

# Tick-borne infectious diseases of dogs

Susan E. Shaw, Michael J. Day, Richard J. Birtles and Edward B. Breitschwerdt

Tick-transmitted infections are an emerging problem in dogs. In addition to causing serious disease in traditional tropical and semi-tropical regions, they are now increasingly recognized as a cause of disease in dogs in temperate climates and urban environments. Furthermore, subclinically infected companion animals could provide a reservoir for human tick-transmitted infectious agents, such as *Ehrlichia chaffeensis*, *Ehrlichia ewingii*, the *Ehrlichia phagocytophila* group and *Rickettsia conorii*. Here, we discuss the emergence of new canine tick-transmitted diseases, which results from several factors, including the expansion of the tick range into urban and semi-urban areas worldwide, the movement of infected dogs into previously non-endemic areas, and the advent of novel molecular techniques for diagnosis and pathogen identification.

Being haematophagous, ticks are well designed to transmit disease agents such as viruses, bacteria and protozoa. Historically, they have been considered second only to mosquitoes in this ability<sup>1</sup>. Ticks attach securely to their hosts, facilitating not only effective transmission of infectious agents, but also the spread of both ticks and microorganisms to different geographical habitats via travelling pets. Pathogens ingested by ticks can be spread trans-stadially and/or trans-ovarially. As female ticks are extremely fecund, this allows effective dissemination of infectious agents in reservoir populations with which pets and their owners interact.

Arthropods in general, and ticks in particular, have evolved as ectoparasites of wild animals<sup>1</sup>. Only a minority of tick species, generally those with a wide host range, transmit diseases to domestic animals and humans. The increasing prevalence of tick-transmitted diseases of pets and their owners has been associated with increased accessibility of traditional 'wilderness' environments and an increase in the reservoir of wild host species (deer, small mammals and foxes) that now have a closer association with human activity. In addition, there has been a rapid evolution of molecular-based techniques, which has allowed more sensitive and accurate detection of tick-borne pathogens. For example, granulocytic ehrlichiosis in dogs, cats, horses and humans is now considered to be a disease of major importance in Sweden<sup>2</sup>, the USA<sup>3</sup>, Scotland, Switzerland and Slovenia.

Ticks and the diseases they transmit have a zoogeographical range restricted by host movement and, to some extent, climatic factors. However, the increased mobility of pets has resulted in rapid extension of the zoogeographical ranges for many species. In the UK, this will be exacerbated by recent alterations to the animal quarantine regulations (Box 1). Between 1995 and 1998, 36% of cases of monocytic ehrlichiosis reported in Germany occurred in dogs that had travelled for short periods to the

Mediterranean area<sup>4</sup>. In addition, during the same period, both *Ehrlichia canis* infection and infestation with *Rhipicephalus sanguineus*, the ehrlichiosis vector traditionally found in southern Europe, were found in dogs that had never been outside Germany<sup>4</sup>. The zoogeographical range is also increasing because tick species are finding niches in different climatic conditions. Since the 1980s, *Ixodes ricinus* has extended its range in Sweden to include more northern and western areas<sup>5</sup> and *Dermacentor variabilis*, the major vector of Rocky Mountain spotted fever (RMSF), has extended its range to include the northeastern USA<sup>6</sup>.

## Dogs as sentinels for human infection

The medical and veterinary importance of tick infestation in dogs lies in the transmission of a wide variety of infectious agents (Table 1). The most important tick-transmitted infectious diseases causing severe clinical illness in dogs are babesiosis, ehrlichiosis and, in the USA, RMSF and hepatozoonosis. However, although *Borrelia burgdorferi* and *Rickettsia conorii* infections commonly produce subclinical infection, as evidenced by seropositivity, their association with clinical disease in dogs is more difficult to evaluate<sup>7</sup>. Dogs also appear to be susceptible to infection with *Coxiella burnetii* (Q fever) and tick-borne viral encephalitides<sup>8</sup>, but reports of clinical illness are uncommon. Other canine tick-transmitted infections include haemobartonellosis, bartonellosis, tularaemia (*Francisella tularensis*) and, rarely, louping ill (Flaviviridae).

Several of the tick-borne infections that affect dogs can cause serious disease in humans, notably borreliosis, ehrlichiosis, RMSF, *R. conorii* infection and tick-borne encephalitis. However, the potential zoonotic threat posed by dogs is strongly influenced by the natural cycle of the specific agent with which the dog is infected. Three general epidemiological scenarios can be described. First, if transmission of an infectious agent involves ticks with a broad host range (such as *I. ricinus*), dogs can act directly as sentinels for infection of humans. Second, by acting as natural hosts for certain nidicolous ticks (such as *R. sanguineus* and *Ixodes canisuga*), dogs significantly increase contact between these species and humans, thereby increasing the risk of transmission<sup>9</sup>. Finally, there is a limited risk of transmission by exposure to infected-tick contents following damage to ticks during grooming of infested animals. This scenario has been reported for *R. conorii*<sup>10</sup>.

### Susan E. Shaw

Dept of Clinical Veterinary Sciences, University of Bristol, Langford House, Langford, Bristol, UK BS40 5DU  
e-mail: susan.e.shaw@bris.ac.uk

### Michael J. Day

Dept of Pathology and Microbiology, University of Bristol, Langford House, Langford, Bristol, UK BS40 5DU.

### Richard J. Birtles

Dept of Pathology and Microbiology, School of Medical Sciences, University of Bristol, Medical Walk, Bristol, UK BS8 1TD.

### Edward B. Breitschwerdt

College of Veterinary Medicine, North Carolina State University, 4700 Hillsborough St, Raleigh, NC 27606, USA.

### Box 1. The UK Pet Travel Scheme

Since February 2000, the pilot Pet Travel Scheme (PETS) has provided an alternative to the mandatory six-month period of quarantine for dogs and cats entering the UK. Such animals can now travel freely from particular EU countries providing they satisfy the requirements of the scheme (microchip identification, rabies vaccination confirmed by serology and ecto- and endoparasiticide treatment 24–48 h before re-entry to the UK). The scheme is to be extended in April 2001. More than 9000 dogs and cats travelled under the scheme between February and October 2000.

#### Pathogenesis of tick-borne infectious diseases

Although the cutaneous pathology induced by tick bites has been described, there have been a limited number of studies on the process of transmission of infectious agents by ticks to dogs, and even fewer studies on factors that determine vector competency. Most recent studies relate to canine borreliosis, where

infection has been reproduced experimentally for vaccine development<sup>11</sup>.

Recent work using laboratory animals suggests that tick salivary components or intestinal secretions introduced during feeding produce profound changes in the immunological activity of the surrounding skin and draining lymph nodes<sup>12</sup>. Direct suppressive effects of *I. ricinus* and *Dermacentor andersoni* saliva on the host immune system have been reported, including inhibition of T-cell responses to concanavalin A. Immunoglobulin-binding and high-affinity histamine-binding proteins have been isolated from the saliva of several tick species (*Rhipicephalus appendiculatus*, *Amblyomma veriegatum*, *I. hexagonus*); both of these proteins might reduce tick rejection and facilitate disease transmission. There is strong evidence in *R. sanguineus* infestation for a switch from the normal protective T helper 1 (Th1) cytokine profile

Table 1. Emerging tick-transmitted diseases of dogs

Disease agent	Geographical distribution	Tick vectors	Refs
<b>Babesiosis</b>			
<i>Babesia canis canis</i>	Tropical/semiotropical worldwide	<i>Rhipicephalus sanguineus</i> , <i>Dermacentor reticulatus</i> , <i>Dermacentor marginatus</i>	16,19,21–26,45
<i>B. canis vogeli</i>	Tropical/semiotropical worldwide	<i>R. sanguineus</i>	
<i>B. canis rossii</i>	Southern Africa	<i>Haemaphysalis leachi</i>	
<i>B. gibsoni</i>	Africa, Asia, USA, southern Europe, Middle East	<i>Haemaphysalis bispinosa</i> , <i>R. sanguineus</i>	
<b>Hepatozoonosis</b>			
<i>Hepatozoon canis</i>	Southern Europe, Middle East, Far East, Africa	<i>R. sanguineus</i> , <i>Amblyomma maculatum</i> , <i>Haemaphysalis longicornis</i>	27–29,31–35,38
<i>H. americanum</i>	Southern USA	<i>Amblyomma americanum</i>	
<b>Ehrlichia canis genogroup</b>			
<i>E. canis</i>	Southern USA, southern Europe, Africa, Middle East, eastern Asia	<i>R. sanguineus</i>	4,17–20,31–35,38
<i>E. chaffeensis</i>	USA	<i>A. americanum</i> , <i>Dermacentor variabilis</i>	
<i>E. ewingii</i>	USA	Unconfirmed	
<b>E. phagocytophila genogroup</b>			
<i>E. phagocytophila</i>	Northwestern Europe		3,36,37
<i>E. equi</i>	USA	<i>Ixodes</i> species <sup>a</sup>	
Human granulocytic ehrlichiosis agent	Midwestern and northeastern USA, northwestern Europe		
<i>E. platys</i>	USA, southern Europe, Middle East	Unconfirmed	
<b>Borreliosis (Lyme disease)</b>			
<i>Borrelia burgdorferi sensu lato</i>			7,11,15,20,39–42, 46, 47
Genogroups:			
<i>B. burgdorferi sensu stricto</i>	North America, Europe, Middle East		
<i>B. garinii</i>	Europe, Asia	<i>Ixodes</i> species <sup>a</sup>	
<i>B. afzelii</i>	Europe, Asia		
<i>B. japonica</i>	Japan		
<b>Spotted fever group</b>			
<i>Rickettsia conorii</i> (Boutonneuse fever)	Southern Europe, Middle East, Africa	<i>R. sanguineus</i>	9,17,19,30,43
<i>Rickettsia rickettsii</i> (Rocky Mountain spotted fever)	USA	<i>Dermacentor andersoni</i> , <i>D. variabilis</i>	

<sup>a</sup>Species dependent on geographical location, unconfirmed for *B. japonica*.

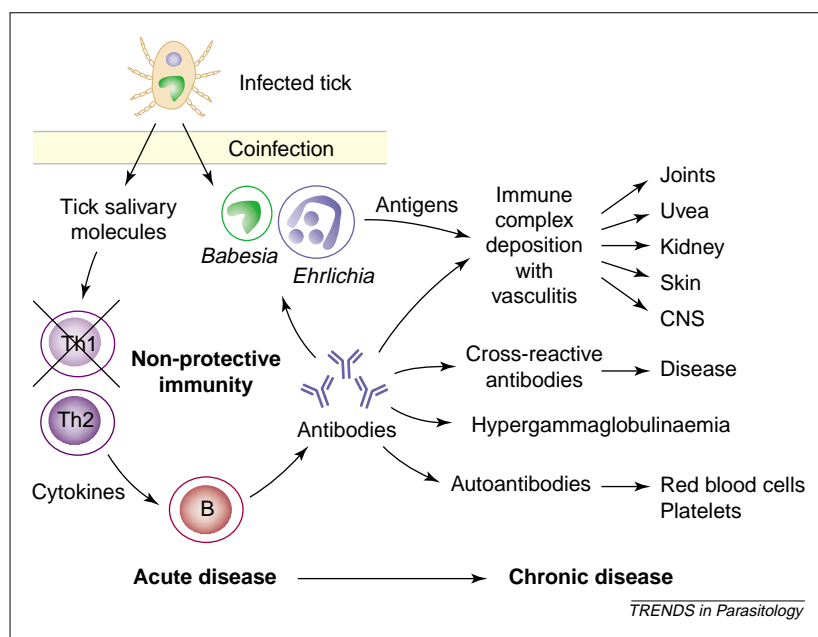


Fig. 1. A hypothetical model for the immunopathogenesis of tick-transmitted disease in non-immune dogs. The bite of an infected tick transmits microorganisms to the host. Tick salivary proteins are able to subvert the skin-associated immune system from the normally protective T helper 1 (Th1) immunity to a Th2 response, which permits establishment of systemic infection. This non-protective immune response is dominated by antibody production, and these antibodies can be significant mediators of the chronic clinical symptoms of disease, via: (1) the vascular effects of hypergammaglobulinaemia; (2) the formation of autoantibodies or antibodies that cross-react with self tissue; and (3) the formation of immune complexes that deposit in predisposed capillary beds and initiate tissue damage. Abbreviation: CNS, central nervous system.

associated with resistance to tick-borne disease to an aberrant Th2 cytokine profile<sup>13</sup>. In addition, a component of the *I. ricinus* salivary gland that inhibits the antiviral effect of interferon has recently been isolated. These local immunomodulatory changes enhance transmission of infectious organisms by *I. ricinus* not only to the host but also to co-feeding ticks<sup>14</sup>.

After successful transmission, tick-transmitted pathogens can induce profound immunological abnormalities (Fig. 1). In fully susceptible dogs, infection can produce clinical syndromes easily recognized in endemic areas. However, despite therapy, animals with an incomplete protective immune response commonly develop persistent subclinical infection which can recrudesce with stress or concurrent disease<sup>15</sup>. In these cases, low numbers of organisms and cryptic infections make diagnosis difficult using routine methods. It is likely that tick-transmitted pathogens contribute significantly to the considerable numbers of dogs with 'pyrexia of unknown origin', lymphadenopathy, chronic cyclic anaemia and thrombocytopenia, polyarthropathy, uveitis or vasculitis. Continued antigenaemia can lead to the induction of polysystemic immune-mediated disease. Infected animals commonly have high serum globulin levels, with monoclonal or polyclonal gammopathies and immune-complex formation. Immunological mechanisms can result in Coombs' positive haemolytic anaemia,

thrombocytopenia, leukopenia or non-erosive polyarthropathy<sup>11,16,17</sup>.

#### Coinfection

Infection with multiple tick-transmitted pathogens, or with multiple genotypes of the same pathogenic species, can occur in an individual animal following heavy exposure to ticks<sup>18,19</sup>. The same tick species can be a vector for several pathogens (Table 1) and coinfection of individual ticks can occur<sup>20</sup>. Infection with tick-borne pathogens can also be complicated by other arthropod-borne diseases that share the tick biohabitat, such as leishmaniasis. In dogs, coinfection with combinations of *Ehrlichia*, *Bartonella*, *Babesia*, *Hepatozoon*, *Leishmania* and *Rickettsia* species occurs in endemic areas. The role of such coinfection is being clarified by PCR<sup>19</sup>. Coinfection could partially explain variations in clinical presentation, pathogenicity and response to therapy.

#### Protozoan diseases transmitted by ticks

##### Babesiosis

Canine babesiosis is caused by the intraerythrocytic protozoan parasite *Babesia canis*, and increasingly by *Babesia gibsoni*, which is extending its range in the USA<sup>21</sup> and Europe. Molecular studies have recently identified novel *Babesia* spp. that infect dogs<sup>22</sup>. Although the pathogenicity of *B. gibsoni* is uniformly high, pathogenicity varies among strains of *Babesia canis*. *Babesia canis rossi*, the prevalent strain in South Africa, causes severe clinical disease; *B. canis canis* in Europe is moderately pathogenic; and *Babesia canis vogeli* infection causes relatively mild disease worldwide<sup>23</sup>. The relative importance of tick species in the transmission of canine babesiosis varies with geographical location.

The clinico-pathogenesis of babesiosis caused by *B. c. canis* and *B. gibsoni* involves progressive haemolytic anaemia. By contrast, the more severe disease caused by *B. c. rossi* can involve hypoxic, hypotensive shock with disseminated intravascular coagulation (DIC), systemic inflammatory response syndrome and multiple organ dysfunction syndrome<sup>24</sup> (Table 2). The severity of disease also varies with the species of vector, and the age, breed and immune status of the dog<sup>23</sup>. Erythrocyte-bound autoantibodies are involved in the haemolytic form of the disease<sup>16</sup>. There can be erythrocyte autoagglutination and many dogs with babesiosis give a positive Coombs' test. Methaemoglobinaemia and methaemoglobinuria occur secondary to oxidative damage in parasitized red blood cells<sup>25</sup>. As a consequence of the methaemoglobinaemia, there is enhanced damage by anti-erythrocyte antibodies and erythrophagocytosis. Persistent infection with *B. canis* or *B. gibsoni* is common in endemic areas<sup>26</sup>. Although these animals appear healthy unless subjected to stress, they provide a reservoir of infection for susceptible animals and have suboptimal athletic performance<sup>26</sup>.

Table 2. Clinical features and chemotherapy of canine tick-transmitted diseases

Disease	Major clinical syndrome	Therapy <sup>a</sup>	Refs
<b>Babesiosis</b>	<b>Uncomplicated disease</b> Intravascular haemolytic anaemia	Imidocarb dipropionate Diminazene aceturate	16,21–26
	<b>Complicated disease</b> Hypotensive shock Multiple organ dysfunction syndrome	Phenamidine isethionate Pentamidine isethionate	
<b>Hepatozoonosis</b>	Myositis with muscle atrophy and paresis	Imidocarb dipropionate Doxycycline	27–29
	Polysystemic immune complex disease	Minocycline Oxytetracycline Tetracycline Sulfonamide/pyrimethamine/ clindamycin in combination	
<b>Ehrlichiosis</b> <i>Ehrlichia canis</i> genogroup	Thrombocytopenia causing bleeding diathesis	Doxycycline Minocycline	18,19,32–35
	Irreversible bone marrow destruction (chronic disease)	Oxytetracycline Tetracycline	
	Polysystemic immune complex disease	Chloramphenicol Rifampin Fluquinolones Imidocarb dipropionate	
<i>E. phagocytophila</i> genogroup	Severe lethargy and weakness Polyarthritits Central nervous signs	Doxycycline Minocycline Oxytetracycline Tetracycline	37
<b>Borreliosis</b>	Non-erosive polyarthropathy Central and peripheral neurological signs	Doxycycline Amoxicillin Azithromycin Penicillin Ceftriaxone Cefotaxime Chloramphenicol	5,7,11,15,39–42
<b>Rocky Mountain Spotted Fever</b>	Subcutaneous oedema owing to necrotising vasculitis Central and peripheral neurological signs	Doxycycline Oxytetracycline Chloramphenicol Enrofloxacin	9,17,43

<sup>a</sup>Therapeutic agents are listed in order of decreasing efficacy, with drug of choice listed first.

### Hepatozoonosis

Canine hepatozoonosis is a tick-transmitted, protozoan disease caused by species of the intraleukocytic parasite *Hepatozoon*. Unlike most other tick-borne diseases, *Hepatozoon* is transmitted by ingestion of an infected tick, rather than tick bites. *Hepatozoon canis* commonly infects dogs in Africa, southern Europe, the Middle East and Asia, reflecting the geographical distribution of its major vector, *R. sanguineus*. However, unique clinical features suggest that a separate species (*Hepatozoon americanum*) transmitted by the tick *Amblyomma maculatum* causes disease in dogs in the southern USA and that this disease is spreading<sup>27,28</sup>. The clinical spectrum of *H. canis* infection ranges from subclinical to severe life-threatening disease<sup>28,29</sup>. *Hepatozoon canis* is commonly associated with coinfection with other diseases, in particular ehrlichiosis and leishmaniasis in endemic areas, and clinical presentations are variable (Table 2).

### Bacterial diseases transmitted by ticks

#### *Ehrlichiosis*

Canine ehrlichiosis is caused by tick-transmitted intracellular bacteria of the genus *Ehrlichia*, which, in dogs, have been identified parasitizing monocytes, granulocytes and platelets. Three genogroups of ehrlichiae have now been identified by 16S rRNA<sup>30</sup> phylogenetic analysis (Table 1). Genogroup III includes *E. canis*, which is responsible for widespread disease in tropical and temperate areas of the world. The geographical distribution of *E. canis* has expanded with the distribution of *R. sanguineus*. More recently, other genogroup III species, *E. chaffeensis* and *Ehrlichia ewingii*, have been identified by PCR in naturally infected dogs, with or without ehrlichiosis symptoms<sup>18,19</sup>. These species have more restricted geographical distributions but a potentially wider range of tick vectors than *E. canis*. A subspecies of *Ehrlichia risticii* (Genogroup I) has also been reported to cause an atypical syndrome of monocytic ehrlichiosis in dogs in the USA<sup>31</sup>.

Disease manifestations caused by members of the *E. canis* genogroup (genogroup III) infecting dogs can be indistinguishable<sup>18</sup> (Table 2), and there can be strain variation in pathogenicity<sup>32</sup>. At present, information on the pathogenesis of experimental monocytic ehrlichiosis relates primarily to *E. canis*. Immunological destruction of platelets occurs in acute disease and anti-platelet antibodies have been found in naturally occurring and experimental cases<sup>33</sup>. Autoantibodies decrease platelet life-span, and interfere with platelet membrane glycoproteins, causing inhibition of aggregation. Other factors such as splenic sequestration and the production of a cytokine, platelet migration-inhibition factor, are also involved in the pathogenesis of thrombocytopenia. Hyperviscosity owing to hyperproteinaemia adds further to platelet dysfunction and can result in ocular and central nervous system (CNS) abnormalities.

Subclinical persistent infection owing to splenic sequestration of organisms is common<sup>34</sup>. Severe life-threatening chronic ehrlichiosis can develop following persistent infection and can be associated with irreversible bone marrow destruction. Factors that predispose to myelofibrosis are not understood. Ehrlichiosis is more severe in certain breeds (e.g. German shepherd) and in younger animals. However, coinfection, immune status and strain variation could all play a role<sup>35</sup>.

Genogroup II Ehrlichiae of pathogenic significance include the *Ehrlichia phagocytophila* group and *Ehrlichia platys*. Strains (currently species) within the *E. phagocytophila* genogroup (*Ehrlichia equi*, human granulocytic ehrlichiosis agent and *E. phagocytophila*) are transmitted by *Ixodes* spp. ticks, and are the major causative agents of canine ehrlichiosis in the northern and western USA<sup>3</sup> and in northern and central Europe<sup>2,36</sup>.

Infection with species of the *E. phagocytophila* group is generally associated with less severe clinical signs than ehrlichiosis caused by *E. canis* (Table 2). Persistent sub-clinical infection has recently been identified with the granulocytic *Ehrlichia* species in dogs in Sweden<sup>37</sup>. However, the severe chronic disease seen with *E. canis* in susceptible dogs has not been reported. As granulocytic ehrlichiosis is transmitted by *Ixodes* spp. ticks, coinfection with *Borrelia* in dogs is probable, as reported in humans.

Canine cyclic thrombocytopenia (Table 2), caused by *E. platys*, was first reported in the USA and appears to be an emerging problem in several southern European countries, Israel, Taiwan and Venezuela<sup>38</sup>. Although it is assumed to be transmitted by *R. sanguineus*, its natural mode of transmission is uncertain. Pathogenicity is generally low but *E. platys* infection might play a role in coinfection with other arthropod-borne diseases.

#### *Borreliosis*

Canine borreliosis is caused by a spirochete, *B. burgdorferi sensu lato*, which is transmitted by

ticks of the genus *Ixodes* (Table 1). At least four genospecies, with geographical distributions primarily in the northern hemisphere, cause disease in humans<sup>39</sup> (Table 1). In northern Europe, the distribution of borreliosis is expanding, and infected *I. ricinus* ticks are now commonly found in urban areas<sup>5,40</sup>. Much of the published borreliosis research relates to the Genogroup I species, *B. burgdorferi sensu stricto*, which is the primary isolate in humans and dogs in the USA. There is considerable genetic heterogeneity between North American and European isolates of *B. burgdorferi*<sup>41</sup>. In Europe, human borreliosis is also caused by *Borrelia garinii* and *Borrelia afzelii*. and in Japan, by *Borrelia japonica*, a separate genospecies. The degree to which *B. garinii*, *B. afzelii* and *B. japonica* contribute to canine infection and disease is currently unclear. However, infection with these *Borrelia* spp. could explain the disparity in pathogenicity and clinical syndromes described<sup>42</sup>.

Although a high proportion of dogs are seropositive in endemic areas, relatively few develop clinical signs<sup>7</sup> (Table 2). Several mechanisms have been incriminated in causing joint damage. The production of the inflammatory mediator nitric oxide is upregulated, as is interleukin 8, a cytokine that recruits neutrophils into infected synovial membranes<sup>11</sup>. Neurological abnormalities occur in some cases of borreliosis, most particularly in Japan<sup>42</sup> (Table 2). Immunological cross-reactions to bacterial and self-antigens are important in human neuroborreliosis (Fig. 1), and antibodies to flagellin, one of the most immunogenic *Borrelia* antigens, have been shown to cross-react with neuroaxonal proteins. In canine borreliosis, cutaneous and cardiac disease are rare.

Persistent infection with *Borrelia* even after antibiotic therapy is reportedly common in dogs<sup>15</sup>. The organism is sequestered in the skin, connective tissue, joints and CNS. Reactivation of infection with recrudescence of disease can occur in immunocompromised individuals or in coinfection.

#### *The spotted fever group*

RMSF is a potentially fatal rickettsial disease of dogs and humans caused by the intracellular bacterium *Rickettsia rickettsii*. It has been reported throughout the USA, Central and South America, and is transmitted by ticks of the *Dermacentor*, *Rhipicephalus* and *Amblyomma* genera, respectively (Table 1). *Rickettsia conorii*, the agent of boutonneuse fever in humans in southern Europe, the Middle East and southern Africa (Table 1), is reported to infect dogs<sup>9</sup>, but clinical signs of disease have not been reported.

The major pathogenic mechanism involved in canine RMSF is rickettsial invasion of, and damage to, endothelial cells of small arteries and venules (Table 2). Although platelet consumption is considered the primary cause of the thrombocytopenia in clinical

cases, anti-platelet antibodies have also been identified in infected dogs<sup>17</sup>. A tendency towards more fulminant disease has been reported in Springer spaniels with phosphofructokinase deficiency<sup>43</sup> and in German shepherds.

#### Diagnosis of tick-borne diseases

Diagnosis of tick-transmitted diseases requires a combination of compatible clinical and laboratory findings, direct microscopic visualization or immunodetection of infective organisms in blood or infected tissue, microbial culture, serological testing, immunoblotting and PCR. Although in endemic areas, direct visualization (using light microscopy) of *Babesia*, *Ehrlichia* and *Hepatozoon* spp. is valuable, the tendency for chronic and/or subclinical disease to be associated with cryptic infection reduces its sensitivity and does not allow intraspecies differentiation. However, the use of direct immunohistochemical or immunocytochemical methods can increase sensitivity. Cultivation of tick-borne bacteria is generally difficult as these are fastidious organisms, and eukaryotic cell co-culture systems are often required.

Serological testing is the most commonly used diagnostic methodology, and the indirect fluorescent antibody and ELISA tests are most widely used. However, serological testing is limited by reduced ability to identify acute infection, difficulty in differentiating infection from prior exposure or vaccinal titres, and species cross-reactivity. Western immunoblotting has been used to characterize and distinguish different species involved. However, PCR assays are becoming increasingly practicable for all tick-borne infections already described. The sensitivity of PCR makes it particularly appropriate for diagnosis of this group of diseases, although this technique does not distinguish between acute infection and a carrier state.

#### Strategies for control

##### Vector control

Tick control using an effective long-acting acaricide, for example fipronil<sup>44</sup>, permethrin or amitraz in spot-on, spray or collar formulation according to manufacturers instructions, remains the most effective preventative measure for this group of diseases. This is particularly the case where unexposed dogs are travelling into endemic areas or where they are involved in activities where exposure to ticks can be high, such as hunting and herding. Tick eradication is impossible in most situations because of maintenance of the tick life cycle on reservoir hosts.

##### Chemotherapy and chemoprophylaxis

The treatment of tick-transmitted diseases is a challenge for a variety of reasons (Table 2). Many of the target organisms are intracellular and sequestration limits drug penetration and,

consequently, drug efficacy. Therapy with a single chemotherapeutic agent might not be sufficient to eliminate infection. There are species variations in response to therapy; for example, *B. gibsoni* is relatively unresponsive to the drugs used to treat *B. canis*. In addition, some drugs have toxic side-effects. Poor or partial responses to single-agent chemotherapy might also reflect the presence of coinfection.

##### Vaccination

Although vaccination is a potentially important means of control, there are inherent problems in designing effective vaccines against organisms that can evade host immunity effectively and that have species or strain variability. Moreover, relatively little is known of what constitutes a protective immune response to these agents in dogs, and such endogenous immune responses can be subverted by tick-derived soluble factors, as discussed. Despite this, commercially produced vaccines are available for canine babesiosis and borreliosis.

The canine babesiosis vaccine is based on exoantigens of *B. canis* obtained from cell culture, and is available in Europe<sup>45</sup>. The vaccine does not prevent infection, but reduces parasitaemia and limits the severity of anaemia and splenomegaly. Moreover, the vaccine is capable of protection only from homologous strains of *B. canis*<sup>45</sup>. By contrast, inactivated bacterins and a recombinant vaccine based on the outer surface protein OspA (Ref. 46) are commercially available for canine borreliosis. OspA is expressed by *Borrelia* in unfed ticks, and, following transmission to the host, is replaced by OspC expression. Anti-OspA antibodies induced by vaccination and ingested by feeding ticks halt the growth and migration of *Borrelia* to the salivary gland. A combined OspA–OspB subunit vaccine has also been evaluated experimentally in the dog<sup>47</sup>.

In experimental challenge studies, these vaccines appear to protect from spirochetaemia and clinical signs (polyarthritis) relative to unvaccinated control dogs. The vaccines are recommended for use in endemic areas but, as they are based on *B. burgdorferi sensu stricto*, they might not crossprotect against other *Borrelia* species. Although not commercially available, experimental vaccines for canine monocytic ehrlichiosis (*E. canis*) have also been reported. Antigen extracts from cell-cultured *E. canis*, administered with adjuvant, induced antibody-mediated but not protective immunity, whereas a vaccine based on inactivated whole *E. canis* organisms was able to protect dogs from experimental challenge<sup>35</sup>.

#### Conclusions

The tick-transmitted infectious diseases of dogs that are described in this review have recently become a major focus of interest in areas of the world in which they have traditionally been considered non-endemic.

## Acknowledgements

We apologize to all the authors whose primary contributions we were unable to reference because of editorial limitations on reference numbers, and hope that original citations will be accessible through the review papers.

This relates to both their significance to canine health, and to the possible reservoir status of the dog for potentially zoonotic disease. The concern that these diseases might become established in new geographical locations arises from the increased international mobility of pet dogs and increased contact of these animals with non-urban environments and wildlife disease reservoirs. These factors, coupled with the trend for global climatic change, create real risks for animal and human health. These changes come at a time when PCR now

enables rapid screening of blood samples for multiple tick-transmitted pathogens, and large-scale epidemiological surveys of disease prevalence. There is an urgent need for the provision of baseline data on the prevalence of these diseases in dogs (and ticks) in traditionally non-endemic areas (e.g. northern Europe), so future trends can be monitored. Further research is required to understand fully the immunopathogenesis of these diseases in dogs, to develop more effective chemotherapy and prophylactic vaccines.

## References

- Hillyard, P.D. (1996) Diseases carried by ticks in NW Europe: their medical and veterinary importance. In *Ticks of North-West Europe. Synopses of the British Fauna (New Series)* (Barnes, R.S.K. and Coles, J.H., eds), pp. 22–23, FSC Publications
- Egenvall, A. *et al.* (1994) Tickborne infections in dogs in Sweden. *Svensk Veterinær Tidning* 46, 321–329
- Greig, B. *et al.* (1996) Geographic, clinical, serologic and molecular evidence of granulocytic ehrlichiosis, a likely zoonotic disease in Minnesota and Wisconsin dogs. *J. Clin. Microbiol.* 34, 44–48
- Gothe, R. (1999) *Rhipicephalus sanguineus* (Ixodidae): frequency of infestation and ehrlichial infections transmitted by this tick in dogs in Germany; an epidemiological study and consideration. *Wiener Tierärztliche Monatsschrift* 86, 49–56
- Talleklint, L. and Jaenson, T.G.T. (1998) Increasing geographical distribution and density of *Ixodes ricinus* in central and northern Sweden. *J. Med. Entomol.* 35, 521–526
- Snetsinger, R. *et al.* (1993) Extension of the range of *Dermacentor variabilis* (Acari, Ixodidae) in Pennsylvania. *J. Med. Entomol.* 30, 795–798
- Levy, S.A. and Magnarelli, L.A. (1992) Relationship between development of antibodies to *Borrelia burgdorferi* in dogs and the subsequent development of limb/joint borreliosis. *J. Am. Vet. Med. Assoc.* 200, 344–347
- Weissenböck, H. and Holtzmann, H. (1996) Tick-borne encephalitis in Austrian dogs. *Vet. Rec.* 139, 575–576
- Mumcuoglu, K.Y. *et al.* (1993) Ecological studies on the brown dog tick *Rhipicephalus sanguineus* (Acari, Ixodidae) in southern Israel and its relationship to spotted-fever group rickettsiae. *J. Med. Entomol.* 30, 114–121
- Senneville, E. *et al.* (1991) *Rickettsia conorii* isolated from ticks introduced to northern France by a dog. *Lancet* 337, 676
- Straubinger, R.K. *et al.* (1997) *Borrelia burgdorferi* migrates into joint capsules and causes up-regulation of interleukin-8 in synovial membranes of dogs experimentally infected with ticks. *Infect. Immun.* 65, 1273–1285
- Willadsen, P. and Jongejan, F. (1999) Immunology of the tick–host interaction and the control of ticks and tick-borne diseases. *Parasitol. Today* 15, 258–262
- Ferreira, B.R. and Silva, J.S. (1999) Successive tick infestations selectively promote a T-helper 2 cytokine profile in mice. *Immunology* 96, 434–439
- Labuda, M. *et al.* (1997) Tick borne encephalitis virus transmission between ticks co-feeding on specific immune natural rodent hosts. *Virology* 235, 138–143
- Straubinger, R.K. *et al.* (1997) Persistence of *Borrelia burgdorferi* in experimentally infected dogs after antibiotic treatment. *J. Clin. Microbiol.* 35, 111–116
- Adachi, K. *et al.* (1992) Anti-erythrocyte membrane antibodies detected in sera of dogs naturally infected with *Babesia gibsoni*. *J. Vet. Med. Sci.* 54, 1081–1084
- Grindem, C.B. *et al.* (1999) Platelet-associated immunoglobulin (antiplatelet antibody) in canine Rocky Mountain spotted fever and ehrlichiosis. *J. Am. Anim. Hosp. Assoc.* 35, 56–61
- Breitschwerdt, E.B. *et al.* (1998) Sequential evaluation of dogs naturally infected with *Ehrlichia canis*, *Ehrlichia chaffeensis*, *Ehrlichia equi*, *Ehrlichia ewingii* or *Bartonella vinsonii*. *J. Clin. Microbiol.* 36, 2645–2651
- Kordick, S.K. *et al.* (1999) Coinfection with multiple tick-borne pathogens in a Walker Hound kennel in North Carolina. *J. Clin. Microbiol.* 37, 2931–2938
- Schouls, L.M. *et al.* (1999) Detection and identification of *Ehrlichia*, *Borrelia burgdorferi sensu lato* and *Bartonella* species in Dutch *Ixodes ricinus* ticks. *J. Clin. Microbiol.* 37, 2215–2222
- Birkenheuer, A.J. *et al.* (1999) *Babesia gibsoni* infections in dogs from North Carolina. *J. Am. Anim. Hosp. Assoc.* 35, 125–128
- Zahler, M. *et al.* (2000) Detection of a new pathogenic *Babesia microti*-like species in dogs. *Vet. Parasitol.* 89, 241–248
- Irwin, P.J. and Hutchinson, G.W. (1991) Clinical and pathological findings of *Babesia* infection in dogs. *Aust. Vet. J.* 68, 204–209
- Welz, C. *et al.* (2000) Systemic inflammatory response syndrome and secondary multiple organ dysfunction syndrome in canine babesiosis. *J. Vet. Intern. Med.* 14, 244
- Morita, T. *et al.* (1996) Erythrocyte oxidation in artificial *Babesia gibsoni* infection. *Vet. Parasitol.* 63, 1–7
- Taboada, J. *et al.* (1992) Seroprevalence of babesiosis in greyhounds in Florida. *J. Am. Vet. Med. Assoc.* 200, 47–50
- Mathew, J.S. *et al.* (1998) Experimental transmission of *Hepatozoon americanum* (Vincent-Johnson *et al.*, 1997) to dogs by the Gulf Coast tick, *Amblyomma maculatum*. *Vet. Parasitol.* 80, 1–14
- Macintire, D.K. *et al.* (1997) Canine hepatozoonosis: a retrospective study of 22 naturally occurring cases (1989–1994). *J. Am. Vet. Med. Assoc.* 210, 916–922
- Baneth, G. *et al.* (1996) *Hepatozoon canis*: prevalence of antibodies and gametocytes in dogs in Israel. *Vet. Res. Commun.* 20, 41–46
- Drancourt, M. and Raoult, D. (1994) Taxonomic position of the Rickettsiae: current knowledge. *FEMS Microbiol. Rev.* 13, 13–24
- Kakoma, I. *et al.* (1994) Cultural, molecular and immunological characterisation of the agent for atypical canine ehrlichiosis. *J. Clin. Microbiol.* 32, 170–175
- Hegarty, B.C. *et al.* (1997) Immunoblot analysis of the immunoglobulin G response to *Ehrlichia canis* in dogs: an international survey. *J. Vet. Diag. Invest.* 9, 32–38
- Gaunt, S.D. *et al.* (1996) Platelet associated IgG and antibodies to platelet proteins in dogs with *Ehrlichia canis* infection. *Vet. Pathol.* 33, 557
- Harrus, S. *et al.* (1998) Amplification of ehrlichial DNA from dogs 34 months after infection with *Ehrlichia canis*. *J. Clin. Microbiol.* 36, 73–76
- Harrus, S. *et al.* (1999) Recent advances in determining the pathogenesis of canine monocytic ehrlichiosis. *J. Clin. Microbiol.* 37, 2745–2749
- Johansson, K.E. *et al.* (1995) Identification of the causative agent of granulocytic ehrlichiosis in Swedish dogs and horses by direct solid phase sequencing of PCR products. *Res. Vet. Sci.* 58, 109–112
- Egenvall, A. *et al.* (2000) Detection of granulocytic *Ehrlichia* species DNA by PCR in persistently infected dogs. *Vet. Rec.* 146, 186–190
- Harrus, S. *et al.* (1997) Clinical manifestations of infectious canine cyclic thrombocytopaenia. *Vet. Rec.* 141, 247–250
- Fillipuzzi-Jenny, E. *et al.* (1993) Genetic diversity among *Borrelia burgdorferi* isolates: more than three genospecies? *Res. Microbiol.* 144, 295–304
- Junttila, J. *et al.* (1999) Prevalence of *Borrelia burgdorferi* in *Ixodes ricinus* ticks in urban recreational areas of Helsinki. *J. Clin. Microbiol.* 37, 1361–1365
- Lorvich, S.D. *et al.* (1994) Seroprotective groups of Lyme borreliosis spirochaetes from North America and Europe. *J. Infect. Dis.* 170, 115–121
- Azuma, Y. *et al.* (1994) Canine Lyme disease: clinical and serological evaluations in 21 dogs in Japan. *Vet. Rec.* 134, 369–372
- Weiser, I.B. and Greene, C.E. (1989) Dermal necrosis associated with Rocky Mountain spotted fever in four dogs. *J. Am. Vet. Med. Assoc.* 195, 1756–1758
- Searle, A. *et al.* (1995) Results of a trial of fipronil as an adulticide on ticks (*Ixodes holocyclus*) naturally attached to animals in the Brisbane area. *Aust. Vet. Practit.* 25, 157–158
- Schettler, T.H. *et al.* (1997) Vaccination of dogs against *Babesia canis* infection. *Vet. Parasitol.* 73, 35–41
- Ma, J.N. *et al.* (1996) Safety, efficacy and immunogenicity of a recombinant Osp subunit canine Lyme disease vaccine. *Vaccine* 14, 1366–1374
- Chang, Y.F. *et al.* (1995) Recombinant OspA protects dogs against infection and disease caused by *Borrelia burgdorferi*. *Infect. Immun.* 63, 3543–3549