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Primer

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Lyme borreliosis

Franc Strle[1] ✉, Klemen Strle[2], Adriana Marques[3], Anna J. Henningsson[4,5], Randi Eikeland[6,7], Jacob E. Lemieux[8], Jean I. Tsao[9], Paul S. Mead[10] & Gary P. Wormser[11]

Abstract

Lyme borreliosis is the most common tick-borne disease in the northern hemisphere. It is a zoonosis caused by several species of *Borrelia burgdorferi* sensu lato and transmitted by the bite of infected ticks of the *Ixodes ricinus* complex. Lyme borreliosis in North America and Europe differs in certain respects, likely reflecting the different *Borrelia* species that cause human disease in these locations. The earliest manifestation of Lyme borreliosis is the skin lesion erythema migrans, which develops at the tick bite site, typically 7–14 days after the bite. Some untreated patients will then (within the first few weeks or months after onset of the infection) develop additional erythema migrans skin lesions or other clinical manifestations such as borrelial lymphocytoma, nervous system involvement or carditis. Several months or even years after infection onset, Lyme arthritis or acrodermatitis chronica atrophicans may develop. The diagnosis of typical erythema migrans is clinical, whereas for all other manifestations the diagnosis is supported via serological testing. Treatment with an appropriate antibiotic will result in resolution of clinical symptoms in most patients; however, some patients experience prolonged subjective symptoms, which usually improve over time. Repeated courses of antimicrobials are not beneficial except in rare cases when there is objective evidence of treatment failure.

Sections

[1]Department of Infectious Diseases, University Medical Centre Ljubljana, Ljubljana, Slovenia. [2]Tufts University School of Medicine, Boston, MA, USA. [3]Lyme Disease Studies Unit, Laboratory of Clinical Immunology and Microbiology, National Institute of Allergy and Infectious Diseases, National Institute of Health, Bethesda, MD, USA. [4]Division of Inflammation and Infection, Department of Biomedical and Clinical Sciences, Linköping University, Linköping, Sweden. [5]Division of Clinical Microbiology, Department of Laboratory Medicine, Region Jönköping County, Jönköping, Sweden. [6]Department of Health and Nursing Science, University of Adger, Kristiansand, Norway. [7]Norwegian Advisory Unit for Tick-borne Diseases, Sørlandet Hospital Trust, Kristiansand, Norway. [8]Division of Infectious Diseases, Massachusetts General Hospital, Boston, MA, USA. [9]Department of Fisheries and Wildlife, Department of Large Animal Clinical Sciences, Michigan State University, East Lansing, MI, USA. [10]Bacterial Diseases Branch, Division of Vector-Borne Diseases, National Center for Emerging and Zoonotic Infectious Diseases, Centers for Disease Control and Prevention, Fort Collins, CO, USA. [11]New York Medical College, Valhalla, NY, USA. ✉e-mail: franc.strle@kclj.si

Introduction

Lyme borreliosis is the most common tick-borne disease in the northern hemisphere. It is a zoonosis caused by several species of the spirochaete *Borrelia burgdorferi* sensu lato (Lyme borreliae) and is transmitted by the bite of infected ticks. The animal reservoirs are mainly small rodents and birds. This Primer focuses mainly on Europe and the USA, since there are only very limited published data from other geographical areas. In the USA, this spirochaetal infection is called Lyme disease, and was named after the town of Lyme, Connecticut, located in the geographical area where the disease was first investigated. However, in Europe, certain clinical manifestations of this infection, such as erythema migrans (EM), were known as clinical entities decades earlier. The disease in North America and Europe differs in certain respects, likely reflecting the different species of Lyme borreliae that cause disease in North America compared with Europe[1,2]. In 2021, these spirochaetes were renamed *Borreliella*[3] but this change is controversial[4]; therefore, these spirochaetes will still be referred to as Lyme borreliae in this article.

Lyme borreliosis usually starts with the characteristic EM skin lesion that develops at the tick bite site, a few days to a few weeks following the bite of an infected tick. Some patients will then develop manifestations of early disseminated Lyme borreliosis, such as additional EM skin lesions, borrelial lymphocytoma, carditis or nervous system involvement, either while the primary EM skin lesion is still present or after its spontaneous resolution, which may occur after a few weeks to months. Clinical manifestations of early disseminated Lyme borreliosis typically occur within the first few months after onset of the infection. Late clinical manifestations of Lyme borreliosis include arthritis and acrodermatitis chronica atrophicans (ACA), which may develop several months or even years after onset of infection[1,2]. Of note, disseminated manifestations may occur without a recognized EM skin lesion.

This Primer discusses the epidemiology of Lyme borreliosis, the tick vectors and animal hosts that maintain Lyme borreliae, prevention strategies, the clinical features, diagnosis, management and outcome of Lyme borreliosis, and the pathophysiology of the disease, including the impact of the immune responses of the host and the genomic variability of Lyme borreliae on the course and outcome of the infection. We also highlight knowledge gaps and propose relevant research and public health priorities to address these gaps.

Epidemiology

Factors that influence the epidemiology of Lyme borreliosis include the regional clades of Lyme borreliae, the ecology of the vector ticks, and the human behaviours that increase exposure to ticks. Statistics on Lyme borreliosis incidence are influenced by surveillance practices, which vary widely across jurisdictions and time periods, confounding comparisons. For example, some countries have active, sentinel-based systems while others rely on passive reporting; some track a particular disease manifestation (for example, Lyme neuroborreliosis) while others consider a range of features; some utilize clinician reports while others rely solely on electronic reporting of positive laboratory results[5,6].

Tick vector and animal host

Lyme borreliae are transmitted to humans by the bite of tick species from the *Ixodes ricinus* complex: in the USA, by *Ixodes scapularis* and less often by *Ixodes pacificus* ticks; in Europe by *I. ricinus* and *Ixodes persulcatus* ticks[7] (Fig. 1). Of the >20 genospecies of Lyme borreliae thus far detected in nature, the four that most commonly infect humans are: *Borrelia afzelii*, *Borrelia garinii*, *Borrelia bavariensis* and *Borrelia burgdorferi* sensu stricto (hereafter termed *B. burgdorferi*)[8–12].

I. ricinus complex ticks are hard ticks that predominantly inhabit temperate and boreal forests. Enzootic maintenance of Lyme borreliae depends mainly on horizontal transmission between the juvenile stages (larvae and nymphs) of the ticks and the reservoir animal hosts. Larvae hatch uninfected and may then acquire Lyme borreliae through a blood meal from an infected animal host. The infected nymphs may then transmit Lyme borreliae to a susceptible animal host or to a human during their next blood meal[7,13] (Fig. 2). For the two tick vectors responsible for the largest number of cases of Lyme borreliosis, the average nymphal tick infection prevalence in areas where Lyme borreliosis is endemic ranges from 13% to 24% in the eastern USA (*I. scapularis*) and is ~10% in Europe (*I. ricinus*), with a range of 0–50% in individual studies[14,15]. *I. persulcatus* nymphs rarely bite humans and most disease is transmitted by adult ticks[16].

I. ricinus complex ticks feed on numerous species of mammals, songbirds and lizards but only a subset, mainly small mammals and songbirds, serve as reservoirs for Lyme borreliae[17]. Some species of migratory songbirds may also disperse ticks and Lyme borreliae, but the quantitative contribution of these birds to the dispersal of ticks and Lyme borreliae is likely geographically variable. While deer are incompetent hosts for Lyme borreliae, they are the most important hosts for the adult tick stage, and the increasing abundance and spread of deer has been associated with the range expansion of vector ticks and of Lyme borreliosis foci[13,14].

Incidence

Geographical distribution. The geographical distribution of Lyme borreliosis follows that of the principal tick vectors (Fig. 1). The incidence of Lyme borreliosis varies widely depending on the local ecology. In North America, Lyme borreliosis occurs primarily in the northeast, mid-Atlantic and upper Midwest regions of the USA and in the adjoining provinces of Canada, where the northern clade of *I. scapularis* (the blacklegged tick) is present[18]. In some localities, the reported incidence can exceed 200 cases per 100,000 population per year[6]. Smaller geographical areas of less intense transmission are found in western coastal regions, where *I. pacificus* is the tick vector. Although the distribution of the blacklegged tick extends throughout the eastern USA, the risk of disease is lower or absent in the southern USA due to differences in the host-seeking behaviour of the southern blacklegged tick, including its predilection to feed on *B. burgdorferi*-incompetent lizard hosts[19,20].

Reported cases have increased markedly over time to reach nearly 90,000 cases in the USA in 2023 (ref. 6). This increase is influenced by changes in surveillance practices and by the geographical expansion of *I. scapularis* and of high incidence areas for Lyme borreliosis[6,21]. Because surveillance is passive, medically attended cases may go unreported. An analysis of medical claims data estimated that ~476,000 people are treated for Lyme disease annually in the USA, including patients without infection treated empirically[22]. *B. burgdorferi* causes most infections in North America, with a second genospecies, *Borrelia mayonii*, causing some infections in the upper Midwest[14,23,24].

In Europe, Lyme borreliosis is caused much more commonly by *B. afzelii* and *B. garinii*/*B. bavariensis* than by *B. burgdorferi*[10,11]. Infection is widespread, but risk is highly variable and depends on the habitat. Differences in surveillance practices make direct comparisons across countries difficult[25]. In general, the incidence seems

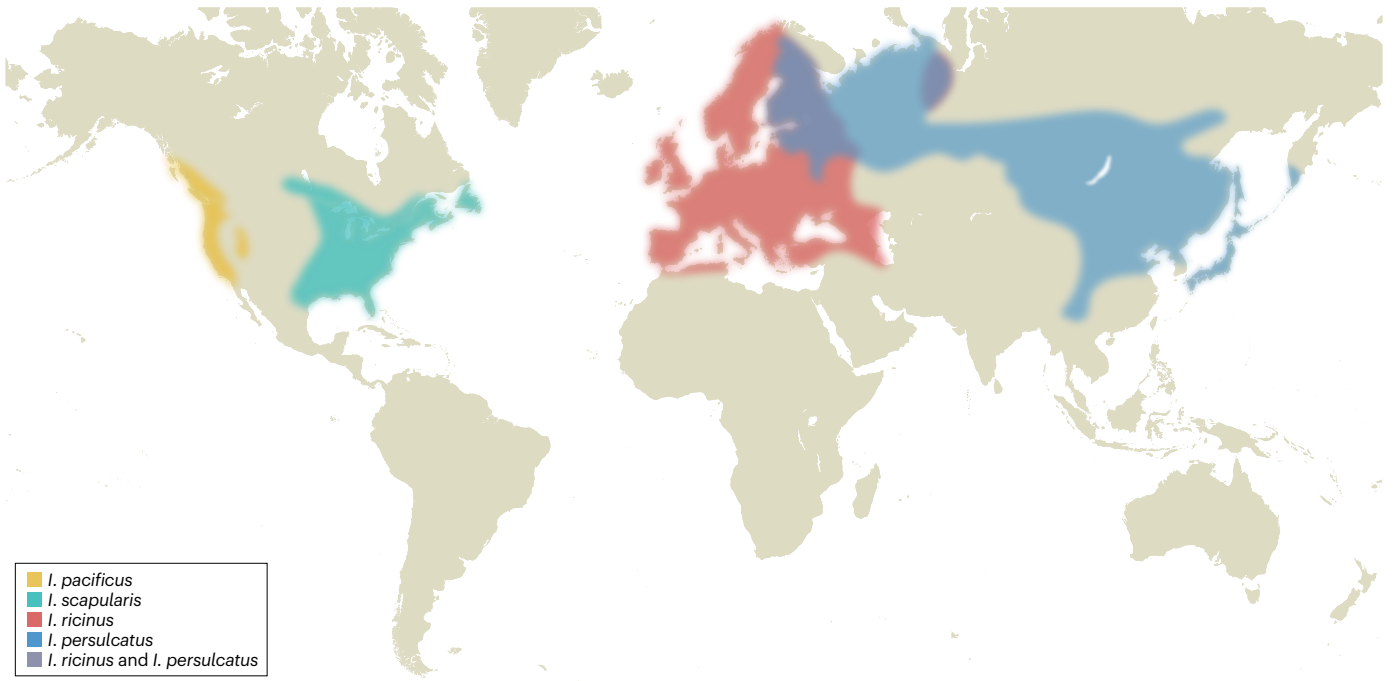


Fig. 1 | Geographical distribution of principal tick species that transmit Lyme borreliae to humans. The approximate geographical distributions of the principal tick vectors of Lyme borreliosis. The blacklegged tick, *Ixodes scapularis* (turquoise), is the primary vector in the eastern USA, while *Ixodes pacificus* (yellow), the western blacklegged tick, is the primary vector in the coastal Pacific states of the USA. Differences in questing behaviour and host preferences between northern and southern populations of *I. scapularis* account for the much lower or absent incidence of Lyme borreliosis in the southern USA. *Ixodes ricinus* (red), the castor bean or sheep tick, is the main vector in Europe, while *Ixodes persulcatus* (blue), the taiga tick, is the primary vector in Eurasia. Both *I. ricinus* and *I. persulcatus* overlap (purple) in eastern Europe and the Baltic countries[7,19,20,47,345–348]. Reprinted with permission from ref. 349, CABI.

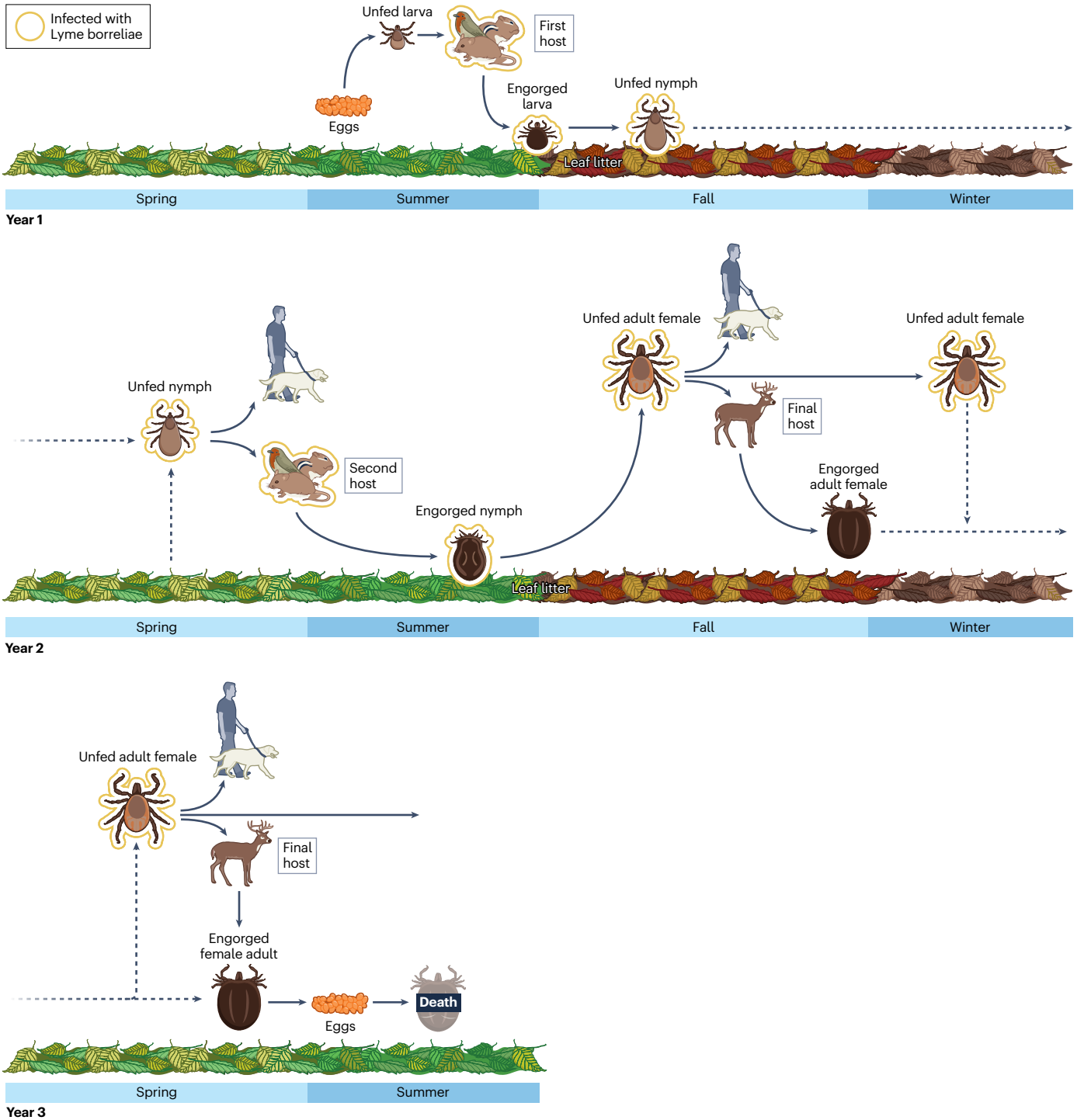
greatest in northeastern and central Europe and lower in the British Isles, Spain, and Italy[26,27]. National or sub-national rates exceeding 100 cases per 100,000 population per year have been reported from Finland, Estonia, Lithuania, Poland, Czech Republic, Slovenia, Switzerland, the Netherlands and Belgium[26,28–32]. Annual rates ranging from 10 to 100 per 100,000 population have been reported for Latvia, Sweden, France, Germany, Scotland and Slovakia, while rates in Romania, Bulgaria, Serbia, Ireland and England are generally under 10 per 100,000 population[26,33–36].

Although the USA and Europe are best studied, Lyme borreliosis also occurs in other regions. Based on the distribution of *I. persulcatus* ticks, risk for Lyme borreliosis extends eastward from western Russia through Mongolia to China and Japan[37]. Reported annual incidence rates range from 5 to 10 cases per 100,000 population in western Russia[26], while human infection has been identified through serological testing in the northern Chinese provinces of Xinjiang, Gansu, Inner Mongolia, Jinlin and Heilongjiang in particular[38,39], where *Ixodes granulatus* and *Ixodes sinensis* have been implicated as vectors[39]. Enzootic cycles have been detected in Korea[40], and a small number of culture-confirmed cases have been reported in Japan and Taiwan[41,42]. Although a Lyme-like illness has occasionally been reported from other regions of the world, including Central and South America, India, and Australia, efforts to isolate Lyme borreliae from clinical specimens have been unsuccessful, and thus the presence of Lyme borreliosis in these areas has not been established[43–46].

The past century has observed population growth and range expansion of several *I. ricinus* complex tick species, as well as an increase in tick numbers in peri-urban and urban environments. In both North America and Europe, these changes have occurred because of multiple interacting anthropogenic factors, including changes in land use, land cover, weather patterns, and wildlife conservation and management practices[47]. Future changes, such as decreased precipitation with increasing temperatures, or those regarding changes in the management of natural resources, could lead to range contraction or to a reduction of suitable tick habitats and animal hosts[13].

Age/sex demographics. The age distribution of reported Lyme borreliosis cases varies among countries. In general, a bimodal pattern is observed, with one peak in incidence among children <15 years of age and a second peak among adults aged ≥50 years[31]. Women and girls account for 50–60% of reported cases in several European countries, while the incidence is higher among men and boys in nearly all age groups in North America. These patterns likely reflect sex-related and age-related differences in exposure, care-seeking behaviour and surveillance practices[6,34,48].

Seasonality. Seasonally, early-stage infections in humans peak in June–July in North America and in July–August in parts of Europe. These peaks correlate with the questing (host-seeking behaviour) of the relevant vector stage most associated with transmission of Lyme borreliae to humans[6,29,49–51]. Although subject to local and seasonal variation, nymphal activity of *I. scapularis* is generally greatest from May to July, of *I. pacificus* from April to June[51], and of *I. ricinus* from April to July[13] and adult *I. persulcatus* activity peaks in April–May[16]. Compared



with early-stage infections, cases of later-stage Lyme borreliosis tend to peak slightly later and show less seasonal fluctuation due to longer and more variable incubation periods[29,49,50,52–54].

Risk factors

Where homes are in a suitable habitat for vector ticks, most infections are the result of peri-domestic exposures[55]. In other areas, infection is more often associated with recreational activities such as hiking, camping, hunting and orienteering[48]. Occupational risk among forestry workers, farmers, linemen and soldiers is well documented[56,57]. North American studies indicate that transmission of *B. burgdorferi* by *I. scapularis* ticks requires more than 24 h of tick attachment, supporting a preventive role for daily tick checks[58,59]. However, the same time delay does not seem to hold for European *I. ricinus* ticks[60,61]. Although the available data are relatively limited, transmission from these ticks may occur in <24 h (refs. 2,60,61). No cases of transfusion-associated Lyme

Fig. 2 | Generalized life cycle of *Ixodes ricinus* complex ticks. The *Ixodes ricinus* complex life cycle comprises four life stages (egg, larva, nymph and adult), where each post-egg stage needs a blood meal to develop into the next stage or reproduce. Larvae and nymphs feed on many vertebrate species, including mammals, songbirds and lizards, whereas adults feed on a narrower range of hosts, most commonly deer. Lyme borreliae are not passed from infected female ticks to their offspring; thus, the bacteria are maintained in an enzootic cycle between immature ticks (larvae and nymphs) and reservoir hosts, typically small mammals and songbirds. Climate has a large role in the duration of the life cycle, which commonly takes 2–3 years to complete but may take substantially longer in higher latitudes. Similarly, the seasonal activity of these ticks is greatest in spring and early summer and less in fall but varies within and among tick species. For illustrative purposes, this figure typifies the life cycle of *Ixodes scapularis* in the northeastern USA. Eggs are laid in the late spring; larvae will hatch in mid-summer and then seek their first host in late summer. Engorged larvae will detach, drop off the host and moult into nymphs in the leaf litter. Unfed nymphs will emerge and quest for their second host the following late spring through summer, which is therefore the epidemiologically most risky time of the year. Engorged nymphs will detach, drop off the host and moult into adults in the leaf litter. Beginning in the fall, to mate and feed, adult ticks will quest for a third host, most commonly deer. Engorged females will detach from the host and drop into the leaf litter, where they will overwinter. Adult ticks that are unable to locate hosts in the fall will also overwinter and resume questing in early spring. Both fall-fed and spring-fed *I. scapularis* females will oviposit a single egg mass (up to 1,500–2,000 eggs) in late spring or summer and then die. Larvae hatch from eggs later in the summer, continuing the cycle. Although both nymphs and adult females may be infected, because of their small size (about the size of a poppy seed), their large numbers, and a spring/summer activity period that coincides with increased outdoor human activity, nymphs are responsible for most human infections. Because they are larger (about the size of a sesame seed) and have red coloration, adult female ticks are easier to notice and remove before the bacteria can be transmitted[13,51].

borreliosis have been documented, and there is no credible evidence of transmission through sexual contact, semen, urine or breast milk[62,63].

Mechanisms/pathophysiology

The pathogenesis of Lyme borreliosis involves a complex interplay between Lyme borreliae and the host, resulting in activation of innate and adaptive immune responses[64]. Lyme borreliae use several strategies to infect, disseminate and persist in the vertebrate host[65]. Following a tick bite, Lyme borreliae proteins, such as BBK32 as well as REVA, REVB and BB0347, bind fibronectin and glycosaminoglycans to initiate vascular attachment, promote migration along endothelium and facilitate tissue exit[66]. Decorin-binding proteins (DbpA and DbpB) promote binding to decorin, which is expressed in collagen-rich tissues and likely contributes to Lyme borreliae tropism to joints[66]. Dissemination and transmigration through tissues are further facilitated by the ability of Lyme borreliae to recruit and activate host proteases like plasmin and matrix metalloproteinases, which degrade extracellular matrix components. Lyme borreliae use several strategies to evade the immune system[1,65]. For example, outer-surface proteins, such as complement regulator-acquiring surface proteins and complement-sensitive protein Z (CspZ), bind to the human complement regulator Factor H, effectively cloaking the spirochaete from complement-mediated killing[65]. The antigenic variation primarily in the vlsE locus results in a diverse and variable antigenic repertoire that helps the evasion of antibodies and antibody clearance[1,67]. The production of antioxidant enzymes, such as the manganese-dependent superoxide dismutase (MnSOD or SodA), counteracts reactive oxygen species that could cause lipid peroxidation and compromise membrane integrity[68]. The incorporation of host lipids into the spirochaete's membranes may reduce immunological recognition by the host[65]. Collectively, these factors promote the ability of Lyme borreliae to infect, disseminate and, ultimately, persist in untreated vertebrate hosts.

Much of this knowledge was gained from animal models that have generated a substantial amount of information on how Lyme borreliae interact with and evade immune responses and thereby persist in reservoir hosts[65]. However, animal models do not reproduce the full spectrum of clinical features of Lyme borreliosis observed in humans[64]. The remainder of this section focuses on the immune responses to Lyme borreliae and how these responses potentially impact disease presentation in humans.

Immune responses in Lyme borreliosis

Immune events during infection. The initial human encounter with Lyme borreliae occurs at the tick-bite site in the skin. The innate immune system in skin, composed of macrophages, Langerhans cells and neutrophils, serves as the first line of defence by recognizing multiple Lyme borreliae lipoproteins via Toll-like receptor 1 (TLR1) and TLR2 (refs. 69–71), flagellar proteins via TLR5 (ref. 72), nucleic acids via TLR7, TLR8 and TLR9 (refs. 73–75), and peptidoglycans via intracellular NOD1 and NOD2 receptors[64,76]. This facilitates phagocytosis, antigen presentation, and robust production of pro-inflammatory cytokines and chemokines, leading to recruitment and activation of adaptive immune cells to sites of infection[1,64,77]. The inflammatory immune responses are further bolstered by activation of resident stromal cells, including dermal fibroblasts and endothelial cells, that sense and respond to Lyme borreliae[78–80]. In humans, the relatively broad-range immune responses to spirochaetes result in the production of a wide range of cytokines and chemokines, including TNF, IL-6, CCL2 and CXCL8, which are important in the early innate immune responses, IFNγ and IFNγ-inducible chemokines[81], which orchestrate T helper 1 (T_H1) responses, and IL-17 and IL-23, which promote T_H17 responses[82–84]. Elevated levels of these inflammatory mediators are apparent in skin and blood within days of the onset of EM skin lesions[84,85]. However, antibody reactivity to Lyme borreliae is slower to develop, requiring several weeks. This delayed antibody response poses diagnostic challenges during the first weeks of infection[86]. As the B cell response matures, robust antibody reactivity is observed against an expanded array of borrelia outer surface proteins. These elevated antibody resposes are thought to have an important role in controlling the infection and contribute to high seropositivity in patients at the late stage of infection[70,87].

From the inoculation site in the skin, borreliae may disseminate through blood and lymphatics, and possibly along the nerve canal, to other organ tissues[1,88–90]. Lymphatic spread is thought to contribute to early dissemination without immediately triggering strong systemic inflammation, giving Lyme borreliae time to adapt. Once Lyme borreliae cross into the bloodstream, the pathogen gains access to other areas of the skin or to distant non-skin sites, such as joints, heart and the nervous system, where they can persist for months to years. However, dissemination seems to be a transient process, and spirochaetes do not appear to multiply to large numbers or survive long-term in the systemic circulation. As the disease progresses, immune responses become increasingly localized to tissues, such as joints or cerebrospinal fluid (CSF), with little other systemic involvement[91,92]. However, Lyme borreliae are rarely detected by either PCR or culture in CSF and very rarely by culture in synovial fluid.

Although the host immune response contributes to protection against Lyme borreliae, timely resolution of infection generally requires appropriate antibiotic therapy[93]. Without antibiotic therapy, Lyme borreliae may persist in tissues for months to years. For example, Lyme borreliae have been isolated from the skin of patients with ACA more than a decade after the presumed initial infection and despite the presence of an expanded anti-borrelia antibody response[73,94]. By contrast, particularly in Europe, a substantial proportion of Lyme borreliae infections are asymptomatic[1,2], a phenomenon that is likely largely attributable to an effective host immune response.

Immune response in disease pathogenesis. Because Lyme borreliae do not produce any known toxins, the clinical signs and symptoms of infection are largely attributed to the host immune response. A growing number of studies have also linked variation in the immune response with the clinical heterogeneity of the disease[1,82–84,95–97]. For example, in patients with EM, high levels of inflammatory mediators in blood, including IL-6, IFNγ, CXCL9 and CXCL10, are associated with a greater number of systemic symptoms[84] (Figs. 3 and 4). In patients with Lyme neuroborreliosis, high levels of inflammatory mediators in CSF, particularly those related to activation and recruitment of T_H1, T_H17 and B cells, including CXCL13, are associated with painful meningoradiculoneuritis, termed Bannwarth syndrome (nervous system impairment characterized by severe pain due to inflammation of the nerve roots), and a higher frequency of constitutional symptoms[90,91,98]. Similarly, high levels of these mediators in joint fluid are associated with persistent synovitis despite antibiotic therapy, a condition now referred to as post-infectious Lyme arthritis[77,84,99,100]. Thus, certain clinical manifestations and the duration of illness seem to be largely shaped by the host immune response to Lyme borreliae.

There is also increasing evidence that inappropriate, sustained immune activation contributes to long-term sequelae after antibiotic therapy for Lyme borreliosis. Although persistent infection is often proposed as an explanation for lingering symptoms, microbiological measures of infection in the post-antibiotic period have been negative for almost all patients[101–103], and six double-blind, placebo-controlled treatment trials in Europe and the USA have not shown a risk-to-benefit ratio that favours antibiotic retreatment[104–108]. Of note, longitudinal studies of patients followed from initial infection into the post-antibiotic period have linked immunological or metabolic abnormalities with persistent symptoms after various manifestations of Lyme borreliosis[84,87,95,99,100,109–111]. In particular, several studies demonstrated heightened levels of inflammatory cytokines, including IFNα, and IL-17A and IL-23, which are associated with T_H17 responses, in patients with persistent symptoms following Lyme neuroborreliosis or EM[91,95,109,111,112].

The reasons for immune dysregulation are not well understood and are likely multifactorial. One hypothesis is that retained spirochaetal antigens, such as the Lyme borreliae peptidoglycan, continue to trigger immune activation[76,97,113]. Another hypothesis is that a subset of patients is (genetically) predisposed to aberrant immune responses that persist even after resolution of infection. This is illustrated in patients with post-infectious Lyme arthritis, in whom sustained levels of T_H1 and T_H17 mediators months after antibiotic therapy correlate strongly with autoreactive T cell and B cell responses and joint pathology[77,81,84,114–122]. Collectively, several studies suggest that dysregulated immune responses triggered early in the infection may contribute to more severe disease, including persistent symptoms in the post-treatment period.

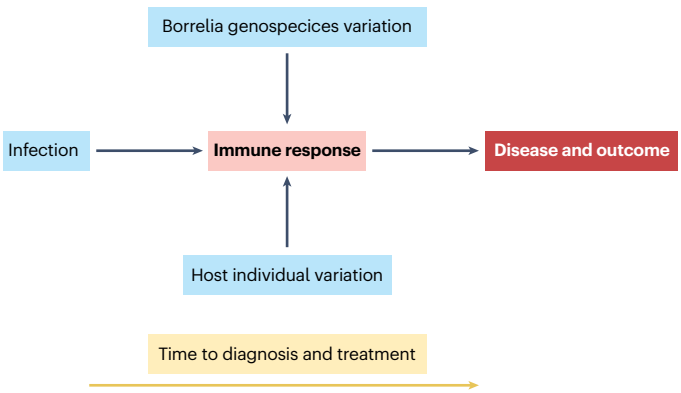


Fig. 3 | Factors affecting disease outcome. The clinical presentation of Lyme borreliosis is primarily due to the host immune response to Lyme borreliae; Lyme borreliae are not known to express toxins. Robust multi-prong innate and adaptive immune responses are already observed within days to weeks of infection onset and may be associated with culture-negative results indicative of Lyme borreliae killing. However, heightened immune responses also correlate with greater symptomatology. Moreover, sustained elevated immune responses have been associated with unfavourable long-term outcomes in some patients. As is the case for many infections, both microbial and host factors are thought to contribute to these responses. In addition, timely diagnosis and treatment are the major determinants of the clinical course and outcome of Lyme borreliosis[84].

Microbial genetic variation in Lyme borreliosis

Clinical heterogeneity in Lyme borreliosis is also linked to genetic differences in the infecting strain of Lyme borreliae. The most compelling evidence comes from studies of Lyme borreliae species recovered from European patients with various presentations of Lyme borreliosis. These studies demonstrated that *B. afzelii* is the species most commonly associated with skin manifestations, *B. garinii* and *B. bavariensis* with nervous system involvement[2,94,123–126] (Fig. 5), and *B. burgdorferi* with arthritis[127]. In addition to tissue tropism, differences in symptomatology have also been observed. Namely, infection with *B. burgdorferi* is associated with a more symptomatic early infection in the USA than infection with either *B. afzelii*, *B. garinii* or *B. burgdorferi* in Europe[12,128–130]. *B. mayonii* potentially reaches higher levels of spirochaetaemia and can present with diffuse rashes due to multiple very small EM skin lesions[23].

These differences in virulence are likely due to a combination of factors, including variations in adhesion proteins and tissue tropism, the ability to establish infection in various organs, and the capacity to elicit host immune responses. Efforts to understand microbial factors in Lyme borreliosis pathogenesis have focused on mechanistic studies in vitro and in animal models, particularly laboratory mice, using a few genetically manipulatable laboratory strains of *B. burgdorferi* (for example, B31 and 297). These studies have yielded relevant information on genes that influence *B. burgdorferi* infectivity and/or tissue tropism, including DbpA[131], outer surface protein C (OspC)[132], BBK32, BBA33, BB074 (refs. 133–136), and complement regulators such as complement regulator-acquiring surface proteins and Erps[137–139]. Nevertheless, the commonly used *B. burgdorferi* strains used in such studies do not represent the full Lyme borreliae heterogeneity in human disease, and laboratory mice recapitulate only certain aspects of Lyme borreliosis (for example, arthritis). Consequently, it has not been possible to link mechanistic insights from these models to disease in patients[12,140–142]. Advances in genotyping through whole-genome

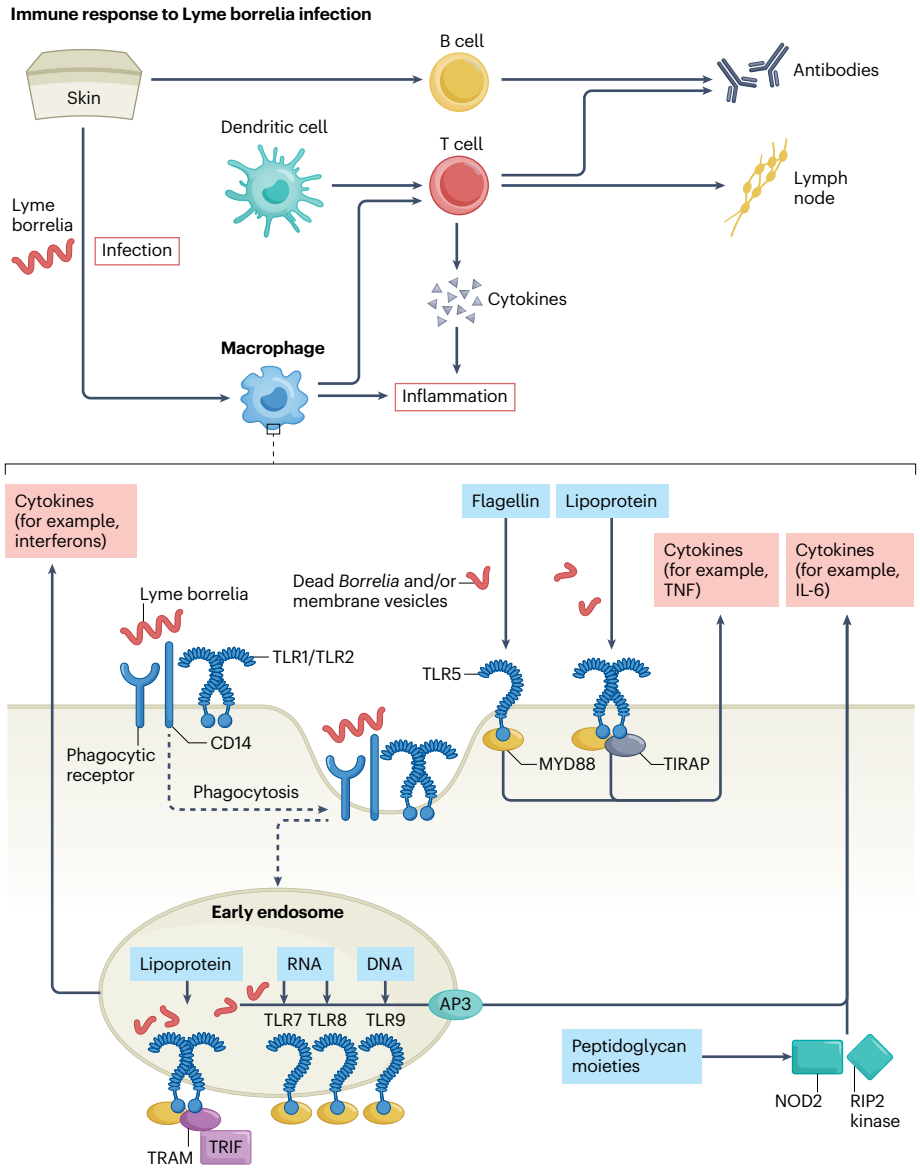


Fig. 4 | Pathogenesis of Lyme borreliosis. Immune response to Lyme borreliae: the innate immune system in the skin serves as the first line of defence against Lyme borreliae by recognizing multiple Lyme borreliae lipoproteins (via Toll-like receptor 1 (TLR1) and TLR2), flagellar proteins (via TLR5 present on the cell surface of immune cells), nucleic acids (via TLR7, TLR8 and TLR9) and peptidoglycans (via intracellular NOD1 and NOD2 receptors, which are present in intracellular compartments). This multi-antigen recognition facilitates efficient presentation of spirochaetal antigens by dendritic cells and macrophages and robust production of pro-inflammatory cytokines and chemokines, which can result in recruitment and activation of adaptive immune cells, including T cells and B cells, to sites of infection. The inflammatory immune responses are bolstered by the activation of resident stromal cells in skin, including dermal fibroblasts and endothelial cells, which sense and respond to Lyme borreliae. Elevated levels of a broad range of cytokines and chemokines are observed in the blood of patients within days of infection. Although humoral immunity is slow to develop, antibody reactivity to numerous borrelial outer-surface proteins is observed within a few weeks and is thought to be important in controlling the infection. Adapted from ref. 1, Springer Nature Limited.

sequencing will help to facilitate microbial genetic association studies to identify Lyme borreliae genetic elements linked to virulence, tissue tropism and particular clinical manifestations[12,143].

The Lyme borreliae genome has a unique architecture that affects the organism's virulence. In the *B. burgdorferi* B31 reference strain, ~40% of its genetic content is located on 12 linear and 9 circular plasmids. Plasmids encode several surface lipoproteins with tissue-adhesive and immunomodulatory functions[70,144], including a system for antigenic variation of an immunodominant surface lipoprotein, vlsE[145]. In addition, plasmids undergo recombination, leading to genetic exchanges[146]. Because of their homology, plasmids have been difficult to characterize with next-generation short-read sequencing platforms because they do not readily distinguish diverse and homologous regions[9,12,142,147,148]. Long-read sequencing technologies, along with open source bioinformatics tools, have enabled the systematic annotation of plasmid diversity across different strains of Lyme borreliae; efforts to correlate microbial genetic diversity with clinical symptoms have begun to provide insights into the genes associated with virulence[12]. Systematic association studies with thousands of isolates spanning the diversity of Lyme borreliosis clinical manifestations, as well as the genetic diversity of genospecies that cause Lyme borreliosis, are now needed. Such studies have the potential to reveal the specific microbial genes and allelic variants that underlie microbial genetic contributions to Lyme borreliosis, uncovering new aspects of spirochaete biology and disease pathogenesis.

Host genetics in Lyme borreliosis

Most host genetic associations in Lyme borreliosis to date involve variations in genes with immune function. The first evidence for this was the association of persistent post-infectious Lyme arthritis with certain HLA-DR alleles, namely DR4 and DR2 (refs. 149,150). HLA-DR molecules have a central role in antigen presentation and are associated

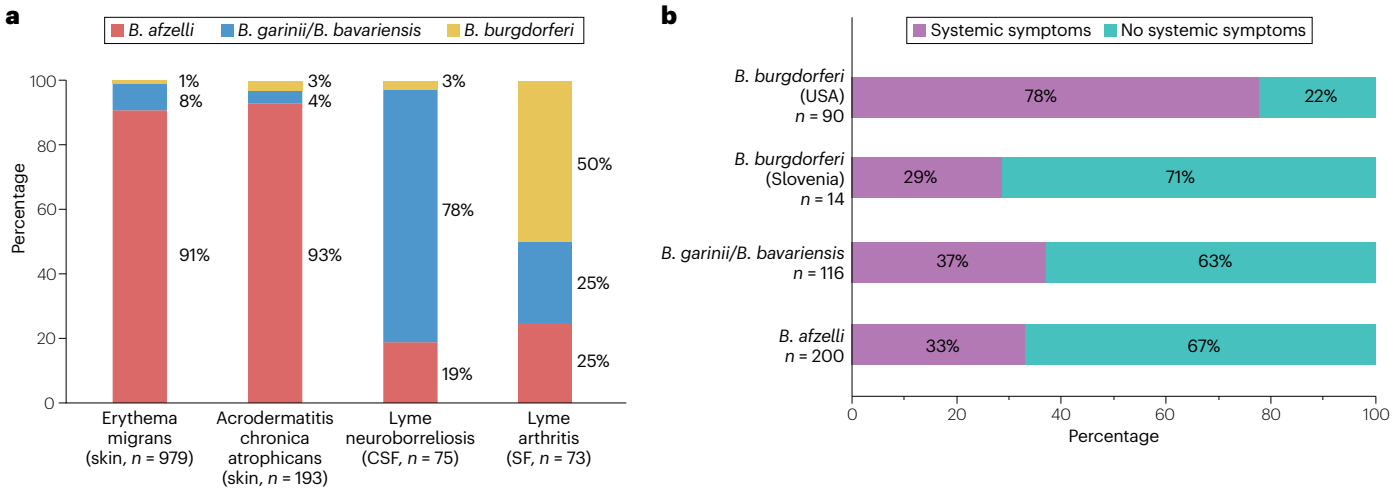


Fig. 5 | Genetic differences in Lyme borreliae and clinical heterogeneity in Lyme borreliosis. a, Distribution of Lyme borreliae species recovered from patients with different manifestations of Lyme borreliosis in Europe. Lyme borreliae were obtained from skin biopsy specimens from European patients with erythema migrans (n = 979) or acrodermatitis chronica atrophicans (n = 193), cerebrospinal fluid (CSF) from patients with Lyme neuroborreliosis (n = 75) and synovial fluid (SF) DNA from patients with Lyme arthritis (n = 73). The data shown are the percentages of Lyme borreliae species for each clinical manifestation. *Borrelia garinii* and *Borrelia bavariensis* are combined as most studies did not differentiate between the two species. Data from refs. 2,94,123–125,127,184–186. **b**, Differences in presentation of early Lyme borreliosis between European and North American patients. Comparison of the presence of systemic symptoms in 420 patients with erythema migrans in Europe and the USA according to the type of infecting Lyme borreliae[129] shows that infection with *Borrelia burgdorferi* in North America is associated with more symptomatic illness than infection with *Borrelia afzelii*, *B. garinii*/*B. bavariensis*, or *B. burgdorferi* in Europe.

with a high risk of several inflammatory and autoimmune conditions, including rheumatoid arthritis. Mutations in other innate immune genes, including the *TLR1*, a major sensor for *B. burgdorferi*, and the LCE3 gene family, which have defensin-like properties, were found to be associated with excessive inflammatory responses and a greater risk of developing post-infectious Lyme arthritis[99,100]. In addition, mutations including in the secretoglobulin gene *SCGB1D2* (refs. 151,152) and in the KCTD20 and ETV7 loci[152] have been linked to increased susceptibility to developing Lyme borreliosis. These findings in humans have been corroborated by elucidation of multiple risk loci that regulate immune responses and arthritis severity in infected mice[153]. Collectively, these findings suggest that host genetic variation may impact not only the susceptibility to developing Lyme borreliosis but also shape the clinical course and severity of disease, presumably by modulating host immune responses to infection. A more complete understanding of the microbial and host determinants of virulence and immunogenicity is likely to usher in a new era of knowledge with potential implications for diagnosis, management, and treatment of Lyme borreliosis.

Diagnosis, screening and prevention

Lyme borreliosis begins with infection of the skin at the site of the tick bite, usually presenting as a single EM skin lesion[1,2]. From the initial site, Lyme borreliae can disseminate to other body sites through soft tissues and lymphatic and circulatory systems within days to weeks. Although EM will completely resolve without antibiotic treatment, untreated patients may go on to develop later objective manifestations such as lymphocytoma, neuroborreliosis, carditis, arthritis and/or ACA. Clinically, Lyme borreliosis is categorized as early localized, early disseminated and late stages (Fig. 6), although manifestations can overlap and not all stages will necessarily develop in an individual patient. Differences in the infecting Lyme borreliae species and in virulence within the same species influence dissemination and tissue tropism, factors that lead to variations in clinical features[8,141]. Early diagnosis and antibiotic treatment of Lyme borreliosis can prevent development or progression of the disease.

Clinical manifestations

Erythema migrans. EM is the most common clinical manifestation of Lyme borreliosis in both the USA and Europe, occurring in ≥80% of patients[8]. Most EM cases occur from June through August. Of importance, patients are often not aware of a preceding tick bite. EM starts as a small macule or papule that appears at the tick bite site, usually 7–14 days later, and enlarges over several days to weeks. Initially, EM lesions are typically a round or oval, homogeneous erythematous patch (Fig. 7a,b,c,f,g). As the lesion grows, it can develop a ring-like or target-like appearance with central clearing. Despite these stereotypical characteristics, lesions may vary in their appearance. Sometimes, the central part of an EM skin lesion is darker, a central papule is often present, and occasionally lesions may contain scale or central vesiculation. While mild local symptoms, such as itching, burning or pain, may occur at the EM site, lesions are often asymptomatic. Associated systemic symptoms may include fatigue, headache, neck pain, fever, musculoskeletal pain and regional lymphadenopathy[154,155]. Within days to weeks, haematogenous dissemination from the initial site can lead to multiple secondary EM lesions (Fig. 7d,e). These lesions resemble the initial primary lesion but are typically smaller. For early detection of EM, a complete skin examination is essential since the patient may be unaware of an EM skin lesion. EM lesions in dark-skinned persons (Fig. 7c) can be more difficult to recognize as the colour contrast between the erythematous areas and normal skin is less noticeable[156].

In North America, patients presenting with EM lesions have a shorter incubation period, faster lesion expansion and less central

clearing compared with EM skin lesions in Europe. Patients with EM in North America are also more likely to have systemic symptoms, multiple skin lesions, and regional lymphadenopathy and are less likely to recall a tick bite at the site of the skin lesion (25% versus 60%)[130,155,157–160]. *B. mayonii* infection, which thus far has only been detected in the upper Midwest USA, can cause numerous small erythematous macular lesions[23]. In Europe, EM caused by *B. garinii* spreads faster and more often has local symptoms than EM caused by *B. afzelii*[160]. A non-specific febrile illness during late spring and summer without an EM skin lesion can be the initial presentation of up to 18% of patients diagnosed with Lyme borreliosis in the USA[161,162].

Borrelial lymphocytoma. Borrelial lymphocytoma (Fig. 7h) is an uncommon, early localized cutaneous manifestation only observed in Europe, as it is mainly caused by *B. afzelii*. It appears as a single bluish-red nodule or plaque, up to a few centimetres in size, often found alongside an EM skin lesion. It occurs more frequently in children than in adults[163]. In adults, these lesions commonly involve the breast nipple and areola, while they often appear on the earlobe in children. About 50% of patients report a tick bite near or at the site of the lymphocytoma. Without antibiotic treatment, this skin manifestation may persist for several months to a year[163].

Early Lyme neuroborreliosis. Early Lyme neuroborreliosis occurs in ~10% of Lyme borreliosis cases[52,164]. The most common manifestations are cranial neuropathy (particularly a peripheral facial nerve palsy), lymphocytic meningitis and painful radiculitis, each of which can occur individually or together. In adult patients with Lyme neuroborreliosis acquired in Europe, where most cases are caused by *B. garinii* and *B. bavariensis*[10,165–169], Lyme neuroborreliosis most frequently presents as a painful meningoradiculoneuritis that typically worsens at night and does not respond well to analgesics. Lyme neuroborreliosis presenting with radiculopathy is often initially misdiagnosed[170]. In the USA, peripheral facial palsy is the most common clinical manifestation, with fewer patients presenting with severe radicular pain[54,171]. Up to 30% of patients with Lyme neuroborreliosis with facial palsy present with bilateral nerve involvement. Lyme meningitis typically has a subacute onset, beginning with a mild headache and minimal or absent neck stiffness. Less commonly, patients with Lyme neuroborreliosis may also develop multifocal polyneuritis, including brachial and lumbosacral plexopathies, and acute mononeuritis that may involve multiple nerves[54,166–169,172–174]. Rarely, patients may present with cerebral vasculitis and/or stroke or a disease resembling Guillain–Barré syndrome[175–177].

Children with Lyme neuroborreliosis usually present with acute peripheral facial palsy and CSF lymphocytic pleocytosis[173,174,178]. Clinical predictors of Lyme meningitis versus viral meningitis in children include duration of headache ≥7 days, papilloedema, the presence of a concomitant sixth or seventh cranial nerve palsy, and a proportion of mononuclear cells in CSF of ≥70%[179]. Pseudotumour cerebri is an unusual presentation of Lyme borreliosis that occurs predominantly in children[178].

Lyme carditis. Lyme carditis occurs in up to 2% of patients with early disseminated disease and is ~1.75 times more common in men[180]. Lyme carditis usually presents as a fluctuating acute atrioventricular

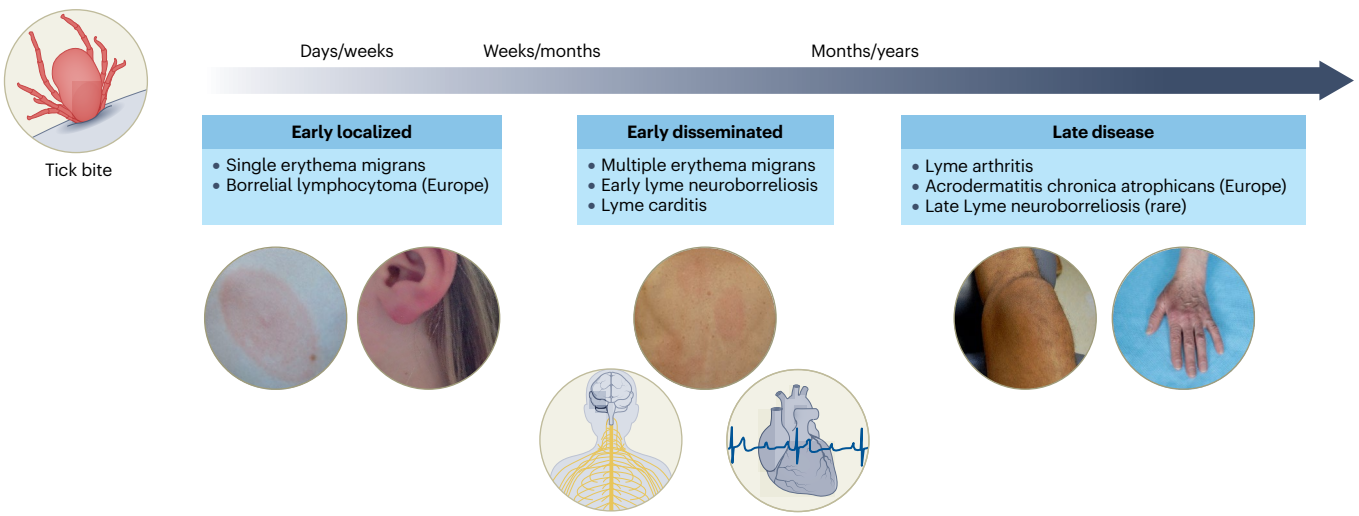


Fig. 6 | Clinical manifestations of Lyme borreliosis. Lyme borreliosis is categorized into early localized, early disseminated and late stages, although manifestations can overlap. The infection starts at the tick bite site, presenting as an erythema migrans skin lesion. From the initial skin site, Lyme borreliae can disseminate to other skin and extracutaneous sites within days to weeks. Borrelial lymphocytoma is an uncommon, early localized manifestation most often observed in children that appears as a single bluish-red nodule usually on the earlobe. Early disseminated manifestations include multiple erythema migrans skin lesions, early Lyme neuroborreliosis and Lyme carditis, usually occurring within 1–2 months after infection. Multiple erythema migrans skin lesions resemble the initial primary lesion but are usually smaller in size and do not have a punctum (because a punctum only occurs at the tick bite site). Early Lyme neuroborreliosis typically presents with cranial neuropathy (especially peripheral facial nerve palsy), lymphocytic meningitis and/or painful radiculopathy. Lyme carditis most commonly presents with fluctuating acute atrioventricular block. Late-stage manifestations occur months after the initial infection. Lyme arthritis is the most common late manifestation in the USA. Lyme arthritis presents with intermittent or persistent joint swelling and pain affecting large joints, particularly the knee. Acrodermatitis chronica atrophicans, the most common late manifestation in Europe, is characterized by reddish-blue discoloration and swelling of the skin, usually on the distal parts of the extremities, that slowly expand and progress to an atrophic stage. Borrelial lymphocytoma and acrodermatitis chronica atrophicans are manifestations of Lyme borreliosis that only occur in Europe. Late Lyme neuroborreliosis presenting with encephalitis, myelitis or encephalomyelitis has been reported in Europe but is very rare in the USA.

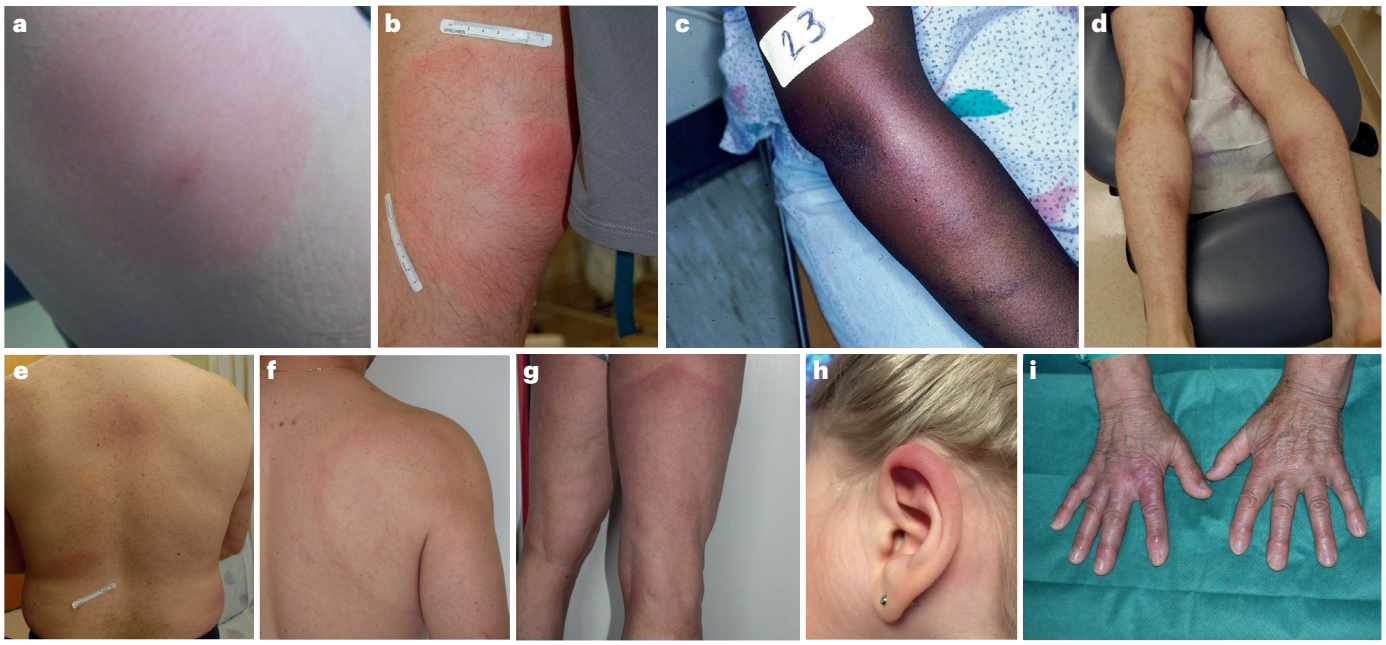


Fig. 7 | Dermatological manifestations of Lyme borreliosis. Solitary erythema migrans skin lesions in patients in the USA (parts **a** and **b**) and Europe (parts **f** and **g**). Erythema migrans lesion in a patient from the Caribbean who acquired the infection in Westchester County, New York, USA (part **c**). Multiple erythema migrans in patients in the USA (parts **d** and **e**). Borrelial lymphocytoma on the ear helix (part **h**), acrodermatitis chronica atrophicans on the dorsal side of the right hand (part **i**). Part **c** reprinted with permission from ref. 350, Elsevier.

block. Other manifestations, such as myocarditis, pericarditis and cardiomegaly, can occur. Signs and symptoms may include palpitations, syncope, dyspnoea, oedema, lightheadedness and chest pain. Lyme carditis can occur together with EM as well as with early Lyme neuroborreliosis. Even without antibiotic treatment, cardiac abnormalities usually resolve within 1–2 weeks[181]. There is no association between seropositivity for Lyme borreliae and heart failure[182].

Lyme arthritis. Lyme arthritis occurs most commonly with *B. burgdorferi* infection[53,127,183–186], and is therefore more common in the USA than elsewhere. In the USA, it is the most common late manifestation of Lyme borreliosis, occurring in ~60% of untreated patients with EM at a mean of 6 months after onset of the skin lesion[187]. Patients experience intermittent or persistent episodes of joint swelling involving one or a few large joints, especially the knee[188]. Children in the USA can have an acute presentation of Lyme arthritis that resembles septic arthritis, presenting with fever, joint pain and effusion, and inability to bear weight on the limb with the affected joint. Lyme arthritis can present at any time of the year, is more common among men and can be the only recognized manifestation of Lyme borreliosis[189].

Acrodermatitis chronica atrophicans. ACA, primarily caused by *B. afzelii* infection, is a common late manifestation in Europe. Patients with ACA are more often women (approximately two-thirds of all patients with ACA) and older in age (typically >50 years)[94]. The initial oedematous stage appears as a reddish-blue discoloration and swelling of the skin (Fig. 7i), which slowly enlarges and mostly involves the distal extensor parts of the extremities. If untreated, ACA may progress to the atrophic stage, with loss of body hair, connective tissue and fatty tissue, leading to thinning and wrinkling of the skin at the involved site. Juxta-articular fibrous nodules and swelling of joints on the involved extremity can also occur. A peripheral neuropathy is frequently associated with long-standing ACA. The median duration of ACA before diagnosis has been reported to be 12 months[94].

Late Lyme neuroborreliosis. Late Lyme neuroborreliosis, presenting as encephalitis, myelitis or encephalomyelitis, is observed uncommonly in Europe (~4% of Lyme neuroborreliosis cases) and is extremely rare in the USA[166,172]. The association of two other neurological syndromes attributed to Lyme borreliosis has been debated in the USA. The first is a mild encephalopathy with memory and concentration complaints but without CSF abnormalities or microbiological evidence of Lyme borreliae infection in the central nervous system[190]. The second is a primarily axonal and chronic symmetric distal sensory neuropathy[191–193].

Coinfections

The *Ixodes* ticks that transmit Lyme borreliae to humans also transmit other human pathogenic bacteria, viruses and parasites. In the USA, *I. scapularis* is also the vector for *Anaplasma phagocytophilum* (anaplasmosis), *Babesia microti* (babesiosis), *Borrelia miyamotoi* (*B. miyamotoi* disease), *Ehrlichia muris eauclairensis* (ehrlichiosis) and Powassan virus (Powassan virus disease/encephalitis)[194]. *A. phagocytophilum* and *B. microti* are the most frequent coinfections in patients with Lyme borreliosis in the USA[195]. In Europe, *I. ricinus* also transmits tick-borne encephalitis virus, *A. phagocytophilum*, *Rickettsia* species, *Neoehrlichia mikurensis*, *B. miyamotoi* and *Babesia* protozoans. Tick-borne encephalitis virus is the most common coinfection in endemic areas in Europe[196]. Emerging infections with novel viruses have been reported from Eurasia, including Alongshan virus, Beiji nairovirus, Songling virus, Jingmen

tick virus and Yezo virus[197–205]. These potential coinfections typically cause a non-specific febrile illness with malaise, myalgia, headache and anorexia. High fever, leukopenia, neutropenia, thrombocytopenia or anaemia are uncommon in Lyme borreliosis and, if present, might indicate an alternative diagnosis or a coinfection. More data on the impact of coinfections on the diagnosis and disease course of Lyme borreliosis are needed.

Southern tick-associated rash illness. Southern tick-associated rash illness (STARI) is a skin lesion that resembles EM, associated with the bite of the *Amblyomma americanum* tick, also known as the lone star tick[206–208]. The cause of STARI and the natural history of the illness are unknown. *A. americanum*, the most common human biting tick in the southeastern USA, is an aggressive species[209] and its range is steadily expanding northward, overlapping *I. scapularis* areas. Distinguishing between STARI and EM in these areas is difficult, unless the tick has been identified[210–212]. While STARI and EM have different serum metabolic profiles[213], there are no available tests to diagnose STARI. STARI does not occur in Europe, as *A. americanum* is not found in that geographical area. However, cases of a tick-associated rash illness associated with bites of *Amblyomma testudinarium* have been reported from Japan[214,215]. STARI is a substantial problem complicating Lyme borreliosis diagnosis in the USA, impacting the evaluation of the sensitivity of laboratory tests for diagnosing early Lyme borreliosis and the evaluation of the efficacy of Lyme borreliosis vaccines.

Laboratory testing

Because direct methods of detection of Lyme borreliae have limited sensitivity on most clinical samples, laboratory tests for Lyme borreliosis primarily detect antibodies to the bacteria. However, detection of Lyme borreliae DNA by PCR can be a valuable supplement to serology in certain cases, especially for testing of synovial fluid samples if Lyme arthritis is suspected, skin biopsy samples in cases of suspected ACA or atypical cases of EM, and, possibly, CSF in unusual cases of Lyme neuroborreliosis. However, owing to the low sensitivity of PCR in CSF, a negative result would not exclude the diagnosis. Recommended approaches[86,216–219] to support the diagnosis of different manifestations of Lyme borreliosis are shown on Table 1. Patients with EM should be diagnosed and treated based on clinical presentation alone.

Serology. Modern commercial tests based on recombinant borrelial antigens or on synthetic peptides generally show high analytic sensitivity and specificity[220,221]. The diagnostic sensitivity of antibody-based assays increases with duration of infection[216,218,222], with low sensitivity in patients with an EM skin lesion and high sensitivity in both Lyme arthritis and ACA.

To improve the specificity of serological testing, a two-tier approach is often applied. The standard algorithm typically uses an enzyme immunoassay to screen for IgM and/or IgG antibodies as the first-tier test, followed by a confirmatory immunoblot if the first-tier test is positive or equivocal[216,218,222,223]. In the USA, criteria for interpretation of immunoblots have been established by the Centers for Disease Control and Prevention[224], whereas no general criteria have been formulated in Europe, probably due to the presence of several different species of Lyme borreliae causing disease[225]. The IgM immunoblot result is only applicable for patients with an illness duration of ≤30 days. A modified algorithm using two first-tier tests, ideally with different antigenic constituents, can be used either in parallel or sequentially[86,224]. This modified two-tier testing algorithm has higher sensitivity in early Lyme borreliosis, while maintaining high specificity[226], and can be used

Table 1 | Diagnostic strategy for different clinical manifestations of Lyme borreliosis

Manifestation	Diagnostic strategy	Serology	Other tests
Erythema migrans (single and multiple)	Clinical presentation	For atypical presentations, antibody testing on an acute phase sample; if negative, retest in 2–6 weeks	PCR for Lyme borreliae on skin biopsy samples can be helpful in atypical cases
Borrelial lymphocytoma	Clinical presentation with serological testing	Serological testing is usually positive at presentation; if negative, retest in 2–6 weeks	Histopathology and PCR for Lyme borreliae on skin biopsy samples can be helpful
Early LNB	Clinical presentation with serological testing CSF examination is necessary for diagnosis in Europe and recommended for consideration in the USA Skin examination for erythema migrans	Serological tests are usually positive at presentation; if negative, retest in 2–6 weeks In Europe, Lyme borreliae-specific CSF/serum-antibody index should be determined	CSF lymphocytic pleocytosis and/or evidence of intrathecal antibody production against Lyme borreliae corroborate the diagnosis but may be negative in very early LNB CSF CXCL13 levels are elevated PCR for Lyme borreliae in CSF has very low sensitivity but may be helpful in very early LNB, unusual cases and in immunosuppressed individuals with impaired antibody production
Lyme carditis	Clinical presentation with serological testing Skin examination for erythema migrans	Serological tests are usually positive at presentation; if negative, retest in 2–6 weeks	None
Lyme arthritis	Clinical presentation with serological testing	Serological testing is positive at presentation, with high levels of specific IgG antibodies	PCR in synovial fluid has moderate-to-high sensitivity (40–96%) and can be helpful to establish the diagnosis of Lyme arthritis; by contrast, borrelial antibody testing of synovial fluid is not useful or recommended
Acrodermatitis chronica atrophicans	Clinical presentation with serological testing Histopathology	Serological testing is positive at presentation, with high levels of specific IgG antibodies	PCR for Lyme borreliae in skin samples can be helpful (sensitivity ~30–40%)

For further information, see refs. 217–219. CSF, cerebrospinal fluid; LNB, Lyme neuroborreliosis.

in both the USA and Europe[227], but the increased sensitivity is still not sufficient for reliably diagnosing EM[86,226,228].

The diagnosis of Lyme borreliosis cannot be based on serological laboratory testing alone, and serological testing should only be performed when there is clinical suspicion of Lyme borreliosis. Assays must have high specificity, as most testing is being performed in situations with a low pretest probability of Lyme borreliosis, increasing the chance of false-positive results[86,222,228,229]. Non-specific antibody cross-reactivity due to other infections or autoimmune diseases occurs and can cause false-positive Lyme serological test results, particularly with IgM-based tests[86,228,230]. Persistence of IgM reactivity without subsequent development of IgG reactivity in untreated patients with symptoms for longer than 4–6 weeks makes Lyme borreliosis unlikely[216]. The use of non-validated tests or non-validated interpretation criteria is not recommended.

Serological assays have important limitations that must be considered when interpreting the test results. Current antibody-based assays do not distinguish between active and past Lyme borreliae infections; antibodies, including IgM, can persist for many years after successful antibiotic therapy[231]. Repeat testing is not recommended as follow-up after antibiotic treatment[86,222]. In highly endemic regions, the background seroprevalence rate, which is between 2% and 20% in Europe[230,232,233] but often less than 2% in the USA[234–237], needs to be considered when interpreting serological test results. Detected antibodies may also be from previous asymptomatic exposure to Lyme borreliae with subsequent seroconversion[238]. Asymptomatic seroconversion is rare in the USA, occurring in <7% of cases[239,240]. By contrast, ~60% of infections in Europe result in asymptomatic seroconversion[238,241].

Intrathecal production of antibodies against *B. burgdorferi* sensu lato. Investigation of possible Lyme neuroborreliosis with CSF analysis, including CSF leukocyte count and testing for intrathecal production of Lyme borreliae-specific antibodies, is the mainstay of laboratory diagnosis in Europe[217] and is also recommended in the USA[218]. To demonstrate intrathecal antibody production, a Lyme borreliae-specific antibody index is calculated using both CSF and serum to correct for passive diffusion of antibodies across the blood–brain barrier. Interpretation of results depends on the specific assay[242,243]. In Europe, the sensitivity of the antibody index is <80% during the first 2 weeks of neurological symptoms, increasing to 90–100% in cases with longer symptom duration[242]. In some patients, antibodies are detected in CSF earlier than in serum, possibly due to the sensitivity of the assay used and the algorithm used to define seropositivity[166,244–248]. Importantly, a positive CSF antibody index may persist for several months to years after adequate treatment[244,249,250]. There is limited research on the use of the antibody index to diagnose Lyme neuroborreliosis and there is no standardization of assays and methods between laboratories in the USA; however, a positive CSF antibody index seems to be less common in the USA than in Europe[54,251–253].

PCR and culture. The sensitivity of PCR to detect Lyme borreliae DNA varies depending on the type of sample assessed and on the assay methodology. Sensitivity of PCR testing is low in CSF[217] during Lyme neuroborreliosis (5–30%) but is much higher in synovial fluid samples of patients with Lyme arthritis (40–96%) and on skin biopsy samples in suspected cases of EM (30–89%) and ACA (20–100%)[254]. It is possible to culture Lyme borreliae from clinical specimens, especially from skin biopsies taken from EM or ACA skin lesions; however, the sensitivity of culture from blood, CSF and synovial fluid for extracutaneous manifestations is very low. Because performing a culture is time-consuming and requires special culture medium and expertise, its use is mostly restricted to research studies[216].

Other diagnostic modalities. Although in clinical practice histopathological findings are — in general — not important for the diagnosis of EM, they may be valuable in patients with suspected borrelial lymphocytoma located outside of the earlobe to exclude neoplasia and are a part of the diagnostic algorithm for those with suspected ACA.

In patients with Lyme borreliosis, routine laboratory blood tests are typically in the normal range. However, an elevated erythrocyte sedimentation rate, increased liver enzyme levels, and/or lymphopenia can occur, although more commonly in patients in North America[130,157–159].

Synovial fluid in patients with Lyme arthritis shows leukocytosis with a neutrophil predominance. By contrast, in Lyme neuroborreliosis, CSF pleocytosis is predominantly due to the presence of lymphocytes, and most patients also have an elevated CSF protein level with evidence of oligoclonal IgG bands[217]. The B cell-attractant chemokine CXCL13 is elevated in the CSF of most patients with Lyme neuroborreliosis and decreases faster than either cell count or antibody index after antibiotic treatment[255]. However, moderately to highly elevated levels of CXCL13 in CSF are not specific for Lyme neuroborreliosis and can be found in other neuroinflammatory conditions such as multiple sclerosis, central nervous system lymphoma and viral meningitis[256].

Screening and prevention

There are no recommended screening strategies for Lyme borreliosis. Preventive measures include avoiding tick exposure by limiting outdoor activities in tick-infested locations, using tick repellents, tucking in clothing to decrease exposed skin surfaces, bathing or showering within 2 h after work or recreation in a tick habitat, and frequent skin inspections for early detection and removal of ticks[218,257]. Use of acaricides on property and construction of deer fences have also been proposed[218]. A phase II study of TP-05, an oral preparation of the isoxazoline drug lotilaner, has been completed in humans in the USA and was reported to kill most attached *I. scapularis* ticks within 24 h of attachment[258].

Antibiotic prophylaxis with single-dose doxycycline given within 72 h after a recognized *I. scapularis* tick bite has been shown to be 87% effective in reducing the risk of acquiring Lyme borreliosis from that tick bite in the USA[240]. A single dose of doxycycline can be offered to adult patients and to children in the USA when: (1) the attached tick is an adult or nymphal *Ixodes* species tick estimated to be attached for ≥36 h based on the degree of tick engorgement or time of exposure; (2) prophylaxis can be started within 72 h from tick removal; (3) ecological information indicates tick infection rates with *B. burgdorferi* of ≥20%; and (4) doxycycline is not contraindicated[218]. For patients who do not fulfil all these criteria, observation is recommended. A similar single-dose doxycycline prophylaxis study conducted in the Netherlands showed a relative risk reduction for prevention of Lyme borreliosis of 67%[259]. However, in most European countries, the principle of 'watch and wait' is recommended for *Ixodes* tick bites[2].

Vaccines. OspA is expressed by Lyme borreliae during the tick phase and downregulated in the vertebrate host[260]. Circulating anti-OspA antibodies in vaccinated hosts ingested by the tick during feeding will bind to spirochaetes in the tick midgut, preventing transmission to the host[261,262]. The efficacy of OspA-based vaccines correlates with high anti-OspA antibody levels[263,264]. Vaccination with a multi-dose regimen of a single recombinant OspA preparation was found to be safe and

effective for preventing Lyme borreliosis in the USA, but this vaccine has not been available since 2002 (ref. 265). VLA15 is a multivalent OspA vaccine that covers the additional Lyme borreliae types present in Europe. Results of the phase I and II studies have found VLA15 to be safe, well-tolerated and immunogenic[266–270]. Two phase III clinical trials of VLA15 have completed enrolment[271]. Regimens include four doses, at ~0, 2, 6 and 18 months, with the third and fourth doses given just before the start of tick season. Results from these trials are expected to be available soon. Pre-exposure prophylaxis using an anti-OspA monoclonal antibody prevented transmission of *B. burgdorferi* in animal models[272]. The results of a phase I human clinical trial for this monoclonal antibody have not yet been reported. Canine vaccines for the prevention of Lyme borreliosis are available, effective and widely used in both North America and Europe[273].

Management

General approaches and basic principles for antibiotic treatment

Treatment with an appropriate antibiotic (Table 2) should result in both microbiological cure and resolution of the clinical symptoms in most patients[218]. Nevertheless, some patients have residual tissue or organ damage that occurred during the infection prior to antibiotic treatment (for example, a residual facial palsy or skin atrophy in patients with ACA) or the result of inflammatory processes that began during the course of the infection but then continued independently of the presence of the causative agent (for example, post-infectious Lyme arthritis). The success of antibiotic treatment in symptom resolution is influenced by the duration of inflammation before antibiotic treatment, the type of tissue affected and any residual tissue damage[1,219]. Early manifestations of Lyme borreliosis should be treated promptly to shorten the duration of illness and to prevent the development of later manifestations[94,274,275].

Results of testing in vitro have shown that Lyme borreliae are susceptible to most penicillins, many second-generation and third-generation cephalosporins, tetracyclines, and certain macrolides; conversely, they are resistant to specific fluoroquinolones, rifampin and first-generation cephalosporins[218,219,276]. Available evidence suggests that acquired antibiotic resistance does not represent a substantial clinical problem[218,219,276].

The shortest duration of effective treatment for patients with Lyme borreliosis has never been systematically evaluated for any antimicrobial agent. However, based on clinical trial data, patients with early Lyme borreliosis manifested by EM are usually effectively treated with 10 days of doxycycline[277,278]. In a European study, a 7-day course of doxycycline was shown to be non-inferior to a 14-day course of doxycycline for patients with a single EM skin lesion[279]. Alternative antibiotic treatments for patients with EM typically include amoxicillin (14 days), cefuroxime axetil (14 days) or phenoxymethylpenicillin (10–14 days). By contrast, for patients with late Lyme borreliosis manifested by Lyme arthritis, a 4-week course of an appropriate oral antibiotic is recommended as the initial treatment[1,218,219], and for the late skin manifestation, ACA, a 3–4-week course of an appropriate oral antibiotic is recommended[2].

Treatment of Lyme borreliosis manifestations

Recommendations for antibiotic treatment of the most common clinical manifestations of Lyme borreliosis are shown in Table 3.

Erythema migrans. For treatment of patients with EM, doxycycline, amoxicillin and cefuroxime axetil are each highly effective and are the preferred agents in the USA[218]; in Europe, in addition to these agents, phenoxymethylpenicillin is recommended for treatment of EM[2] as well as for borrelial lymphocytoma[163] in some of the guidelines. Macrolides, such as azithromycin, are considered somewhat less effective and are therefore recommended as a second-line therapy[1,218,219].

Lyme neuroborreliosis. Early Lyme neuroborreliosis is usually treated with a 14–21-day course of oral doxycycline or, alternatively, with intravenous administration of ceftriaxone or another recommended parenterally administered β-lactam antibiotic. Studies conducted in Europe have demonstrated that oral doxycycline is equally as effective as intravenous ceftriaxone for treatment of early Lyme neuroborreliosis[167,280–287]. However, in patients with parenchymal involvement of the brain or spinal cord (typically observed only with late Lyme neuroborreliosis), intravenous ceftriaxone is currently recommended, rather than oral doxycycline[218].

Lyme carditis. Hospitalization for continuous cardiac monitoring is recommended for patients with Lyme carditis who have electrocardiogram abnormalities such as PR prolongation of >300 ms, second-degree or third-degree heart block, or severe arrhythmias, as well as for patients with other clinical manifestations of myopericarditis[218]. Temporary cardiac pacing should be used for symptomatic patients with bradycardia that cannot be managed medically. Hospitalized patients with Lyme carditis are treated with intravenous ceftriaxone. When clinical improvement occurs, the patient may be switched to an appropriate oral antibiotic, such as doxycycline, to complete a 14–21-day total course of treatment. Outpatients with Lyme carditis are treated with a 14–21-day course of an appropriate oral antibiotic[218].

Lyme arthritis. A 28-day course of oral antibiotic therapy is usually effective for treating patients with Lyme arthritis. However, patients whose arthritis has improved but not resolved may be retreated with

Table 2 | Antibiotics recommended for the treatment of patients with Lyme borreliosis

Route of administration	Preference	Drug
Oral	Preferred antibiotics	Doxycycline[a] Amoxicillin Cefuroxime axetil Phenoxymethylpenicillin[b]
	Alternative antibiotics	Azithromycin[c]
Parenteral	Preferred antibiotic	Ceftriaxone
	Alternative antibiotics	Cefotaxime Penicillin G

For further information, see refs. 218,344. [a]Tetracyclines are relatively contraindicated in pregnant women (previously listed as pregnancy category D). In most circumstances, oral β-lactam antibiotics are preferred for the treatment of children <8 years of age. Short courses of doxycycline are less likely to cause dental staining than earlier tetracycline antibiotics in children <8 years of age and may be considered for treatment of young children with neurological involvement, β-lactam antibiotic allergy or possible coinfection with microbial agents sensitive only to doxycycline. [b]Recommended for the treatment of skin manifestations of Lyme borreliosis in Europe. Ranges in the dosages recommended vary somewhat in different European countries. [c]Owing to lower efficacy compared with other antibiotics in one study conducted in the USA, azithromycin is reserved for patients who are unable to take or who are intolerant to doxycycline, amoxicillin or cefuroxime axetil (and phenoxymethylpenicillin in Europe). Patients treated with azithromycin should be closely followed to ensure resolution of the clinical manifestations.

Table 3 | Recommended therapy for individual manifestations of Lyme borreliosis

Manifestation	Antibiotic	Duration of antibiotic therapy
Erythema migrans	Oral regimen[a,b]	10 days for doxycycline; 14 days for amoxicillin or cefuroxime axetil, or 10–14 days for phenoxymethylpenicillin
Borrelial lymphocytoma	Oral regimen[a,b]	14 days for all regimens
Early extracutaneous disease		
Meningitis or meningoradiculoneuritis[c]	Oral regimen with doxycycline[a] or parenteral regimen[a]	14–21 days for all regimens
Cranial nerve palsy[c,d]	Oral regimen with doxycycline[a]	14–21 days
Cardiac disease	Oral regimen[a,e]	14–21 days for all regimens
	Parenteral regimen[a,e]	14–21 days for all regimens
Late disease		
Arthritis	Oral regimen[a,f]	28 days for all regimens
Recurrent arthritis after oral regimen	Oral regimen[a,f]	28 days for all regimens
	Parenteral regimen with ceftriaxone[a,f]	14 days
Post-infectious Lyme arthritis[f]	See therapies listed in the footnote[f]	Not applicable
Encephalitis, myelitis or encephalomyelitis	Parenteral regimen[a]	14–28 days for all regimens
Acrodermatitis chronica atrophicans	Oral regimen[a]	21–28 days for all regimens
Post-treatment Lyme disease symptoms/syndrome	Assess for alternative explanations for the symptoms; if none identified, proceed with symptomatic therapy	Not applicable

For further information, see refs. 218,344. Complete response to treatment might be delayed beyond the treatment duration, and relapse can occur. Patients with objective signs of relapse may benefit from a second course of antibiotic treatment. [a]See Table 2. [b]For adult patients who are intolerant of amoxicillin, doxycycline, cefuroxime axetil and phenoxymethylpenicillin, azithromycin can be prescribed (Table 2). [c]Data from European studies of early Lyme neuroborreliosis indicate that oral doxycycline and parenteral β-lactam antibiotic therapy are equally effective in Lyme meningitis, although similar studies have not been conducted in the USA. However, if there is also evidence of parenchymal involvement of the brain or spinal cord, then a 14–28-day course of parenteral β-lactam therapy is preferred over doxycycline. [d]Systematic studies of oral antibiotic therapy for patients with cranial nerve palsy have only evaluated doxycycline in patients with seventh cranial nerve palsy. Whether oral antibiotic therapy would be as effective for patients with other cranial neuropathies is unknown. [e]Parenteral antibiotic therapy is recommended at the initiation of therapy for hospitalized patients; an oral regimen can be substituted to complete a course of antibiotic therapy or to initiate treatment of ambulatory patients. A temporary pacemaker may be required for patients with advanced heart block. [f]Initial antibiotic treatment for Lyme arthritis is a 28-day course of either doxycycline, amoxicillin or cefuroxime axetil for adults and either amoxicillin or cefuroxime axetil for children <8 years of age. NSAIDs are often prescribed concomitantly. For patients who do not respond adequately to this therapy, subsequent treatment is less-well defined with several options, including repeating a 28-day course of an oral antibiotic or treating with a 14-day course of ceftriaxone (range 14–28 days). Patients with persistent synovitis despite repeated antibiotic treatment can be managed with NSAIDs and disease-modifying antirheumatic drugs. Synovectomy can be considered in refractory cases. Consultation with a rheumatologist is recommended.

a second 28-day course of an oral antibiotic. Patients without any improvement from the initial treatment with an oral antibiotic may instead be retreated with at least a 14-day course of ceftriaxone[1,219]. However, recommendations on antibiotic retreatment are largely based on expert consensus rather than high-quality evidence. NSAIDs may be administered with antibiotic therapy. NSAIDs may also be useful following antibiotic therapy, since persistence of mild joint inflammation immediately following antibiotic therapy is found in ~25% of patients with Lyme arthritis[218,288]. For patients with Lyme arthritis of the knee, physical therapy may be needed if quadriceps atrophy has developed[1]. In patients not responding to NSAIDs disease-modifying antirheumatic drugs, such as methotrexate, may be given for 6–12 months and are usually effective. Alternatively, for non-responding patients, arthroscopic synovectomy is an option[1,288].

Acrodermatitis chronica atrophicans. Patients with ACA are typically treated with oral antibiotics for 21–28 days[218,257]. Oedematous skin changes usually disappear within the first few weeks after beginning antibiotic treatment, although atrophic skin changes are typically irreversible[94].

Treatment in special populations

Pregnancy. Although miscarriage and stillbirth in pregnant women with Lyme borreliosis have been rarely reported, a causal relationship has not been established[289–292]. Pregnant women who develop Lyme borreliosis have generally had good outcomes if they receive appropriate antimicrobial therapy[292,293]. Antibiotic treatment of Lyme borreliosis in pregnant women is the same as for non-pregnant patients in the USA, except that doxycycline is not recommended as first-line therapy[218]. By contrast, some authorities in Europe instead recommend intravenous ceftriaxone for treatment of all manifestations of Lyme borreliosis during pregnancy, including EM[218,292].

Children. Approaches to treating children with Lyme borreliosis are similar to those for adults[218]; however, in most circumstances, a child <8 years of age is treated with recommended oral β-lactam antibiotics, and a patient ≥8 years of age is treated with oral doxycycline. On average, the recovery rate is faster in children, and the proportion of children with non-specific symptoms after treatment is lower than in adults[166,218,283,294,295]. Short courses of doxycycline are much less likely to cause dental staining than earlier tetracycline antibiotics in children <8 years of age, and treatment with doxycycline may be considered in young children with neurological involvement, drug allergy or possible coinfection with infectious agents sensitive to doxycycline[218].

Immunocompromised patients. The choice of antibiotic, the dosages and duration of treatment for a single EM skin lesion in immunocompromised patients are generally the same as for immunocompetent patients[296–298]. Likely, the same is also true for disseminated Lyme borreliosis such as for patients with multiple EM skin lesions or with Lyme neuroborreliosis. Close follow-up is desirable for patients with Lyme borreliosis who have an underlying haematological malignancy. In published case series, immunocompromised patients with EM more frequently developed signs of disseminated Lyme borreliosis and required antibiotic retreatment than immunocompetent patients with EM, matched for sex, age and antibiotic treatment. However, the outcome was excellent in both groups[296,297].

Quality of life

Death due to Lyme borreliosis is extremely rare and occurs primarily among patients with cardiac involvement[180,299,300]. Unconventional treatments for 'chronic Lyme disease' have also been linked to

mortality[301–304]. Despite these rare fatal outcomes, life expectancy for patients with Lyme borreliosis is comparable with that of the general population[305], and seropositive individuals without a clinical Lyme borreliosis history do not experience more health problems than seronegative populations[306]. However, some patients with Lyme borreliosis may experience impaired quality of life (QOL), mainly due to symptoms from untreated infection, although some may experience symptoms that persist after antibiotic treatment (Box 1).

Typically, signs and symptoms of Lyme borreliosis diminish and resolve during or within days to months following antibiotic therapy. Recovery is generally slower for patients with late manifestations than for those with early disease[165,168,278]. While most patients fully recover, a subset continues to experience persistent symptoms, potentially affecting QOL. Objective physical sequelae, such as persistent facial palsy following Lyme neuroborreliosis, post-infectious Lyme arthritis, neurological deficits post-encephalomyelitis, and skin atrophy due to ACA, may occur but are relatively uncommon. More commonly, patients may report non-specific symptoms, including fatigue, musculoskeletal pain and cognitive difficulties[103,169,278] without objective findings on physical examination.

Studies indicate that 0–27% of patients treated for EM and 10–20% of patients treated for Lyme neuroborreliosis experience symptoms for at least 6–24 months after antibiotic treatment[103,155,169,277,279,287,307–315]. One European study reported that 15% of patients with Lyme neuroborreliosis had residual complaints affecting daily life, and 78% had some remaining symptoms 12 months after doxycycline treatment[168]. Individual studies using the Short Form Health Survey (SF-36) have documented reduced QOL across multiple domains compared with the general population[316–318]. Reduced work capacity has also been reported in patients with Lyme borreliosis[316].

Predictors of poor QOL include delayed antibiotic treatment, persistent complaints at 3 months post-treatment[168,316], patient expectations regarding treatment, pre-existing anxiety and depression, poorer social and physical functioning, and prior cognitive impairment or chronic pain. Both fatigue and reduced QOL generally improve over time[305,317–320]. A prospective study showed that QOL scores increased to just above the USA national average after 3 years, regardless of Lyme borreliosis stage or severity at diagnosis[318]. Similar long-term improvements were observed in patients with EM evaluated at a median of 16 years after diagnosis[317].

Non-specific symptoms, such as fatigue, pain and sleep disturbances, are common in the general population and may be unexplained or caused by various conditions. Several prospective studies comparing patients with Lyme borreliosis to controls have found similar symptoms among both groups[103,278,279,310,311]. However, a large European study reported persistent symptoms at 1 year in 27% of patients with Lyme borreliosis, 23% of people with only tick bites, and 21% of the general population, for an excess of 4–6% of patients with symptoms attributable to Lyme borreliosis[312]. Although the difference was statistically significant, the Lyme borreliosis diagnosis was only a moderate predictor of persistent symptoms; main predictors included poor physical and social functioning, anxiety, depression, maladaptive illness perceptions, and comorbidities[321]. Female sex and history of traumatic life events have been independently associated with post-treatment Lyme disease syndrome[313]. Conversely, higher functioning and positive expectations before treatment correlated with better symptom improvement[104,322]. In an analysis based on >140,000 insurance claims in the USA, about 46% of Lyme borreliosis cases versus 41% of controls had one or more diagnosis codes associated with pain, fatigue, or cognitive symptoms in the year following treatment. The excess coding decreased from 2.5% at 2 months to 0.5% at 6 months and 1% at 12 months. Pain-associated codes were similar between patients and controls by 6 months. Fatigue-associated codes remained slightly elevated (~1%) by 12 months. Cognitive-associated codes were rare in both groups[323].

Patients with Lyme borreliosis with persistent, unexplained symptoms after antibiotic therapy are often diagnosed with post-treatment Lyme disease symptoms or post-treatment Lyme disease syndrome, the latter term applied when the symptoms substantially reduce functionality[219]. The underlying causes remain unclear, but increasing evidence suggests that unbalanced and sustained immune activation contributes to these sequelae. Microbiological studies have consistently failed to detect ongoing Lyme borreliae infection or coinfections transmitted by *Ixodes* ticks in these patients[324]. Furthermore, retreatment with prolonged antibiotics has shown no clear benefit and carries risks that often outweigh any marginal improvements[104–108,219].

Public fear of Lyme borreliosis causing severe brain and organ damage and reduced QOL has led to widespread non-scientific diagnostics and non-evidence-based treatment guidelines[302]. To further complicate the situation, the poorly defined term chronic Lyme disease has been introduced and widely used. It is often applied to patients with persistent pain, neurocognitive complaints, or fatigue despite lacking objective clinical or serological evidence of Lyme borreliae infection. Referral centre studies show that 60–91% of patients evaluated for Lyme borreliosis do not have active infection, and alternative diagnoses are found in up to 79% of cases[325–341]. Chronic Lyme disease is increasingly recognized as a complex syndrome often involving misdiagnosis and unproven, potentially harmful long-term multidrug treatments, delaying correct diagnosis and appropriate care.

Patients with reduced QOL and persistent symptoms after Lyme borreliosis treatment should be offered counselling to help manage their symptoms. Studies show that when patient complaints are taken seriously and patients receive a detailed and comprehensive evaluation, they avoid prolonged insecurity and anxiety. Providing clear

Box 1 | Patient experience

I often do garden work among bushes and thickets, so tick bites are common. When I find one attached, I remove it and check for some days for a red ring to see if I need medical attention. This Easter, I removed a few ticks and saw no signs of infection.

In early May, I developed pain in my lower back, which later spread to the front of my thighs. The doctor suspected sciatica and prescribed painkillers, advising physiotherapy. The physiotherapist ruled out a herniated disc and treated me for muscular issues. Massage and light exercises helped a little.

Over the next weeks, the pain worsened, especially at night, affecting my back, legs and feet. I couldn't train or run. Painkillers didn't help me sleep, and by mid-June, my arms also hurt severely. After ruling out blood clots, the doctor suspected something rheumatic, but tests were negative. I mentioned ticks, and a blood test indicated Lyme borreliosis. I started antibiotics, and the worst pain eased, but my shoulders were weak. At the hospital, tests confirmed neuroborreliosis with shoulder paresis. Recovery is slow; 7 months later, I still struggle to lift my arms, but I hope to fully recover over time.

Box 2 | Knowledge gaps and priorities

Prevention and public health

- Improvement and harmonization of Lyme borreliosis surveillance practices across different geographical jurisdictions to inform case burden and spread
- Assessment of the efficacy of personal protection measures for the prevention of tick bites and on how to increase their adoption by the public
- Determination of the minimum time required for transmission of European Lyme borreliae to humans following tick attachment
- Development of point-of-care technical aides for tick identification and measurement of tick engorgement levels
- Studies to test whether a very short course of amoxicillin or other antibiotics is comparable to single-dose doxycycline for the prophylactic treatment of tick bites
- Vaccines against Lyme borreliosis
- Development of an anti-tick vaccine and continuing studies on the use of certain oral acaricides to interrupt tick feeding and pathogen transmission in humans

Mechanisms

- Understanding the microbial and host determinants of the clinical presentation of Lyme borreliosis
- Understanding the pathogenesis of post-treatment Lyme disease symptoms/syndrome
- Explanation for a dysregulated immune response to infection in some patients with Lyme borreliosis
- Elucidation of the host immune mechanisms that shape Lyme borreliae persistence (or clearance) in reservoir animals and in patients

Clinical (course and outcome)

- Adding knowledge on the comparative frequency, course and outcome of the main clinical manifestations of Lyme borreliosis in different endemic geographical regions
- Further data on the course and outcome of Lyme borreliosis in immunocompromised patients, in pregnant patients, and in patients with Lyme borreliosis and coinfections
- Adding information on reinfections with Lyme borreliae
- Research on the impact of coinfections on the diagnosis and disease course of Lyme borreliosis
- Finding the cause of tick-associated rash illnesses

Diagnostics

- Rapid and accurate diagnostic tests for Lyme borreliosis
- Development of diagnostic tests that can discriminate between active infection versus previous Lyme borreliae infection or exposure
- Assessment and standardization of Lyme neuroborreliosis diagnostic criteria across North America and Europe, including intrathecal antibody index determinations and the value of cerebrospinal fluid CXCL13 in diagnosis and monitoring of the therapeutic response
- Studies to find biomarkers for the simple assessment of therapeutic response
- Studies to find biomarkers to predict post-treatment Lyme disease symptoms or syndrome and to discriminate between diverse causes of symptoms after treatment of Lyme borreliosis

Treatment

- Studies of shorter-course antibiotic regimens as non-inferior options to currently recommended courses
- Assessment of the therapeutic equivalence of oral and intravenous treatment of North American adult and paediatric patients with Lyme neuroborreliosis
- Studies comparing treatment with a second course of oral versus intravenous antibiotic therapy in patients with synovitis who do not respond to the initial 28-day course of oral antibiotic therapy
- Comparative studies to determine the optimal duration of therapy for borrelial lymphocytoma
- Comparative studies to determine the efficacy of different antibiotics and durations of treatment for acrodermatitis chronica atrophicans

explanations helps them cope[331]. A multidisciplinary approach in specialized clinics often identifies alternative diagnoses such as cancer or autoimmune diseases like multiple sclerosis[342,343].

Outlook

Lyme borreliosis is the most common tick-borne disease in the Northern Hemisphere. Despite awareness and numerous studies focused on this disease for nearly 50 years, many uncertainties remain regarding its diagnosis, treatment and prevention. These gaps should serve as an incentive for, and help to guide, appropriate basic, translational and clinical research (Box 2).

Improvements in Lyme borreliosis surveillance practices, with harmonization across different geographical jurisdictions, are needed to accurately quantify the public health burden associated with the disease.

Controlling Lyme borreliosis involves four main strategies: managing landscapes, managing wildlife hosts, using environmental insecticides and biological controls, and using personal protection measures. More research is needed to evaluate the effectiveness of combining multiple strategies for tick control and protection methods. Post-exposure antibiotic prophylaxis after a tick bite could be further refined by point-of-care tools that would help with tick identification and estimates of tick engorgement. In the case of a bite from a fully engorged tick, development of Lyme borreliosis is far more probable than with a tick that is newly attached. A vaccine to protect against Lyme borreliosis is under study and may be available soon in both the USA and Europe.

The development of diagnostic tests that could discriminate between active infection and past exposure and be used to monitor response to treatment would be a major advance in the field, as would be a rapid and accurate point-of-care test, which could be particularly useful in evaluating patients with early disseminated Lyme borreliosis as well as children with arthritis.

In terms of treatment, studies aimed at demonstrating that shorter courses of antibiotics are as effective as currently recommended

longer courses may lead to fewer adverse effects, less disruption of the microbiome, lower healthcare costs, and more sustainable use of antibiotics from an environmental and antimicrobial resistance perspective. As all evidence of equivalent outcome when comparing oral doxycycline with intravenous ceftriaxone for neurological Lyme borreliosis comes from European studies, comparative studies of North American adult and paediatric patients are needed. For patients with Lyme arthritis who have persistent synovitis after an initial course of oral antibiotic therapy, studies comparing intravenous antibiotic treatment to a second course of oral antibiotics would be helpful. Similarly needed are studies comparing the efficacy of different antibiotic regimens for the treatment of ACA and borrelial lymphocytoma.

The underlying mechanisms of symptoms after treatment of Lyme borreliosis are likely to be diverse, with a combination of factors predisposing and perpetuating symptoms in individual patients. Biomarkers are needed to distinguish between different subgroups of patients, enabling interventional research studies and personalized care for those experiencing prolonged symptoms. High-quality research is needed to understand and assist the large and varied group of individuals diagnosed with chronic Lyme disease.

Lastly, combating pseudoscience and false information in an era where unvetted online information abounds, and many people use social media as a primary information source, is a major challenge for today's society. The need to deliver accurate, evidence-based information is a critical issue in safeguarding the integrity of medical science and patient care.

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References

1. Steere, A. C. et al. Lyme borreliosis. *Nat. Rev. Dis. Primers* **2**, 16090 (2016).
2. Stanek, G. & Strle, F. Lyme borreliosis-from tick bite to diagnosis and treatment. *FEMS Microbiol. Rev.* **42**, 233–258 (2018).
3. Barbour, A. G. & Gupta, R. S. The family Borreliaceae (Spirochaetales), a diverse group in two genera of tick-borne spirochetes of mammals, birds, and reptiles. *J. Med. Entomol.* **58**, 1513–1524 (2021).
4. Margos, G. et al. Proposing a subgenus *Borreliella*. *Ticks Tick Borne Dis.* **16**, 102536 (2025).
5. Nagarajan, A. et al. The landscape of Lyme borreliosis surveillance in Europe. *Vector Borne Zoonotic Dis.* **23**, 142–155 (2023).
6. Kugeler, K. J., Earley, A., Mead, P. S. & Hinckley, A. F. Surveillance for Lyme disease after implementation of a revised case definition — United States, 2022. *MMWR Morb. Mortal. Wkly Rep.* **73**, 118–123 (2024).
7. Gray, J. & Kahl, O. in *Lyme borreliosis* (eds Hunfeld, K.-P. & Gray, J.) 31–45 (Springer International, 2022).
8. Marques, A. R., Strle, F. & Wormser, G. P. Comparison of Lyme disease in the United States and Europe. *Emerg. Infect. Dis.* **27**, 2017–2024 (2021).
9. Becker, N. S. et al. Recurrent evolution of host and vector association in bacteria of the *Borrelia burgdorferi* sensu lato species complex. *BMC Genomics* **17**, 734 (2016).
10. Coipan, E. C. et al. Imbalanced presence of *Borrelia burgdorferi* s.l. multilocus sequence types in clinical manifestations of Lyme borreliosis. *Infect. Genet. Evol.* **42**, 66–76 (2016).
11. Li, Z. et al. Genomic characterization of clinical *Borrelia burgdorferi* sensu lato isolates in the Netherlands over a thirty-year period. *BMC Genomics* **26**, 1153 (2025).
12. Lemieux, J. E. et al. Whole genome sequencing of human *Borrelia burgdorferi* isolates reveals linked blocks of accessory genome elements located on plasmids and associated with human dissemination. *PLoS Pathog.* **19**, e1011243 (2023).
13. Kahl, O. & Gray, J. S. The biology of Ixodes ricinus with emphasis on its ecology. *Ticks Tick Borne Dis.* **14**, 102114 (2023).
14. Foster, E., Maes, S. A., Holcomb, K. M. & Eisen, R. J. Prevalence of five human pathogens in host-seeking *Ixodes scapularis* and *Ixodes pacificus* by region, state, and county in the contiguous United States generated through national tick surveillance. *Ticks Tick Borne Dis.* **14**, 102250 (2023).
15. Gray, J., Kahl, O. & Zintl, A. Pathogens transmitted by *Ixodes ricinus*. *Ticks Tick Borne Dis.* **15**, 102402 (2024).
16. Uspensky, I. in *Lyme Disease* 8–16 (SMGroup, 2016).
17. Steinbrink, A., Brugger, K., Margos, G., Kraiczy, P. & Klimpel, S. The evolving story of *Borrelia burgdorferi* sensu lato transmission in Europe. *Parasitol. Res.* **121**, 781–803 (2022).
18. Public Health Agency of Canada. Lyme disease. Surveillance. *Government of Canada* https://www.canada.ca/en/public-health/services/diseases/lyme-disease/surveillance-lyme-disease.html (2024).
19. Arsnoe, I., Tsao, J. I. & Hickling, G. J. Nymphal *Ixodes scapularis* questing behavior explains geographic variation in Lyme borreliosis risk in the eastern United States. *Ticks Tick Borne Dis.* **10**, 553–563 (2019).
20. Ginsberg, H. S. et al. Selective host attachment by *Ixodes scapularis* (Acari: Ixodidae): tick-lizard associations in the southeastern United States. *J. Med. Entomol.* **59**, 267–272 (2022).
21. Kugeler, K. J., Farley, G. M., Forrester, J. D. & Mead, P. S. Geographic distribution and expansion of human Lyme disease, United States. *Emerg. Infect. Dis.* **21**, 1455–1457 (2015).
22. Kugeler, K. J., Schwartz, A. M., Delorey, M. J., Mead, P. S. & Hinckley, A. F. Estimating the frequency of Lyme disease diagnoses, United States, 2010-2018. *Emerg. Infect. Dis.* **27**, 616–619 (2021).
23. Pritt, B. S. et al. Identification of a novel pathogenic *Borrelia* species causing Lyme borreliosis with unusually high spirochaetaemia: a descriptive study. *Lancet Infect. Dis.* **16**, 556–564 (2016).
24. Kingry, L. et al. Targeted metagenomics for clinical detection and discovery of bacterial tick-borne pathogens. *J. Clin. Microbiol.* https://doi.org/10.1128/JCM.00147-20 (2020).
25. Blanchard, L. et al. Comparison of national surveillance systems for Lyme disease in humans in Europe and North America: a policy review. *BMC Public Health* **22**, 1307 (2022).
26. Burn, L. et al. Incidence of Lyme borreliosis in Europe from national surveillance systems (2005-2020). *Vector Borne Zoonotic Dis.* **23**, 156–171 (2023).
27. Vandekerckhove, O., De Buck, E. & Van Wijngaerden, E. Lyme disease in western Europe: an emerging problem? A systematic review. *Acta Clin. Belg.* **76**, 244–252 (2021).
28. Hofhuis, A. et al. Decrease in tick bite consultations and stabilization of early Lyme borreliosis in the Netherlands in 2014 after 15 years of continuous increase. *BMC Public Health* **16**, 425 (2016).
29. Sajanti, E. et al. Lyme borreliosis in Finland, 1995-2014. *Emerg. Infect. Dis.* **23**, 1282–1288 (2017).
30. Geebelen, L. et al. Combining primary care surveillance and a meta-analysis to estimate the incidence of the clinical manifestations of Lyme borreliosis in Belgium, 2015-2017. *Ticks Tick Borne Dis.* **10**, 598–605 (2019).
31. Petrulioniene, A. et al. Epidemiology of Lyme disease in a highly endemic European zone. *Medicina* https://doi.org/10.3390/medicina56030115 (2020).
32. Skufca, J. et al. Incidence of Lyme borreliosis in Germany: exploring observed trends over time using public surveillance data, 2016-2020. *Vector Borne Zoonotic Dis.* **23**, 237–246 (2023).
33. Ermenlieva, N., Tsankova, G. & Todorova, T. T. Epidemiological study of Lyme disease in Bulgaria. *Cent. Eur. J. Public Health* **27**, 235–238 (2019).
34. Nuttens, C. et al. Epidemiology of Lyme borreliosis in France in primary care and hospital settings, 2010-2019. *Vector Borne Zoonotic Dis.* **23**, 221–229 (2023).
35. Cairns, V., Wallenhorst, C., Rietbrock, S. & Martinez, C. Incidence of Lyme disease in the UK: a population-based cohort study. *BMJ Open* **9**, e025916 (2019).
36. Tulloch, J. S. P. et al. The demographics and geographic distribution of laboratory-confirmed Lyme disease cases in England and Wales (2013-2016): an ecological study. *BMJ Open* **9**, e028064 (2019).
37. Dedkov, V. G. et al. The burden of tick-borne diseases in the Altai region of Russia. *Ticks Tick Borne Dis.* **8**, 787–794 (2017).
38. Fang, L. Q. et al. Emerging tick-borne infections in mainland China: an increasing public health threat. *Lancet Infect. Dis.* **15**, 1467–1479 (2015).
39. Stark, J. H. et al. Systematic review and meta-analysis of Lyme disease data and seropositivity for *Borrelia burgdorferi*, China, 2005–2020. *Emerg. Infect. Dis.* **28**, 2389–2397 (2022).
40. Kim, S. Y., Kim, T. K., Kim, T. Y. & Lee, H. I. Geographical distribution of *Borrelia burgdorferi* sensu lato in ticks collected from wild rodents in the Republic of Korea. *Pathogens* https://doi.org/10.3390/pathogens9110866 (2020).
41. Chao, L. L., Chen, Y. J. & Shih, C. M. First detection and molecular identification of *Borrelia garinii* isolated from human skin in Taiwan. *J. Med. Microbiol.* **59**, 254–257 (2010).
42. Wang, J., Masuzawa, T. & Yanagihara, Y. Characterization of *Borrelia garinii* isolated from Lyme disease patients in Hokkaido, Japan, by sequence analysis of OspA and OspB genes. *FEMS Microbiol. Lett.* **154**, 371–375 (1997).
43. Faccini-Martínez, A. A. Call for caution to consider Lyme neuroborreliosis as a frequent neurological disease in Mexico. *Arch. Med. Res.* **50**, 18 (2019).
44. Vinayaraj, E. V. et al. Clinical and laboratory evidence of Lyme disease in North India, 2016-2019. *Travel. Med. Infect. Dis.* **43**, 102134 (2021).
45. Collignon, P. J., Lum, G. D. & Robson, J. M. Does Lyme disease exist in Australia? *Med. J. Aust.* **205**, 413–417 (2016).
46. Labruna, M. B., Faccini-Martinez, A. A., Munoz-Leal, S., Szabo, M. P. J. & Angerami, R. N. Lyme borreliosis in Brazil: a critical review on the Baggio-Yoshinari syndrome (Brazilian Lyme-like disease). *Clin. Microbiol. Rev.* **37**, e0009724 (2024).
47. Medlock, J. M. et al. Driving forces for changes in geographical distribution of *Ixodes ricinus* ticks in Europe. *Parasit. Vectors* **6**, 1 (2013).
48. Earley, A. R., Schiffman, E. K., Wong, K. K., Hinckley, A. F. & Kugeler, K. J. Epidemiologic and tick exposure characteristics among people with reported Lyme disease — Minnesota, 2011-2019. *Zoonoses public. health* **71**, 779–789 (2024).
49. Wilking, H. & Stark, K. Trends in surveillance data of human Lyme borreliosis from six federal states in eastern Germany, 2009-2012. *Ticks Tick Borne Dis.* **5**, 219–224 (2014).

50. Berglund, J. et al. An epidemiologic study of Lyme disease in southern Sweden. *N. Engl. J. Med.* **333**, 1319–1327 (1995).
51. Eisen, L. Seasonal activity patterns of *Ixodes scapularis* and *Ixodes pacificus* in the United States. *Ticks Tick Borne Dis.* **16**, 102433 (2025).
52. Moon, K. A. et al. Risk factors for Lyme disease stage and manifestation using electronic health records. *BMC Infect. Dis.* **21**, 1269 (2021).
53. Enkelmann, J. et al. Incidence of notified Lyme borreliosis in Germany, 2013-2017. *Sci. Rep.* **8**, 14976 (2018).
54. Marques, A., Okpali, G., Liepshutz, K. & Ortega-Villa, A. M. Characteristics and outcome of facial nerve palsy from Lyme neuroborreliosis in the United States. *Ann. Clin. Transl. Neurol.* **9**, 41–49 (2022).
55. Mead, P. et al. Risk factors for tick exposure in suburban settings in the Northeastern United States. *Ticks Tick Borne Dis.* **9**, 319–324 (2018).
56. De Keukeleire, M., Robert, A., Luyasu, V., Kabamba, B. & Vanwambeke, S. O. Seroprevalence of *Borrelia burgdorferi* in Belgian forestry workers and associated risk factors. *Parasit. Vectors* **11**, 277 (2018).
57. Adam-Poupart, A. et al. Occupations at risk of contracting zoonoses of public health significance in Quebec. *Can. Commun. Dis. Rep.* **47**, 47–58 (2021).
58. Connally, N. P. et al. Peridomestic Lyme disease prevention: results of a population-based case-control study. *Am. J. Prev. Med.* **37**, 201–206 (2009).
59. Eisen, L. Pathogen transmission in relation to duration of attachment by *Ixodes scapularis* ticks. *Ticks Tick Borne Dis.* **9**, 535–542 (2018).
60. Crippa, M., Rais, O. & Gern, L. Investigations on the mode and dynamics of transmission and infectivity of *Borrelia burgdorferi* sensu stricto and *Borrelia afzelii* in Ixodes ricinus ticks. *Vector Borne Zoonotic Dis.* **2**, 3–9 (2002).
61. Kahl, O. et al. Risk of infection with *Borrelia burgdorferi* sensu lato for a host in relation to the duration of nymphal *Ixodes ricinus* feeding and the method of tick removal. *Zentralbl Bakteriol.* **287**, 41–52 (1998).
62. McQuiston, J. H., Childs, J. E., Chamberland, M. E. & Tabor, E. Transmission of tick-borne agents of disease by blood transfusion: a review of known and potential risks in the United States. *Transfusion* **40**, 274–284 (2000).
63. Woodrum, J. E. & Oliver, J. H. Jr. Investigation of venereal, transplacental, and contact transmission of the Lyme disease spirochete, *Borrelia burgdorferi*, in Syrian hamsters. *J. Parasitol.* **85**, 426–430 (1999).
64. Bockenstedt, L. K., Wooten, R. M. & Baumgarth, N. Immune response to Borrelia: lessons from Lyme disease spirochetes. *Curr. Issues Mol. Biol.* **42**, 145–190 (2021).
65. Lin, Y. P., Diuk-Wasser, M. A., Stevenson, B. & Kraiczy, P. Complement evasion contributes to Lyme borreliae-host associations. *Trends Parasitol.* **36**, 634–645 (2020).
66. Coburn, J., Leong, J. & Chaconas, G. Illuminating the roles of the *Borrelia burgdorferi* adhesins. *Trends Microbiol.* **21**, 372–379 (2013).
67. Norris, S. J. VLS antigenic variation systems of Lyme disease *Borrelia*: eluding host immunity through both random, segmental gene conversion and framework heterogeneity. *Microbiol. Spectr.* https://doi.org/10.1128/microbiolspec.MDNA3-0038-2014 (2014).
68. Troxell, B. et al. Manganese and zinc regulate virulence determinants in *Borrelia burgdorferi*. *Infect. Immun.* **81**, 2743–2752 (2013).
69. Dowdell, A. S. et al. Comprehensive spatial analysis of the *Borrelia burgdorferi* lipoproteome reveals a compartmentalization bias toward the bacterial surface. *J. Bacteriol.* https://doi.org/10.1128/JB.00658-16 (2017).
70. Barbour, A. G. et al. A genome-wide proteome array reveals a limited set of immunogens in natural infections of humans and white-footed mice with *Borrelia burgdorferi*. *Infect. Immun.* **76**, 3374–3389 (2008).
71. Oosting, M. et al. TLR1/TLR2 heterodimers play an important role in the recognition of *Borrelia spirochetes*. *PLoS ONE* **6**, e25998 (2011).
72. Singh, S. K. & Girschick, H. J. Toll-like receptors in *Borrelia burgdorferi*-induced inflammation. *Clin. Microbiol. Infect.* **12**, 705–717 (2006).
73. Cervantes, J. L. et al. Human TLR8 is activated upon recognition of *Borrelia burgdorferi* RNA in the phagosome of human monocytes. *J. Leukoc. Biol.* **94**, 1231–1241 (2013).
74. Petzke, M. M., Brooks, A., Krupna, M. A., Mordue, D. & Schwartz, I. Recognition of *Borrelia burgdorferi*, the Lyme disease spirochete, by TLR7 and TLR9 induces a type I IFN response by human immune cells. *J. Immunol.* **183**, 5279–5292 (2009).
75. Cervantes, J. L. et al. Phagosomal signaling by *Borrelia burgdorferi* in human monocytes involves Toll-like receptor (TLR) 2 and TLR8 cooperativity and TLR8-mediated induction of IFN-β. *Proc. Natl Acad. Sci. USA* **108**, 3683–3688 (2011).
76. Jutras, B. L. et al. *Borrelia burgdorferi* peptidoglycan is a persistent antigen in patients with Lyme arthritis. *Proc. Natl Acad. Sci. USA* **116**, 13498–13507 (2019).
77. Lochhead, R. B., Strle, K., Arvikar, S. L., Weis, J. J. & Steere, A. C. Lyme arthritis: linking infection, inflammation and autoimmunity. *Nat. Rev. Rheumatol.* **17**, 449–461 (2021).
78. Sellati, T. J., Abrescia, L. D., Radolf, J. D. & Furie, M. B. Outer surface lipoproteins of *Borrelia burgdorferi* activate vascular endothelium in vitro. *Infect. Immun.* **64**, 3180–3187 (1996).
79. Wooten, R. M., Modur, V. R., McIntyre, T. M. & Weis, J. J. *Borrelia burgdorferi* outer membrane protein a induces nuclear translocation of nuclear factor-kappa B and inflammatory activation in human endothelial cells. *J. Immunol.* **157**, 4584–4590 (1996).
80. Schramm, F. et al. Microarray analyses of inflammation response of human dermal fibroblasts to different strains of *Borrelia burgdorferi* sensu stricto. *PLoS ONE* **7**, e40046 (2012).
81. Shin, J. J., Glickstein, L. J. & Steere, A. C. High levels of inflammatory chemokines and cytokines in joint fluid and synovial tissue throughout the course of antibiotic-refractory Lyme arthritis. *Arthritis Rheum.* **56**, 1325–1335 (2007).
82. Codolo, G. et al. *Borrelia burgdorferi* NapA-driven Th17 cell inflammation in Lyme arthritis. *Arthritis Rheum.* **58**, 3609–3617 (2008).
83. Oosting, M. et al. Role of interleukin-23 (IL-23) receptor signaling for IL-17 responses in human Lyme disease. *Infect. Immun.* **79**, 4681–4687 (2011).
84. Strle, K. et al. T-Helper 17 cell cytokine responses in Lyme disease correlate with *Borrelia burgdorferi* antibodies during early infection and with autoantibodies late in the illness in patients with antibiotic-refractory Lyme arthritis. *Clin. Infect. Dis.* **64**, 930–938 (2017).
85. Marques, A. et al. Transcriptome assessment of erythema migrans skin lesions in patients with early Lyme disease reveals predominant interferon signaling. *J. Infect. Dis.* **217**, 158–167 (2017).
86. Branda, J. A. & Steere, A. C. Laboratory diagnosis of Lyme borreliosis. *Clin. Microbiol. Rev.* https://doi.org/10.1128/CMR.00018-19 (2021).
87. Blum, L. K. et al. Robust B cell responses predict rapid resolution of Lyme disease. *Front. Immunol.* **9**, 1634 (2018).
88. Hyde, J. A. *Borrelia burgdorferi* keeps moving and carries on: a review of borrelial dissemination and invasion. *Front. Immunol.* **8**, 114 (2017).
89. Rupprecht, T. A., Koedel, U., Fingerle, V. & Pfister, H. W. The pathogenesis of Lyme neuroborreliosis: from infection to inflammation. *Mol. Med.* **14**, 205–212 (2008).
90. Ogrinc, K. et al. Colocalization of radicular pain and erythema migrans in patients with Bannwarth syndrome suggests a direct spread of *Borrelia* into the central nervous system. *Clin. Infect. Dis.* **75**, 81–87 (2022).
91. Gyllemark, P., Sjowall, J., Forsberg, P., Ernerudh, J. & Henningsson, A. J. Intrathecal Th17-driven inflammation is associated with prolonged post-treatment convalescence for patients with Lyme neuroborreliosis. *Sci. Rep.* **13**, 9722 (2023).
92. Ogrinc, K. et al. Unique clinical, immune, and genetic signature in patients with borrelial meningoradiculoneuritis[1]. *Emerg. Infect. Dis.* **28**, 766–776 (2022).
93. Steere, A. C. et al. Treatment of the early manifestations of Lyme disease. *Ann. Intern. Med.* **99**, 22–26 (1983).
94. Ogrinc, K. et al. Acrodermatitis chronica atrophicans: clinical and microbiological characteristics of a cohort of 693 Slovenian patients. *J. Intern. Med.* **290**, 335–348 (2021).
95. Aucott, J. N. et al. CCL19 as a chemokine risk factor for posttreatment Lyme disease syndrome: a prospective clinical cohort study. *Clin. Vaccine Immunol.* **23**, 757–766 (2016).
96. Chandra, A. et al. Anti-neural antibody reactivity in patients with a history of Lyme borreliosis and persistent symptoms. *Brain Behav. Immun.* **24**, 1018–1024 (2010).
97. Bockenstedt, L. K., Gonzalez, D. G., Haberman, A. M. & Belperron, A. A. Spirochete antigens persist near cartilage after murine Lyme borreliosis therapy. *J. Clin. Invest.* **122**, 2652–2660 (2012).
98. Gyllemark, P., Forsberg, P., Ernerudh, J. & Henningsson, A. J. Intrathecal Th17- and B cell-associated cytokine and chemokine responses in relation to clinical outcome in Lyme neuroborreliosis: a large retrospective study. *J. Neuroinflamm.* **14**, 27 (2017).
99. Strle, K., Shin, J. J., Glickstein, L. J. & Steere, A. C. Association of a toll-like receptor 1 polymorphism with heightened Th1 inflammatory responses and antibiotic-refractory Lyme arthritis. *Arthritis Rheum.* **64**, 1497–1507 (2012).
100. Ehrbar, D. et al. Variants in the late cornified envelope gene locus are associated with elevated T-helper 17 responses in patients with postinfectious Lyme arthritis. *J. Infect. Dis.* **230**, S40–S50 (2024).
101. Li, X. et al. Burden and viability of *Borrelia burgdorferi* in skin and joints of patients with erythema migrans or Lyme arthritis. *Arthritis Rheum.* **63**, 2238–2247 (2011).
102. Stupica, D., Lusa, L., Cerar, T., Ruzic-Sabljic, E. & Strle, F. Comparison of post-Lyme borreliosis symptoms in erythema migrans patients with positive and negative *Borrelia burgdorferi* sensu lato skin culture. *Vector Borne Zoonotic Dis.* **11**, 883–889 (2011).
103. Stupica, D., Lusa, L., Ruzic-Sabljic, E., Cerar, T. & Strle, F. Treatment of erythema migrans with doxycycline for 10 days versus 15 days. *Clin. Infect. Dis.* **55**, 343–350 (2012).
104. Berende, A. et al. Randomized trial of longer-term therapy for symptoms attributed to Lyme disease. *N. Engl. J. Med.* **374**, 1209–1220 (2016).
105. Fallon, B. A. et al. A randomized, placebo-controlled trial of repeated IV antibiotic therapy for Lyme encephalopathy. *Neurology* **70**, 992–1003 (2008).
106. Klempner, M. S. et al. Two controlled trials of antibiotic treatment in patients with persistent symptoms and a history of Lyme disease. *N. Engl. J. Med.* **345**, 85–92 (2001).
107. Krupp, L. B. et al. Study and treatment of post Lyme disease (STOP-LD): a randomized double masked clinical trial. *Neurology* **60**, 1923–1930 (2003).
108. Sjowall, J., Ledel, A., Ernerudh, J., Ekerfelt, C. & Forsberg, P. Doxycycline-mediated effects on persistent symptoms and systemic cytokine responses post-neuroborreliosis: a randomized, prospective, cross-over study. *BMC Infect. Dis.* **12**, 186 (2012).
109. Strle, K., Stupica, D., Drouin, E. E., Steere, A. C. & Strle, F. Elevated levels of IL-23 in a subset of patients with post-Lyme disease symptoms following erythema migrans. *Clin. Infect. Dis.* **58**, 372–380 (2014).
110. Fitzgerald, B. L. et al. Metabolic response in patients with post-treatment Lyme disease symptoms/syndrome. *Clin. Infect. Dis.* **73**, e2342–e2349 (2021).
111. Hernandez, S. A. et al. Association of persistent symptoms after Lyme neuroborreliosis and increased levels of interferon-alpha in blood. *Emerg. Infect. Dis.* **29**, 1091–1101 (2023).
112. Jacek, E. et al. Increased IFNα activity and differential antibody response in patients with a history of Lyme disease and persistent cognitive deficits. *J. Neuroimmunol.* **255**, 85–91 (2013).
113. Wormser, G. P., Nadelman, R. B. & Schwartz, I. The amber theory of Lyme arthritis: initial description and clinical implications. *Clin. Rheumatol.* **31**, 989–994 (2012).

114. Crowley, J. T. et al. Matrix metalloproteinase-10 is a target of T and B cell responses that correlate with synovial pathology in patients with antibiotic-refractory Lyme arthritis. *J. Autoimmunity* **69**, 24–37 (2016).
115. Crowley, J. T. et al. A highly expressed human protein, apolipoprotein B-100, serves as an autoantigen in a subgroup of patients with Lyme disease. *J. Infect. Dis.* **212**, 1841–1850 (2015).
116. Drouin, E. E. et al. A novel human autoantigen, endothelial cell growth factor, is a target of T and B cell responses in patients with Lyme disease. *Arthritis Rheum.* **65**, 186–196 (2013).
117. Kanjana, K. et al. Autoimmunity to synovial extracellular matrix proteins in patients with postinfectious Lyme arthritis. *J. Clin. Invest.* https://doi.org/10.1172/JCI161170 (2023).
118. Londono, D. et al. Antibodies to endothelial cell growth factor and obliterative microvascular lesions in the synovium of patients with antibiotic-refractory Lyme arthritis. *Arthritis Rheumatol.* **66**, 2124–2133 (2014).
119. Pianta, A., Drouin, E. E., Arvikar, S., Costello, C. E. & Steere, A. C. Identification of annexin A-2 as an autoantigen in rheumatoid arthritis and in Lyme arthritis. *Arthritis Rheum.* **66**, S437 (2014).
120. Pianta, A. et al. Annexin A2 is a target of autoimmune T and B cell responses associated with synovial fibroblast proliferation in patients with antibiotic-refractory Lyme arthritis. *Clin. Immunol.* **160**, 336–341 (2015).
121. Shen, S. et al. Treg cell numbers and function in patients with antibiotic-refractory or antibiotic-responsive Lyme arthritis. *Arthritis Rheum.* **62**, 2127–2137 (2010).
122. Vudattu, N. K., Strle, K., Steere, A. C. & Drouin, E. E. Dysregulation of $CD4^{+}CD25^{high}$ T cells in the synovial fluid of patients with antibiotic-refractory Lyme arthritis. *Arthritis Rheum.* **65**, 1643–1653 (2013).
123. Ruzic-Sabljic, E. et al. Characterization of *Borrelia burgdorferi* sensu lato strains isolated from human material in Slovenia. *Wien Klin. Wochenschr.* **114**, 544–550 (2002).
124. Strle, F., Ruzic-Sabljic, E., Cimperman, J., Lotric-Furlan, S. & Maraspin, V. Comparison of findings for patients with *Borrelia garinii* and *Borrelia afzelii* isolated from cerebrospinal fluid. *Clin. Infect. Dis.* **43**, 704–710 (2006).
125. Ogrinc, K. et al. Proportion of confirmed Lyme neuroborreliosis cases among adult patients with suspected early European Lyme neuroborreliosis. *Infection* **53**, 1403–1412 (2025).
126. van Dam, A. P. et al. Different genospecies of *Borrelia burgdorferi* are associated with distinct clinical manifestations of Lyme borreliosis. *Clin. Infect. Dis.* **17**, 708–717 (1993).
127. Grillon, A. et al. Characteristics and clinical outcomes after treatment of a national cohort of PCR-positive Lyme arthritis. *Semin. Arthritis Rheum.* **48**, 1105–1112 (2019).
128. Strle, K. et al. *Borrelia burgdorferi* stimulates macrophages to secrete higher levels of cytokines and chemokines than *Borrelia afzelii* or *Borrelia garinii*. *J. Infect. Dis.* **200**, 1936–1943 (2009).
129. Cerar, T. et al. Differences in genotype, clinical features, and inflammatory potential of *Borrelia burgdorferi* sensu stricto strains from Europe and the United States. *Emerg. Infect. Dis.* **22**, 818–827 (2016).
130. Strle, F. et al. Comparison of culture-confirmed erythema migrans caused by *Borrelia burgdorferi* sensu stricto in New York State and by *Borrelia afzelii* in Slovenia. *Ann. Intern. Med.* **130**, 32–36 (1999).
131. Lin, Y. P. et al. Strain-specific variation of the decorin-binding adhesin DbpA influences the tissue tropism of the Lyme disease spirochete. *PLoS Pathog.* **10**, e1004238 (2014).
132. Lin, Y. P. et al. Strain-specific joint invasion and colonization by Lyme disease spirochetes is promoted by outer surface protein C. *PLoS Pathog.* **16**, e1008516 (2020).
133. Seshu, J. et al. Inactivation of the fibronectin-binding adhesin gene bbk32 significantly attenuates the infectivity potential of *Borrelia burgdorferi*. *Mol. Microbiol.* **59**, 1591–1601 (2006).
134. Hyde, J. A. et al. Bioluminescent imaging of *Borrelia burgdorferi* in vivo demonstrates that the fibronectin-binding protein BBK32 is required for optimal infectivity. *Mol. Microbiol.* **82**, 99–113 (2011).
135. Zhi et al. The BBA33 lipoprotein binds collagen and impacts *Borrelia burgdorferi* pathogenesis. *Mol. Microbiol.* **96**, 68–83 (2015).
136. Wager, B., Shaw, D. K., Groshong, A. M., Blevins, J. S. & Skare, J. T. BB0744 affects tissue tropism and spatial distribution of *Borrelia burgdorferi*. *Infect. Immun.* **83**, 3693–3703 (2015).
137. Kraiczy, P., Skerka, C., Kirschfink, M., Zipfel, P. F. & Brade, V. Mechanism of complement resistance of pathogenic *Borrelia burgdorferi* isolates. *Int. Immunopharmacol.* **1**, 393–401 (2001).
138. Sandholm, K. et al. Early cytokine release in response to live *Borrelia burgdorferi* Sensu Lato Spirochetes is largely complement independent. *PLoS ONE* **9**, e108013 (2014).
139. Marcinkiewicz, A. L. et al. Eliminating factor H-binding activity of *Borrelia burgdorferi* CspZ combined with virus-like particle conjugation enhances its efficacy as a Lyme disease vaccine. *Front. Immunol.* **9**, 181 (2018).
140. Strle, K., Jones, K. L., Drouin, E. E., Li, X. & Steere, A. C. *Borrelia burgdorferi* RST1 (OspC type A) genotype is associated with greater inflammation and more severe Lyme disease. *Am. J. Pathol.* **178**, 2726–2739 (2011).
141. Wormser, G. P. et al. *Borrelia burgdorferi* genotype predicts the capacity for hematogenous dissemination during early Lyme disease. *J. Infect. Dis.* **198**, 1358–1364 (2008).
142. Schwartz, I., Margos, G., Casjens, S. R., Qiu, W. G. & Eggers, C. H. Multipartite genome of Lyme disease *Borrelia*: structure, variation and prophages. *Curr. Issues Mol. Biol.* **42**, 409–454 (2021).
143. Akther, S. et al. Natural selection and recombination at host-interacting lipoprotein loci drive genome diversification of Lyme disease and related bacteria. *mBio* **15**, e0174924 (2024).
144. Brandt, M. E., Riley, B. S., Radolf, J. D. & Norgard, M. V. Immunogenic integral membrane proteins of *Borrelia burgdorferi* are lipoproteins. *Infect. Immun.* **58**, 983–991 (1990).
145. Zhang, J. R., Hardham, J. M., Barbour, A. G. & Norris, S. J. Antigenic variation in Lyme disease *Borreliae* by promiscuous recombination of VMP-like sequence cassettes. *Cell* **89**, 275–285 (1997).
146. Casjens, S. R. et al. Primordial origin and diversification of plasmids in Lyme disease agent bacteria. *BMC Genomics* **19**, 218 (2018).
147. Margos, G. et al. Lost in plasmids: next generation sequencing and the complex genome of the tick-borne pathogen *Borrelia burgdorferi*. *BMC Genomics* **18**, 422 (2017).
148. Tyler, S. et al. Whole genome sequencing and phylogenetic analysis of strains of the agent of Lyme disease *Borrelia burgdorferi* from Canadian emergence zones. *Sci. Rep.* **8**, 10552 (2018).
149. Steere, A. C., Dwyer, E. & Winchester, R. Association of chronic Lyme arthritis with HLA-DR4 and HLA-DR2 alleles. *N. Engl. J. Med.* **323**, 219–223 (1990).
150. Steere, A. C. et al. Antibiotic-refractory Lyme arthritis is associated with HLA-DR molecules that bind a *Borrelia burgdorferi* peptide. *J. Exp. Med.* **203**, 961–971 (2006).
151. Strausz, S. et al. SCGB1D2 inhibits growth of *Borrelia burgdorferi* and affects susceptibility to Lyme disease. *Nat. Commun.* **15**, 2041 (2024).
152. Vrijmoeth, H. D. et al. Genome-wide analyses in Lyme borreliosis: identification of a genetic variant associated with disease susceptibility and its immunological implications. *BMC Infect. Dis.* **24**, 337 (2024).
153. Bramwell, K. K., Teuscher, C. & Weis, J. J. Forward genetic approaches for elucidation of novel regulators of Lyme arthritis severity. *Front. Cell Infect. Microbiol.* **4**, 76 (2014).
154. Nadelman, R. B. et al. The clinical spectrum of early Lyme borreliosis in patients with culture-confirmed erythema migrans. *Am. J. Med.* **100**, 502–508 (1996).
155. Smith, R. P. et al. Clinical characteristics and treatment outcome of early Lyme disease in patients with microbiologically confirmed erythema migrans. *Ann. Intern. Med.* **136**, 421–428 (2002).
156. Hunt, K. M. et al. Racial differences in the diagnosis of Lyme disease in children. *Clin. Infect. Dis.* **76**, 1129–1131 (2023).
157. Maraspin, V. et al. Are differences in presentation of early Lyme borreliosis in Europe and North America a consequence of a more frequent spirochetemia in American patients? *J. Clin. Med.* https://doi.org/10.3390/jcm10071448 (2021).
158. Maraspin, V. et al. Characteristics of spirochetemic patients with a solitary erythema migrans skin lesion in Europe. *PLoS ONE* **16**, e0250198 (2021).
159. Strle, F. et al. Comparison of erythema migrans caused by *Borrelia burgdorferi* and *Borrelia garinii*. *Vector Borne Zoonotic Dis.* **11**, 1253–1258 (2011).
160. Logar, M. et al. Comparison of erythema migrans caused by *Borrelia afzelii* and *Borrelia garinii*. *Infection* **32**, 15–19 (2004).
161. Steere, A. C. et al. Systemic symptoms without erythema migrans as the presenting picture of early Lyme disease. *Am. J. Med.* **114**, 58–62 (2003).
162. Feder, H. M., Gerber, M. A., Krause, P. J., Ryan, R. & Shapiro, E. D. Early Lyme-disease — a flu-like illness without erythema migrans. *Pediatrics* **91**, 456–459 (1993).
163. Maraspin, V. & Strle, F. Borrelial lymphocytoma. *Wien Klin. Wochenschr.* https://doi.org/10.1007/s00508-022-02064-5 (2022).
164. Brestrich, G. et al. Estimation of medically-attended LB incidence in Norway, Finland and Poland using data from national surveillance and published literature. *Ticks Tick Borne Dis.* **16**, 102454 (2025).
165. Ogrinc, K. et al. Course and outcome of early European Lyme neuroborreliosis (Bannwarth syndrome): clinical and laboratory findings. *Clin. Infect. Dis.* **63**, 346–353 (2016).
166. Knudtzen, F. C., Andersen, N. S., Jensen, T. G. & Skarphedinsson, S. Characteristics and clinical outcome of Lyme neuroborreliosis in a high endemic area, 1995-2014: a retrospective cohort study in Denmark. *Clin. Infect. Dis.* **65**, 1489–1495 (2017).
167. Nordberg, C. L. et al. Lyme neuroborreliosis in adults: a nationwide prospective cohort study. *Ticks Tick Borne Dis.* **11**, 101411 (2020).
168. Solheim, A. M. et al. Clinical and laboratory characteristics during a 1-year follow-up in European Lyme neuroborreliosis: a prospective cohort study. *Eur. J. Neurol.* **31**, e16487 (2024).
169. Stupica, D. et al. Clinical manifestations and long-term outcome of early Lyme neuroborreliosis according to the European Federation of Neurological Societies diagnostic criteria (definite versus possible) in central Europe. A retrospective cohort study. *Eur. J. Neurol.* **28**, 3155–3166 (2021).
170. Tetens, M. M. et al. Healthcare-seeking behaviour preceding diagnosis of Lyme neuroborreliosis: population-based nationwide matched nested case-control study. *Clin. Microbiol. Infect.* **30**, 1576–1584 (2024).
171. Pachner, A. R. & Steere, A. C. The triad of neurologic manifestations of Lyme disease: meningitis, cranial neuritis, and radiculoneuritis. *Neurology* **35**, 47–53 (1985).
172. Halperin, J. J., Eikeland, R., Branda, J. A. & Dersch, R. Lyme neuroborreliosis: known knowns, known unknowns. *Brain* **145**, 2635–2647 (2022).
173. Bloch, J. et al. Peripheral facial palsy in children: serum *Borrelia* antibodies may reduce the need for lumbar puncture. *Acta Paediatr.* **114**, 122–130 (2025).
174. Sodermark, L., Sigurdsson, V., Nas, W., Wall, P. & Trollfors, B. Neuroborreliosis in Swedish children: a population-based study on incidence and clinical characteristics. *Pediatr. Infect. Dis. J.* **36**, 1052–1056 (2017).
175. Kaminsky, A.-L. et al. Confirmed cases of Neuroborreliosis with involvement of peripheral nervous system. *Medicine* **99**, e21986 (2020).

176. Winter, Y. et al. Cerebral vasculitis as clinical manifestation of neuroborreliosis: pattern of vascular pathology and prognostic factors of outcome. *Int. J. Stroke* **20**, 205–214 (2024).
177. Wittwer, B. et al. Cerebrovascular events in Lyme neuroborreliosis. *J. Stroke Cerebrovasc. Dis.* **24**, 1671–1678 (2015).
178. Bruinsma, R. A., Zomer, T. P., Skogman, B. H., van Hensbroek, M. B. & Hovius, J. W. Clinical manifestations of Lyme neuroborreliosis in children: a review. *Eur. J. Pediatr.* **182**, 1965–1976 (2023).
179. Garro, A. et al. Validation of the Rule of 7's for identifying children at low-risk for Lyme meningitis. *Pediatr. Infect. Dis. J.* **40**, 306–309 (2021).
180. Shen, R. V., McCarthy, C. A. & Smith, R. P. Lyme carditis in hospitalized children and adults, a case series. *Open Forum Infect. Dis.* **8**, ofab140 (2021).
181. Steere, A. C. et al. Lyme carditis: cardiac abnormalities of Lyme disease. *Ann. Intern. Med.* **93**, 8–16 (1980).
182. Tetens, M. M. et al. Risk of heart failure among individuals tested for *Borrelia burgdorferi* sensu lato antibodies, and serum *Borrelia burgdorferi* sensu lato seropositive individuals; a nationwide population-based, registry-based matched cohort study. *Ticks Tick Borne Dis.* **15**, 102345 (2024).
183. Strle, F. et al. Gender disparity between cutaneous and non-cutaneous manifestations of Lyme borreliosis. *PLoS ONE* **8**, e64110 (2013).
184. Vasiliu, V., Herzer, P., Rossler, D., Lehnert, G. & Wilske, B. Heterogeneity of *Borrelia burgdorferi* sensu lato demonstrated by an ospA-type-specific PCR in synovial fluid from patients with Lyme arthritis. *Med. Microbiol. Immunol.* **187**, 97–102 (1998).
185. Jaulhac, B. et al. Direct molecular typing of *Borrelia burgdorferi* sensu lato species in synovial samples from patients with Lyme arthritis. *J. Clin. Microbiol.* **38**, 1895–1900 (2000).
186. Corre, C. et al. Lyme arthritis in Western Europe: a multicentre retrospective study. *Eur. J. Clin. Microbiol. Infect. Dis.* **41**, 21–27 (2022).
187. Steere, A. C. et al. Lyme arthritis: an epidemic of oligoarticular arthritis in children and adults in three Connecticut communities. *Arthritis Rheum.* **20**, 7–17 (1977).
188. Steere, A. C., Schoen, R. T. & Taylor, E. The clinical evolution of Lyme arthritis. *Ann. Intern. Med.* **107**, 725–731 (1987).
189. Arvikar, S. L. & Steere, A. C. Lyme arthritis. *Infect. Dis. Clin. North Am.* **36**, 563–577 (2022).
190. Eckman, E. A., Pacheco-Quinto, J., Herdt, A. R. & Halperin, J. J. Neuroimmunomodulators in neuroborreliosis and Lyme encephalopathy. *Clin. Infect. Dis.* **67**, 80–88 (2018).
191. Halperin, J. J., Little, B. W., Coyle, P. K. & Dattwyler, R. J. Lyme disease: cause of a treatable peripheral neuropathy. *Neurology* **37**, 1700–1706 (1987).
192. Logigian, E. L. & Steere, A. C. Clinical and electrophysiologic findings in chronic neuropathy of Lyme disease. *Neurology* **42**, 303–311 (1992).
193. Wormser, G. P., Strle, F., Shapiro, E. D., Dattwyler, R. J. & Auwaerter, P. G. A critical appraisal of the mild axonal peripheral neuropathy of late neurologic Lyme disease. *Diagn. Microbiol. Infect. Dis.* **87**, 163–167 (2017).
194. Nelder, M. P. et al. Human pathogens associated with the blacklegged tick *Ixodes scapularis*: a systematic review. *Parasit. Vectors* **9**, 265 (2016).
195. Grimaudo, A. T. et al. Geographic variation in risk of blacklegged tick-borne coinfections in the eastern United States. *Ticks Tick Borne Dis.* **17**, 102610 (2026).
196. Taba, P. et al. EAN consensus review on prevention, diagnosis and management of tick-borne encephalitis. *Eur. J. Neurol.* **24**, 1214–e61 (2017).
197. Gomer, A., Lang, A., Janshoff, S., Steinmann, J. & Steinmann, E. Epidemiology and global spread of emerging tick-borne Alongshan virus. *Emerg. Microbes Infect.* **13**, 2404271 (2024).
198. Zhang, M. Z. et al. A series of patients infected with the emerging tick-borne Yezo virus in China: an active surveillance and genomic analysis. *Lancet Infect. Dis.* **25**, 390–398 (2025).
199. Guan, R. et al. Prevalence of three important tick-borne pathogens in ticks and humans in Shaanxi province, northwest China. *Parasitol. Int.* **104**, 102980 (2025).
200. Lv, X. et al. Yezo virus infection in tick-bitten patient and ticks, northeastern China. *Emerg. Infect. Dis.* **29**, 797–800 (2023).
201. Jia, N. et al. Emergence of human infection with Jingmen tick virus in China: a retrospective study. *EBioMedicine* **43**, 317–324 (2019).
202. Wang, Y. C. et al. A new nairo-like virus associated with human febrile illness in China. *Emerg. Microbes Infect.* **10**, 1200–1208 (2021).
203. Ma, J. et al. Identification of a new orthonairovirus associated with human febrile illness in China. *Nat. Med.* **27**, 434–439 (2021).
204. Kodama, F. et al. A novel nairovirus associated with acute febrile illness in Hokkaido, Japan. *Nat. Commun.* **12**, 5539 (2021).
205. Wang, Z. D. et al. A new segmented virus associated with human febrile illness in China. *N. Engl. J. Med.* **380**, 2116–2125 (2019).
206. Lindell, K. et al. Epidemiologic and clinical characteristics and outcomes of patients diagnosed with southern tick associated rash illness (STARI) — 2018-2019. *Diagn. Microbiol. Infect. Dis.* **113**, 116928 (2025).
207. Wormser, G. P. et al. Prospective clinical evaluation of patients from Missouri and New York with erythema migrans-like skin lesions. *Clin. Infect. Dis.* **41**, 958–965 (2005).
208. Wormser, G. P. et al. Microbiologic evaluation of patients from Missouri with erythema migrans. *Clin. Infect. Dis.* **40**, 423–428 (2005).
209. Stromdahl, E. Y. & Hickling, G. J. Beyond Lyme: aetiology of tick-borne human diseases with emphasis on the south-eastern United States. *Zoonoses Public Health* **59** (Suppl. 2), 48–64 (2012).
210. Molaei, G., Little, E. A. H., Williams, S. C. & Stafford, K. C. Bracing for the worst — range expansion of the lone star tick in the northeastern United States. *N. Engl. J. Med.* **381**, 2189–2192 (2019).
211. Molaei, G., Eisen, L. M., Price, K. J. & Eisen, R. J. Range expansion of native and invasive ticks: a looming public health threat. *J. Infect. Dis.* **226**, 370–373 (2022).
212. Raghavan, R. K., Peterson, A. T., Cobos, M. E., Ganta, R. & Foley, D. Current and future distribution of the lone star tick, *Amblyomma americanum* (L.) (Acari: Ixodidae) in North America. *PLoS ONE* **14**, e0209082 (2019).
213. Molins, C. R. et al. Metabolic differentiation of early Lyme disease from southern tick-associated rash illness (STARI). *Sci. Transl. Med.* https://doi.org/10.1126/scitranslmed.aal2717 (2017).
214. Moriyama, Y. et al. Three cases diagnosed not Lyme disease but "tick-associated rash illness (TARI)" in Japan. *J. Infect. Chemother.* **27**, 650–652 (2021).
215. Natsuaki, M., Takada, N., Kawabata, H., Ando, S. & Yamanishi, K. Case of tick-associated rash illness caused by *Amblyomma testudinarium*. *J. Dermatol.* **41**, 834–836 (2014).
216. Stanek, G. et al. Lyme borreliosis: clinical case definitions for diagnosis and management in Europe. *Clin. Microbiol. Infect.* **17**, 69–79 (2011).
217. Mygland, A. et al. EFNS guidelines on the diagnosis and management of European Lyme neuroborreliosis. *Eur. J. Neurol.* **17**, 8–16, e11–14. (2010).
218. Lantos, P. M. et al. Clinical practice guidelines by the Infectious Diseases Society of America (IDSA), American Academy of Neurology (AAN), and American College of Rheumatology (ACR): 2020 guidelines for the prevention, diagnosis and treatment of Lyme disease. *Clin. Infect. Dis.* **72**, 1–8 (2021).
219. Wormser, G. P. et al. The clinical assessment, treatment, and prevention of Lyme disease, human granulocytic anaplasmosis, and babesiosis: clinical practice guidelines by the Infectious Diseases Society of America. *Clin. Infect. Dis.* **43**, 1089–1134 (2006).
220. Leeflang, M. M. et al. The diagnostic accuracy of serological tests for Lyme borreliosis in Europe: a systematic review and meta-analysis. *BMC Infect. Dis.* **16**, 140 (2016).
221. Waddell, L. A. et al. The accuracy of diagnostic tests for Lyme disease in humans, a systematic review and meta-analysis of North American research. *PLoS ONE* **11**, e0168613 (2016).
222. Dessau, R. B. et al. To test or not to test? Laboratory support for the diagnosis of Lyme borreliosis: a position paper of ESGBOR, the ESCMID study group for Lyme borreliosis. *Clin. Microbiol. Infect.* **24**, 118–124 (2018).
223. Wilske, B., Fingerle, V. & Schulte-Spechtel, U. Microbiological and serological diagnosis of Lyme borreliosis. *FEMS Immunol. Med. Microbiol.* **49**, 13–21 (2007).
224. Mead, P., Petersen, J. & Hinckley, A. Updated CDC recommendation for serologic diagnosis of Lyme disease. *MMWR Morb. Mortal. Wkly Rep.* **68**, 703 (2019).
225. Hauser, U., Lehnert, G. & Wilske, B. Validity of interpretation criteria for standardized Western blots (immunoblots) for serodiagnosis of Lyme borreliosis based on sera collected throughout Europe. *J. Clin. Microbiol.* **37**, 2241–2247 (1999).
226. Marques, A. R. Revisiting the Lyme disease serodiagnostic algorithm: the momentum gathers. *J. Clin. Microbiol.* https://doi.org/10.1128/JCM.00749-18 (2018).
227. Hoeve-Bakker, B. J. A., Kerkhof, K., Heron, M., Thijsen, S. F. T. & van Gorkom, T. Evaluation of different standard and modified two-tier testing strategies for the laboratory diagnosis of Lyme borreliosis in a European setting. *Eur. J. Clin. Microbiol. Infect. Dis.* **43**, 2397–2406 (2024).
228. Kobayashi, T. & Auwaerter, P. G. Diagnostic testing for Lyme disease. *Infect. Dis. Clin. North Am.* **36**, 605–620 (2022).
229. Hinckley, A. F. et al. Lyme disease testing by large commercial laboratories in the United States. *Clin. Infect. Dis.* **59**, 676–681 (2014).
230. Lager, M. et al. Serological diagnostics of Lyme borreliosis: comparison of assays in twelve clinical laboratories in Northern Europe. *Eur. J. Clin. Microbiol. Infect. Dis.* **38**, 1933–1945 (2019).
231. Kalish, R. A. et al. Persistence of immunoglobulin M or immunoglobulin G antibody responses to *Borrelia burgdorferi* 10-20 years after active Lyme disease. *Clin. Infect. Dis.* **33**, 780–785 (2001).
232. Coors, A. et al. Regional seropositivity for *Borrelia burgdorferi* and associated risk factors: findings from the Rhineland study, Germany. *Parasit. Vectors* **15**, 241 (2022).
233. Dehnert, M. et al. Seropositivity of Lyme borreliosis and associated risk factors: a population-based study in children and adolescents in Germany (KiGGS). *PLoS ONE* **7**, e41321 (2012).
234. Garro, A. et al. Positive 2-tiered Lyme disease serology is uncommon in asymptomatic children living in endemic areas of the United States. *Pediatr. Infect. Dis. J.* **38**, e105–e107 (2019).
235. Brummitt, S. I. et al. *Borrelia burgdorferi* and *Borrelia miyamotoi* seroprevalence in California blood donors. *PLoS ONE* **15**, e0243950 (2020).
236. Sfeir, M. M. et al. Multicenter clinical evaluation of modified two-tiered testing algorithms for Lyme disease using Zeus Scientific commercial assays. *J. Clin. Microbiol.* **60**, e0252821 (2022).
237. Wormser, G. P. et al. Single-tier testing with the C6 peptide ELISA kit compared with two-tier testing for Lyme disease. *Diagn. Microbiol. Infect. Dis.* **75**, 9–15 (2013).
238. Wilhelmsson, P. et al. A prospective study on the incidence of *Borrelia burgdorferi* sensu lato infection after a tick bite in Sweden and on the Aland Islands, Finland (2008-2009). *Ticks Tick Borne Dis.* **7**, 71–79 (2016).
239. Steere, A. C., Sikand, V. K., Schoen, R. T. & Nowakowski, J. Asymptomatic infection with *Borrelia burgdorferi*. *Clin. Infect. Dis.* **37**, 528–532 (2003).
240. Nadelman, R. B. et al. Prophylaxis with single-dose doxycycline for the prevention of Lyme disease after an *Ixodes scapularis* tick bite. *N. Engl. J. Med.* **345**, 79–84 (2001).

241. Rojko, T., Ruzic-Sabljic, E., Strle, F. & Lotric-Furlan, S. Prevalence and incidence of Lyme borreliosis among Slovene forestry workers during the period of tick activity. *Wien Klin. Wochenschr.* **117**, 219–225 (2005).
242. Hansen, K. & Lebech, A. M. Lyme neuroborreliosis: a new sensitive diagnostic assay for intrathecal synthesis of *Borrelia burgdorferi*-specific immunoglobulin G, A, and M. *Ann. Neurol.* **30**, 197–205 (1991).
243. Reiber, H. & Peter, J. B. Cerebrospinal fluid analysis: disease-related data patterns and evaluation programs. *J. Neurol. Sci.* **184**, 101–122 (2001).
244. Cerar, T., Ogrinc, K., Strle, F. & Ruzic-Sabljic, E. Humoral immune responses in patients with Lyme neuroborreliosis. *Clin. Vaccine Immunol.* **17**, 645–650 (2010).
245. Tetens, M. M. et al. The diagnostic value of serum *Borrelia burgdorferi* antibodies and seroconversion after Lyme neuroborreliosis, a nationwide observational study. *Clin. Microbiol. Infect.* **28**, 1500.e1–1500.e6 (2022).
246. Orfanos, I., Chytilova, V. & Eklund, E. A. Clinical characteristics of children with Borrelia-related peripheral facial palsy and utility of cerebrospinal fluid testing. *Acta Paediatr.* **113**, 1630–1636 (2024).
247. Zomer, T. P. et al. Lyme neuroborreliosis with antibodies in cerebrospinal fluid but not in serum. *Eur. J. Neurol.* **30**, 741–744 (2023).
248. Naudion, P. et al. Positive intrathecal anti-Borrelia antibody synthesis: what are the implications for clinical practice? Clinical features and outcomes of 138 patients in a French multicenter cohort study. *Eur. J. Clin. Microbiol. Infect. Dis.* **42**, 441–452 (2023).
249. Hammers-Berggren, S., Hansen, K., Lebech, A. M. & Karlsson, M. *Borrelia burgdorferi*-specific intrathecal antibody production in neuroborreliosis: a follow-up study. *Neurology* **43**, 169–175 (1993).
250. Krogen, I., Skarphedinsson, S., Jensen, T. G. & Knudtzen, F. C. No correlation between symptom duration and intrathecal production of IgM and/or IgG antibodies in Lyme neuroborreliosis — a retrospective cohort study in Denmark. *J. Infect.* **85**, 507–512 (2022).
251. Steere, A. C., Berardi, V. P., Weeks, K. E., Logigian, E. L. & Ackermann, R. Evaluation of the intrathecal antibody response to *Borrelia burgdorferi* as a diagnostic test for Lyme neuroborreliosis. *J. Infect. Dis.* **161**, 1203–1209 (1990).
252. Eckman, E. A., Clausen, D. M., Herdt, A. R., Pacheco-Quinto, J. & Halperin, J. J. Specificity and diagnostic utility of cerebrospinal fluid CXCL13 in Lyme neuroborreliosis. *Clin. Infect. Dis.* **72**, 1719–1726 (2021).
253. Theel, E. S., Aguero-Rosenfeld, M. E., Pritt, B., Adem, P. V. & Wormser, G. P. Limitations and confusing aspects of diagnostic testing for neurologic Lyme disease in the United States. *J. Clin. Microbiol.* https://doi.org/10.1128/JCM.01406-18 (2019).
254. Guerin, M. et al. Lyme borreliosis diagnosis: state of the art of improvements and innovations. *BMC Microbiology* **23**, 204 (2023).
255. Rupprecht, T. A. et al. Diagnostic value of cerebrospinal fluid CXCL13 for acute Lyme neuroborreliosis. A systematic review and meta-analysis. *Clin. Microbiol. Infect.* **24**, 1234–1240 (2018).
256. Masouris, I., Klein, M. & Kodel, U. The potential for CXCL13 in CSF as a differential diagnostic tool in central nervous system infection. *Expert Rev. Anti Infect. Ther.* **18**, 875–885 (2020).
257. Eisen, L. Personal protection measures to prevent tick bites in the United States: knowledge gaps, challenges, and opportunities. *Ticks Tick Borne Dis.* **13**, 101944 (2022).
258. Tarsus Pharmaceuticals. Tarsus Announces Positive Topline Results from Carpo, a Phase 2a Proof-of-Concept "Tick-Kill" Trial Evaluating TP-05 (lotilaner) for the Prevention of Lyme Disease. *Tarsus Pharmaceuticals* http://ir.tarsusrx.com/news-releases/news-release-details/tarsus-announces-positive-topline-results-carpo-phase-2a-proof (2024).
259. Harms, M. G. et al. A single dose of doxycycline after an *Ixodes ricinus* tick bite to prevent Lyme borreliosis: an open-label randomized controlled trial. *J. Infect.* **82**, 98–104 (2021).
260. Schwan, T. G., Piesman, J., Golde, W. T., Dolan, M. C. & Rosa, P. A. Induction of an outer surface protein on *Borrelia burgdorferi* during tick feeding. *Proc. Natl Acad. Sci. USA* **92**, 2909–2913 (1995).
261. Gomes-Solecki, M. et al. Protective immunity and new vaccines for Lyme disease. *Clin. Infect. Dis.* **70**, 1768–1773 (2020).
262. de Silva, A. M. et al. *Borrelia burgdorferi* OspA is an arthropod-specific transmission-blocking Lyme disease vaccine. *J. Exp. Med.* **183**, 271–275 (1996).
263. Van Hoecke, C., Lebacq, E., Beran, J. & Parenti, D. Alternative vaccination schedules (0, 1, and 6 months versus 0, 1, and 12 months) for a recombinant OspA Lyme disease vaccine. *Clin. Infect. Dis.* **28**, 1260–1264 (1999).
264. Schoen, R. T. et al. Safety and immunogenicity profile of a recombinant outer-surface protein A Lyme disease vaccine: clinical trial of a 3-dose schedule at 0, 1, and 2 months. *Clin. Ther.* **22**, 315–325 (2000).
265. Wormser, G. P. A brief history of OspA vaccines including their impact on diagnostic testing for Lyme disease. *Diagn. Microbiol. Infect. Dis.* **102**, 115572 (2022).
266. Bezay, N. et al. Safety and immunogenicity of a novel multivalent OspA-based vaccine candidate against Lyme borreliosis: a randomised, phase 1 study in healthy adults. *Lancet Infect. Dis.* **23**, 1186–1196 (2023).
267. Bezay, N. et al. Optimisation of dose level and vaccination schedule for the VLA15 Lyme borreliosis vaccine candidate among healthy adults: two randomised, observer-blind, placebo-controlled, multicentre, phase 2 studies. *Lancet Infect. Dis.* **24**, 1045–1058 (2024).
268. Ghadge, S. K. et al. Immunogenicity and safety of an 18-month booster dose of the VLA15 Lyme borreliosis vaccine candidate after primary immunisation in healthy adults in the USA: results of the booster phase of a randomised, controlled, phase 2 trial. *Lancet Infect. Dis.* **24**, 1275–1286 (2024).
269. Wagner, L. et al. Immunogenicity and safety of an 18-month booster dose of the VLA15 Lyme borreliosis vaccine candidate after primary immunisation in children, adolescents, and adults in the USA: a randomised, observer-blind, placebo-controlled, phase 2 trial. *Lancet Infect. Dis.* https://doi.org/10.1016/S1473-3099(25)00541-9 (2025).
270. Wagner, L. et al. Immunogenicity and safety of different immunisation schedules of the VLA15 Lyme borreliosis vaccine candidate in adults, adolescents, and children: a randomised, observer-blind, placebo-controlled, phase 2 trial. *Lancet Infect. Dis.* **25**, 986–999 (2025).
271. Simon, R., Lamberth, E., Stark, J. H. & Skinner, J. M. A human Lyme disease vaccine: two steps forward on the path to prevention. *Expert Rev. Vaccines* **25**, 2607482 (2026).
272. Schiller, Z. A. et al. Blocking *Borrelia burgdorferi* transmission from infected ticks to nonhuman primates with a human monoclonal antibody. *J. Clin. Invest.* https://doi.org/10.1172/JCI144843 (2021).
273. Izac, J. R. & Marconi, R. T. Diversity of the Lyme disease spirochetes and its influence on immune responses to infection and vaccination. *Vet. Clin. North Am. Small Anim. Pract.* **49**, 671–686 (2019).
274. Asbrink, E., Hovmark, A. & Olsson, I. Clinical manifestations of acrodermatitis chronica atrophicans in 50 Swedish patients. *Zentralbl Bakteriol. Mikrobiol. Hyg. A* **263**, 253–261 (1986).
275. Strle, F. & Stanek, G. Clinical manifestations and diagnosis of Lyme borreliosis. *Curr. Probl. Dermatol.* **37**, 51–110 (2009).
276. Hunfeld, K. P., Ruzic-Sabljic, E., Norris, D. E., Kraiczy, P. & Strle, F. In vitro susceptibility testing of *Borrelia burgdorferi* sensu lato isolates cultured from patients with erythema migrans before and after antimicrobial chemotherapy. *Antimicrob. Agents Chemother.* **49**, 1294–1301 (2005).
277. Wormser, G. P. et al. Duration of antibiotic therapy for early Lyme disease. A randomized, double-blind, placebo-controlled trial. *Ann. Intern. Med.* **138**, 697–704 (2003).
278. Cerar, D., Cerar, T., Ruzic-Sabljic, E., Wormser, G. P. & Strle, F. Subjective symptoms after treatment of early Lyme disease. *Am. J. Med.* **123**, 79–86 (2010).
279. Stupica, D. et al. Treatment of erythema migrans with doxycycline for 7 days versus 14 days in Slovenia: a randomised open-label non-inferiority trial. *Lancet Infect. Dis.* **23**, 371–379 (2023).
280. Borg, R. et al. Intravenous ceftriaxone compared with oral doxycycline for the treatment of Lyme neuroborreliosis. *Scand. J. Infect. Dis.* **37**, 449–454 (2005).
281. Ljostad, U. & Henriksen, T. H. Management of neuroborreliosis in European adult patients. *Acta Neurol. Scand. Suppl.* **188**, 22–28 (2008).
282. Rauer, S. et al. Guidelines for diagnosis and treatment in neurology — Lyme neuroborreliosis. *Ger. Med. Sci.* **18**, Doc03 (2020).
283. Arnason, S. & Skogman, B. H. Effectiveness of antibiotic treatment in children with Lyme neuroborreliosis — a retrospective study. *BMC Pediatr.* **22**, 332 (2022).
284. Solheim, A. M. et al. Six versus 2 weeks treatment with doxycycline in European Lyme neuroborreliosis: a multicentre, non-inferiority, double-blinded, randomised and placebo-controlled trial. *J. Neurol. Neurosurg. Psychiatry* **93**, 1222–1228 (2022).
285. Dersch, R. & Rauer, S. Efficacy and safety of pharmacological treatments for Lyme neuroborreliosis: an updated systematic review. *Eur. J. Neurol.* **30**, 3780–3788 (2023).
286. Ljostad, U. et al. Oral doxycycline versus intravenous ceftriaxone for European Lyme neuroborreliosis: a multicentre, non-inferiority, double-blind, randomised trial. *Lancet Neurol.* **7**, 690–695 (2008).
287. Kortela, E. et al. Oral doxycycline compared to intravenous ceftriaxone in the treatment of Lyme neuroborreliosis: a multicenter, equivalence, randomized, open-label trial. *Clin. Infect. Dis.* **72**, 1323–1331 (2021).
288. Arvikar, S. L. & Steere, A. C. Diagnosis and treatment of Lyme arthritis. *Infect. Dis. Clin. North Am.* **29**, 269–280 (2015).
289. Figueroa, R. et al. Confirmation of *Borrelia burgdorferi* spirochetes by polymerase chain reaction in placentas of women with reactive serology for Lyme antibodies. *Gynecol. Obstet. Invest.* **41**, 240–243 (1996).
290. Markowitz, L. E., Steere, A. C., Benach, J. L., Slade, J. D. & Broome, C. V. Lyme disease during pregnancy. *JAMA* **255**, 3394–3396 (1986).
291. Walsh, C. A., Mayer, E. W. & Baxi, L. V. Lyme disease in pregnancy: case report and review of the literature. *Obstet. Gynecol. Surv.* **62**, 41–50 (2007).
292. Maraspin, V. et al. Course and outcome of erythema migrans in pregnant women. *J. Clin. Med.* https://doi.org/10.3390/jcm9082364 (2020).
293. Waddell, L. A., Greig, J., Lindsay, L. R., Hinckley, A. F. & Ogden, N. H. A systematic review on the impact of gestational Lyme disease in humans on the fetus and newborn. *PLoS ONE* **13**, e0207067 (2018).
294. van Samkar, A. et al. Clinical characteristics of Lyme neuroborreliosis in Dutch children and adults. *Eur. J. Pediatr.* **182**, 1183–1189 (2023).
295. Daikh, B. E., Emerson, F. E., Smith, R. P., Lucas, F. L. & McCarthy, C. A. Lyme arthritis: a comparison of presentation, synovial fluid analysis, and treatment course in children and adults. *Arthritis Care Res.* **65**, 1986–1990 (2013).
296. Maraspin, V. et al. Erythema migrans in solid-organ transplant recipients. *Clin. Infect. Dis.* **42**, 1751–1754 (2006).
297. Maraspin, V., Ruzic-Sabljic, E., Lusa, L. & Strle, F. Course and outcome of early Lyme borreliosis in patients with hematological malignancies. *Clin. Infect. Dis.* **61**, 427–431 (2015).
298. Furst, B., Glatz, M., Kerl, H. & Mullegger, R. R. The impact of immunosuppression on erythema migrans. A retrospective study of clinical presentation, response to treatment and production of Borrelia antibodies in 33 patients. *Clin. Exp. Dermatol.* **31**, 509–514 (2006).

299. Forrester, J. D. et al. Notes from the field: update on Lyme carditis, groups at high risk, and frequency of associated sudden cardiac death — United States. *MMWR Morb. Mortal. Wkly Rep.* **63**, 982–983 (2014).
300. Uzomah, U. A. et al. Incidence of carditis and predictors of pacemaker implantation in patients hospitalized with Lyme disease. *PLoS ONE* **16**, e0259123 (2021).
301. Patel, R., Grogg, K. L., Edwards, W. D., Wright, A. J. & Schwenk, N. M. Death from inappropriate therapy for Lyme disease. *Clin. Infect. Dis.* **31**, 1107–1109 (2000).
302. Lantos, P. M. et al. Unorthodox alternative therapies marketed to treat Lyme disease. *Clin. Infect. Dis.* **60**, 1776–1782 (2015).
303. Marzec, N. S. et al. Serious bacterial infections acquired during treatment of patients given a diagnosis of chronic Lyme disease — United States. *MMWR Morb. Mortal. Wkly Rep.* **66**, 607–609 (2017).
304. Strizova, Z., Patek, O., Vitova, L., Horackova, M. & Bartunkova, J. Internet-based self-diagnosis of Lyme disease caused death in a young woman with systemic lupus erythematosus. *Jt Bone Spine* **86**, 650–651 (2019).
305. Obel, N. et al. Long term survival, health, social functioning, and education in patients with European Lyme neuroborreliosis: nationwide population based cohort study. *BMJ* **361**, k1998 (2018).
306. Thortveit, E. T. et al. Subjective health complaints and exposure to tick-borne infections in southern Norway. *Acta Neurol. Scand.* **142**, 260–266 (2020).
307. Bechtold, K. T., Rebman, A. W., Crowder, L. A., Johnson-Greene, D. & Aucott, J. N. Standardized symptom measurement of individuals with early Lyme disease over time. *Arch. Clin. Neuropsychol.* **32**, 129–141 (2017).
308. Eliassen, K. E., Hjetland, R., Reiso, H., Lindbaek, M. & Tschudi-Madsen, H. Symptom load and general function among patients with erythema migrans: a prospective study with a 1-year follow-up after antibiotic treatment in Norwegian general practice. *Scand. J. Prim. Health Care* **35**, 75–83 (2017).
309. Borsic, K., Blagus, R., Cerar, T., Strle, F. & Stupica, D. Clinical course, serologic response, and long-term outcome in elderly patients with early Lyme borreliosis. *J. Clin. Med.* https://doi.org/10.3390/jcm7120506 (2018).
310. Stupica, D. et al. Oral doxycycline versus intravenous ceftriaxone for treatment of multiple erythema migrans: an open-label alternate-treatment observational trial. *J. Antimicrob. Chemother.* **73**, 1352–1358 (2018).
311. Wormser, G. P. et al. Prospective evaluation of the frequency and severity of symptoms in Lyme disease patients with erythema migrans compared with matched controls at baseline, 6 months, and 12 months. *Clin. Infect. Dis.* **71**, 3118–3124 (2020).
312. Ursinus, J. et al. Prevalence of persistent symptoms after treatment for Lyme borreliosis: a prospective observational cohort study. *Lancet Reg. Health Eur.* **6**, 100142 (2021).
313. Aucott, J. N. et al. Risk of post-treatment Lyme disease in patients with ideally-treated early Lyme disease: a prospective cohort study. *Int. J. Infect. Dis.* **116**, 230–237 (2022).
314. Geebelen, L. et al. Non-specific symptoms and post-treatment Lyme disease syndrome in patients with Lyme borreliosis: a prospective cohort study in Belgium (2016-2020). *BMC Infect. Dis.* **22**, 756 (2022).
315. Oksi, J. et al. Duration of antibiotic treatment in disseminated Lyme borreliosis: a double-blind, randomized, placebo-controlled, multicenter clinical study. *Eur. J. Clin. Microbiol. Infect. Dis.* **26**, 571–581 (2007).
316. Eikeland, R., Mygland, A., Herlofson, K. & Ljostad, U. European neuroborreliosis: quality of life 30 months after treatment. *Acta Neurol. Scand.* **124**, 349–354 (2011).
317. Wormser, G. P. et al. Long-term assessment of health-related quality of life in patients with culture-confirmed early Lyme disease. *Clin. Infect. Dis.* **61**, 244–247 (2015).
318. Wills, A. B. et al. Long-term follow-up of patients with Lyme disease: longitudinal analysis of clinical and quality-of-life measures. *Clin. Infect. Dis.* **62**, 1546–1551 (2016).
319. Andreassen, S. et al. Assessment of cognitive function, structural brain changes and fatigue 6 months after treatment of neuroborreliosis. *J. Neurol.* **270**, 1430–1438 (2023).
320. Eikeland, R. et al. Patient-reported outcome after treatment for definite Lyme neuroborreliosis. *Brain Behav.* **10**, e01595 (2020).
321. Vrijmoeth, H. D. et al. Determinants of persistent symptoms after treatment for Lyme borreliosis: a prospective observational cohort study. *EBioMedicine* **98**, 104825 (2023).
322. van Middendorp, H. et al. Expectancies as predictors of symptom improvement after antimicrobial therapy for persistent symptoms attributed to Lyme disease. *Clin. Rheumatol.* **40**, 4295–4308 (2021).
323. Nawrocki, C. C. et al. Nonspecific symptoms attributable to Lyme disease in high-incidence areas, United States, 2017-2021. *Emerg. Infect. Dis.* **31**, 30–37 (2025).
324. Lantos, P. M. & Wormser, G. P. Chronic coinfections in patients diagnosed with chronic Lyme disease: a systematic review. *Am. J. Med.* **127**, 1105–1110 (2014).
325. Sigal, L. H. Summary of the first 100 patients seen at a Lyme disease referral center. *Am. J. Med.* **88**, 577–581 (1990).
326. Steere, A. C., Taylor, E., McHugh, G. L. & Logigian, E. L. The overdiagnosis of Lyme disease. *JAMA* **269**, 1812–1816 (1993).
327. Rose, C. D., Fawcett, P. T., Gibney, K. M. & Doughty, R. A. The overdiagnosis of Lyme disease in children residing in an endemic area. *Clin. Pediatrics* **33**, 663–668 (1994).
328. Reid, M. C., Schoen, R. T., Evans, J., Rosenberg, J. C. & Horwitz, R. I. The consequences of overdiagnosis and overtreatment of Lyme disease: an observational study. *Ann. Intern. Med.* **128**, 354–362 (1998).
329. Aucott, J. N., Seifter, A. & Rebman, A. W. Probable late Lyme disease: a variant manifestation of untreated *Borrelia burgdorferi* infection. *BMC Infect. Dis.* **12**, 173 (2012).
330. Kobayashi, T. et al. Misdiagnosis of Lyme disease with unnecessary antimicrobial treatment characterizes patients referred to an academic infectious diseases clinic. *Open Forum Infect. Dis.* https://doi.org/10.1093/ofid/ofz299 (2019).
331. Gynthersen, R. M. M. et al. Classification of patients referred under suspicion of tick-borne diseases, Copenhagen, Denmark. *Ticks Tick Borne Dis.* **12**, 101591 (2021).
332. Kortela, E., Kanerva, M., Kurkela, S., Oksi, J. & Jarvinen, A. Suspicion of Lyme borreliosis in patients referred to an infectious diseases clinic: what did the patients really have? *Clin. Microbiol. Infect.* **27**, 1022–1028 (2021).
333. Kobayashi, T., Higgins, Y., Melia, M. T. & Auwaerter, P. G. Mistaken identity: many diagnoses are frequently misattributed to Lyme disease. *Am. J. Med.* **135**, 503–511 e505 (2022).
334. Cottle, L. E., Mekonnen, E., Beadsworth, M. B., Miller, A. R. & Beeching, N. J. Lyme disease in a British referral clinic. *QJM* **105**, 537–543 (2012).
335. Coumou, J. et al. Ticking the right boxes: classification of patients suspected of Lyme borreliosis at an academic referral center in the Netherlands. *Clin. Microbiol. Infect.* **21**, 368.e11–20 (2015).
336. Bouiller, K., Klopfenstein, T. & Chirouze, C. Consultation for presumed Lyme borreliosis: the need for multidisciplinary management. *Clin. Infect. Dis.* **68**, 1974 (2019).
337. Haddad, E. et al. Holistic approach in patients with presumed Lyme borreliosis leads to less than 10% of confirmation and more than 80% of antibiotic failures. *Clin. Infect. Dis.* **68**, 2060–2066 (2019).
338. Jacquet, C. et al. Multidisciplinary management of patients presenting with Lyme disease suspicion. *Med. Mal. Infect.* **49**, 112–120 (2019).
339. Djukic, M. et al. The diagnostic spectrum in patients with suspected chronic Lyme neuroborreliosis — the experience from one year of a university hospital's Lyme neuroborreliosis outpatients clinic. *Eur. J. Neurol.* **18**, 547–555 (2011).
340. Qureshi, M. Z., New, D., Zulqarni, N. J. & Nachman, S. Overdiagnosis and overtreatment of Lyme disease in children. *Pediatr. Infect. Dis. J.* **21**, 12–14 (2002).
341. van de Schoor, F. R. et al. Evaluation and 1-year follow-up of patients presenting at a Lyme borreliosis expertise centre: a prospective cohort study with validated questionnaires. *Eur. J. Clin. Microbiol. Infect. Dis.* **43**, 937–946 (2024).
342. Raffetin, A. et al. Acceptance of diagnosis and management satisfaction of patients with "suspected Lyme borreliosis" after 12 months in a multidisciplinary reference center: a prospective cohort study. *BMC Infect. Dis.* **23**, 380 (2023).
343. Raffetin, A. et al. Multidisciplinary management of suspected Lyme borreliosis: clinical features of 569 patients, and factors associated with recovery at 3 and 12 months, a prospective cohort study. *Microorganisms* https://doi.org/10.3390/microorganisms10030607 (2022).
344. Conlon, C. P., Wormser, G. P. & Strle, F. in *Oxford Textbook of Medicine* (eds Firth, J., Conlon, C. & Cox, T.) 1182–1187 (Oxford University Press, 2020).
345. ECDC. Tick Species Distribution Maps: Ixodes ricinus/persultatus — known distribution September 2021. *ECDC* https://www.ecdc.europa.eu/en/disease-vectors/surveillance-and-disease-data/tick-maps (2024).
346. Eisen, R. J., Eisen, L. & Beard, C. B. County-scale distribution of *Ixodes scapularis* and *Ixodes pacificus* (Acari: Ixodidae) in the continental United States. *J. Med. Entomol.* **53**, 349–386 (2016).
347. Wang, S. S. et al. Geographical distribution of *Ixodes persulcatus* and associated pathogens: analysis of integrated data from a China field survey and global published data. *One Health* **16**, 100508 (2023).
348. Rar, V. et al. *Ixodes persulcatus*/pavlovskyi natural hybrids in Siberia: occurrence in sympatric areas and infection by a wide range of tick-transmitted agents. *Ticks Tick Borne Dis.* **10**, 101254 (2019).
349. Mead, P. in *Lyme Disease: An Evidence-based Approach* (ed. Halperin, J.) 53–73 (CABI, 2025).
350. Nadelman, R. B. Erythema migrans. *Infect. Dis. Clin. North Am.* **29**, 211–239 (2015).

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Author contributions

Introduction (F.S. and G.P.W.); Epidemiology (P.S.M. and J.I.T.); Mechanisms/pathophysiology (K.S. and J.E.L.); Diagnosis, screening and prevention (A.M. and A.J.H.); Management (F.S. and G.P.W.); Quality of life (R.E.); Outlook (F.S., K.S., A.M. and G.P.W.); overview of the Primer (F.S., K.S., A.M., A.J.H., R.E., J.E.L., J.I.T., P.S.M. and G.P.W.).

Competing interests

F.S. served on the scientific advisory board for Roche on Lyme disease serological diagnostics and on the scientific advisory board for Pfizer on Lyme disease vaccines and served as a research investigator for Pfizer and Roche; he is an unpaid member of the steering committee of the European Society of Clinical Microbiology and Infectious Disease Study Group on Lyme Borreliosis and other tick-borne diseases. K.S. served as a consultant for Roche, bioMérieux and New York State Biodefense Fund for the

development of diagnostic assays in Lyme borreliosis; he is a member of the European Society of Clinical Microbiology and Infectious Disease Study Group on Lyme Borreliosis and other tick-borne diseases. A.M. has a patent (USA 8,926,989) issued and is an unpaid scientific adviser to the Global Lyme Alliance and to the American Lyme Disease Foundation. A.J.H. has a research collaboration agreement with Pfizer for seroprevalence studies of Lyme borreliosis in Sweden but does not receive any personal honoraria; she is an unpaid member of the executive committee of the European Society of Clinical Microbiology and Infectious Diseases Study Group on Lyme Borreliosis and other tick-borne diseases. R.E. has received travel reimbursement and has received money for lectures for Pfizer. She is a member of the scientific committee of the European Society of Clinical Microbiology and Infectious Disease Study Group on Lyme Borreliosis and other tick-borne diseases, and co-chair of the managing group of infectious diseases in the European Academy of Neurology. J.I.T. is an unpaid board member of the nonprofit American Lyme Disease Foundation. She is a member of the Centers for Disease Control and Prevention's Advisory Committee on Immunization Practices Lyme disease vaccines working group. G.P.W. reports receiving a research grant from Biopeptides, Corp. He has been an expert witness in malpractice cases involving Lyme disease and babesiosis and is an unpaid board member of the nonprofit American Lyme Disease Foundation. J.E.L. and P.S.M. declare no competing interests.

Informed consent

The authors affirm that human research participants provided informed consent for publication of the images in Fig. 7 and experience in Box 1.

Additional information

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