CHAPTER 51

Lyme Borreliosis

Jane E. Sykes

Overview of Lyme Borreliosis

First Described: Connecticut, USA, 1982 (Burgdorfer)[1]

Cause: *Borrelia burgdorferi* sensu lato (a spirochete)

Primary Mode of Transmission: *Ixodes* spp. ticks

Affected Hosts: Humans and a large variety of animals; disease occurs in dogs, humans, horses, cattle

Geographic Distribution: North America, Europe, Asia

Major Clinical Signs: Fever, lethargy, inappetence, lameness due to polyarthritis. Signs of Lyme nephropathy include vomiting, weight loss, and polyuria and polydipsia.

Differential Diagnoses: Differential diagnoses for suspected Lyme polyarthritis includes bilateral cruciate ligament rupture, primary immune-mediated polyarthritis, septic polyarthritis, and polyarthritis secondary to infection with other pathogens such as *Ehrlichia ewingii, Bartonella* spp., *Ehrlichia canis, Anaplasma phagocytophilum, Rickettsia rickettsii,* and fungal organisms. Differential diagnosis for suspected Lyme nephritis include leptospirosis, bacterial pyelonephritis, primary immune-mediated glomerulonephritis, familial nephropathies, amyloidosis, and glomerulonephritis secondary to other chronic infections such as *Dirofilaria immitis, Babesia canis, E. canis,* and *Leishmania* spp. infections.

Human Health Significance: Dogs and cats are not a direct source of human infection but may bring unfed infected ticks into the house. Evidence of canine exposure to *B. burgdorferi* is a sentinel for human exposure.

Etiologic Agent and Epidemiology

Lyme borreliosis, or Lyme disease, is a vector-borne spirochetosis caused by the motile, corkscrew-shaped bacterium *Borrelia burgdorferi* sensu lato. Lyme disease occurs in North America, Europe, and Asia. It was named after Old Lyme in Connecticut, where clusters of the disease were first recognized in children with juvenile rheumatoid arthritis in 1976.[2] However, chronic cutaneous manifestations of infection were recognized in human patients in Germany as far back as 1883.[3] The spirochete was first detected in ticks by Burgdorfer in 1982.[1] A variety of different mammalian hosts and humans may be infected. The disease has been increasingly recognized and is now the most common vector-borne infectious disease of humans in the United States and Europe. Reforestation of farmland and proliferation of deer and tick populations may have contributed to emergence of the disease in humans.[4] Cats can be infected and seroconvert, but appear to be relatively resistant to development of disease.[5] Dogs can develop fever, arthritis, and renal disease, but most infected dogs show no signs of illness.

More than 15 different species of *B. burgdorferi* sensu lato have been described, some of which are nonpathogenic (Table 51-1). *Borrelia turicatae,* the cause of tick-borne relapsing fever in humans, has been detected in sick dogs from Texas.[6] The DNA of *Borrelia lonestari,* which is thought to be nonpathogenic, has been detected in dogs from Arkansas.[7] In North America, Lyme disease is caused by *Borrelia burgdorferi* sensu stricto, whereas in Europe and Asia, other species that belong to *Borrelia burgdorferi* sensu lato are more important, specifically *Borrelia afzelii* and *Borrelia garinii*. This may account for differences in clinical manifestations that occur in human patients in Europe compared with those in North America (see Public Health Significance, later). Some strains of *B. burgdorferi* appear to have increased virulence, such as *B. burgdorferi* OspC type A within the United States.[8]

B. burgdorferi sensu lato is transmitted by *Ixodes ricinus-persulcatus* complex ticks (Table 51-2). The geographic distribution of Lyme disease reflects that of the vector ticks as well as the competency of the reservoir hosts involved. In the United States, the vectors are *Ixodes scapularis* in the east and upper Midwest, and *Ixodes pacificus* in the West (see Chapter 29). Major foci of infection exist in the upper Midwest, Northeast, mid-Atlantic, and parts of northern California (Figure 51-1).[9] In some endemic areas, seroprevalence in dogs is nearly 90%,[10] although seroprevalence in a study that used a C6 ELISA assay (see Diagnosis, later) was lower than this in endemic areas such as Connecticut and Massachusetts, around 20%.[11] Infection is passed transstadially within the tick (i.e., from larva to nymph to adult), and not transovarially (from adult to egg). Reservoirs for the spirochete in the Northeast and the upper Midwest are *Peromyscus leucopus,* the white-footed mouse, which can harbor large numbers of the organism without overt signs of illness,[12] as well as shrews and chipmunks.[13] In the western United States, the western gray squirrel is the primary reservoir host.[14] Lyme disease is less prevalent in the western United States because *I. pacificus* prefers to feed on the western fence lizard, a poor reservoir for the spirochete. Similarly, the low prevalence of Lyme disease in the southeastern United States, is because *I. scapularis* ticks feed primarily on lizards in this region.[15] *B. burgdorferi* is primarily transmitted to humans by nymphal ticks, because they are extremely small and often enough go unnoticed. Adult ticks may be more important for transmission of infection to dogs.[16] The nymphs of *I. scapularis* quest in the late spring and summer, when humans and dogs are often outdoors and become exposed. The peak questing times for *I. scapularis* adult ticks are in the spring and fall. Other

TABLE 51-1

Pathogenic and Nonpathogenic Species That Belong to *Borrelia burgdorferi* sensu lato and Their Geographic Distributions

	United States	Europe	Asia
Pathogenic	*B. burgdorferi* sensu stricto	*B. burgdorferi* sensu stricto *B. afzelii* *B. garinii*	*B. afzelii* *B. garinii*
Non-pathogenic or questionable pathogenicity	*B. bissettii* *B. andersonii* *B. californiensis* *B. carolinensis* *B. americana* *B. kurtenbachii*	*B. valaisiana** *B. bissettii** *B. spielmanii** *B. lusitaniae* *B. bavariensis*	*B. bissettii** *B. japonica* *B. turdi* *B. tanukii* *B. sinica* *B. yangtze*

**B. valaisiana* and *B. bissettii* have been isolated from single cases of Lyme borreliosis; *B. spielmanii* has been detected in early skin lesions (Stanek G, Reiter M. The expanding Lyme *Borrelia* complex—clinical significance of genomic species? Clin Microbiol Infect 2011;17:487-493).

TABLE 51-2

Vectors of Lyme Disease Worldwide

Geographic Location	Tick Species	Common Name	Reservoir host for *Borrelia*
Western United States	*Ixodes pacificus*	Pacific black-legged tick	Western gray squirrel
Northeastern and upper Midwestern United States	*Ixodes scapularis*	Black-legged tick	White-footed mouse, shrews, chipmunks
Europe	*Ixodes ricinus*	Castor bean or sheep tick	Squirrels, thrushes, chipmunks, mice, shrews, hedgehogs, rats, hares, pheasants, voles
Asia	*Ixodes persulcatus*	Taiga tick	Voles

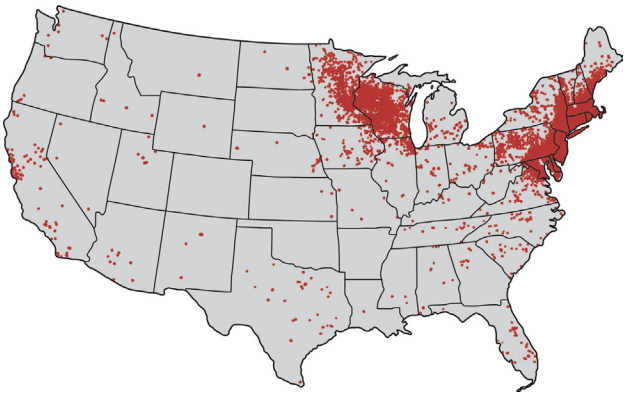

FIGURE 51-1 Approximate geographic distribution of Lyme borreliosis in humans (and dogs) in the United States.

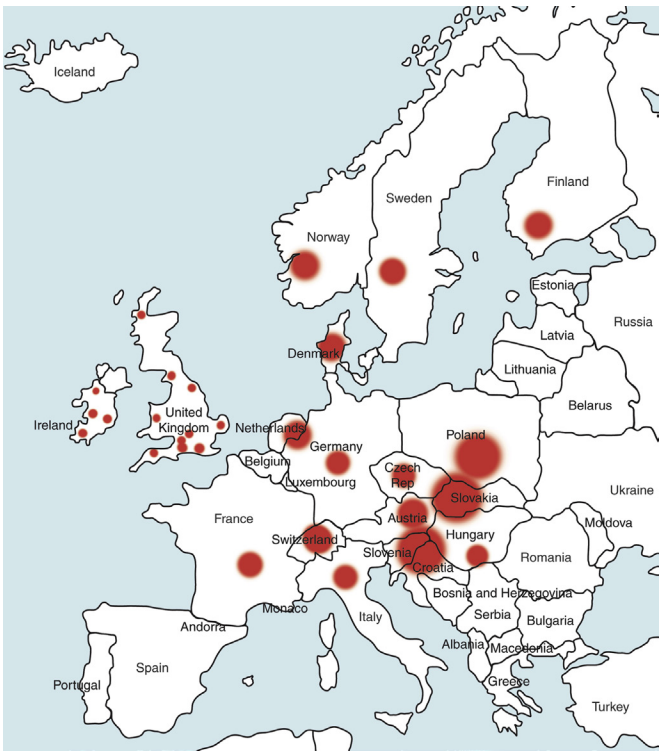

FIGURE 51-2 Countries and regions where Lyme borreliosis occurs in Europe. The size of the red dot correlates with the relative prevalence of the infection in those areas.

vector-borne pathogens, such as *Anaplasma phagocytophilum,* may be co-transmitted and complicate the clinical picture. There is some evidence that co-infection with *A. phagocytophilum* can enhance the pathogenicity of *B. burgdorferi*.[17]

In continental Europe, Lyme disease is most prevalent in central and eastern Europe, especially Poland, Slovakia, and Slovenia, but also Germany, Switzerland, Austria, and southern Scandinavia (Figure 51-2). *B. garinii* was detected in a dog from the Czech republic with meningoencephalitis,[18] and *B. afzelii* has been detected in dogs from Poland.[19] Evidence of infection has also been found in dogs from the British Isles,[20] where endemic areas include the Scottish Highlands, Ireland, Wales, the Lake District, the Yorkshire Moors, Exmoor, Wiltshire, Berkshire, and particularly the New Forest, the South Downs, and the Thetford Forest region. A huge variety of reservoir hosts appear to be involved in Europe. Small rodents such as squirrels appear to be an important reservoir host for *B. afzelii*, and birds such as blackbirds and song thrushes are important reservoir hosts for *B. garinii*.[21]

The complete genome of *B. burgdorferi* has been sequenced. The organism expresses different outer-surface lipoproteins at different stages of infection, which allows it to adapt to dramatically different environments within the arthropod vector and the mammalian host. This property of the organism has been exploited for the purpose of diagnostic assay and vaccine development. Outer surface proteins of importance include OspA, OspC, and VlsE. Over the fall, winter, and spring, the spirochete remains dormant within the nymphal tick and expresses OspA, which allows it to adhere to the tick midgut. When the tick ingests mammalian blood in the late spring and summer, OspA expression is downregulated by the spirochete, and OspC expression is upregulated.[22] The spirochete then moves to the tick hemolymph

and salivary glands. OspC binds a tick salivary gland protein, which may help the organism evade the host immune response. OspC also binds to mammalian plasminogen and helps the spirochete to disseminate within the mammalian host.[23] VlsE undergoes recombinational shuffling of its genetic code, which further allows the spirochete to evade the immune response.[24] Finally, *B. burgdorferi* may undergo metamorphosis into a spherical shape when it encounters unfavorable conditions within the host, such as when it is exposed to antibiotics, nutrient deprivation, and changes in pH.[25] This may also contribute to its ability to evade the immune system and resist antimicrobial drug treatment.

Clinical Features

Signs and Their Pathogenesis

Transmission is thought to require tick attachment for a minimum of 24 hours,[26] but in some cases transmission may occur at earlier time points.[27] The spirochete replicates at the site of tick attachment, then disseminates to multiple locations. Although it can be transiently found in blood, the organism primarily replicates and spreads through connective tissue (Figure 51-3). It binds proteins such as plasminogen, β_3 integrins such as the platelet integrin $\alpha_{IIb}\beta_3$, glycosaminoglycans, fibronectin, laminin, and decorin (a collagen proteoglycan).[28] After invasion, the organism can persist in dogs for over a year, through evasion of host immune responses.[29]

In contrast to human Lyme disease, dogs do not develop an early skin rash (erythema migrans). It has been estimated that only 10% of affected dogs show overt signs of illness. Initial signs in dogs occur 2 to 5 months after a tick bite and consist of variable fever, inappetence, thrombocytopenia, and lameness due to neutrophilic polyarthritis. In a study of experimentally infected dogs, the first joint to be affected was the joint closest to the site of the tick bite, which supports the notion that spirochetes reach the joints as a result of spread through connective tissue.[29,30] Subsequently, other joints can be affected, and dogs may develop shifting lameness. It is not clear whether dogs develop other clinical manifestations seen in humans such as persistent, antibiotic-refractory Lyme arthritis; uveitis; carditis; or encephalopathy. Complete heart block was described in a seropositive dog from Connecticut that had pathologic changes at necropsy consistent with Lyme carditis.[31]

Lyme nephritis is a syndrome of membranoproliferative glomerulonephritis that has been recognized in dogs in association with exposure to *B. burgdorferi*. Rare reports of a similar disease in human patients exist.[32] Golden and Labrador retriever breeds appear to be overrepresented,[33] and it has been suggested that Shetland sheepdogs and Bernese mountain dogs might also be predisposed.[34-36] Dogs with Lyme nephritis are younger than dogs with other glomerular diseases. In one study, more than 50% of dogs with Lyme nephritis were 5 years of age or younger.[33] Many affected dogs are also thrombocytopenic, and some also have polyarthritis.[33] The pathogenesis of Lyme nephritis is uncertain; immune-mediated mechanisms have been proposed.[37] The DNA or antigen of the spirochete cannot be consistently detected within renal biopsies.[37,38] Subendothelial deposits of IgM, IgG, and C3 can be detected within the glomeruli.[33] Clinical signs result from proteinuria and renal failure and include inappetence, lethargy, weight loss, vomiting, and polyuria and polydipsia.

Physical Examination Findings

Physical examination findings in dogs with Lyme arthritis include lethargy, fever, lameness, and swollen, warm, painful joints. Mild generalized peripheral lymphadenopathy may be present. Dogs with Lyme nephritis may have a thin body condition, dehydration, and show evidence of peripheral edema, pleural effusion, or ascites, especially after crystalloid fluid administration. Complications of hypertension can sometimes occur, such as retinal detachment.

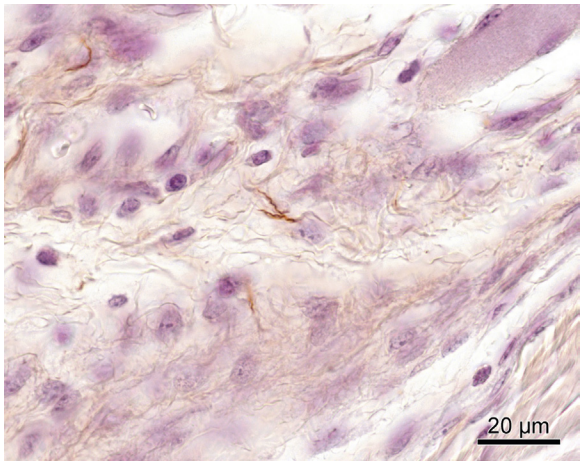

FIGURE 51-3 *Borrelia burgdorferi* in the metatarsal tendon sheath of a mouse with severe combined immunodeficiency. (Courtesy Dr. Stephen Barthold, Center for Comparative Medicine, University of California, Davis.)

Diagnosis

Diagnosis of Lyme disease is notoriously difficult. The overwhelming majority of infected dogs experience subclinical infection, after which positive antibody titers may persist for months or years. Because only a small percentage (e.g., <10%) of infections result in disease, it is important to distinguish between diagnosis of *infection* or previous exposure to the spirochete and diagnosis of Lyme *disease*. Dogs with positive antibody titers may develop unrelated illnesses, which results in diagnostic confusion. As in human patients, the diagnosis must be established based on characteristic clinical signs, a history of exposure in an endemic area, and a positive antibody response to *B. burgdorferi*.[39] A history of tick exposure may not always be present.

Laboratory Abnormalities

Complete Blood Count

The most common finding on the CBC of dogs with Lyme polyarthritis or nephritis is mild to moderate thrombocytopenia. The mechanism of thrombocytopenia is not known, but it may be immune mediated, because treatment of dogs with Lyme nephritis with immunosuppressive drugs can result in normalization of the platelet count. Mild to moderate anemia and leukocytosis due to a mature neutrophilia may be present in dogs with Lyme nephritis, but white cell counts may also be normal. Lymphopenia and mild neutropenia can also occur.

Serum Biochemical Tests

Dogs with polyarthritis typically have minimal changes on serum biochemistry testing. Dogs with Lyme nephritis may show azotemia, mild to marked hypoalbuminemia, metabolic acidosis, and electrolyte changes such as hyperphosphatemia, hypochloremia, and mild hyperkalemia or hypokalemia. Uncommonly, hypercholesterolemia is present.

Urinalysis

The urinalysis in dogs with Lyme nephritis generally reveals isosthenuria and proteinuria. Pyuria and microscopic hematuria

may also be present. Urine protein to creatinine ratios are often greater than 5 and may be above 15 in some dogs.

Coagulation Profile

Dogs with Lyme nephritis may have low antithrombin activities as a result of glomerular antithrombin loss, which may lead to hypercoagulability syndromes. Sometimes, abnormalities such as a shortened PT, prolonged APTT, and increased fibrinogen and D-dimer concentrations are present.[40]

Synovial Fluid Cytology

Dogs with Lyme arthritis may have normal synovial fluid cytology,[30] or there may be markedly increased numbers of nondegenerate neutrophils within the synovial fluid of distal joints (>5000 and frequently >10,000 cells/μL). Because not all joints may be affected, synovial fluid should be collected from at least three and preferably four peripheral joints.

Diagnostic Imaging

Plain Radiography

Lyme arthritis is a nonerosive polyarthritis, so the only changes visible on plain radiography of the joints are increased periarticular soft tissue opacity. Thoracic radiography is generally unremarkable, but pleural effusion is occasionally identified in severely hypoalbuminemic dogs with Lyme nephritis.

Sonographic Findings

Abdominal sonographic examination is normal in dogs with Lyme arthritis. In dogs with nephritis, thickening and increased echogenicity of the renal cortices, decreased renal corticomedullary distinction, and peritoneal effusion may be seen.

Microbiologic Tests

Specific diagnostic assays available for Lyme borreliosis in dogs are described in Table 51-3.

TABLE 51-3

Diagnostic Assays Available for Lyme Borreliosis in Dogs

Assay	Specimen Type	Target	Performance
Bacterial isolation	Skin biopsy close to the tick bite site; synovial fluid	*B. burgdorferi* spirochete	Low sensitivity, requires special media, and may take several weeks. Isolation does not imply that *B. burgdorferi* is the cause of disease.
Serology (C6 assay)	Serum	Antibodies against *B. burgdorferi* C6 protein	Positive serology does not equate with Lyme disease, so test results must be interpreted in light of clinical findings. False positives can occur in regions of low prevalence. False negatives are rare, because antibodies are present by the time dogs develop illness. Cross-reactivity with vaccine antibodies does not occur.
Serology (whole cell IFA and ELISA)	Serum	Antibodies against *B. burgdorferi* antigens	As for C6 serology except that cross-reactivity with vaccine antibodies occurs, and false positives may also occur with other inflammatory diseases, and other spirochete infections.
Serology (Western immunoblot)	Serum	Antibodies against *B. burgdorferi* antigens	Used to confirm the serological response to natural infection in dogs that test positive with whole-cell ELISA and IFA. Technically difficult to perform and requires expertise to interpret. Positive serology does not equate with Lyme disease, so test results must be interpreted in light of clinical findings.
Serology (multiplex fluorescent bead assay)	Serum	Antibodies against *B. burgdorferi* OspA, OspC, and OspF	Antibodies to OspC appear only early in infection, whereas those to OspF reflect chronic infection and appear to correlate with the C6 antibody response. Sensitivity and specificity in dogs with naturally occurring Lyme disease unknown. Positive serology does not equate with Lyme disease, so test results must be interpreted in light of clinical findings.
Serology (multi-target silicon disc–based assay)	Serum	Antibodies against *B. burgdorferi* OspA, OspC, and OspF, SLP, and P39	Sensitivity and specificity in dogs with naturally occurring Lyme disease requires further study. Positive serology does not equate with Lyme disease, so test results must be interpreted in light of clinical findings.
PCR	As for isolation	*B. burgdorferi* DNA	Rapid but insensitive. Synovial fluid from dogs with polyarthritis may be the best specimen, but further study is needed. Assay performance can vary between laboratories.

Culture

B. burgdorferi can be isolated from tissue specimens in Barbour-Stoenner-Kelly medium. The optimal specimen for culture is a skin biopsy collected from a site adjacent to the tick bite, which is rarely identifiable in affected dogs. Incubation of cultures for several weeks may be required. Because of these factors, culture is not generally used on a routine basis for diagnosis.

Serologic Diagnosis

Serologic tests for antibody are the main assays used for diagnosis of Lyme disease in humans and also in dogs, but it is critical to recognize that positive serology does not necessarily equate with the presence of Lyme disease. In Europe, infection with nonpathogenic variants of *B. burgdorferi* sensu lato can result in positive serologic results, which further complicates diagnosis. Because the infection is chronic and persistent, and the incubation period is long, paired serology is generally not performed, because seroconversion may not occur. However, paired serology using serologic panels that include *B. burgdorferi* may be required for diagnosis of other vector-borne diseases that might be present. Positive test results in regions of low prevalence are more likely to be false positives than in regions of high prevalence, so interpretation of positive results should be performed with care in low-prevalence regions.

Currently, one of the most widely used serodiagnostic tests for canine Lyme disease is based on a C6 ELISA, which detects antibodies against a portion of the VlsE lipoprotein. The advantages of the C6 ELISA assay are that (1) it detects IgG antibodies 3 to 5 weeks after the time of infection, so by the time dogs develop clinical signs they are virtually always seropositive,[41] and (2) it is negative in dogs that have been vaccinated for Lyme disease, because the antigen is not expressed by organisms used in Lyme vaccines. The C6 ELISA is available as an in-practice lateral-flow assay, in combination with serodiagnostic spots for *Ehrlichia canis/Ehrlichia ewingii* antibody, *Anaplasma* spp. antibody, and *Dirofilaria immitis* antigen (SNAP 4Dx Plus, IDEXX Laboratories, ME) and as a quantitative ELISA (Quant C6), which is performed at IDEXX central veterinary diagnostic laboratories. There is no correlation between the magnitude of the C6 ELISA titer and disease severity. In a study from Europe, positive test results did not correlate with disease.[36] Another rapid immunochromatographic ELISA assay has recently become available in the United States (Abaxis VetScan Canine Lyme Rapid Test) that detects antibodies to a different portion of the VlsE lipoprotein, OspC and p41; these are combined on a single line.

Other serologic assays that are available include whole-cell ELISA or immunofluorescent antibody (IFA) assays and Western blotting (WB; see Chapter 2 for a description of these techniques). WB has traditionally been considered the gold standard assay. False positives using whole-cell ELISA and IFA have the potential to occur in patients with other spirochete infections, with other inflammatory disorders, and in vaccinated dogs. In human patients, a two-tiered approach is recommended for diagnosis.[39] Serum from patients who test positive for IgM or IgG with ELISA is then subjected to WB, because WB has increased specificity. WB has been used to differentiate the response to immunization and natural infection in dogs (Figure 51-4).[42] It has also been used to identify "dual status" dogs, that is, dogs that have been both immunized and naturally infected. WB is more time-consuming to perform than ELISA and IFA and experience is required to interpret it correctly.[43]

A multiplex fluorescent bead assay has been marketed in North America for detection of antibodies to three antigens of *B. burgdorferi:* OspA, OspC, and OspF.[43] The assay uses tiny beads to which OspA, OspC, and OspF are coupled. Dog serum is added to the beads, and if present, antibodies in the serum bind to the antigens and can be detected using a fluorescent conjugate. The pattern of reactivity to each antigen can be used to differentiate among the response to vaccination, early infection, and chronic infection. The presence of anti-OspA antibodies suggests previous vaccination, because vaccines contain OspA, and the spirochete rarely expresses OspA within

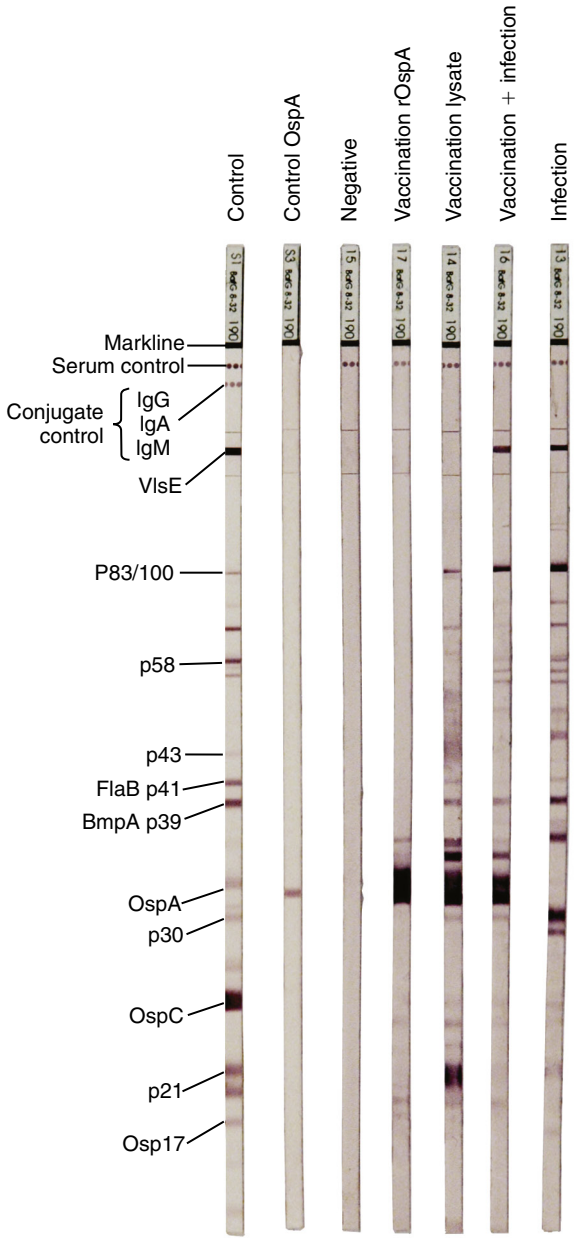

FIGURE 51-4 Western immunoblots of sera from dogs not infected with *B. burgdorferi* (Negative); immunized with recombinant OspA (vaccination rOspA); Immunized with a whole cell lysate vaccine (vaccination lysate); immunized and infected with *B. burgdorferi* (vaccination + infection); and infected only with *B. burgdorferi* (infection). The left strip shows all major signals available on the blot (control); the second strip from the left is stained with a monoclonal antibody against OspA (control OspA). (Courtesy R. Straubinger, Ludwig-Maximillians-Universitat, Munich, Germany. In: Greene CE: Infection Diseases of the Dog and Cat. 4th ed. St. Louis: Elsevier/Saunders; 2012.)

the host. OspC is expressed as the spirochete moves to the tick salivary glands and shortly after it enters the host. Thus an antibody response to OspC may suggest recent infection; titers decline and become undetectable beyond 3 months after infection.[44] Antibodies to OspC appear as early as 3 weeks after experimental infection of dogs, and OspF as early as 5 weeks.[44] OspF is expressed in more chronic infections, and can be detected together with the C6 antibody response in naturally exposed dogs (Figure 51-5).[44,45] In dogs, when WB was used as the gold standard, the sensitivities of the OspA, OspC, and OspF assays were 83%, 62%, and 82%, and the specificities were 90%, 89%, and 86%, respectively. The use of WB as the gold standard was questioned, and it was suggested that in fact, the fluorescent bead assay may have greater sensitivity and specificity than WB. The performance of the fluorescent bead assay is yet to be thoroughly evaluated in dogs with naturally occurring Lyme disease.

A novel silicon disc–based serologic assay is available in the United States for detection of antibodies to *B. burgdorferi, E. canis,* and *A. phagocytophilum* and antigen to *Dirofilaria immitis* (Accuplex 4, Antech Diagnostics, Irvine, CA). The assay for *B. burgdorferi* detects antibodies to the spirochete proteins OspA, OspC, OspF, P39, and SLP and is performed at central veterinary diagnostic laboratories. Preliminary data suggests that laboratory analysis of the response to these proteins correlates with the results of WB and can differentiate between the responses to natural infection and vaccination, and acute and chronic infection.[46]

Molecular Diagnosis Using the Polymerase Chain Reaction

As with culture, PCR assays are best performed on skin biopsy specimens collected from a region adjacent to the tick bite site. In human patients, the use of PCR assays for diagnosis of Lyme borreliosis on blood has low sensitivity, and so false negatives are common when *B. burgdorferi* PCR assay is used as part of a whole-blood vector-borne infection PCR panel. Synovial fluid or synovial membrane biopsies may be the optimum specimen for diagnosis of Lyme arthritis in dogs. This is also true in human patients.[47] However, in a study of experimentally infected dogs, PCR assay of the synovial fluid was insensitive.[30] The sensitivity and specificity of PCR assays when used on specimens such as synovial fluid for diagnosis of canine Lyme borreliosis requires further study.

Pathologic Findings

Gross Pathologic Findings

Gross pathologic findings in dogs with Lyme arthritis include peripheral lymphadenomegaly, joint swelling, and synovial effusion.[29] The synovial fluid may be yellow-tinged and cloudy, with decreased viscosity. In dogs with nephritis, the kidneys are diffusely light tan and may have pinpoint red foci over the cortical surfaces.[33] In addition, evidence of systemic edema, thrombosis and infarction, and uremia (such as parathyroid hyperplasia, ulcerative stomatitis, and gastritis) may be present.

Histopathologic Findings

Histopathology of the joints of dogs that have been experimentally infected with *B. burgdorferi* reveals fibrinosuppurative or lymphoplasmacytic inflammation of the synovial membranes, joint capsules, and tendon sheaths.[29,30] Inflammation can sometimes be found in the joints even when a history of lameness is not present.[29] In human patients, organisms have been demonstrated in the synovium using immunostains and electron microscopy.[48] Other findings in experimentally infected dogs include periarteritis and perineuritis, especially within joint capsules and the skin, as well as lymphoid hyperplasia within peripheral lymph nodes. Mild, focal meningitis and encephalitis was identified in dogs that were infected with *B. burgdorferi* and immunosuppressed with dexamethasone.[49]

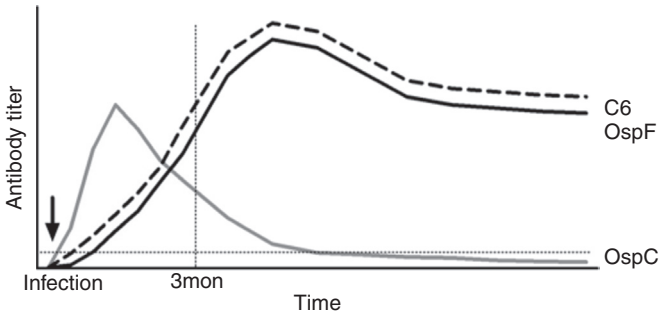

FIGURE 51-5 Proposed canine antibody response to OspC, OspF, and C6 antigens of *B. burgdorferi* during early and late infection. Data were obtained by fluorescent bead multiplex analysis. The lines for the first 3 months after infection are based on multiplex results from experimentally infected dogs. After 3 months the lines are projected from data obtained from patient sera. The horizontal dotted line shows the cutoff value for the multiplex assay. The vertical dotted line indicates 3 months after infection. (Modified from Wagner B, Freer H, Rollins A, et al. Antibodies to *Borrelia burgdorferi* OspA, OspC, OspF, and C6 antigens as markers for early and late infection in dogs. Clin Vaccine Immunol 2012;19:527-535.)

Renal histopathology in dogs with Lyme nephritis reveals diffuse membranoproliferative glomerulonephritis (Figure 51-6), dilation of the cortical renal tubules, tubular necrosis and regeneration, and mild to moderate, diffuse interstitial lymphoplasmacytic inflammation.[33] Periglomerular or diffuse interstitial fibrosis may be present in dogs with end-stage disease.

Treatment and Prognosis

Antimicrobial Treatment

Antibiotic treatment is recommended for seropositive dogs that have clinical illness consistent with Lyme disease. There is no evidence that treatment of healthy seropositive dogs is beneficial and it may lead to drug adverse effects and contribute to antimicrobial resistance in other bacteria and antimicrobial shortages. The antibiotic of choice for Lyme arthritis is doxycycline. The optimal dose and duration of treatment is unknown. Recommended doses have included 5 mg/kg PO q12h and 10 mg/kg PO q12h or 10 mg/kg PO q24h.[34,50] Four weeks of treatment has been recommended[34] because the clinical manifestations of disease resemble those of late-stage disease in human patients, for which relapses occur when treatment durations of less than 30 days are used.[39] Dogs with polyarthritis generally respond clinically to doxycycline treatment within 24 to 48 hours. Other differential diagnoses for polyarthritis should be considered if an inadequate response to treatment occurs. On the other hand, a clinical response to doxycycline treatment is not sufficient to make a diagnosis of Lyme arthritis. This is because there are other doxycycline-responsive causes of infectious polyarthritis in dogs; signs of primary immune-mediated polyarthritis can wax and wane; and doxycycline has antiinflammatory properties that may contribute to clinical improvement. For dogs that do not tolerate doxycycline, amoxicillin can be used, which also has activity against *B. burgdorferi* (Table 51-4). Azithromycin and third-generation cephalosporins have also been used to treat Lyme disease in human patients.[39] Doxycycline treatment

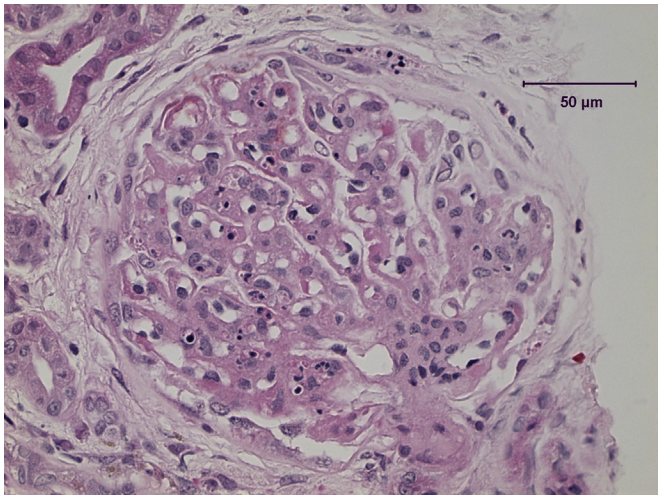

FIGURE 51-6 Membranoproliferative glomerulonephritis in a 6-year-old, intact male golden retriever dog that was seropositive for *B. burgdorferi* C6 protein antibodies and had clinical and biochemical evidence of protein-losing nephropathy. The morphologic diagnosis was moderate to severe, diffuse, global membranoproliferative glomerulonephritis with moderate chronic-active tubulointerstitial nephritis. The capillary walls are thickened and there is mesangial cell proliferation. There was also evidence of focal arteriolar mural disorganization, lamination, and sclerosis in this biopsy specimen. (Image courtesy Dr. George Lees, Texas A&M University.)

is also recommended for dogs with Lyme nephritis, although clinical improvement generally does not occur with antimicrobial treatment alone (see Supportive Care).

Although treatment leads to clinical improvement and a reduction in antibody titers, *B. burgdorferi* can persist in tissues after treatment is discontinued. Studies in mouse and primate models have shown that despite negative cultures after treatment, PCR assays remain positive, and infection can be transmitted from PCR-positive to naïve animals.[51,52] Regardless of the serologic test used, antibody titers can persist for months or years after resolution of clinical signs and antimicrobial drug treatment, so the results of antibody testing are not consistently useful to guide further treatment when clinical resolution of arthritis has occurred.

Supportive Care

Dogs with arthritis usually respond rapidly to treatment with antibiotics, and additional treatment may not be necessary. Severe pain associated with arthritis may be treated with nonsteroidal antiinflammatory drugs or opiate analgesics such as tramadol. Because Lyme arthritis can be reactivated in some dogs by administration of glucocorticoids more than a year after exposure,[53] glucocorticoids are not recommended for their antiinflammatory effects.

Dogs with Lyme nephritis require management for protein-losing nephropathy, which may include treatment with intravenous crystalloids and colloids, angiotensin-converting enzyme inhibitors such as enalapril or benazepril, low-dose aspirin in an attempt to reduce thrombotic events, and nutritional support with a reduced protein diet. Placement of an esophagostomy or gastrostomy tube for feeding purposes may be required. Blood pressure should be monitored and if necessary, antihypertensive drugs such as amlodipine should be administered and titrated to effect. Anecdotally, improved outcomes have been noted in dogs with Lyme nephritis after immunosuppressive drug treatment (Table 51-5). The optimum protocol is yet to be determined. Dogs treated with immunosuppressive drugs should be carefully monitored for adverse effects of drug therapy.

TABLE 51-4

Suitable Antimicrobials for Treatment of Lyme Disease in Dogs

Drug	Dose	Route	Interval (hours)	Duration (days)
Doxycycline	5 to 10 mg/kg	PO	12	28
Amoxicillin	20 mg/kg	PO	8	28

TABLE 51-5

Immunosuppressive Drug Protocols That Could Be Considered for Treatment of Lyme Nephritis in Dogs in Conjunction with Antimicrobial Drug Treatment

Drug	Dose	Route	Interval and Duration
Methylprednisolone sodium succinate with either cyclophosphamide, mycophenolate mofetil, or azathioprine below	5 mg/kg	IV	24 hours for 2 days
Cyclophosphamide	200 mg/m^2	IV	Every 14 days for a maximum of 6 cycles. Recheck CBC 1 week after each treatment.
Mycophenolate mofetil	10 mg/kg	IV or PO	12 hours until remission occurs, then consider tapering
Azathioprine	1-2 mg/kg	PO	24 hours for 7 days, then every 48 hours until remission occurs, then consider tapering

Prognosis

It is estimated that more than 90% of dogs that are infected with *B. burgdorferi* show no signs of illness. Dogs with Lyme arthritis generally recover rapidly with antimicrobial treatment and do not develop relapse of disease. It is not clear whether the "antibiotic-refractory arthritis" that occurs in a small percentage of genetically predisposed human patients also occurs in dogs. Seropositive dogs with antibiotic-refractory polyarthritis may have other unrelated causes of their disease, such as primary immune-mediated polyarthritis.

The prognosis for dogs with Lyme nephritis is guarded to poor. In the past, it was noted that most dogs die or are euthanized within days to weeks.[34] Death often results from systemic thrombosis or oliguric or anuric renal failure. Anecdotally, longer survival times of months to over a year have been noted in

some dogs treated with immunosuppressive drugs in addition to doxycycline and other supportive treatments.

Immunity and Vaccination

Borrelia spp. can evade the host immune response through modification of outer surface proteins and possibly metamorphosis to a resistant spherical form.[54] Interference with B-cell responses by the spirochete also seems to occur.[55] Humoral immunity is most critical for resolution of infection, although T-cell responses may be important for resolution of cell-mediated consequences of infection, such as carditis in human patients.[56]

Several vaccines are available in North America for reduction of Lyme disease in dogs. Lyme vaccines stimulate the formation of borreliacidal antibodies that are directed against the surface proteins normally expressed by the spirochete when it resides within the tick. When the tick ingests dog blood that contains these antibodies, complement-mediated lysis of the bacteria occurs *within the tick*. Thus, bacteria are inactivated before they invade the host. A canine recombinant OspA vaccine is available that induces the formation of antibodies only against OspA. This subunit vaccine is similar to a Lyme disease vaccine that was previously available for prevention of human Lyme borreliosis (LYMErix, GlaxoSmithKline). In humans, the vaccine was shown to be safe and efficacious, conferring immunity on 76% of adults and 100% of children with a low prevalence of local injection site reactions and flu-like symptoms.[57] However, this vaccine was withdrawn in February 2002 because of concerns that it may trigger autoimmune disease as a result of molecular mimicry in sensitive individuals, despite a lack of scientific justification for these concerns.[58,59]

Two inactivated whole spirochete vaccines are also available for dogs. One contains two strains of *B. burgdorferi*, one of which expresses high levels of OspC. The other vaccine is a single-strain vaccine. The potential advantage of these vaccines is that they stimulate immunity not only to OspA, but also to other surface proteins expressed by the spirochete. This may provide the opportunity to neutralize *B. burgdorferi* when OspA is downregulated, such as when the organism is in the salivary glands and after it enters the host. Concern has been expressed that whole-cell vaccines may be more likely to trigger autoimmune consequences in dogs than the subunit vaccine, especially in dogs that have been previously exposed to *B. burgdorferi*.[34] However, no strong evidence exists that these types of adverse reactions occur. Both OspA and whole-cell vaccines provide protection against infection and disease that results from experimental challenge.[60,61] A study in an endemic area for Lyme disease found a reduced prevalence of C6 seropositivity in dogs that had been vaccinated with a whole-cell bacterin when compared with dogs that had not been vaccinated, suggesting the possibility of protection from natural infection.[62] The dual-strain bacterin has been shown to have a 1-year duration of immunity.[63] Although no Lyme vaccine can be relied on to provide complete protection, side-by-side comparisons of canine Lyme vaccines that are available on the market have not been performed. In addition, whether Lyme vaccination protects against, or contributes to, the most severe consequence of infection, Lyme nephritis, is unknown. Finally, whether there are any advantages or disadvantages of vaccinating dogs that are already seropositive as a result of natural infection remains to be elucidated. The recent identification of reinfection (as opposed to relapse) as a cause of recurrent clinical signs of borreliosis in human patients[64] suggests that vaccination of previously exposed individuals may offer some benefit, because the protection induced by vaccination (anti-OspA antibodies) differs from that induced by natural infection (no anti-OspA antibodies). However, whether recurrent Lyme disease occurs in dogs as a result of reinfection is not known.

Prevention

Avoidance of tick-infested areas, use of topical ectoparasiticides, and routine inspection of dogs for ticks after outdoor activities can help to prevent Lyme disease. Ticks should be removed within 24 hours of attachment, before transmission of the spirochete can occur, but removal up to 60 hours after attachment may still reduce the chance of transmission.[65] Refer to Chapter 28 for general information on tick prevention and removal. Although treatment with a single dose of doxycycline after a known *Ixodes* tick bite can prevent Lyme disease in humans[66] and mice,[67] it is controversial because of the very low risk of infection after a tick bite.[68] A study in mice showed that treatment 2 or more days after tick removal was ineffective.[67]

For healthy dogs that test positive for antibodies to *B. burgdorferi* during a heartworm screen with assays that include a *B. burgdorferi* ELISA, a urinalysis could be offered in order to assess for proteinuria.[34] If proteinuria is detected, further work-up that includes a urine protein to creatinine ratio, aerobic bacterial culture of the urine, serum biochemistry panel, and imaging may be indicated. However, whether treatment of seropositive, proteinuric dogs with doxycycline influences the progression of Lyme nephropathy is not known, so this is controversial. Similarly, a CBC or platelet count could be performed, but whether treatment of thrombocytopenic dogs is necessary is also unknown. At a minimum, tick control for seropositive, apparently healthy dogs should be recommended.

Public Health Aspects

Lyme disease is the most common vector-borne disease of humans in the Northern Hemisphere, and the prevalence of the disease has increased progressively.[69] In humans, the first sign of disease in 80% to 90% of infected individuals is erythema migrans, which is a characteristic "bull's-eye" rash that occurs 3 to 30 days after a tick bite and moves outward from the bite site at a rate of approximately 1 cm/day. In some people, the rash is pruritic or painful, and it may be accompanied by headache and malaise. Early disseminated disease can appear as multiple erythema migrans rashes; myocardial disease that is usually characterized by atrioventricular block; or neuroborreliosis. Neuroborreliosis is more common in Europe and may be characterized by the development of cranial nerve palsies or signs of meningitis and polyradiculoneuritis. Arthritis is a manifestation of late Lyme disease. A small percentage of infected individuals, especially those of certain human leukocyte antigen (HLA) types, can develop chronic, antibiotic-refractory arthritis, which may result from persistence of spirochete residues in tissues.[70] European strains, particularly *B. afzelii*, can also induce a chronic skin manifestation known as acrodermatitis chronica atrophicans.[71]

Dogs do not pose a direct zoonotic risk to humans in the household. However, they may carry infected, unfed ticks into the household that could subsequently attach to humans. The presence of antibodies in dogs can also indicate increased human risk as a result of common exposure to infected ticks in the environment (sentinel exposure).

CASE EXAMPLE

Signalment: "Dako", a 7-year-old MC Doberman Pinscher dog from Amador County in northern California

History: Dako was evaluated for a 2-day history of lethargy, apparent blindness, inappetence, and inability to walk. He was taken to a local emergency clinic within 24 hours of illness, where a neurologic examination was considered abnormal and an intracranial lesion was suspected. He was treated with intravenous fluids and given a single dose of ampicillin (1 g). The following day his mentation and thoracic limb strength had improved, but he remained unable to walk and developed urinary incontinence. He lived on a farm and had contact with sheep, horses, llamas, chickens, geese, cats, and four other dogs, all of which were currently healthy, although an additional dog had died of acute renal failure 1 month earlier. There was no travel history or toxin exposure, but frequent exposure to ticks occurred. Dako's diet consisted of commercial dry dog food, and occasionally pieces of cooked steak and raw goose eggs. He had not received a Lyme vaccine in the past but had been vaccinated regularly for distemper, adenovirus, parvovirus, parainfluenza, and rabies. Dako had been diagnosed several years previously with color dilution alopecia.

Current Medications: Monthly topical flea and tick preventative (fipronil and *S*-methoprene), monthly oral heartworm preventative (ivermectin and pyrantel).

Physical Examination:

Body Weight: 43.1 kg

General: Quiet, alert, and responsive. Hydrated. Ambulatory on all four limbs but appeared very painful. T = 101.9°F (38.8°C), HR = 78 beats/min, RR = 24 breaths/min, mucous membranes pink, CRT = 1 s.

Integument, Eyes, Ears, Nose, and Throat: A thin haircoat with scaling was noted. Mild episcleral injection was present bilaterally. Moderate dental calculus and gingivitis were also present.

Musculoskeletal: Body condition score was 5/9 and the dog was symmetrically well muscled. Multiple distal joints were severely swollen and painful on palpation, including both carpi, tarsi, stifle, and elbow joints. The dog appeared painful when rising from recumbency and when walked, with pronounced right pelvic limb lameness.

Cardiovascular, Respiratory, Gastrointestinal, and Genitourinary: No clinically significant abnormalities were detected. The dog had a large bladder, and actively urinated a large amount during the examination. Rectal examination was unremarkable.

Lymph Nodes: Bilateral popliteal lymphadenomegaly (3 cm in diameter) was noted. The remaining peripheral lymph nodes measured 2 cm in diameter. All nodes were soft on palpation.

Neurologic Examination: No neurologic abnormalities were detected.

Laboratory Findings:

CBC:

HCT 48.4% (40%-55%)
MCV 69.6 fL (65-75 fL)
MCHC 34.9 g/dL (33-36 g/dL)
WBC 17,100 cells/μL (6000-13,000 cells/μL)
Neutrophils 15,561 cells/μL (3000-10,500 cells/μL)
Lymphocytes 171 cells/μL (1000-4000 cells/μL)
Monocytes 1026 cells/μL (150-1200 cells/μL)
Eosinophils 342 cells/μL (0-1500 cells/μL), platelets clumped.

Serum Chemistry Profile:

Sodium 148 mmol/L (143-151 mmol/L)
Potassium 3.5 mmol/L (3.6-4.8 mmol/L)
Chloride 115 mmol/L (108-116 mmol/L)
Bicarbonate 20 mmol/L (20-29 mmol/L)
Phosphorus 3.1 mg/dL (2.6-5.2 mg/dL)
Calcium 10.0 mg/dL (9.6-11.2 mg/dL)
BUN 11 mg/dL (11-33 mg/dL)
Creatinine 0.8 mg/dL (0.8-1.5 mg/dL)
Glucose 102 mg/dL (86-118 mg/dL)
Total protein 6.1 g/dL (5.4-6.9 g/dL)
Albumin 3.6 g/dL (3.4-4.3 g/dL)
Globulin 2.5 g/dL (1.7-3.1 g/dL)
ALT 17 U/L (21-72 U/L)
AST 36 U/L (20-49 U/L)
ALP 60 U/L (14-91 U/L)
Creatine kinase 217 U/L (55-257 U/L)
GGT 1 U/L (0-6 U/L)
Cholesterol 167 mg/dL (139-353 mg/dL)
Total bilirubin 0.1 mg/dL (0-0.2 mg/dL)
Magnesium 1.8 mg/dL (1.9-2.5 mg/dL).

Urinalysis: SGr 1.018; pH 8.0, negative protein (SSA), negative bilirubin, negative glucose, 0-1 WBC/HPF, 0-2 RBC/HPF.

Imaging Findings:

Thoracic Radiographs: Unremarkable.

Abdominal Ultrasound: The liver was diffusely hypoechoic but was normal in size. There was mild bilateral adrenomegaly (0.9 cm). The urinary bladder was very large and was in a pelvic location.

Microbiologic Testing: 4Dx SNAP test (IDEXX Laboratories): Positive for *B. burgdorferi* antibodies; negative for antibodies to *Anaplasma* spp. and *E. canis*; negative for *Dirofilaria immitis* antigen.

Cytology of Synovial Fluid Obtained via Arthrocentesis

	Right Carpus	Left Carpus	Right Tarsus	Left Stifle
Cell count (cells/μL)	23,730	29,370	ND	1020
Neutrophils (%)	85	94	76	12
Small mononuclear cells (%)	2	0	1	41
Large mononuclear cells (%)	13	6	23	47
Interpretation	Marked purulent inflammation with nondegenerate neutrophils			Very mild purulent inflammation with nondegenerate neutrophils

ND, not done.

Lyme C6 Quant antibody ELISA (IDEXX Laboratories): 153 U/mL (normal, <30 U/mL).
PCR for *B. burgdorferi* on synovial fluid: Negative.

Diagnosis: Neutrophilic polyarthritis; possibly secondary to *B. burgdorferi* infection.

Treatment: Dako was treated with doxycycline (5 mg/kg PO q12h) and tramadol (2 mg/kg PO q8h) and showed dramatic clinical improvement within 24 hours of initiating treatment. A physical examination was normal 2 weeks later.

Comments: The positive serology and response to doxycycline suggested the possibility of Lyme arthritis, although it is possible there was some other cause that was not identified and the presence of C6 antibodies was not related to this dog's illness. It may have been too early in the course of illness for this dog to have seroconverted to other vector-borne pathogens such as *A. phagocytophilum*. Other diagnostic assays that might have been useful in this dog (and in this geographic location) include serology for *Bartonella* spp.; PCR on blood for vector-borne pathogens such as *A. phagocytophilum, Rickettsia* spp., and *Ehrlichia canis;* aerobic bacterial culture of the synovial fluid; and convalescent serology for vector-borne pathogens. The negative synovial fluid PCR result did not rule out the possibility of Lyme arthritis, because organism numbers within the synovial fluid of animals with Lyme arthritis may be extremely low. The dog had been treated with ampicillin before it was seen, which also may have reduced organism numbers further.

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