSR, hedgehogs in Nigeria (Causey et al., 1970), and a multimammate mouse in the Central African Republic (Digoutte and Heme, 1985). Serological evidence from wild animals shows that numerous species of wild vertebrates are infected with CCHF virus in nature. Watts et al. (1989) lists the following wild vertebrates that had detectable antibody to CCHF virus: hares (*Lepus capensis, L. europaeus, L. saxgtillus*); common red fox (*Vulpes vulpes*); great, and naked-soled gerbil (*Rhombomys o. opimus, Tatera indica, M. hurrianae*); ground squirrel (*Xerus inauris*) and long-clawed ground squirrel (*Spermophilopsis l. leptodacylous*; long-eared hedgehog (*Hemiechinus auritus*); pallas cat (*Felis manual*); Swinhoe's (*Meriones crassus swinhoei*) and red-tailed (*Meriones libycus*) jird; black (*Rattus rattus*), field (*Arvicanthus n. niloticus*), multimammate (*Praomys* (*Mastomys*) *natalensis*), and Norway rat (*R. norvegicus*); common noctule (*Nyctalus n. noctula*); house mouse (*Mus musculus bactrianus*); large mouse-earned bat (*Myotis blythi omari*); Williams's jerboa (*Allactaga euphatica williamsi*); multimammate mouse (*Mastomys erythroleucus*); Genet cat (*Genetta g. senegalensis*); and eland antelope (*Taurotragus orynx*). In South Africa, Shepherd et al. (1987b) found antibody to CCHF virus very prevalent in both large and small mammals. Prevalence rates of antibody to CCHF virus

in large mammals was as follows: 100% ($n = 3$) of giraffe (*Giraffa cameleopardalis*); 54% ($n = 13$) of rhinoceros *Ceratotherium simium* and *Diceros bicornis*); 43% ($n = 137$) of eland (*I. oryx*); 20% ($n = 287$) of buffalo (*Syncerus caffer*); 22⩽ ($n = 87$) of kudu (*Tragelaphus strepsiceros*); and 17% ($n = 93$) of zebra (*Equus burchelli*). In small animals, antibody to CCHF virus was most prevalent in hares (14%, $n = 293$) followed by rodents (1.7%, $n = 1{,}305$), and wild carnivores (1.4%, $n = 74$). No antibodies to CCHF virus were found in primates ($n = 522$), insectivores ($n = 176$), and hyrax ($n = 19$). They concluded that *Hyalomma* spp. are the principal CCHF vectors in nature because hares and large herbivores, the hosts of immature and adult *Hyalomma* ticks, respectively, had the highest prevalence of antibody to CCHF virus. *H. truncatum*, *Dermacentor rhinocerinus*, and *Amblyomma sparsum* have been found on black rhinoceroses imported into Texas from South Africa (USDA, 1989c).

The role of birds in the ecology of CCHF virus has been discussed by Hoogstraal (1979). Various species of birds are important hosts for the immature stages of many *Hyalomma* spp. There is strong evidence against avian involvement in enzootic areas of Russia. CCHF virus was isolated from engorged *H. marginatum* nymphs taken from rooks (*Corvus frugilegus*) while the birds were serologically negative for antibody to CCHF virus. Experimental infection of birds is discussed in the Section 2.3.4 on laboratory studies of vertebrates. In an enzootic area of South Africa, Shepherd et al. (1987a) found reversed passive hemagglutination-inhibition antibodies to CCHF virus in the sera of 24% ($n = 92$) of ostriches, but not in the sera of 37 other bird species ($n = 460$ birds). This study raises the significance for determining the importance of ostrich involvement in CCHF virus ecology. In 1989, *A. gemma* and *Hyalomma* species were found on ostriches in Illinois and Texas, USA, that were imported from Tanzania (USDA, 1989a, 1989b).

2.3.3. Laboratory Studies: Vectors

There are limited studies on the role of ticks to serve as vectors and reservoirs of CCHF virus. The most important vectors to man are species of *Hyalomma*, although species of certain other ixodid genera are susceptible to infection and probably contribute in a subsidiary manner. Experimental data from these studies suggest that many ixodid ticks can be infected and transmit CCHF virus; however, argasid ticks do not appear to become infected. Table 13.2 lists species for which experimental studies on CCHF viral infection have been attempted. CCHF viral infection has been demonstrated for the following species feeding upon viremic vertebrates: *Amblyomma hebraeum* (Shepherd et al., 1991); *H. marginatum* (Zgurskaya et al., 1971; Levi and Vasilenko, 1972; Blagoveschchenskaya et al., 1975; Kondratenko, 1976); *H. truncatum* (Logan et al., 1989a; Shepherd et al., 1991); *H. rufipes* (Shepherd et al., 1991); *R. appendiculatus* (Shepherd et al., 1991); *R. evertsi evertsi* (Shepherd et al., 1991); *R. rossicus* (Kondratenko, 1976); *D. marginatus* (Kondratenko, 1976); and *B. decoloratus* (Shephard et al., 1991). CCHF virus disappeared in *Ornithodoros sonrai* 24 h after it ingested $10^{2.5}$ plaque-forming units (PFU) while feeding

on a viremic suckling mouse (Durden et al., 1993). Shepherd et al. (1991) were not able to infect adult *H. rufipes* fed on viremic cattle (virus titer = $10^{1.5-2.7}$ LD_{50}/ml), larvae, and nymphal *R. evertsi evertsi* on viremic scrub hares (virus titer = $10^{2.6-4.2}$ LD_{50}/ml), nor any larval and nymphal *A. hebraeum*, *H. truncatum*, and *R. evertsi mimeticus* fed on viremic guinea pigs and white-tailed rats. Adult *A. hebraeum*, *H. rufipes*, *H. truncatum*, *R. e. evertsi*, *R. appendiculatus*, and *R. simus* did not become infected after feeding on a viremic sheep (virus titer = $10^{2.5-3.2}$ LD_{50}/ml). Wilson et al. (1991) were able to infect *H. truncatum* adults on sheep inoculated with 5×10^6 LD_{50} of a Mauritanian strain of CCHF. Ticks were infected on sheep regardless of whether or not the sheep developed viremia.

CCHF viral infection was demonstrated in adult *H. rufipes* (Lee and Kemp, 1970; Okorie, 1980, 1991; Okorie and Fabiyi, 1980). *H. dromedarii* (Logan et al., 1990), *H. impeltatum* (Logan et al., 1990), *H. truncatum* (Logal et al., 1990; Gonzalez et al., 1991), *R. appendiculatus* (Logan et al., 1990), and *A. variegatum* (Okorie, 1980, 1991; Gonzalez et al., 1991) after intracoelomic inoculation of CCHF virus. Engorged nymphs of *H. rufipes*, *H. truncatum*, and *R. e. evertsi* became infected after intracoelomic inoculation (Shepherd et al., 1989b). Transmission of CCHF virus to a vertebrate host has been demonstrated for all these species. Okorie (1991) reported interesting studies on the intracoelomic inoculation of immature and adult *H. rufipes* and *A. variegatum* with different titers of CCHF virus. After inoculation with the highest dose (virus titer = $10^{3.5}$ mice LD_{50}) 95%, 100%, and 87% of larval, nymphal, and adult *A. variegatum*, respectively, became infected; and CCHF virus was transmitted transstadially and horizontally to rabbits used as hosts. At the same virus dose, infection rates were 100% and 92% for inoculated nymphal and adult *H. rufipes*, respectively, and the virus was transmitted transstadially and horizontally. CCHF did not replicate in *Argas walkerae*, *O. porcinus*, or *O. savignyi* after intracoelomic inoculation of virus (Shepherd et al., 1989b).

Larvae of *H. truncatum* and *H. impeltatum* were infected while co-feeding with corresponding CCHF virus-infected adults on non-viremic guinea pigs. CCHF virus was transstadially transmitted to nymphs and adults, and horizontally to guinea pigs serving as host of nymphs and adults (Gordon et al., 1993). This phenomenon has also been observed in uninfected female *H. truncatum* co-feeding with infected males on rabbits (Gonzalez et al., 1992). Gonzalez et al. (1992) also observed venereal transmission of CCHF virus from male to female *H. truncatum*.

Experimental studies on the role of transovarial transmission in infected ticks are not conclusive and may depend upon the strain of CCHF virus used in the studies (Table 13.2). In ticks orally infected with CCHF virus, transovarial transmission of virus to progeny was demonstrated for *H. marginatum*, *R. rossicus*, and *D. marginatus* in the former Soviet Union (Kondratenko, 1976). Other studies have failed to demonstrate transovarial transmission in *H. truncatum* (Logan et al., 1989a) after oral infection. Lee and Kemp (1970) described transovarial transmission in the larval progeny of adults infected transstadially from intracoelomically inoculated *H. rufipes* nymphs. In parentally

Table 13.2. Ticks experimentally exposed to CCHF virus

Species	Stage	Method of exposure	Infection[a]	Transmission to	Reference
H. dromedarii Koch	Adult	Intracoelomic inoculation	+	Guinea pigs	Logan et al. (1990)
H. impeltatum Schulze and Schlottke	Larva	Co-fed with infected adults on a non-viremic guinea pig	+	Nymphs, adults, guinea pig	Gordon et al. (1993)
	Nymph	Co-fed with infected adults on a non-viremic guinea pig	+	Adults, guinea pig	Gordon et al. (1993)
	Adult	Intracoelomic inoculation	+	Guinea pigs	Logan et al. (1990)
H. m. marginatum Koch	Larva	Viremic European hare, long-eared hedgehog	+	Nymphs, adults, rabbits, guinea pigs, F_1 larvae and nymphs	Zgurskaya et al. (1971)
	Larva	Viremic little suslik	+	Nymphs, adults	Kondratenko (1976)
	Larva	Viremic Belgian hare	+	F_1 larvae	Levi and Vasilenko (1972), Zarubinsky et al. (1976)
	Adult	Viremic little suslik, rabbit	+	F_1, F_2 larvae, nymphs, and adults, little suslik, rabbits	Kondratenko (1976)
H. m. rufipes Koch	Larva	Viremic scrub hare	+	Nymphs, adults, sheep	Shepherd et al. (1991)
	Nymph	Intracoelomic inoculation	+	Adults, sheep	Shepherd et al. (1989a, b)
	Nymph	Intracoelomic inoculation	+	Calf, adults, F_1 larvae	Lee and Kemp (1970)
	Nymph	Viremic scrub hare	+	Adults, sheep	Shepherd et al. (1991)
	Nymph	Intracoelomic inoculation	+	Rabbits	Okorie (1991)
	Adult	Intracoelomic inoculation	+	Rabbits	Okorie (1980, 1992), Okorie and Fabiya (1880)
	Adult	Viremic cow	+	—	Causey et al. (1970)
	Adult	Viremic sheep	−	—	Shepherd et al. (1991)

Species	Stage	Method of exposure	Infection[a]	Transmission to	Reference
	Adult	Viremic cattle	–	—	Shepherd et al. (1991)
H. truncatum Koch	Larva	Viremic suckling mouse	+	Nymphs, adults	Logan et al. (1989a, b)
	Larva	Viremic scrub hare	+	Adults	Shepherd et al. (1991)
	Larva	Viremic guinea pigs	–	—	Shepherd et al. (1991)
	Larva	Viremic white-tailed rat	–	—	Shepherd et al. (1991)
	Larva	Viremic Co-fed with infected adults on a non-viremic guinea pig	+	Nymphs, adults, guinea pig	Gordon et al. (1993)
	Nymph	Intracoelomic inoculation	+	Adults, sheep	Shepherd et al. (1989)
	Nymph	Viremic scrub hare	+	Adults	Shepherd et al. (1991)
	Nymph	Viremic guinea pigs	–	—	Shepherd et al. (1991)
	Adult	Intracoelomic inoculation	+	Guinea pigs	Logan et al. (1990)
	Adult	Viremic cattle	+	—	Shepherd et al. (1991)
	Adult	Viremic sheep	–	—	Shepherd et al. (1991)
	Adult	Intracoelomic inoculation	+	–	Gonzalez et al. (1991)
	Adult	Viremic sheep	+	F_1 eggs	Wilson et al. (1991)
	Adult	Venereal from infected male tick	+	F_1 eggs, larvae	Gonzalez et al. (1992)
	Adult	Co-fed with infected tick on a rabbit	+	Rabbit	Gonzalez et al. (1992)
R. appendiculatus Neumann	Adult	Intracoelomic inoculation	+	Guinea pigs	Logan et al. (1990)
	Adult	Viremic sheep	–	—	Shepherd et al. (1991)
	Adult	Viremic cattle	+	—	Shepherd et al. (1991)
R. evertsi evertsi Neumann	Larva	Viremic sheep	–	—	Shepherd et al. (1991)
	Larva	Viremic scrub hare	–	—	Shepherd et al. (1991)
	Nymph	Viremic scrub hare	–	—	Shepherd et al. (1991)
	Nymph	Viremic sheep	–	—	Shepherd et al. (1991)
	Adult	Viremic sheep	–	—	Shepherd et al. (1991)
	Adult	Viremic cattle	+	—	Shepherd et al. (1991)

(continued)

Table 13.2. (continued)

Species	Stage	Method of exposure	Infection[a]	Transmission to	Reference
R. evertsi mimeticus Donitz	Larva	Viremic guinea pig	–	—	Shepherd et al. (1991)
	Larva	Viremic white-tailed rat	–	—	Shepherd et al. (1991)
	Nymph	Intracoelomic inoculation	+	Adults, sheep	Shepherd et al. (1989a, b)
R. pulchellus Gerstacker	Adult	Intracoelomic inoculation	+	Guinea pigs	Linthicum et al. (unpublished observations)
R. rossicus Yakimov and Kohl-Yakimova	Nymph	Viremic little suslik	+	Adults	Kondratenko (1976)
	Adult	Viremic little suslik, rabbit	+	F_1, F_2 larvae, nymphs, and adults, little suslik, rabbits	Kondratenko (1976)
	Adult	Viremic calf	–	—	Zarubinsky et al. (1976)
R. simus Koch	Larva	Viremic sheep	–	—	Shepherd et al. (1991)
	Nymph	Viremic sheep	–	—	Shepherd et al. (1991)
	Adult	Viremic sheep	–	—	Shepherd et al. (1991)
A. hebraeum Koch	Larva	Viremic sheep	–	—	Shepherd et al. (1991)
	Larva	Viremic guinea pig	–	—	Shepherd et al. (1991)
	Larva	Viremic white-tailed rat	–	—	Shepherd et al. (1991)
	Nymph	Intracoelomic inoculation	+	Adults, sheep	Shepherd et al. (1991)
	Nymph	Viremic sheep	–	—	Shepherd et al. (1991)
	Nymph	Viremic guinea pig	–	—	Shepherd et al. (1991)
	Adult	Viremic sheep	–	—	Shepherd et al. (1991)
	Adult	Viremic cattle	+	—	Shepherd et al. (1991)

Species	Stage	Method of exposure	Infection[a]	Transmission to	Reference
D. marginatus (Sulzer)	Nymph	Viremic little suslik	+	Adults	Kondratenko (1976)
	Adult	Viremic little suslik, rabbit	+	F_1, F_2 larvae, nymphs, and adults, little suslik, rabbits	Kondratenko (1976)
	Adult	Viremic calf	−	—	Zarubinsky et al. (1976)
A. variegatum (Fabricius)	Adult	Intracoelomic inoculation	+	—	Gonzalez et al. (1991)
B. decoloratus (Koch)	Adult	Viremic cattle	+	—	Shepherd et al. (1991)
Argas walkerae Clifford, Kohls and Hoogstraal	Nymph	Intracoelomic inoculation	−	—	Shepherd et al. (1989a, b)
	Adult	Intracoelomic inoculation	−	—	Shepherd et al. (1989a, b)
O. porcinus Walton	Nymph	Intracoelomic inoculation	−	—	Shepherd et al. (1989a, b)
	Adult	Intracoelomic inoculation	−	—	Shepherd et al. (1989a, b)
O. savignyi (Auduoin)	Nymph	Intracoelomic inoculation	−	—	Shepherd et al. (1989a, b)
	Adult	Intracoelomic inoculation	−	—	Shepherd et al. (1989a, b)
O. sonrai Sautet and Witkowski	Nymph	Viremic suckling mouse	−	—	Durden et al. (1993)
	Adult	Viremic suckling mouse	−	—	Durden et al. (1993)

[a] + = Infection present; − = no infection.

infected adult ticks, transovarial transmission of CCHF virus (strain 10200) was not observed in the following F_1 progeny: 11,068 *H. truncatum*, 17.141 *H. impeltatum*, 13,744 *H. dromedarii*, and 36,400 *R. appendiculatus* (Logan et al., 1990). Transovarial transmission was not found in the F_1 progeny of intracoelomically inoculated *H. rufipes*, *H. truncatum*, and *R. evertsi evertsi* nymphs (Shepherd et al., 1989b). Shepherd et al. (1991) did not detect CCHF virus in the F_1 progeny of infected adult *A. hebraeum*, *B. decoloratus*, *H. rufipes*, *H. truncatum*, *R. appendiculatus*, and *R. evertsi evertsi*. In Senegal, Gonzalez et al. (1992) reported transovarial transmission by *H. truncatum* females infected venereally from males inoculated with a Mauritanian strain (HD49199) of CCHF. Wilson et al. (1991) also reported transovarial transmission in 17% ($n = 17$) of egg batches laid by *H. truncatum* infected while feeding on sheep inoculated with 5×10^6 LD_{50} of the HD49199 strain of CCHF virus.

In experimental studies, transstadial transmission of CCHF virus has been demonstrated consistently in all ticks infected by feeding on a viremic vertebrate.

2.3.4. Laboratory Studies: Vertebrates

Information derived from the relationships between tick vectors and their hosts, and serological data from vertebrates in endemic areas have incriminated certain vertebrates as potential maintenance and transmission hosts of CCHF virus. Experimental data on CCHF viral infection in these potential vertebrates hosts have been confined to a very few studies.

Birds serve as important hosts for the immatures of potential tick vectors. Berezin et al. (1969, 1971) reported that rooks (*Corvus f. frugilegus*) and rock doves (*Columba livea*) experimentally inoculated with CCHF virus did not develop a viremia and did not develop a measurable antibody response. Logan et al. (unpublished observations) also found that virus-inoculated chickens did not develop detectable viremia or antibody. Although Shepherd et al. (1987a) found that chickens were refractory to CCHF viral infection, they found the first evidence that a bird species can become infected with the virus when blue-helmeted guinea fowl (*Numidia meleagris*) developed a viremia of $10^{2.5}$ mouse LD_{50}/ml and a transient antibody response.

Hedgehogs have been incriminated in the transmission of CCHF virus by the isolation of the virus from this species in Nigeria (Causey et al., 1970). Antibody was detected in the same species in South Africa (Shepherd et al., 1987b). Blagoveshchenskaya et al. (1975) experimentally infected the European hedgehog (*E. europaeus*) and the long-eared hedgehog (*Hemiechinus auritus*), and found that only the long-eared hedgehog developed a viremia. CCHF virus was detected in *H. m. maginatum* nymphs that had fed as larvae on infected *H. auritus*. In other studies, Zgurskaya et al. (1971) found that infected European and long-eared hedgehogs could serve as an infectious blood-meal source of CCHF virus for *H. m. marginatum*. South African hedgehogs (*Atelerix frontalis*) were found to develop antibody response to CCHF viral infection but not viremia (Shepherd et al., 1989a).

Hares experimentally infected with CCHF virus developed a viremia that

persisted for more than 15 days (Zgurskaya et al., 1975). Scrub hares (*Lepus saxatilis*) developed a viremia of $10^{1.7-4.2}$ mouse LD_{50}/ml after experimental infection with CCHF virus (Shepherd et al., 1989a). Kondratenko (1976) found that the little suslik (*Citeel*(*Citellus pygmaeus*), when experimentally infected could provide an infectious blood-meal to *H. m. marginatum* for 2–7 days. Shepherd et al. (1989a) found that CCHF virus-infected Cape ground squirrels (*Xerus inauris*), red veld rats (*Aethomys chrysophilus*), white-tailed rats (*Mystromys albicaudatus*) bushveld gerbils (*Tatera leucogaster*), striped mice (*Rhabdomys pumilio*), and guinea pigs developed low-titer viremia followed by development of antibodies. However, CCHF-infected highveld gerbils (*T. brantsii*), Namaqua gerbils (*Desmodillus auricularis*), two species of a multimammate mouse (*Mastomys natalensis*, *M. couchs*), and Syrian hamsters only developed antibody responses.

Logan et al. (1989a) were able to demonstrate that *H. truncatum* larvae could become infected after feeding on an experimentally infected suckling mouse. Gordon et al. (1993) did not observe viremia in guinea pigs on which *H. truncatum* larvae, and *H. impeltatum* larvae and nymphs became infected while co-feeding with adults ticks infected with CCHF virus. Gonzalez et al. (1992) reported virus transmission to *H. truncatum* adult females co-feeding with CCHF virus-infected males on a rabbit.

Causey et al. (1970) demonstrated viremia in two calves infected with a Nigerian strain on CCHF virus. Calves inoculated with a Rostov strain of CCHF did not infect *H. m. marginatum*, *D. marginatus*, and *R. rossicus*, even though virus was detected in at least one of the calves. All inoculated calves had detectable antibodies to CCHF virus.

Simpson et al. (1967) reported that a Uganda strain of CCHF virus could produce viremias in sheep sufficient to infect ticks. Juvenile, but not adult, sheep inoculated with a Mauritanian strain of CCHF virus developed a high viremia; however, ticks became infected on sheep with or without detectable viremia (Wilson et al., 1991).

Milyutin et al. (1969) and Blagoveshchenskaya et al. (1969) reported that horses inoculated with CCHF virus developed only antibodies. Humans experimentally infected with CCHF virus developed disease (Chumakov, 1974). CCHF viremia levels observed in infected humans in South Africa are probably sufficient to infect ticks; however, the role of humans in the maintenance and transmission is not understood (Swanepoel et al., 1987). The ability of a vertebrate to serve as a maintenance or transmission host of CCHF virus may be considerably influenced by the strain of CCHF involved.

2.3.5. Life Cycle of CCHF Virus

Limited experimental and field observations suggest that ticks, primarily *Hyalomma* species, are the principal reservoirs of CCHF virus (Fig. 13.4). The hypothetical life cycle shown in Fig. 13.4 describes a series of complex interrelationships among various *H. truncatum* tick stages, and various small and

large mammal hosts; however, it can also apply to other two-host and three-host *Hyalomma* ticks. In the absence of virus amplification by a vertebrate host, CCHF virus can be maintained for long periods of time in *Hyalomma* ticks and be transmitted transovarially and transstadially (Logan et al., 1989a; Wilson et al., 1991). In addition, while co-feeding with infected ticks on small non-viremic vertebrate hosts, uninfected ticks can become infected during feeding or venereally during mating (Gonzalez et al., 1992; Gordon et al., 1993). Infection during co-feeding on large, non-viremic vertebrates may also occur; however; this has not been demonstrated in laboratory studies. Although transstadial transmission of CCHF virus in *Hyalomma* species appears to be relatively efficient in all strains tested, transovarial transmission is inefficient and appears to occur relatively infrequently (Wilson et al., 1991) or not at all with some strains of CCHF virus studied (Shepherd et al., 1989b; Logan et al., 1990). Infection during co-feeding and mating is also relatively inefficient; however, the number of uninfected ticks potentially exposed in endemic areas is large, as estimated by the number of both small and large vertebrates that have been found to contain antibody to CCHF virus (see Section 2.3.2 on field studies of vertebrates). Ticks are somewhat more efficient in becoming infected while feeding on a viremic vertebrates host as opposed to a non-viremic host (Wilson et al., 1991; Gordon et al., 1993); however, the number of species of vertebrates that develop high viremias is relatively low (see Section 2.3.4 on laboratory studies of vertebrates). In South Africa the scrub hare does appear to function as an amplifying host (Shepherd et al., 1991).

Both transovarial and transstadial transmission of CCHF virus in ticks permits the maintenance of the virus during winter in temperate areas, and during times of tick inactivity or scarcity of hosts in tropical regions. Ticks serve as overwintering hosts in Eurasia, as evidence by the isolation of CCHF virus from unfed nymphs and adult female *H. marginatum* (Chumakov, 1972), and from unfed adult *H. anatolicum* collected in the spring (Pak et al., 1974). Kondratenko (1976) also demonstrated that persistent CCHF viral infection is maintained by *H. marginatum*, *R. rossicus*, and *D. marginatus* under winter temperatures. The recent reports by Wilson et al. (1991) and Gonzalez et al. (1992) of transovarial transmission of CCHF virus by *H. truncatum* provides evidence that this method of transmission might be important in the maintenance of the virus in Africa.

Species other than *Hyalomma*, particularly *D. marginatus*, *I. ricinus*, and several species of *Rhipicephalus*, may be important secondary vectors and possible maintenance hosts of CCHF virus. *Rhipicephalus rossicus* may have replaced *H. marginatum* as the principal vector of CCHF after the severe 1968–1969 winter in the Rostov Oblast, and *I. ricinus*, *D. marginatus*, and *H. punctata* may have replaced *H. marginatum* in Moldavia after a change in farming practices (Watts et al., 1989). The role of soft ticks in CCHF virus ecology is poorly understood. Although CCHF virus has been isolated from two species of soft ticks, experimental studies of other species of soft ticks demonstrated that CCHF virus did not replicate (Shepherd et al., 1989b; Durden et al., 1993). Additional experimental studies on species that are

potential vectors are needed to assess the role of secondary species in the life cycle of CCHF virus.

Other than serving as blood-meal sources for a number of vectors of CCHF, the role of birds in the ecology of the disease is unknown. Experimental studies are inconclusive.

3. ECOLOGY

3.1. Environmental Conditions

The principal environmental factors responsible for the enzootic distribution of CCHF virus within the steppe, savanna, semi-desert, and foothill ecotypes of the Paleartic, Oriental, and Ethiopian faunal regions are those that enhance the prevalence of *Hyalomma* ticks. The only exceptions are in the deciduous forests of Moldavia and on the island of Madagascar. In Moldavia, *I. ricinus*, and *Dermacentor* and *Rhipicephalus* species have replaced *Hyalomma* species, possibly because of changes in the practices of handling cattle herds (Hoogstraal, 1979). In Madagascar, where *Hyalomma* ticks do not occur, the virus has been isolated only from *B. microplus*, and there are no human infections (Mathiot et al., 1988).

The geographic range of *Hyalomma* ticks extends eastward through India, ending in the fauna of Burma, where the CCHF virus activity also apparently ends. In enzootic foci, *Hyalomma* ticks appear to be restricted to lowlands, foothills, and low mountain belts with arid to semi-arid climates, often with long dry seasons. These areas include deserts, semi-deserts, and steppes in Eurasia, and tropical savanna and grasslands, arid grasslands, and the Sahalian and desert regions in Africa. In a given region, CCHF virus transmission is greatest in limited foci where climatic factors and/or environmental changes are conducive to the survival of *Hyalomma* ticks and their hosts. These areas may be along river floodplains with rich grasslands (Astrakhan Oblast), shrub and tree vegetation (Uzbekistan), desert or semi-desert, sparse forests scattered in desert and (Kazakhstan), and forests and thickets of rough steppe lands (Rostov Oblast) (Watts et al., 1989). Various changes in environmental factors such as those associated with the war-time neglect of agricultural lands and the introduction of susceptible military personnel into disease foci have significantly increased CCHF viral transmission (Hoogstraal, 1979). More gradual environmental changes such as wide-scale changes in agricultural practices, changing pasture patterns, conversion of flood plains and marshy deltas to farmland and pastures, and flood control measures can create new habitats for vectors and hosts, and lead to increased disease transmission.

3.2. Meteorological Influences

In temperate regions the foci of enzootic CCHF are typically in areas characterized by warm summers and relatively mild winters (Hoogstraal, 1979).

In Eurasia and northern Africa, these areas can be arid deserts and semi-deserts, and in South Africa semi-arid high-altitude regions. In tropical areas in Africa, CCHF enzootic areas can range from wet Central African forests to the very sparsely vegetated, arid Sahelian areas of West Africa, and the semi-arid high-altitude scrub vegetation areas of East Africa.

In Eurasia, CCHF foci are found in the transitional atmospheric humidity zone between the forest-steppe and desert in which the sum of the effective annual temperature above 10°C is between 2,800 and 5,000°C (Watts et al., 1989). In the lowland desert and semi-desert area of southern Tadzhikistan, enzootic foci are in the lowland desert and semi-desert area of southern Tadzhikistan, where the sum of annual temperatures ranges from 3,000°C to as high as 6,000°C in warm years. In southern Tadzhikistan, the annual rains range from 150 to 300 mm.

Hyalomma marginatum, the primary tick vector in Eurasia, cannot survive when winter temperatures fall below a monthly mean temperature of −20°C (Hoogstraal, 1979). A decline in vector populations leads to a decline in CCHF viral activity. In the winter of 1968–1969, temperatures dropped to −30°C and remained at −20°C or lower for more thah 2 months, and the ground froze to a depth of 1 m in the enzootic area of the Republic of Kirzig. As a consequence, the adult tick abundance index per cow fell from approximately 20 in 1968 to less than 0.1 in May 1969, and no CCHF cases were reported during the summer of 1969. CCHF virus did remain enzootic in two species (*R. rossicus* and *D. marginatus*) that are better adapted at surviving at cold temperatures than *H. marginatum* (Watts et al., 1989).

In the Middle East and Africa, the climatological factors affecting populations of vector ticks and CCHF virus transmission are not well studied. In Mauritania, Saluzzo et al. (1986) reported that the geographic distribution of CCHF virus closely fits the distribution of *H. m. rufipes*. *Hyalomma marginatum rufipes* (and sometimes *H. impeltatum*) is commonly infected with CCHF virus in southern Mauritania; however, it is replaced in the very arid semi-desertic regions of northern Mauritania by *Hyalomma dromedarii*, which is not infected with the virus *H. dromedarii* is thought to be very well adapted to the desert environment (Hoogstraal, 1956), and able to withstand low humidity and extremes in temperatures (Delpy and Gouchey, 1937). Hagras and Khalil (1988) found that oviposition in *H. dromedarii* at 75% relative humidity is just as efficient at 34°C as it is at lower temperatures. Presumably *H. rufipes* is not so well adapted to xeric conditions. In a serosurvey of cattle herds in South Africa and Zimbabwe, Swanepoel et al. (1987) reported that antibody prevalence to CCHF virus was higher in areas with higher densities of *Hyalomma* species. Antibodies to CCHF virus were absent from most herds along the southern coast, and there was a tendency for the proportion of positive herds and positive cattle within herds to increase northwards to Zimbabwe, where there were abundant tick populations. Similar findings have been reported for wild mammals in South Africa by Shepherd et al. (1987b). The climate along the coast where *Hyalomma* ticks are less abundant is defined as dry sub-tropical with hot, dry summers and cool, moderately rainy winters (The New Inter-

national Atlas, 1986). In more northern South Africa and Zimbabwe, where *Hyalomma* tick populations are high, the climate is defined by The New International Atlas (1986) as semi-arid tropical characterized by light precipitation, rapid evaporation, and always warm or hot.

Walker (1974) found that in Kenya, *Hyalomma* species are usually found in dry woodland, bushland, and wooded and/or bushed grassland in Ecological Zones IV, V, and VI, as defined by Pratt et al. (1966). These areas range from semi-arid to very arid with annual rainfall ranging from 250 to 750 inches. Only *H. truncatum* is found (rarely) in the wetter Ecological Zones II and III.

In Uganda, *H. m. rufipes* is usually collected from locations with an annual rainfall of 500–1,000 mm with a 4–7-month continuous dry season (Matthysse and Colbo, 1987). Yeoman and Walker (1967) reported that *H. m. rufipes* is usually found in areas with an annual rainfall of 380–900 mm in Tanzania. In west Africa, it is found in areas with a mean annual rainfall of 150–750 mm and it usually disappears in regions with rainfall above 1,250 mm, with the exception of southern Senegal and western Guinea, which have a very intense rainy season (up to 2,000 mm) followed by a long and severe dry season (Morel, 1969).

Hyalomma truncatum is normally found in the dry savanna and steppe regions of Africa characterized by *Combretum* and *Acacia* species of vegetation, and is more restricted to drier regions than *H. m. rufipes* (Theiler, 1964). In Uganda, it is restricted to regions of long continuous dry seasons over 3–7 months long, and with a mean annual rainfall of 650–1,300 mm (Matthysse and Colbo, 1987). In Tanzania, it is found in areas with an annual rainfall of 650–1,500 mm (Yeoman and Walker, 1967). In Kenya, it can be found in areas of less than 250 mm of rainfall (Walker, 1974).

In Africa, *H. impeltatum* also occurs in very dry regions. In Kenya, it is reported to occur in areas receiving less than 250 mm of rainfall per year (Walker, 1974). Yeoman and Walker (1967) found it occurred where the annual mean rainfall was 500–750 mm.

The occurrence of *Hyalomma* species and other potential tick vectors of CCHF virus in restricted ecological habitats determined by reasonably well-defined meteorological parameters makes it possible to map and predict the occurrence of vector species by using vegetation index data produced by National Oceanic Atmospheric Administration meteorological satellites (Linthicum et al., unpublished observations). Similar techniques have been used to estimate the distribution and abundance of *R. appendiculatus* in East Africa (Perry et al., 1990, 1981). Daniel and Kolar (1990) also demonstrated that the occurrence of *I. ricinus* in Czechoslovakia can be forecasted by using Multispectral Scanner data from the Landsat 5 satellite to detect vegetation types associated with the tick.

3.3. Biology of Ticks

The biology of tick species incriminated in the ecology of CCHF virus is poorly known, if at all. Most of what is known has been determined from studies in the laboratory with very little field research completed.

3.3.1. Oviposition and Fecundity

Species of the family Ixodidae (hard ticks) oviposit once in a lifetime. Eggs are laid without regard to location at one time approximately 1–3 weeks after detaching from the host (Logan et al., 1989b; Linthicum et al., 1991).

The period of oviposition in *H. impeltatum* can range from 16 to 50 days (Logal et al., 1989b). Usually, numerous eggs are laid, although the number can vary tremendously. Nuttall (1915) found that a *H. marginatum* female can lay from 4,300 to 15,500 eggs, while Knight et al. (1978) found that *H. m. rufipes* females can lay from 3,184 to 13,180 eggs. *Hyalomma impeltatum* females can lay from 713 to 15,904 eggs with a mean hatch rate of 84% (Logan et al., 1989b). *Hyalomma truncatum* females laid from 4,434 to 8,210 eggs with a mean hatch rate of 48% (Linthicum et al., 1991).

The conversion efficiency index (g eggs/g female) of 56% for *H. truncatum* (Linthicum et al., 1991) was lower than that reported for *H. dromedarii* (Hagras and Khalil, 1988) but equal to that for *H. impeltatum* (Logan et al., 1989b) and *H. schulzei* (Al-Asgah, 1992). The reproduction efficiency index (number eggs/g female) of 12,614 for *H. truncatum* is higher than that reported for *H. asiaticum* (Balashov, 1968) and *H. schulzei* (Al-Asgah, 1992), but equivalent to that for *H. impeltatum* (Logan et al., 1989b). Oviposition parameters measured in laboratory studies may be affected by many factors, including the host species used in the studies and/or the environmental conditions of the ticks while they are free living. Oviposition may be delayed or prevented during cold periods.

Soft ticks of the family Argasidae (*Argas* and *Ornithodoros*) oviposit at intervals in small numbers, usually laying only a few hundred, after each of several bloodmeals (Hoogstraal, 1985). Oviposition and mating occurs off the host in areas where females remain free living. Hooker et al. (1912) found that female *A. persicus* oviposit up to 6–7 times in a lifetime. Oviposition starts 3–10 days after feeding in the summer and 195–646 eggs are laid after the first blood-meal, with decreasing numbers after subsequent blood-meals. Oviposition can be delayed for weeks or months in the winter. Most soft ticks are highly adapted for survival in dry habitats. Eggs of *A. persicus* have a great tolerance to fluctuating climatic factors and *O. moubata* envelops its eggs with a particularly desiccation-resistant waxy, waterproof coating that allows the eggs to withstand very dry conditions.

Ornithodoros lahorensis is very unusual among the soft ticks in that it has a two-host life cycle (Hoogstraal, 1985). Unfed adult females can lay two viable egg batches of 300–500 eggs They need to feed to develop the third and subsequent egg batches. Eggs incubate for 2 to 6 weeks before hatching.

3.3.2. Fecundity

The overall fecundity of a tick population in a given year can be expressed as a product of the total number of eggs oviposited by individual females of a generation that successfully hatch and the number of generations completed during that year. As discussed in Section 3.3.1 on oviposition and fecundity,

the number of eggs oviposited by individual females of different species can vary greatly. In general, the Argasidae lay many fewer eggs than do the Ixodidae.

In nature, the shortest time required to complete a tick generation may be 4 or 5 months; however, climatic conditions and host availability may increase generation time and decrease the overall fecundity of a given population. Under optimal conditions in a laboratory, both *H. impeltatum* (Logan et al., 1989b) and *H. truncatum* (Linthicum et al., 1991) required 108 days to complete their life cycles. Rechav (1986) reported that *H. rufipes* and *H. truncatum* complete two generations per year in the western Transvaal of South Africa. Minshull (1981), however, found that *H. m. rufipes* only completes one generation per year in southeastern Zimbabwe. Hoogstraal (1979) reported that *H. m. marginatum*, *H. truncatum*, and other *Hyalomma* species in Eurasia complete only one generation per year because of the severe winter climate.

3.3.3. Density

Epidemics of CCHF coincide with an elevated population density and increased feeding of *Hyalomma* ticks. During the outbreaks in Bulgaria, adult *H. marginatum*, the principal vector, appeared, reached maximum population densities, and declined in close association with the occurrence of human cases, peak numbers of cases, and decreasing number of cases, respectively (Hoogstraal, 1979). In the Central Asian republics of the former USSR, the seasonal distribution of CCHF cases were closely associated with the seasonal dynamics of *H. anatolicum*, the suspected vector.

In Africa, vector densities exhibit distinct seasonal variation. In Zambia (Pegram et al., 1986) and Zimbabwe (Matson and Norval, 1977), the highest population densities of *H. rufipes* and *H. truncatum* adults occur during the warm seasons. These species have been implicated as vectors of CCHF virus in Mauritania, Nigeria, the Republic of South Africa, and Senegal. Rechav (1986) found that these two species exhibit peak abundance during summer and winter months. Clifford et al. (1976) reported that in Kenya, immature *Hyalomma* species parasitized hares during most of the year; however, peak populations were attained just before the long rainy season (March–June) and then declined significantly during the rainy period. *Hyalomma* and other tick species' abundance and activity can be dramatically affected by temperature, humidity, predators, fire, flooding, and host densities.

3.3.4. Longevity

Adult ticks can survive for very long periods of time, months or years, without blood-feeding, if climate or host populations are not conducive to successful feeding or the survival of immatures. This adaptive behavior maximizes the likelihood of taking a blood-meal and of completing a gonotrophic cycle. Although the long developmental cycle of a tick may also reduce its chances of survival, it provides a stable environment for the maintenance of CCHF

virus when conditions do not favor successful transmission of the virus. CCHF viral infection in ticks could have an effect on the survival of the tick; however, this has yet to be examined by investigators in the field. Nuttall (1915) held *H. marginatum* for more than 800 days without a blood-meal under optimal laboratory conditions. Adult *H. truncatum*, *H. impeltatum*, *H. rufipes*, *A. variegatum*, *R. appendiculatus*, *R. e. evertsi*, *R. pulchellus*, and *R. sanguineus* have been held in the laboratory without blood-feeding for up to 1 year (Linthicum et al., unpublished observations).

3.3.5. Life Cycle

The life cycle of numerous species of ticks either incriminated or potential vectors in the transmission and maintenance of CCHF virus have been studied. The relationship between the life cycle of *H. truncatum* and the transmission and maintenance of CCHF virus is shown in Fig. 13.4. Laboratory investigations of *H. truncatum* ticks include studies on the complete or partial life cycles of *H. aegyptium* (Sweatman, 1968), *H. asiaticum* (Balashov, 1968), *H. detritum* (Pospelova-Shtrom and Pettrova-Piontkovskaya, 1949), *H. dromedarii* (Hagras and Khahil, 1988), *H. excavatum* (Serdyukova, 1946, as *anatolicum*; Rechav, 1986), *H. impeltatum* (Dipeolu, 1983; Logan et al., 1989b), *H. impressum* (Dipeolu, 1983), *H. lusitanicum* (Ouhelli and Pandey, 1986), *H. marginatum* (Nuttall, 1915, as *aegyptium*; Ganiev, 1956, as *plumbeum*; Hueli, 1979), *H. m. rufipes* (Theiler, 1943; Ammah-Attoh, 1966; Knight et al., 1978), *H. schulzei* (Al-Asgah, 1992), *H. truncatum* (Bezuidenhout and Malherbe, 1981; Linthicum et al., 1991), and *H. yakimovi* (Pospelova-Shtrom, 1935). Numerous field studies have also examined different stages in the development of *Hyalomma* ticks and some of these are reviewed by Hoogstraal (1956). Studies on the life cycles of other Ixodidae incriminated in the transmission of CCHF virus include the following species: *A. variegatum* (Garris, 1984), *B. annulatus* (Davey et al., 1980a; Ouhelli et al., 1982), *B. microplus* (Davey et al., 1980b), *D. marginatus* (Honzakova, 1973), *I. ricinus* (Honzakova, 1973), *R. bursa* (Hueli et al., 1986), *R. e. evertsi* (Rechav et al., 1977), and *R. sanguineus* (Srivastava and Varma, 1964; Hafez and Bassal, 1980). The life cycles of *Argas* (*Argas*) and *Argas* (*Persicargas*) are similar and the literature on the life cycle of *A. persicus* is reviewed by Kraiss and Gothe (1982). The two-host life cycle of *O. lahorensis* was studied by Brumpt (1936) and Filippova (1966).

3.3.6. Biting Activity

Seasonal biting activity of ticks in nature is based upon data gathered from ticks questing on vegetation or ticks attached to their hosts. This topic is discussed in Section 3.3.3. In laboratory studies under optimal conditions, Logan et al. (1989b) found that the proportion of the larval and adult female stages spent in the pre-feeding period (the time after molt before responding to a host) was $<5\%$ and $<2\%$, respectively. The pre-feeding period occupied 7.5% of the nymphal stage. Linthicum et al. (1991) found equivalent results for

larval and adult female *H. truncatum*; however, the nymphs' pre-feeding time was shorter. The proportion of time that the larval, nymphal, and adult female stages of *H. impeltatum* spent feeding was 30.6, 17.4, and 13.0%, respectively. These proportions were similar for *H. truncatum*.

3.3.7. Host Preferences

The host preferences of potential tick vectors of CCHF virus are important in understanding the natural ecology of the disease. It is important to understand biological parameters such as number of hosts parasitized by an individual tick during its lifetime, the potential variety of hosts parasitized, and the degree of host specificity. Table 13.1 groups ticks incriminated as vectors of CCHF virus, based upon virus isolation, by the normal number of hosts parasitized during one lifetime. The distinctions made in this table do not always hold true, as under some environmental pressures (i.e., different species of host), the number of hosts required for a given tick to develop can vary.

CCHF virus isolations have been made from one-, two-, three-, and multi-host ticks (Table 13.1). All stages of *Boophilus* species ticks are completed on the same vertebrate host and they rarely attack man (Strickland et al., 1976). The preferred hosts of *B. annulatus* are cattle and, to a lesser extent, horses; they rarely feed on sheep and goats. Attachment to humans and dogs are known, but they are thought to be unsuitable hosts. *Boophilus decoloratus* has almost the same host preference as *B. annulatus* with antelope as the most important wild host. *Boophilus microplus* has a wider host preference that *B. annulatus*. It is found more commonly on goats, sheep, and deer, as well as cattle and horses, than is *B. annulatus*. The role of one-hosts ticks in the ecology of CCHF virus is still unclear; however, the recently demonstrated laboratory importance of non-viremic hosts (such as cattle in some cases) in the transmission of the virus suggests that *Boophilus* species may well be involved in virus amplification.

Two-host ticks start feeding on their host as larvae, molt to nymphs on the same host, and complete nymphal feeding before dropping off the host. After free-living nymphs molt, the adults reattach to another vertebrate host. As described by Hoogstraal (1979), two-host ticks can be regarded as two subgroups based on whether or not they utilize the same or different species of hosts to develop into the immature and adult stages. Species that attach as larvae and adults to the same species of host include *R. bursa*, *R. e. evertsi*, *H. detritum*, *H. anatolicum*, and sometimes *H. truncatum*. *Rhipicephalus bursa* is found on cattle, horses, and other domestic animals (Anastos, 1957). All stages of *R. e. evertsi* normally infest domestic or wild herbivores, but larvae may sometimes infest small mammals such as rodents and hares (Hoogstraal, 1956). *Hyalomma detritum* commonly feed on cattle and horses. The adults feed in the summer and the nymphs undergo a winter diapause (Hoogstraal, 1956). *Hyalomma truncatum* is normally thought to be a three-host tick; however, individual larval ticks have regularly been observed to molt and reattach as nymphs on the same laboratory guinea pig (Linthicum et al., unpublished

observations). Wilson et al. (personal communication) have observed in Senegal that the life cycle of *H. truncatum* depends upon the host on which it feeds. Most individuals will behave like a two-host tick when fed on hares. Tick species in which the immatures and adults feed on two dissimilar host species include *H. m. marginatum*, *H. turanicum*, and *H. m. rufipes*. Larvae and nymphs of *H. m. marginatum* feed primarily on small wild mammals and birds, while adults are commonly found on cattle, horses, sheep, goats, and camels (Hoogstraal, 1956). *Hyalomma marginatum rufipes* adults are most commonly found on domestic cattle but also on horses, sheep, goats, and large wild animals like buffalo and giraffe. Immatures feed on a variety of birds and also on hares. Wilson et al. (personal communication) have also observed that the *H. m. rufipes*, normally considered a two-host tick, will behave like a three-host tick when fed on a guinea pig. It was previously thought that two-host ticks may not contribute as much to the transmission and maintenance of CCHF virus as do three-host ticks because they do not feed on as many different hosts. Now, in light of the demonstration that virus transmission can occur during co-feeding, we must re-evaluate the importance of two-host ticks in CCHF ecology. In addition to being particularly important as epidemic vectors of CCHF virus, *H. detritum*, *H. anatolicum*, *H. m. marginatum*, *H. m. rufipes*, and *H. turanicum* may also be important in the endemic transmission and maintenance of the virus.

The three-host ticks are comprised of species that feed on a different host for each of their development stages. In Eurasia, the following three-host ticks have been associated with CCHF: *I. ricinus*, *H. punctata*, *D. marginatus*, *D. daghestanicus*, *H. asiaticum*, *R. pumilio*, *R. rossicus*, *R. sanguineus*, and *R. turanicum*. In Africa, *H. impeltatum*, *H. nitidum*, *H. truncatum*, *A. variegatum*, *R. pulchellus*, and *R. appendiculatus* have been associated with CCHF virus. These three-host species are considered to be primarily involved in the ecology of CCHF virus as enzootic vectors of the disease and not in transmission to humans. There are some observations that suggest that at least some of these species can serve as epizootic vectors of CCHF virus. In Moldavia, where *H. m. marginatum* is rarely found, CCHF virus transmission has been attributed to *I. ricinus*, and possibly *D. marginatus* and *H. punctata*. In areas of South Africa where CCHF infections in humans has been documented, *H. truncatum* was the predominant *Hyalomma* species found on humans (Swanepoel et al., 1987).

Among the multiple-host ticks of the family Argasidae associated with CCHF virus, *A. persicus* typically feed once as larva, three times as a nymph, and as many as seven times as an adult (Strickland et al., 1976). *Ornithodoros lahorensis* is a parasite of sheep, camels, and cattle in Tibet, Kashmir, southern republics of the former USSR, southwest Asia, and southeast Europe. Its larvae remain on the host for 3–6 weeks and detach as engorged third-instar nymphs, which then molt to adults off the host (Hoogstraal, 1985). No particular vector or reservoir role has been postulated for the two species of argasid ticks from which CCHF virus has been isolated. Limited experimental studies on the ability of other species of soft ticks to become infected with CCHF virus have

all been unable to demonstrate if the virus can replicate in soft ticks (Shepherd et al., 1989b; Durden et al., 1993).

Although very few tick species have been incriminated in the transmission of CCHF virus to humans, almost all the species associated with the virus are known to feed on humans under some circumstances (Hoogstraal, 1979).

3.3.8. Host Immunity

The immune status of a host to tick feeding can be an important factor in reducing or preventing CCHF virus transmission. The subject of acquired resistance has been studied most intensely in the Ixodidae since the initial observation of Trager (1939). The effects of resistance range from simple rejection (Brown and Askenase, 1981), lowered engorged weight (Wikel and Allen, 1976), prolongation of feeding time (Kohler et al., 1967), and interference with feeding (McTier et al., 1981) to death of the tick on the host (Bagnall and Rothwell, 1974).

Many ixodid ticks can induce a host reaction to repeated tick feedings, which results in resistance (Wikel and Whelen, 1986); however, observations in *Hyalomma* species conflict. Brumpt and Chabaud (1947) and Chabaud (1950) found that domestic rabbits did not develop resistance to *H. excavatum* and *H. dromedarii*, but they did develop resistance to *D. pictus* and *R. sanguineus*. On the other hand, Kohler et al. (1967) found that rabbits did develop resistance to *H. excavatum*. We found that guinea pigs developed a limited resistance to *H. truncatum* larvae (Linthicum et al., 1992).

Host resistance to argasid ticks has been studied to a much lesser extent than that reported for the Ixodidae. Trager (1940) demonstrated that host resistance develops when chickens are fed upon by slow-feeding *A. persicus* larvae, but not when fed upon by rapidly feeding nymphs and adults. Brown et al. (1983) studied resistance in guinea pigs sensitized to *O. tartakovskyi*, and found that there was a greater basophil-associated response in hosts fed on by nymphs and adults of this soft tick than by those of *R. appendiculatus*.

3.3.9. Dispersion

Because all stages of ticks associated with CCHF virus are parasitic, and because all ixodid and some argasid ticks feed for extended periods of time, they can be dispersed over long distances while attached to their hosts. Hoogstraal (1979) reported that many bird species are responsible for the intra- and intercontinental dissemination of ticks associated with CCHF. The dispersal of ticks by birds may be restricted to a short distance during local post-breeding flights or extremely long distance during migration flights. In studies conducted on birds migrating through Egypt between 1955 and 1973, it was discovered that the birds migrating from Eurasia to Africa in the fall

carried tick species that were characteristic of the fauna of Europe and Asia. More than 90% of the immature ticks found on birds migrating to the south between 1959 and 1981 were species associated with CCHF virus. *Hyalomma marginatum rufipes* was the most common tick found on birds migrating north from sub-Saharan Africa to Eurasia in the spring.

The movements of domestic animals to new feeding areas, markets, and abattoirs, and the migrations of wild mammals also contribute to the dissemination of CCHF virus from enzootic areas. It was demonstrated by Morrill et al. (1990) that camels imported from Sudan and Kenya into Egypt were previously infected with CCHF virus. In Mauritania, Saluzzo et al. (1985) described the need for studies directed towards the role of camels in the spread of CCHF virus to northern Mauritania during migrations. In Senegal, Wilson et al. (1990) reported that, although the spatial distribution pattern of IgG antibody to CCHF virus could be affected by host migrations resulting from nomadic migrations, the distribution of the tick vectors may be an overriding factor. In Tadzhikistan, Pak and Mikhailova (1973) reported that apparently infected ticks were introduced into the cooler central region, and into the Pamir Mountains with cattle and sheep herded from the south to summer pastures. Causey et al. (1970) found in Nigeria that many CCHF virus-infected ticks were found on domestic animals that had been driven to an abattoir.

3.3.10. Vector Competence

For a tick to be a competent biological vector of CCHF virus, it must become infected by ingesting the virus during feeding or by transstadial or transovarial transmission. Once infected, the tick must be able to transmit the virus to another stage of development and/or to a susceptible vertebrate host during feeding. Vector competence is usually determined in the laboratory, either for species incriminated in nature based upon virus isolation, or from species associated with vertebrates implicated as reservoirs and amplifying hosts. As described in Section 2.3.3 on laboratory studies of vectors, vector competence studies have been conducted on numerous tick species associated with CCHF virus.

The biological characteristics of ticks are especially suitable and unique among hematophagous arthropods with respect to their potential to serve as efficient vectors and reservoirs of CCHF and other arboviruses (Hoogstraal, 1973). Ticks are always parasitic, very long-lived with a great reproductive potential, able to ingest large quantities of blood from a variety of vertebrate species, and very adaptable to changing and different ecological habitats.

3.4. Biology of Vertebrate Hosts

The biology of vertebrates implicated in the ecology of CCHF virus is important in understanding the transmission cycles of this zoonosis. Humans become

infected when they interrupt the natural tick–tick, tick–animal, and animal–animal CCHF virus enzootic cycles. Because of the diversity of ecological and zoogeographic habitats of CCHF foci, the qualitative and quantitative aspects of the biology of each vertebrate involved in maintaining the enzootic cycle of the virus must be determined (Hoogstraal, 1979). CCHF viral infection studies in vertebrates in field and laboratory studies have been discussed previously.

3.4.1. Host Density

It has not been demonstrated conclusively that horizontal transmission of the virus from vertebrates to tick vectors is the primary mechanism for the maintenance of CCHF virus. Theoretically, transovarial and transstadial transmission of the virus from tick to tick could be sufficient to perpetuate the virus without significant vertebrate involvement in virus transmission. However, field and laboratory studies have not been able to quantify the importance of this mechanism in the ecology of CCHF virus. If horizontal transmission of the virus is important in the ecology of the virus, the density of susceptible vertebrate species is a critical factor in determining the prevalence of ticks infected and capable of transmitting the virus.

High population densities of susceptible vertebrates and arthropod vectors were demonstrated to be important for epidemics of mosquito-borne viruses (Reeves, 1967). CCHF disease epidemiology appears to be controlled by ticks with a long life cycle. The virus can remain in ticks for extended periods of time in the absence of susceptible vertebrate populations. Human infections with CCHF virus are infrequent and irregular, but may be related to human association with increased densities of vertebrates and their associated tick species, as has been observed in the Crimean epidemics.

As discussed previously, the immature stages of many species of ticks feed on small mammals or ground-dwelling birds, which have high population turnovers and hence are comprised of animals with no previous exposure to either virus or tick.

3.4.2. Immunological Status

The immune status of vertebrate hosts impact on the ecology of CCHF virus transmission with respect to both previous exposure to the virus and to the extent of previous exposure to vector ticks. As discussed previously, ticks can become infected with CCHF virus while feeding on infected vertebrates before the host develops antibodies to the virus. If there are no vertebrates susceptible to CCHF viral infection, horizontal transmission cannot occur. For horizontal transmission of CCHF virus to occur, vertebrates must be susceptible to both the virus and to the tick vectors. Immunity to ticks can also intefere with the normal development of ticks, irrespective of viral infection, and potentially reduce the number of ticks in a population. Host immunity may dramatically impact on the ability of ticks to transmit and maintain CCHF virus.

3.4.3. Migration

As discussed previously in Section 3.3.9 on dispersal of vectors, the close association between ticks and their hosts is an important factor in the geographic distribution of CCHF virus. It has been demonstrated that bird and wild mammal migrations, and domestic animal movements can have an important impact on the geographic dispersion of CCHF virus by the movement of infected ticks.

4. REFERENCES

Al-Asgah, N.A. (1992) Biology of *Hyalomma schulzei* (Acari: Ixodidae) on rabbits under laboratory conditions. *J. Med. Entomol* **29**, 19–24.

Ammah-Attoh, V. (1966) Reproduction in the tick *Hyalomma marginatum rufipes* Koch, 1844 under laboratory conditions, with notes on mating and insemination. *Ghana J. Sci.* **6**, 9–14.

Anastos, G. (1957) The ticks or Ixodides of the U.S.S.R. A review of the literature. U.S. Dept. Health, Education, and Welfare, Public Health Service Publication 548, 397 pp.

Bagnall, B.G. and Rothwell, T.L.W. (1974) Responses in guinea pigs to larvae of the tick, *Ixodes holocyclus*. Vol. 2, pp. 1082–1083. *In: Proceedings of the 3rd International Congress of Parasitology*. Facta Publication, Munich.

Balashov, Y. S. (1968) Bloodsucking ticks (Ixodidea)—vectors of diseases of man and animals. *Nauka, Leningrad Department, Leningrad* (translation 500, Medical Zoology Department, United States Naval Medical Research Unit No. 3, Cairo, Egypt).

Berezin, V.V., Chumakov, M.P., Rubin, S.G., Stolbov, D.N., Butenko, A.M., Bashkirtsev, V.A. (1969) Contribution to the ecoloty of Crimean hemorrhagic fever virus in the lower Volga River. *Mater. 16. Nauchn. Sess. Inst. Polio Virus. Entsefalitov (Moscow)* **2**, 120–122 (in Russian), English translation NAMRU3-T836).

Berezin, V.V., Chumakov, M. P., Reshetnikov, I. A., and Zgurskaya, G. N. (1971) Study of the role of birds in the ecology of Crimean hemorrhagic fever virus. *Mater. 6. Simp. Izuch. Virus. Ekol. Svyazan. Ptits. (Omsk)*, 94–95 (in Russian, English translation NAMRU3-T721).

Bezuidenhout, J.D. and Malherbe, A. (1981) Sweating sickness: a comparative study of virulent and avirulent strains of *Hyalomma truncatum*. pp. 7–12. *In:* Whitehead, G.B. and Gibson, J.D. (eds) *Tick Biology and Control. Proceedings of an International Conference*, Rhodes University, Grahamstown, South Africa.

Blackburn, N.K., Besselaar, T.G., Shepherd, A.J., and Swanepoel, R. (1987) Preparation and use of monoclonal antibodies for identifying Crimean–Congo hemorrhagic fever virus. *Am. J. Trop. Med. Hyg* **37**, 392–397.

Blagoveshchenskaya, N.M., Butenko, A.M., Vyshnivetskaya, L.K., Zavodova, T.I., Zarubina, L.Z., Karinskaya, G.A., Kuchin, V.V., Milyutin, V.N., Novikova, E.M., Rubin, S.G., and Chumakov, M.P. (1969) Experimental infection of horses with Crimean hemorrhagic fever virus. Report 2. Virological and serological observations. *Mater. 16. Nauchn. Sess. Inst. Polio. Virus. Entsefalitov (Moscow)* **2**, 126–127 (in Russian, English translation NAMRU3-T840).

Blagoveshchenskaya, N.M. Donets, M.A., Zarubina, L.V., Kondratenko, V.F., and

Kuchin, V.V. (1975) Study of susceptibility to Crimean hemorrhagic fever (CHF) virus in European and long-eared hedgehogs. *Tezisy Konf. Vop. Med. Virus. (Moscow)* 269–270 (in Russian, English translation NAMRU3-T985).

Brown, S.J. and Askenase, P.W. (1981) Cutaneous basophil responses and immune resistance of guinea pigs to ticks: passive transfer with peritoneal exudate cells or serum. *J. Immunol.* **127**, 2163–2167.

Brown, S.J., Worms, M.J., and Askenase, P.W. (1983) *Rhipicephalus appendiculatus*: larval feeding sites in guinea pigs actively sensitized and receiving immune serum. *Exp. Parasitol.* **55**, 111–120.

Brumpt, E. (J.A.) (1936) Evolution experimentale de "*Ornithodoros lahorensis*. Similitude biologique des stades post-embryonnaires de cet argasine et de ceuz de l' *O. megnini*. Role pathogene eventuel. *Ann Parasitol. Hum. Comp.* **14**, 632–639.

Brumpt, E. and Chabaud, A.G. (1947) L'infestation par des ixodines provoque-t-elle une immunite chez l'note? I. Note preliminaire. *Ann. Parasitol.* **22**, 348–356.

Burney, M.I., Ghafoor, A., Saleen, M., Webb, P.A., Casals, J. (1980) Nosocomial outbreak of viral hemorrhagic fever caused by Crimean hemorrhagic fever—Congo virus in Pakistan, January 1976. *Am. J. Trop. Med. Hyg.* **29**, 941–947.

Casals, J. (1969) Antigenic similarity between the virus causing Crimean hemorrhagic fever and Congo virus. *Proc. Soc. Exp. Biol. Med.* **131**, 233–236.

Casals, J. (1981) *Report on a Visit to Iraq to Investigate a Viral Hemorrhagic Fever of Man Caused by Crimean Hemorrhagic Fever—Congo Virus.* World Health Organization, Geneva.

Casals, J. and Tignor, G.H. (1980) A new set of antigenic relationships that includes Crimean–Congo hemmorrhagic fever, Nairobi sheep disease and other arboviruses: the nairovirus supergroup. pp. 271–281. *International Symposium on Tropical Arboviruses and Hemmorhagic Fevers.* Academic Brasileira de Ciencia, Rio de Janeiro.

Causey, O.R., Kemp, G.E., Madbouly, M.H., and David-West, T.S. (1970) Congo virus from domestic livestock, African hedgehog, and arthropods in Nigeria. *Am. J. Trop. Med. Hyg.* **19**, 846–850.

Chapman, L.E., Wilson, M.L., Hall, D.B., LeGuenno, B., Dykstra, E.A., Ba, K., and Fisher-Hoch, S.P. (1991) Risk factors for Crimean–Congo hemorrhagic fever in rural northern Senegal. *J. Infect. Dis.* **164**, 686–692.

Chabaud, A.G. (1950) L'infestation par des ixodines provoque-t-elle une immunite chez l'hote (eme note). *Ann. Parasitol.* **25**, 474–479.

Chumakov, M.P. (1945) A new tick-borne virus disease—Crimean hemorrhagic fever. pp. 13–43. *In:* Sokolov, A.A., Chumakov, M.P. and Kolachev, A.A. (eds) *Crimean hemorrhagic fever (Acute Infectious Capillary Toxicosis).* Izd. Otd. Primorskoi Armii, Simferopol. (in Russian).

Chumakov, M.P. (1946) *Crimean Hemorrhagic Fever (Acute Infectious Capillary Toxicosis).* Short reports. Krymskiy Oblastnoi Otdel Zhravookhraneniya "Krymizdat," Simferopol., 27 pp. (in Russian, English translation NAMRU3-T910).

Chumakov, M.P. (1965). A short study of the investigation of the virus of Crimean hemorrhagic fever. *Tr. Inst. Polio. Virusn. Entsefalitov Akad. Med. Nauk SSSR* **7**, 193–196 (in Russian, English translation NAMRU3-T189).

Chumakov, M.P. (1971) Some results of investigation of the etiology and immunology of Crimean hemorrhagic fever. *Tr. Inst. Polio. Virusn. Entsefalitov Akad. Med. Nauk SSSR* **19**, 7–20 (in Russian, English translation, NAMRU3-T953).

Chumakov, M.P. (1972) Investigations of arboviruses in the USSR and the question

of possible association through migratory birds between natural arbovirus infection foci in the USSR and warm-climate countries. *Mater. 5. Izuch. Roli Pereletn. Ptits. Rasp. Arbovirus.*, Novosibirsk, July 1969, pp. 133–138 (in Russian, English translation NAMRU-T876).

Chumakov, M.P. (1974) On 30 years of investigation of Crimean hemorrhagic fever. *Tr. Inst. Polio. Virusn. Entsefalitov Akad. Med. Nauk SSSR* **22**, 5–18 (in Russian, English translation NAMRU3-T950).

Chumakov, M.P., Belyaeva, A.P., Voroshilova, M.K., Butenio, A.M., Shalunova, N.V., Semashko, I.V., Mart'yanova, L.I., Smirnova, S.E., Bashkirtsev, V.N., Zanvodova, T.I., Rubin, S.G., Tkachenko, E. A., Karmysheva, V.Ya., Reingol'd, V.N., Popov, G.V., Kirov, I., Stolbov, D.N., and Perelatov, V.D. (1968) Progress in studying the etiology, immunology, and laboratory diagnosis of Crimean hemorrhagic fever in the USSR and Bulgaria. *Mater. 15. Nauchn. Sess. Inst. Polio. Virus. Entsefalitov (Moscow)* **3**, 100–103 (in Russian, English translation, NAMRU3-T613).

Chumakov, M.P., Smirnova, S.E., Tkachenko, E.A. (1969) Antigenic relationships between the Soviet strains of Crimean hemorrhagic fever virus and the Afro-Asian Congo virus strains. *Mater. 16. Nauchn. Sess. Inst. Polio. Virus. Entsefalitov (Moscow)* **2**, 152–154 (in Russian, English translation NAMRU3-T614).

Chumakov, M.P., Butenko, A.M., Rubin, S.G., Berezin, V.V., Karinskaya, G.A., Vasilenko, S.M., Smirnova, S.E., Bashkirtxev, V.V., Berbedeneva, M.P., Badalov, M.V., and Stolbov, D.N. (1972). Question on the ecology of Crimean Congo hemorrhagic fever virus. pp. 222–229. *Mater. 5. Simp. Izuch. Roli Perelletn. Ptits. Rasp. Arbovirus*, Novosibirsk, July, 1969. (In Russian, English translation NAMRU3-T877).

Chumakov, M.P., Smirnova, S.E., Shalunova, N.Y., Martianova, L.I., Fleer, G.P., Zgurskaya, G.N., Maksumov, S.S., Kasymov, K. Y., and Pak, T.P. (1976) Proofs of etiological identity to Crimean hemorrhagic fever in Central Asian hemorrhagic fever. *IX Internat. Congress on Tropical Medicine and Malaria*, Athens, Vol. 1, pp. 33–34.

Clerx, J.P. and Bishop, D.H.L. (1981) Qalyub virus, a member of the newly proposed *Nairovirus* genus (Bunyaviridae). *Virology* **108**, 361–372.

Clerx, J.P.M., Casals, J., and Bishop, D.H.L. (1981) Structural characteristics of Nairoviruses (genus *Nairoviruses*, Bunyaviridae). *J. Gen. Virol.* **55**, 165–178.

Clifford, C.M., Flux, J.E.C., Hoogstraal, H. (1976) Seasonal and regional abundance of ticks (Ixodidae) on hares (Leporidae) in Kenya. *J. Med. Entomol.* **13**, 40–47.

Daniel, M. and Kolar, J. (1990) Using satellite data to forecast the occurrence of the common tick *Ixodes ricinus* (L.). *J. Hyg. Epidemiol. Microbiol. Immunol.* **3**, 243–252.

Davey, R.B., Garza, J., Jr, Thompson, G.D., and Drummond, R.O. (1980a) Ovipositional biology of the cattle tick, *Boophilus annulatus* (Acari: Ixodidae), in the laboratory. *J. Med. Entomol.* **17**, 287–289.

Davey, R.B., Garza, J., Jr, Thompson, G.D., and Drummond, R.O. (1980b) Ovipositional biology of the southern cattle tick, *Boophilus microplus* (Acari: Ixodidae), in the laboratory. *J. Med. Entomol.* **17**, 117–121.

Delphy, L.P. and Gouchey, S.H. (1937) Biologie de *Hyalomma dromedarii* (*Koch*, 1844). *Ann. Parasitol. Hum. Comp.* **15**, 487–499.

Digoutte, J.P. and Heme, G. (1985) *Rapport annuel 1984.* Rapport Institut Pasteur, Dakar.

Dipeolu, O.O. (1983) Studies on ticks of veterinary importance in Nigeria. VI. Comparisons of oviposition and the hatching of eggs of *Hyalomma* species. *Vet. Parasitol.* **13**, 251–265.

Donchev, D., Kebedzhiev, G., and Rusakiev, M. (1967) Hemorrhagic fever in Bulgaria. *Bulg. Akad. Nauk. Mikrobiol. Inst. 1. Kongr. Mikrobiol.*, 1965, pp. 777–784 (in Bulgarian, English translation NAMRU3-T465).

Durden, L., Logan, T.M., Wilson, M.L., and Linthicum, K.J. (1993) Experimental vector incompetence of the soft tick *Ornithodorus sonrai* (Acari, Argasidae), for Crimean–Congo hemorrhagic fever virus. *J. Med. Entomol.* **30**, 493–496.

Filippova, N.A. (1966) Argasid ticks (Argasidae). *Fauna SSSR, Paukoobraznye* **4**, 1–255 (in Russian, English translation NAMRU3-T600).

Ganiev, I.M. (1956) Biology of *Hyalomma plumbeum* Panzer, 1795 under conditions of southern Dagestan. *Tr. Inst. Zhivot. Dagestan. Fil. Akad. Nauk SSSR* **3**, 27–33 (in Russian).

Garris, G.I. (1984) Colonization and life cycle of *Amblyomma variegatum* (Acari: Ixodidae) in the laboratory in Puerto Rico. *J. Med. Entomol.* **21**, 86–90.

Gear, J.H.S. (1982) The hemorrhagic fevers of southern Africa with special references to studies in the South African Institute for Medical Research. *Yale J. Biol. Med.* **55**, 207–212.

Gear, J.H.S., Thomson, P.D., Hopp, M., Andronikou, S., Cohn, R.J., Ledger, J., and Berkowitz, F.E. (1982) Congo–Crimean haemorrhagic fever in South Africa. Report of a fatal case in the Transvaal. *S. Afr. Med. J.* **62**, 576–580.

Goldfarb, L.G., Chumakov, M.P., Myskin, A.A., Kondratenko, V.F., and Reznikova, O.Y. (1980) An epidemiological model of Crimean hemorrhagic fever. *Am. J. Trop. Med. Hyg.* **29**, 260–264.

Gonzalez, J.P., Cornet, J.P., Wilson, M.L., and Camicas, J.L. (1991) Crimean–Congo haemorrhagic fever virus replication in adult *Hyalomma truncatum* and *Amblyomma variegatum* ticks. *Res. Virol.* **142**, 483–488.

Gonzalez, J.P., Camicas, J.L., Cornet, J.P., Faye, O., and Wilson, M.L. (1992) Sexual and transovarian transmission of Crimean–Congo haemorrhagic fever virus in *Hyalomma truncatum* ticks. *Res. Virol.* **143**, 23–28.

Gordon, S.W., Linthicum, K.J., and Moulton, J.R. (1993) Transmission of Crimean–Congo hemorrhagic fever virus in two species of *Hyalomma* ticks, from infected adults to cofeeding immature forms. *Am. J. Trop. Med. Hyg.* **48**, 576–580.

Hafez, M. and Bassal, T.T.M. (1980) The nutrition and ambient temperature effect on the oviposition of *Rhipicephalus sanguineus* Latr. (Ixodidae, Acarina). *J. Egypt. Soc. Parasitol.* **10**, 295–300.

Hagras, E.H. and Khalil, G.M. (1988) Effect of temperature on *Hyalomma dromedarii* Koch (Acari: Ixodidae). *J. Med. Entomol.* **25**, 354–359.

Honzakova, E. (1973) Survival and development of ticks *Ixodes ricinus*, *Ixodes hexagonus* and *Dermacentor marginatus* in the period of maximum summer temperatures. *Folia Parasitol. (Prague)* **20**, 263–266.

Hoogstraal, H. (1956) African Ixodoidea, Vol I: Ticks of the Sudan. *USAMU-3 Res. Rep. Nm. 005.* **29**, 1101 pp, Cairo.

Hoogstraal, H. (1973) Viruses and ticks. pp. 349–390. *In:* Gibbs, A.J. (ed) *Viruses and Invertebrates.* North Holland Publishing Co., Amsterdam.

Hoogstraal, H. (1979) The epidemiology of tick-borne Crimean–Congo hemorrhagic fever in Asia, Europe, and Africa. *J. Med. Entomol.* **15**, 307–417.

Hoogstraal, H. (1985) Argasid and nuttalliellid ticks as parasites and vectors. *Adv. Parasitol.* **24**, 135–238.

Hooker, W.A., Bishopp, F.C., and Wood, H.P. (1912) The life history and bionomics of some North American ticks. *Bull. U.S. Bur. Entomol.* **106**, 239 pp.

Hueli, L.E. (1979) Estudio del ciclo biologico de *Hyalomma marginatum marginatum* Koch. 1844 (Acarina: Ixodidae) bajo, condiciones estandar der laboratorio. *Rev. Iber. Parasitol.* **39**, 143–152.

Hueli, L.E., Guevara-Benitz, D.C., and Garcia-Fernandez, P. (1986) Oviposition de *Rhipicephalus (Digineus) bursa* Canestrini et Fanzago, 1877 (Acarina, Ixodidae) dans les conditions de laboratorie. *Acarologia* **27**, 117–120.

Jelinkova, A., Benda, R., and Novak, M. (1975) Electron microscopic demonstration of Crimean hemorrhagic fever virus in CV-1 cells. *Acta Virol.* (English edn) **19**, 369–373.

Kalunda, M., Lule, M., Sekyalo, E., Mukuye, A., and Mujomba, E. (1983) Virus isolation and identification. *Rep. East Afr. Virus Res. Inst.* **27**, 17–20.

Karabatsos, N. (1985) *International Catalogue of Arboviruses, including Certain Other Viruses of Vertebrates*, 3rd edn. American Committee on Arthropod-borne Viruses, American Society of Tropical Medicine and Hygiene, Washington, D.C.

Kemp, G.E., Causey, O.R., Setzer, H.W., and Moore, D.L. (1974) Isolation of viruses from wild mammals in West Africa, 1966–1970. *J. Wildl. Dis.* **10**, 279–293.

Knight, M.M., Norval, R.A.I., and Rechav, Y. (1978) The life cycle of the tick *Hyalomma marginatum rufipes* Koch (Acarina: Ixodidae) under laboratory conditions. *J. Parasitol* **64**, 143–146.

Kohler, G., Hoffman, G., Horcher, F., and Weiland, G. (1967) Immunobiologische untersuchungen an kaninchen mit Ixoden-infestation. *Berl. Muench. Tierarztl. Wochenschr.* **80**, 396–400.

Kondratenko, V.F. (1976) Importance of ixodid ticks in transmission and preservation of the causative agent of Crimean hemorrhagic fever agent in the foci of infection. *Parazitologiya* **10**, 297–302 (in Russian, English translation NAMRU3-T1116).

Kondratenko, V.F., Blagoveschenskaya, N.M., Butenko, A.M., Vyshnivetskaya, L.K., Zarubina, L.V., Milyutin, V.N., Kuchin, V.V., Novikova, E. M., Rabinovich, V. D., Shevchenko, S. F., and Chumakov, M. P. (1970) Results of virological investigation of ixodid ticks in Crimean hemorrhagic fever focus in Rostov Oblast. *Mater. 3. Oblast. Nauchn. Prakt. Konf. (Rostov-on-Don)*, pp. 29–35 (in Russian, English translation NAMRU3-T524).

Korolev, M.B., Donets, M.A., Rubin, S.G., and Chumakov, M.P. (1976) Morphology and morphogenesis of Crimean hemorrhagic fever virus. *Arch. Virol.* **50**, 169–172.

Kraiss, A. and Gothe, R. (1982) The life cycle of *Argas (Argas) africolumbae* under constant abiotic and biotic conditions. *Vet. Parasitol* **11**, 365–373.

Lee, E.H. and Kemp, G.E. (1970) Congo virus: experimental infection of *Hyalomma rufipes* and transmission to a calf. *Bull. Entomol. Soc. Niger.* **2**, 133–135.

Levi, V. and Vasilenko, S. (1972) Study on the Crimean hemorrhagic rever (CHF) virus transmission mechanism in *Hyalomma pl. plumbeum* ticks. *Epidemiol. Mikrobiol. Infekts. Boles.* **9**, 182–185 (in Bulgarian).

Linthicum, K.J., Logan, T.M., Kondig, J.P., Gordon, S.W., and Bailey, C.L. (1991) Laboratory biology of *Hyalomma truncatum* (Acari: Ixodidae). *J. Med. Entomol.* **28**, 280–283.

Linthicum, K.J., Dickson, D.L., and Logan, T.M. (1992) Feeding efficiency of larval *Hyalomma truncatum* (Acari: Ixodidae) on hosts previously exposed to ticks. *J. Med. Entomol.* **29**, 310–313.

Logan, T.M., Linthicum, K.J., Bailey, C.L., Watts, D.M., and Moulton, J.R. (1989a) Experimental transmission of Crimean–Congo hemorrhagic fever virus by *Hyalomma truncatum* Koch. *Am. J. Trop. Med. Hyg.* **40**, 207–212.

Logan, T.M., Linthicum, K.J., Kondig, J.P., and Bailey, C.L. (1989b) Biology of *Hyalomma impeltatum* (Acari: Ixodidae) under laboratory conditions. *J. Med. Entomol.* **26**, 479–483.

Logan, T.M., Linthicum, K.J., Bailey, C.L., Watts, D.M., Dohm, L.J., and Moulton, J.R. (1990) Replication of Crimean–Congo hemorrhagic fever virus in four species of ixodid ticks (Acari: Ixodidae) infected experimentally. *J. Med. Entomol.* **27**, 537–542.

Logan, T. M., Linthicum, K.J., Moulton, J.R., and Ksiazek, T.G. (1993) Antigen-capture enzyme-linked immunosorbent assay for detection and quantification of Crimean–Congo hemorrhagic fever virus in the tick, *Hyalomma truncatum*. *J. Virol. Meth.* **42**, 33–44.

Marriott, A.C., Ward, V.K., Higgs, S., and Nuttall, P.A. (1990) RNA probes detect nucleotide sequence homology between members of two different nairovirus serogroups. *Virus Res.* **16**, 77–82.

Mathiot, C. C., Fontenille, D., Digoutte, J.P., and Coulanges, P. (1988) First isolation of Congo–Crimean haemorrhagic fever virus in Madagascar. *Ann. Inst. Pasteur/Virol.* **139**, 239–241.

Matson, B.A. and Norval, R.A.I. (1977) The seasonal occurrence of adult ixodid ticks on cattle on a Rhodesian highveld farm. *Rhodexian Vet. J.* **8**, 2–6.

Matthysse, J.G. and Colbo, M.H. (1987) *The Ixodid Ticks of Uganda.* Entomological Society of American, College Park, Maryland.

McTier, T.L., George, J.E., and Bennett, S.N. (1981) Resistance and cross-resistance of guinea pigs to *Dermacentor andersoni* (Stiles), *D. variabilis* (Say), *Ambloyomma americaum* (Linnaeus), and *Ixodes scapularis* (Say). *J. Parasitol.* **67**, 813–822.

Mikhailov, G.I. (1946) On the epidemiology of an acute infectious hemorrhagic disease. *Klin. Med.* (*Moscow*) **24**, 67–69 (in Russian).

Milyutin, V.N., Butenio, A.M., Artyushenko, A.A., Bliznichenko, A.G., Zavodova, T.I., Zarubina, L.V., Novikova, E. M., Rubin, S.G., Cherynshev, N.I., and Chumakov, M.P. (1969) Experimental infection of horses with Crimean hemorrhagic fever virus. Report I. Clinical observations. *Mater. 16. Nauchn. Sess. Inst. Polio. Virus. Entsefalitov* (*Moscow*). **2**, 145–146 (in Russian, English translation NAMRU3-T851).

Minshull, J.I. (1981) Seasonal occurrence, habitat, distribution and host range of four ixodid tic, species at Kyle Recreational Park in southeastern Zimbabwe. *Zimbabwe Vet. J.* **12**, 58–63.

Morel, P.C. (1969) Contribution a la connaisance de la distribution des tiques (Acariens, Ixodidae et Ambloyommidae) en Afrique Ethiopienne Continentale. These de Soct. Sci. Nat., Fac. Sci d'Orsay, Univ. Paris. Institut d'Elevage et de Medicine Veterinaire des Pays Tropicaux. Maisons-Alfort, France.

Morrill, J. C., Soliman, A.K., Imam, I.A., Botros, B.A., Moussa, M.I., and Watts, D.M. (1990) Serological evidence of Crimean–Congo haemorrhagic fever viral infection among camels imported in Egypt. *J. Trop. Med. Hyg.* **93**, 201–204.

Murphy, F.A., Whitfield, S.G., Coleman, P.H., Calisher, C.H., Rabin, E.R., Jenson, A.B., Melnick, J.L., Edwards, M.R., and Whitney, E. (1968) California group arboviruses: electron microscopic studies. *Exp. Molec. Pathol.* **8**, 44–56.

Murphy, F.A., Harrison, A.K., and Whitfield, S.G. (1973) Bunyaviridae: morphologic

and morphogenetic similarities of Bunyamwera serologic supergroup viruses and several other arthropod-borne viruses. *Intervirology* **1**, 297–316.

The New International Atlas (1986) Rand McNally and Company, Chicago.

Nuttall, G.H.F. (1915) Observations on the biology of Ixodidae. *Parasitology* **7**, 408–456.

Okorie, T.G. (1980) Congo virus—the development in the ixodid ticks—*Hyalomma rufipes* and *Amblyomma variegatum* Fabricius. *International Congress on Entomology*, Kyoto, August 1980, Abstract 16, p. 331.

Okorie, T.G. (1991) Comparative studies on the vector capacity of the different stages of *Ambloyomma variegatum* Fabricius and *Hyalomma rufipes* Koch for Congo virus, after intracoelomic inoculation. *Vet. Parasitol* **38**, 215–223.

Okorie, T.G. and Fabiyi, A. (1980) The replication of Congo virus in *Hyalomma rufipes* Koch following intracoelomic inoculation. *Vet. Parasitol* **7**, 369–374.

Ouheilli, H. and Pandey, V.S. (1986) Development of *Hyalomma lusitanicum* under laboratory conditions. *Vet. Parasitol.* **15**, 57–66.

Ouhelli, H., Pandey, V.S., and Choukri, M. (1982) The effects of temperature, humidity, photoperiod and weight of the engorged female on oviposition of *Boophilus annulatus* (Say, 1821). *Vet. Parasitol.* **11**, 231–239.

Pak, T.P. (1975) Seasonal circulation dynamics of Crimean hemorrhagic fever virus in Tadzhikistan. pp. 35–37. *Mater. 9. Simp. Ekol. Virus* (*Dunshanbe*) (in Russian, English translation NAMRU3-T1126).

Pak, T.P. and Mikhailova, L.I. (1973) Crimean hemorrhagic fever in Tadzhikistan. *IZD. 'Irfon,' Dushanbe*, 154 pp. (in Russian, English translation NAMRU3-T1000).

Pak, T.P., Daniyarov, O.A., Kostyukov, M.A., Bulcyhev, V.P. and Kuima, A.U. (1974) Ecology of Crimean hemorrhagic fever in Tadzhikistan. *Mater. Resp. Simp. Kamenyuki 'Belovezh, Puscha'* (*Minsk*), pp. 93–94 (in Russian, English translation NAMRU3-T968).

Pak, T.P., Kostyukov, M.A., Daniyarov, O.A., and Bulychev, V.P. (1975) A combined focus of arbovirus infections in Tadzhikistan. *Mater. 9. Simp. Ekol. Virus.* (*Dushanbe*), pp. 38–39 (in Russian, English translation NAMRU3-T1127).

Pegram, R.G., Perry, B.D., Musisi, F.L., and Mwanaumo, B. (1986) Ecology and phenology of ticks in Zambia: seasonal dynamics on cattle. *Exp. Appl. Acarol.* **2**, 24–25.

Perry, B.D., Lessard, P., Norval, R.A.I., Kundert, K., and Kruska, R. (1990) Climate, vegetation and the distribution of *Rhipicephalus appendiculatus* in Africa. *Parasitol. Today* **6**, 100–104.

Perry, B.D., Kruska, R., Lessard, P., Norval, R.A.I., and Kundert, K. (1991) Estimating the distribution and abundance of *Rhipicephalus appendiculatus* in Africa. *Prev. Vet. Med.* **11**, 261–268.

Pospelova-Shtrom, M.V. (1935) Biological survey of *Hyalomma yakimovi* Ol. under laboratory conditions. pp. 195–233. *In:* Pavlovsky, E.N. (ed) *Cattle Pests.* Izd. Akad. Nauk SSSR.

Pospelova-Shtrom, M.V. and Pettrova-Piontkovskaya, S.P. (1949) On the biology of *Hyalomma marginatus, H. detritum,* and *H. asiaticum* under laboratory conditions. *Vopr. Kraev. Obshch. Eksp. Parazitol.* **6**, 87–97 (in Russian).

Pratt, D.J., Greenway, P.G., and Gwynne, M.D. (1966) A classification of East African rangeland, with an appendix on terminology. *J. Appl. Ecol* **3**, 369–382.

Rechav, Y. (1986) Seasonal activity and hosts of the vectors of Crimean–Congo hemorrhagic fever in South Africa. *S. Afr. Med. J.* **69**, 364–368.

Rechav, Y., Knight, M.M., and Norval, R.A.I. (1977) Life cycle of the tick *Rhipicephalus*

evertsi evertsi Neumann (Acarina: Ixodidae) under laboratory conditions. *J. Parasitol.* **63**, 575–579.

Reeves, W.C. (1967) Factors that influence the probability of epidemics of western equine, St. Louis, and California encephalitis in California. *Calif. Vector Views* **14**, 13–18.

Robin, Y. (1977) *Centre Collaborateur O.M.S. de Reference et de Recherche pour les arbovirus.* Rapport Institut Pasteur, Dakar, 16 pp.

Saluzzo, J.F. and Le Guenno, B. (1987) Rapid diagnosis of human Crimean–Congo hemorrhagic fever and detection of the virus in naturally infected ticks. *J. Clin. Microbiol.* **25**, 922–924.

Saluzzo, J.F., Digouette, J.P., Camicas, J.L., and Chauvancy, G. (1985) Crimean–Congo haemorrhagic fever and Rift Valley fever in south-eastern Mauritania. *Lancet* **2**, 116.

Saluzzo, J.F., Camicas, J.L., Chartier, C., Martinez, D., and Digoutte, J.P. (1986) Le virus de la fievre hemorragique de Crimee–Congo (CCHF) en Mauritanie. *Zah. ORSTOM Ent. med. Parasitol.* **24**, 129–137.

Saluzzo, J.F., LeGuenno, B., and Van der Groen, G. (1988) Use of heat inactivated viral haemorrhagic fever antigens in serological assays. *J. Virol. Method.* **22**, 165–172.

Semashko, I.V., Dobritsa, P.G., Bashkirtsev, V.N., and Chumakov, M.P. (1975) Results from investigating blood sera from healthy persons, animals, and birds collected in southern Kazakhstan for antibodies to CHF-Congo virus. *Mater. 9. Simp. Ekol. Virus. (Dushanbe)*, pp. 43–44 (in Russian, English translation NAMRU3-T1128).

Serdyukova, G.V. (1946) The development cycle of *Hyalomma antatolicum* Koch. *Izv. Akad. Nauk USSR. S. Biol.* **2–3**, 199–202 (in Russian, English translation NAMRU3-T236).

Shepherd, A.J., Swanepoel, R., Shepherd, S.P., Leman, P.A., Blackburn, N.K., Hallett, A.F. (1985) A nosocomial outbreak of Crimean–Congo haemorrhagic fever at Tygerberg Hospital. Part V. Virological and serological observations. *S. Afr. Med. J.* **68**, 733–736.

Shepherd, A.J., Swanepoel, R., Leman, P.A., and Shepherd, S.P. (1987a) Field and laboratory investigation of Crimean–Congo haemorrhagic fever virus (Nairovirus, family Bunyaviridae) infection in birds. *Trans Roy. Soc. Trop. Med. Hyg.* **81**, 1004–1007.

Shepherd, A.J., Swanepoel, R., Shepherd, S.P., McGillivray, G.M., and Searle, L.A. (1987b) Antibody to Crimean–Congo hemorrhagic fever virus in wild mammals from southern Africa. *Am. J. Trop. Med. Hyg.* **36**, 133–142.

Shepherd, A.J., Swanepoel, R., and Gill, D.E. (1988). Evaluation of enzyme-linked immunosorbent assay and reversed passive hemagglutination for detection of Crimean–Congo hemorrhagic fever virus antigen. *J. Clin. Microbiol.* **26**, 347–353.

Shepherd, A.J., Leman, P.A., and Swanepoel, R. (1989a) Viremia and antibody response of small Africa and laboratory animals to Crimean–Congo hemorrhagic fever virus infection. *Am. J. Trop. Med. Hyg.* **40**, 541–547.

Shepherd, A.J., Swanepoel, R., Cornel, A.J., and Mathee, O. (1989b) Experimental studies on the replication and transmission of Crimean–Congo hemorrhagic fever virus in some African tick species. *Am. J. Trop. Med. Hyg.* **40**, 326–331.

Shepherd, A.J., Swanepoel, R., Shepherd, S.P., Leman, P.A., and Mathee, O. (1991)

Viraemic transmission of Crimean–Congo haemorrhagic fever virus to ticks. *Epidemiol. Infect.* **106**, 373–382.

Simpson, D.I.H., Knight, H.E.M., Courtois, G., Williams, M.C., Weinbren, M.P., and Kibukamusoke, J.W. (1967) Congo virus: a hitherto undescribed virus occurring in Africa. I. Human isolations—clinical notes. *East Afr. Med. J.* **44**, 87–92.

Srivastava, S.C. and Varma, M.G.R. (1964) The culture of the tick *Rhipicephalus sanguineus* (Latreille) Ixodidae) in the laboratory. *J. Med. Entomol.* **1**, 154–157.

Stolbov, D.N., Butenko, A.M., Egorova, P.S., Leshchinskaya, E.V., and Chumakov, M.P. (1965) Crimean hemorrhagic fever (CCHF) in Astrakhan Oblast. *Tr. Inst. Polio. Virusn. Entsefalitov Akad. Med Nauk. SSSR.* **7**, 271–278 (in Russian, English translation MANRU3-604).

Strickland, R.K., Gerrish, R.R., Hourrigan, J.L., and Schubert, G.O. (1976) *Ticks of Veterinary Importance.* USDA, APHIS, Agricultural Handbook, no. 485.

Swanepoel, R., Struthers, J.K., Shepherd, A.J., McGillivray, G.M., Nel, M.J., and Jupp, P.G. (1983) Crimean–Congo hemorrhagic fever in South Africa. *Am. J. Trop. Med. Hyg.* **32**, 1407–1415.

Swanepoel, R., Shepherd, A.J., Leman, P.A., and Shepherd, S.P. (1985a) Investigations following initial recognition of Crimean–Congo haemorrhagic fever in South Africa and the diagnosis of 2 further cases. *S. Afr. Med. J.* **68**, 638–641.

Swanepoel, R., Shepherd, A.J., Leman, P.A., Shepherd, S.P., and Miller, G.B. (1985b) A common-source outbreak of Crimean–Congo haemorrhagic fever on a dairy farm. *S. Afr. Med. J.* **68**, 635–637.

Swanepoel, R., Shepherd, A.J., Leman, P.A., Shepherd, S.P., McGillivray, G.M., Erasmus, M.J., Searle, L.A., and Gill, D.E. (1987) Epidemiologic and clinical features of Crimean–Congo hemorrhagic fever in Southern Africa. *Am. J. Trop. Med. Hyg.* **36**, 120–132.

Sweatman, G.K. (1968) Temperatures and humidity effects on the oviposition of *Hyalomma aegyptium* ticks of different engorgement weights. *J. Med. Entomol.* **5**, 429–439.

Theiler, G. (1943) *Notes on the Ticks of Domestic Stock from Portugese, East Africa.* Estae Anti Malaria, Lourenco Marques, 55.

Theiler, G. (1964) Ecogeographical aspects of tick distribution. pp. 284–299. *In:* Davis, D.H.S. (ed) *Ecological Studies in Southern Africa.* Monographie Biol. Junk, The Hague.

Tignor, G.H., Smith, A.L., Casals, J., Ezeokoli, C.D., and Okoli, J. (1980) Close relationships of Crimean hemorrhagic fever-Congo (CHF-C) virus strains by neutralizing antibody assays. *Am. J. Trop. Med. Hyg.* **29**, 676–685.

Trager, W. (1939) Acquired immunity to ticks. *J. Parasitol.* **25**, 57–81.

Trager, W. (1940) A note on the problem of acquired immunity to argasid ticks. *J. Parasitol.* **26**, 71–74.

USDA (1989a) Ostriches with exotic ticks. *Foreign Animal Disease Report* **17(3)**, 5–6.

USDA (1989b) Update on exotic ticks. Foreign Animal Disease Report **17(4)**, 2–3.

USDA (1989c) Ectoparasites on imported rhinocoses. *Foreign Animal Disease Report* **17(4)**, 3.

Van Eeden, P.J., Van Eeden, S.F., Joubert, J.R., King, J.B., Van De Wal, B.W., and Michell, W.L. (1985) A nosocomial outbreak of Crimean–Congo haemorrhagic fever at Tygerberg Hospital. I. Clinical features. *S. Afr. Med. J.* **68**, 711–717.

Walker, J.B. (1974) *The Ixodid Ticks of Kenya. A Review of Present Knowledge of their Hosts and Distribution.* Agricultural Bureau, Commonwealth Institute of Entomology, London, 220 pp.

Watts, D.M., Ksiazek, T.G., Linthicum, K.J., and Hoogstraal, H. (1989) Crimean–Congo hemorrhagic fever. Vol. II. *In:* Monath, T.P. (ed.) *The Arboviruses: Epidemiology and Ecology.* CRC Press, Boca Raton, FL.

Wikel, S.K. and Allen, J.R. (1976) Acquired resistance to ticks. I. Passive transfer of resistance. *Immunology* **30**, 311–316.

Wikel, S.K. and Whelen, A.C. (1986) Ixodid–host immune interaction. Identification and characterization of relevant antigens and tick-induced host immunosuppression. *Vet. Parasitol.* **20**, 149–174.

Wilson, M.L., LeGuenno, B., Guillaud, M., Desoutter, D., Gonzalez, J.P., and Camicas, J.L. (1990) Distribution of Crimean–Congo hemorrhagic fever viral antibody in Senegal: environmental and vectorial correlates. *Am. J. Trop. Med. Hyg.* **43**, 557–566.

Wilson, M.L., Gonzalez, J.P., Cornet, J.P., and Camicas, J.L. (1991) Transmission of Crimean–Congo hemorrhagic fever virus from experimentally infected sheep to *Hyalomma truncatum* ticks. *Rev. Virol.* **142**, 395–404.

Woodall, J.P., Williams, M.C., Simpson, D.I.H., Ardoin, P., Lule, M., and West, R. (1965) The Congo group of agents. *Rep. E. Afr. Virus Res. Inst. (1963–64)* **14**, 34–36.

Yeoman, G.H. and Walker, J.B. (1967) *The Ixodid Ticks of Tanzania. A Study of the Zoogeography of the Ixodidae of an East African Country.* Commonwealth Agricultural Bureau, Commonwealth Institute of Entomology, London.

Yu-Chen, Y., Ling-Xiong, K., Ling, L., Yu-Qin, Z., Feng, L., Bao-Jian, C., and Shou-Yi, G. (1985) Characteristics of Crimean–Congo hemorrhagic fever virus (Xinjiang strain) in China. *Am. J. Trop. Med. Hyg.* **34**, 1179–1182.

Zarubinsky, V.Ya., Klisenko, G.A., Kuchin, V.V., Timchenko, V.V., and Shanoyan, N.K. (1975) Application of the indirect hemagglutination inhibition test for serological investigation of Crimean hemorrhagic fever focus in Rostov Oblat. *Sb. Tr. Inst. Virus. imeni D.I. Ivanovsky, Akad. Med. Nauk SSSR* **2**, 73–77 (in Russian, English translation NAMRU3-T1145).

Zarubinsky, V.Ya., Kondratenko, V.F., Blagoveschchenskaya, N.M., Zarvbina, L.V., and Kuchin, V.V. (1976) Susceptibility of calves and lambs to Crimean hemorrhagic fever virus. *Tezisy Dokl. 9. Vses. Konf. Prirod. Ochag. Bolez. Chelov. Zhivot.* (Omsk), pp. 130–131 (in Russian, English translation NAMRU3-T1178).

Zgurskaya, B.N., Berezin, S.E., Smirnova, S.E., and Chumakov, M.P. (1971) Investigation of the question of Crimean hemorrhagic fever virus transmission and interepidemic survival in the tick *Hyalomma plumbeum plumbeum. Panzer. Tr. Inst. Polio. Virusn. Entsefalitov Akad. Med. Nauk SSSR.* **19**, 217–220 (in Russian, English translation NAMRU3-T911).

Zgurskaya, G.N., Berezin, V.V., and Smirnova, S.E. (1975) Threshold levels of blood infectiousness for *Hyalomma p. plumbeum* tick during viremia in hares and rabbits caused by CHF virus. *Tezisy Konf. Vop. Med. Virus. (Moscow)*, pp. 291–292 (in Russian), English translation NAMRU3-T997).

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Index