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Review

Review of European and American guidelines for the diagnosis of Lyme borreliosis

Revue des recommandations européennes et américaines concernant le diagnostic de la maladie de Lyme

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Abstract

Lyme disease is a tick-borne bacterial disease with polymorphic clinical manifestations (cutaneous, rheumatological, and neurological). In recent years the issue of the diagnosis of this infection has been highly publicized on the Internet and other media in Europe and America. Some patients and physicians may share the perception that the diagnosis of the infection is not reliable in France. We reviewed current European and American guidelines on Lyme disease and performed a methodological evaluation of all guidelines. We retrieved 16 guidelines from seven countries. Our analysis revealed a global consensus regarding diagnosis at each stage of the infection. All guidelines indicate that the diagnosis is currently based on a two-tier serology at all stages of the infection, except for the early localized dermatological presentation known as Erythema migrans. One text of so-called guidelines has discordant recommendations when compared with the other guidelines, possibly explained by its low quality score. Contrary to the intense debate taking place on the Internet and in the European and American media, our analysis shows that the great majority of medical scientific guidelines with a high quality score, agree on the clinical diagnostic methods of Lyme disease.

Résumé

La maladie de Lyme est une maladie bactérienne transmise par les tiques aux présentations cliniques polymorphes (cutanées, rhumatologiques et neurologiques). Ces dernières années, la problématique du diagnostic de cette infection a été largement médiatisée sur Internet ainsi que dans d'autres médias en Europe et aux États-Unis. Certains patients et médecins pensent que le processus diagnostique de l'infection n'est pas fiable en France. Nous avons donc examiné les recommandations européennes et américaines actuelles portant sur la maladie de Lyme et nous avons réalisé une évaluation méthodologique de toutes ces recommandations. Nous avons identifié 16 recommandations issues de sept pays. Notre analyse a mis en évidence un consensus général sur le diagnostic à chaque stade de l'infection. Toutes les recommandations indiquent que le diagnostic repose actuellement sur un diagnostic sérologique en deux temps à tous les stades de l'infection, à l'exception de la manifestation dermatologique

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localisée précoce connue sous le nom d'érythème migrant. Parmi ces 16 directives, une soi-disant recommandation préconise une approche diagnostique différente des autres recommandations, ce qui peut expliquer son faible score qualitatif. Contrairement au débat animé qui a lieu sur Internet et dans les médias européens et américains, notre analyse montre que la majorité des recommandations scientifiques médicales associées à un score qualitatif élevé s'accordent sur les méthodes diagnostiques cliniques de la maladie de Lyme.

Keywords : Guidelines ; Lyme ; *Borrelia*

Mots clés : Recommandations ; Lyme ; *Borrelia*

1. Introduction

Lyme disease is a tick-borne disease, transmitted by hard tick of the *Ixodes* genus (*Ixodes ricinus* in Europe). The infection is caused by spirochetes of the *Borrelia burgdorferi sensu lato* complex, mainly *B. burgdorferi sensu stricto, B. afzelii*, and *B. garinii* [1]. New members of this complex have been added over the years, thanks to the advances of genotyping techniques. It now includes around 20 genomic species [2]. Following the description of the disease in Connecticut [3] and the discovery of its agent in the 1980s [4], the issue of the diagnosis rapidly emerged. Clinicians and microbiologists were confronted with several barriers. First, the infection is polymorphic and thus involves various organs (skin, neurological system, bones, eyes, heart). Many physicians of different medical specialties should therefore be trained to recognize Lyme disease. Second, despite this multisystemic nature, spirochetemia is highly transient after the primary infection, which makes the isolation of *B. burgdorferi sensu lato* from blood almost inexistent [2]. Finally, the culture of bacteria—the gold standard of microbiological diagnosis—requires special media for *B. sensu lato* complex and laboratory expertise [2]. As a consequence, serology—an indirect method—has rapidly emerged as the cornerstone for the diagnosis of Lyme disease in routine practice [2]. The most common option is to perform a two-tier testing using an ELISA as a screening test, followed by an immunoblot. Molecular tools have emerged, but the PCR sensitivity varies depending on the sample tested (blood, skin, synovial fluid, cerebrospinal fluid) [1].

However, in recent years, numerous pieces of information about Lyme disease have emerged on the Internet and other media, mostly as patients' testimonials [5]. Many patients, associations of patients, and some physicians share the perception that the laboratory diagnosis of Lyme disease in France and other European countries is not relevant and that they should be tested abroad (mainly in Germany) to benefit from reliable tests. This phenomenon can result in a mistrust of patients towards the French medical community. Moreover, for the general population and physicians unfamiliar with the subject, this volume of information can be confusing, and it may be difficult to know whom to trust in this debate. Because it is often challenging for clinicians to have an updated overview of scientific papers, guidelines on Lyme disease have been regularly developed to synthetize the existing evidence and translate it into recommendations for clinical practice.

We aimed to provide an overview of the existing guidelines on the diagnosis of Lyme disease in countries where the disease is prevalent. We reviewed and compared the evidence-based guidelines from North America and Europe currently available in the literature on the diagnosis of Lyme disease. We also aimed to carry out an evaluation of the methodological quality of existing guidelines.

2. Materials and methods

We conducted a search on Medline, Google, and Google Scholar in French, English, and German languages using the keywords "Guidelines" and "Lyme disease" and "diagnosis". We analyzed German guidelines with a special interest because patients in France are often convinced that German physicians have a different approach of the disease. Articles published before 2004 were excluded. When two guidelines from the same authors or organizations were found, only the most recent one was included in the analysis. Guidelines only dealing with treatment and not with diagnostic criteria were also excluded from the analysis. Sixteen guidelines were included in the analysis (Table 1).

Six German guidelines were retrieved. Five of them were issued by academic societies and available on the website of the Association of Scientific Medical Societies in Germany [6]. The sixth guideline was issued by an organization named German Borreliosis Society, which is defined as a "transdisciplinary medical association" of physicians and researchers working on Lyme and tick-borne diseases. This society is not officially recognized by the German authorities as an academic society. As for other countries and regions, we retrieved guidelines from France ($n = 1$), the United States ($n = 1$), Canada ($n = 1$), Switzerland ($n = 1$), Belgium ($n = 1$), Poland ($n = 1$), the United Kingdom ($n = 2$), and Europe ($n = 2$).

The evaluation of the methodology used for each guideline was performed using an in-house score adapted from Siering et al. [7] with the following criteria: presence of reference citations in the guidelines (1 point); presence of a description of the methodology for searching evidence (1 point); systematic method for searching evidence (1 point); explicit link between recommendations and evidence (1 point); presence of a system of recommendation gradation (1 point); single or multiple learned societies involved in developing the guidelines (1 point). The total score for each guideline was obtained by the addition of the number of points for each item.

Table 1
Quality score of guidelines.
Score qualitatif des recommandations.

Guidelines	References	Method for searching evidence	Systematic search of evidence	Explicit link between recommendation and evidence	Gradation	Single or multiple organism(s)	Total score
SPILF 2006	Yes	No	No	Yes	Yes	Multiple	4
IDSA 2006 United States	Yes	No	No	Yes	Yes	Single	3
British Infection Association 2011	Yes	Yes	No	Yes	No	Single	3
Swiss Infectious Diseases Society 2006	Yes	No	No	Yes	No	Single	2
Canadian Public Health Laboratory Network 2006	Yes	No	No	Yes	No	Single	2
Committee for infectious diseases and vaccinations of the German academy for pediatrics and adolescent health 2012	Yes	No	No	Yes	No	Single	2
German Borreliosis Society 2010	Yes	No	No	No	No	Single	1
EFNS 2010	Yes	Yes	Yes	Yes	Yes	Multiple	6
Polish Society of epidemiology and infectious diseases 2015	Yes	No	No	No	No	Multiple	2
Belgian Society of Infectious Diseases and Clinical Microbiology 2016	Yes	No	No	Yes	No	Multiple	3
ESGBOR 2017	Yes	Yes	No	Yes	Yes	Multiple	5
NICE guidelines draft 2017	Yes	Yes	Yes	Yes	Yes	Multiple	6
Rheumatology Society and German Association of Children and Adolescent Health 2013	Yes	Yes	Yes	Yes	No	Multiple	5
German Neurology Society 2012	Yes	Yes	Yes	Yes	No	Multiple	5
German Society of Hygiene and Microbiology 2017	Yes	Yes	Yes	Yes	No	Multiple	5
German Dermatology Society 2016	Yes	Yes	Yes	Yes	No	Multiple	5

SPILF: French Infectious Diseases Society; IDSA: Infectious Diseases Society of America; EFNS: European Federation of Neurological Societies; ESGBOR: ESCMID study group for Lyme borreliosis; NICE: British National Institute for health Care and Excellence.

3. Results of the quality analysis of guidelines

Table 1 synthetizes the evaluation of guidelines. The highest quality score was 6 and was obtained by the European Federation of Neurological Societies (EFNS) guidelines and the British National Institute for health Care and Excellence (NICE) guidelines (Table 1). The German Borreliosis Society showed the lowest quality score (score of 1 point).

For each clinical presentation of Lyme disease, we chose to detail the recommendations of the guidelines regarding diagnosis. We specified at the end of each section the consensual recommendations (included in the majority of guidelines) and the discordant points. These items are also summarized in Table 2.

4. Diagnosis of early localized infection: *erythema migrans* (EM)

4.1. Clinical description

Most guidelines describe EM as a cutaneous lesion appearing between a few days and several weeks after the tick bite, at the site of the bite [8–14] (Fig. 1). This is the first sign of localized infection with *B. burgdorferi sensu lato*. It is an erythematous annular rash with a centrifugal extension [8,9,13]. After several days, the center of the lesion tends to brighten with an infiltration of the borders. It can spread for several weeks up to 30 cm of diameter and spontaneously disappear after several months. In case of EM suspicion, the Infectious Diseases Society of America (IDSA) guidelines recommend tracing the borders of the lesion with ink to measure the extension [9]. The Committee for Infectious Diseases and Vaccinations of the German Academy for Pediatrics and Adolescent Health also recommends tracing the borders with a pen to confirm or rule out the extension of the lesion [15]. The guidelines of the German Dermatological Society stress that EM can be atypical: not marginated, infiltrated, centrally vesicular, hemorrhagic, irregular blotches, only visible when heat is applied to the skin [13]. The British Infection Association guidelines mention that EM caused by *B. garinii* may be more erythematous and homogeneous than EM caused by *B. afzelii* [10]. Many guidelines state that less than 24–48 hours for the rash onset, disappearing within a few days without extension, should rule out the diagnosis of EM [8,9,12,13].

4.2. Diagnosis

Early serology is not sensitive enough (40% to 60%) to confirm Lyme diagnosis at the EM stage and the following guidelines do not recommend early sampling: French Infectious Diseases Society (French acronym SPILF) (2006), IDSA guidelines (2006), British Infection Association guidelines (2011), Committee for Infectious Diseases and Vaccinations of the German Academy for Pediatrics and Adolescent Health (2012), Polish Society of Infectious Diseases (2015), Belgian Antibiotic Policy Coordination Committee (BAPCOC) (2016), ESCMID Study Group for Lyme Borreliosis (ESGBOR) (2017), German Dermatological Society and German Society of Hygiene and Microbiology (2017) [8–18].

However, several guidelines recommend a baseline serum sample to allow for the seroconversion diagnosis [11]. The German Dermatological Society also recommends a serological test in case of atypical EM [13]. The Canadian Public Health Laboratory Network Guidelines differentiate two situations: they do not recommend serology for EM with a compatible seasonal occurrence in an established tick area with a compatible history of tick bite [19]. In that case, the diagnosis of EM is clinical. In case of occurrence out of season or in an area without ticks, a two-tier serology should be performed and repeated four weeks after symptom onset and treatment is at the physician's discretion [19].

PCR on a skin biopsy of EM is suggested by some guidelines, as an option and mainly in case of atypical EM. Its sensitivity is around 70%. In case of atypical EM with negative serology, the German Dermatological Society states that patients should be referred to a dermatologist and a biopsy performed for PCR and culture [13].

The German Borreliosis Society guidelines state that serology may be "falsely negative" in case of EM. However, they are the only ones to recommend the one-tier serology, (IgM Ab, IgG Ab enzymatic immunoassay, or IgM blot, IgG blot) and a lymphocyte transformation test for *Borrelia* in case of "early infection with or without EM" [20]. This latter test is not recommended in any other guideline because of a lack of standardization and reproducibility.

Consensual recommendation: no serology in case of EM suspicion (15/16 guidelines).

Discordant recommendation: the German Borreliosis Society recommends (relative indication) a one-tier serology in case of early infection suspicion with or without EM and a lymphocyte transformation test (1/16 guidelines).

5. Diagnosis of early disseminated infection

5.1. Multiple erythema migrans

5.1.1. Clinical description

Multiple EM is rare according to the SPILF guidelines [8]. The IDSA and British Infection Association guidelines state that secondary hematogenous lesions are usually smaller and more irregular in patients presenting with multiple EM than in those with the initial localized EM [9,10]. The Swiss Infectious Diseases Society guidelines specify that multiple EM is rarer in Europe than in the United States [11]. The Belgian guidelines describe multiple EM as "secondary lesions" appearing several days or weeks after the bite. They are frequently associated with systemic symptoms (fever, myalgia, lymphadenitis) and represent 4% to 20% of EM cases [14].

The German Dermatological Society guidelines have a more detailed section about multiple EM than other guidelines [13]. Multiple EM is described as a hematogenous dissemination of *B. burgdorferi sensu lato* noticeable by sharp, marginated, asymptomatic lesions of various sizes [13]. Children can present symmetrical erythema on their face mimicking fifth

Table 2
Global summary of guidelines content.
Résumé du contenu des recommandations.

Guideline Year Country	Erythema migrans	Lymphocytoma	Early neuroborreliosis	Arthritis	Cardiac features	Ocular features	Acrodermatitis chronica atrophicans	Late neuroborreliosis	Other symptoms
SPILF 2006 France	No serology[a]	Two-tier serology[a], biopsy[b]	CSF cell count and protein, serology in CSF and blood (intrathecal synthesis), PCR may be useful but not very sensitive[a]	Two-tier serology, synovial fluid: cell count and/or PCR[a]	Two-tier serology[a]	Two- tier serology[a]	Two-tier serology[a] and biopsy for histology[b]	Serology in CSF and blood (intrathecal synthesis)[a]	No test for Lyme borreliosis[a]
IDSA 2006 United States	No serology[a]	Two-tier serology[a]	CSF cell count and protein, serology in CSF and blood (intrathecal synthesis), PCR may be useful but not very sensitive[a]	Two-tier serology, synovial fluid: cell count and/or PCR[a]	Two-tier serology[a]	[d]	Two-tier serology[a] and biopsy for histology[b]	Serology in CSF and blood (intrathecal synthesis)[a]	No test for Lyme borreliosis[a]
British Infection Association 2011 United Kingdom	No serology[a]	Two-tier serology[a], possible biopsy[b]	CSF cell count and protein, serology in CSF and blood (intrathecal synthesis), PCR may be useful but not very sensitive[a]	Two-tier serology, synovial fluid: cell count and/or PCR[a]	Two-tier serology[a]	[d]	Two-tier serology[a], possible biopsy for histology[b]	Serology in CSF and blood (intrathecal synthesis)[a]	No test for Lyme borreliosis[a]
Swiss Infectious Diseases Society 2006 Switzerland	No serology[a]	Two-tier serology[a], biopsy[b]	CSF cell count and protein, serology in CSF and blood (intrathecal synthesis), PCR may be useful but not very sensitive[a]	Two-tier serology, synovial fluid: cell count and/or PCR[a]	Two-tier serology[a]	Two-tier serology	Two-tier serology[a], possible biopsy for PCR[b]	Serology in CSF and blood (intrathecal synthesis)[a]	No test for Lyme borreliosis[a]
Canadian Public Health Laboratory Network 2006 Canada	No serology[a]	[d]	Blood serology and/or PCR in CSF[b]	Two-tier serology[a] and PCR on synovial fluid[b]	Two-tier serology[a]	[d]	[d]	Blood serology and/or CSF PCR[b]	No test for Lyme borreliosis[a]
Committee for infectious diseases and vaccinations of the German academy for pediatrics and adolescent health 2012 Germany	No serology[a]	False negative not infrequent in serology[c]	CSF cell count, serology in CSF and blood (intrathecal synthesis)[a]	Two-tier serology[a] and PCR on synovial fluid[b]	Two-tier serology[a]	[d]	Two-tier serology[a]	Serology in CSF and blood (intrathecal synthesis)[a]	No test for Lyme borreliosis[a]

Table 2 (*Continued*)

Guideline Year Country	Erythema migrans	Lymphocytoma	Early neuroborreliosis	Arthritis	Cardiac features	Ocular features	Acrodermatitis chronica atrophicans	Late neuroborreliosis	Other symptoms
German Borreliosis Society 2010 Germany	One-tier serology and/or LTT[c]	One-tier serology and/or LTT[c]	CSF: pleocytosis, high CSF protein levels, intrathecal Ig *Borrelia*[a], PCR in CSF, culture in CSF[b]	One-tier serology and/or LTT and/orc Borrelia PCR on biopsy[b]	One-tier serology and/or LTT[c]	One-tier serology and/or LTT and/or *Borrelia* PCR on biopsy[c]	One-tier serology and/or LTT and/orc *Borrelia* PCR on biopsy[b]	One-tier serology and/or LTTc	Chronic polyorganic symptoms: one-tier serology and/or LTT[c]
EFNS 2010 Europe	[d]	[d]	CSF cell count and protein, serology in CSF and blood (intrathecal synthesis), PCR may be useful but not very sensitive, likewise for culture[a]	[d]	[d]	[d]	[d]	CSF cell count and protein, serology in CSF and blood (intrathecal synthesis), PCR may be useful but not very sensitive, likewise for culture[a]	No test for Lyme borreliosis
Polish Society of epidemiology and infectious diseases 2015 Poland	No serologya PCR on a cutaneous biopsy[b]	Two-tier serology[a]	CSF cell count, serology in CSF and blood (intrathecal synthesis), and/or PCR in CSF[a]	Two-tier serology[a]	Two-tier serology[a]	Two-tier serology	Two-tier serologya Biopsy for histology[b]	CSF cell count, serology in CSF and blood (intrathecal synthesis), and/or PCR in CSF[a]	[d]
Belgian Society of Infectious diseases and clinical Microbiology 2016 Belgium	No serology[a]	Two-tier serology[a], possible biopsy[b]	CSF cell count and protein, serology in CSF and blood (intrathecal synthesis), PCR may be useful but not very sensitive[a]	Two-tier serology[a], PCR on synovial fluid possible[b]	Two-tier serology[a]	[d]	Two- tier serology[a] and biopsy for histology[b]	CSF cell count and protein, serology in CSF and blood (intrathecal synthesis)[a]	No test for Lyme borreliosis[a]
ESGBOR 2017 Europe	No serology[a]	Two-tier serology[a]	CSF cell count and protein, serology in CSF and blood (intrathecal synthesis), PCR may be useful but not very sensitive[a]	Two-tier serology[a]	Two-tier serology[a]	Two-tier serology[a]	Two-tier serology[a]	CSF cell count and protein, serology in CSF and blood (intrathecal synthesis), PCR may be useful but not very sensitive[a]	No test for Lyme borreliosis[a]
NICE guidelines draft 2017 UK	No serology[a]	Two-tier serology[a]	Two-tier serology[a] and referral to a specialist[b]	Two-tier serology[a]	Two-tier serology[a]	Two-tier serology[a]	Two-tier serology[a]	Two-tier serology[b]	No test for Lyme borreliosis[a]

Table 2 (*Continued*)

Guideline Year Country	Erythema migrans	Lymphocytoma	Early neuroborreliosis	Arthritis	Cardiac features	Ocular features	Acrodermatitis chronica atrophicans	Late neuroborreliosis	Other symptoms
German Rheumatology Society and German association of children and adolescent health 2013 Germany	d	d	d	Two-tier serology[a]	d	d	d	d	d
German Neurology Society 2012 Germany	d	d	CSF cell count, serology in blood (IgG or IgM), intrathecal synthesis, PCR may be useful but not very sensitive, CXCL13 chemokine should be better evaluated[a]	d	d	d	d	CSF cell count, serology in blood, intrathecal synthesis, PCR very low sensitivity, CXCL13 chemokine should be better evaluated[a]	d
German Society of Hygiene and Microbiology 2017 Germany	No serology[a] (PCR, culture or antibody rise can be helpful)[b]	Two-tier serology[a], biopsy	CSF cell count and intrathecal synthesis, PCR may be useful but not very sensitive, rising antibody titer or presence of oligoclonal band as secondary criteria[a]	Two-tier serology, synovial fluid: cell counta and/or PCR[b]	Two-tier serology with preferentially rising antibody titer, PCR can be useful but low sensitivity[b]	Two-tier serology with preferentially rising antibody titera, PCR can be useful but low sensitivity[b]	Two-tier serology[a], possible biopsy for histology and PCR[b]	Intrathecal synthesis and CSF cell count, oligoclonal band as secondary criteria[a]	d
German Dermatology Society 2016 Germany	No serology for typical erythema migrans; in cases of atypical erythema migrans, consider serology or PCR[a]	Two-tier serology, consider biopsy for atypical Lyme borreliosis (histology, PCR, culture)[b]	d	d	d	d	Two-tier serology[a] and biopsy for histology[b]		d

SPILF: French Infectious Diseases Society; IDSA:Infectious Diseases Society of America; EFNS:European Federation of Neurological Societies; ESGBOR: European Society of Clinical Microbiology and Infectious Diseases Study Group on Lyme Borreliosis; NICE: National Institute for health and Care Excellence; LTT: lymphocyte transformation test; PCR: polymerase chain reaction; CSF: cerebrospinal fluid.

[a] Consensual recommendation (recommended by the majority of guidelines).

[b] Optional recommendation by some guidelines.

[c] Recommendation present in only one of the guidelines.

[d] Not present in the guidelines.

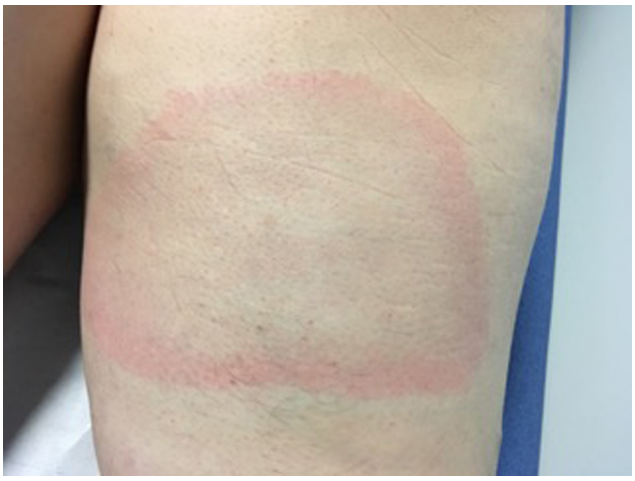

Fig. 1. Erythema migrans.

disease (Parvovirus B19 infection). There is no associated epidermal change, and it can be associated with systemic or acute neurological symptoms [13].

5.1.2. Diagnosis

For most guidelines, multiple EM is not differentiated from typical isolated EM in terms of diagnostic strategy. However, the German Dermatological Society guidelines recommend performing a two-tier serology in case of multiple EM to help the differential diagnosis [13]. If the serology is negative and the clinical suspicion remains high, they recommend performing a biopsy for culture and PCR [13].

Consensual recommendation: same strategy as for isolated EM (12 guidelines).

Discordant recommendation: two-tier serology and if negative and high clinical suspicion, biopsy of the lesion (one guideline: German Dermatological Society).

5.2. Borrelial lymphocytoma

5.2.1. Clinical description

Borrelial lymphocytoma is a very rare but typical manifestation of the early disseminated infection (0.3% to 3% of cases). It is mainly observed in Europe [8,10,11]. The mean time to onset of borrelial lymphocytoma after the tick bite ranges from one to two months [8]. It is more prevalent in children and the preferential localizations are areolar, scrotal, ear lobes, and the helix [8,13,21]. It is a nodular lesion from pink or red to purple [8,9,13]. The histological analysis shows a dermal type B lymphocytic infiltrate which can evoke pseudolymphoma.

5.2.2. Diagnosis

The two-tier serology is recommended by the SPILF, the IDSA, the ESGBOR, the British Infection Association, the NICE, the German Dermatological Society, the Swiss, Belgian, and Polish Society of Infectious Diseases guidelines at this stage, because its sensitivity ranges from 70% to 95% [8–10,12,13,16,17] (better than at the EM stage). However, the Committee for Infectious Diseases and Vaccinations of the German Academy for Pediatrics and Adolescent Health guidelines state that the serology can be negative in lymphocytoma and the diagnosis may be established by clinical means as it is the case for EM [15].

Most guidelines recommend performing a biopsy at this stage for histological analysis and PCR [8–10,13] (Table 2). The Swiss Infectious Diseases Society guidelines recommend a biopsy of the lesion only if there is no improvement after treatment or if there is an atypical localization to rule out cutaneous lymphoma [11]. The Belgian guidelines also suggest a biopsy only in case of atypical lymphocytoma (optional) [14]. The German Dermatological Society stresses that in rare cases, early Borrelial lymphocytoma may be disseminated and that biopsy should be performed to differentiate it from malignant cutaneous lymphomas [13]. The German Borreliosis Society guidelines are the only ones to recommend performing the one-tier serology and a lymphocyte transformation test and do not mention whether or not a biopsy is required [20].

Consensual recommendation: to perform a two-tier serology (10/16).

Discordant recommendation: German Borreliosis Society: to perform a one-tier serology and a lymphocyte transformation test (1/16).

5.3. Neuroborreliosis

5.3.1. Clinical description

The EFNS guidelines provide the most detailed section about neuroborreliosis. This presentation of Lyme disease is more frequent in Europe and is often observed with *B. garinii* infection [22]. Neurological symptoms usually occur 1-12 weeks after the tick bite [8,22]. More than 95% of them can be classified as early Lyme neuroborreliosis (LNB), defined as signs and symptoms lasting for < 6 months after the tick bite [22]. The most common manifestation in Europe is meningoradiculitis, also named Bannwarth's syndrome, [22] with patients experiencing radicular pain and paresis. The pain is usually described as being of a type never experienced before and usually resistant to analgesic treatment [8,22]. The paresis may affect muscles innervated by cranial nerves (especially the facial nerve, less often the abducens or the oculomotor nerves), the abdominal wall, or the limbs. Headaches occur in about 43% of patients, but prominent headaches without radicular pain or paresis is rare in adults [22]. Isolated meningitis is even rarer (5%) [8]. The Committee for Infectious Diseases and Vaccinations of the German Academy for Pediatrics and Adolescent Health specifies that stiffness of the neck is often very mild or absent [15]. These guidelines also state that headaches experienced by patients presenting with neuroborreliosis usually have a clear beginning and are of a short duration [15].

Apart from Bannwarth's syndrome and meningitis, other peripheral neurological presentations are described in 5% to 10% of patients. These presentations are plexus neuritis and mononeuritis multiplex [8,22]. Acute myelitis—reported in less than 5% of neuroborreliosis patients—manifests as paraparesis, sensitive, proprioceptive, and urinary disorders. Encephalitis is, at this

stage, very rare but may be responsible for headaches, confusion, or cognitive focal neurological signs or epileptic seizures [8].

5.3.2. Diagnosis

Cerebrospinal fluid (CSF) examination is the cornerstone of the laboratory diagnosis of LNB [8,22]. A pleocytosis is most frequently observed, with 10 to 1000 leukocytes/mm^3, mainly lymphocytes and elevated protein [22]. According to the EFNS guidelines, a normal cell count or absence of leukocytes in European LNB is rare but possible—especially at the very early stage—in immunosuppressed patients or during LNB caused by *B. afzelii* [22]. Oligoclonal bands and elevated IgG synthesis are commonly reported.

The most important feature is the demonstration of intrathecal production of anti-*B. burgdorferi sensu lato* antibodies (by comparing CSF and serum antibody rates, correcting for blood-brain barrier breakdown). Intrathecal production is the diagnostic gold standard, but has limitations such as low sensitivity at the very early stage of the disease and its persistence for years after eradication of the infection [22]. Almost all guidelines recommend CSF examination (cell count and protein) and search for intrathecal antibody production for the diagnosis of early Lyme neuroborreliosis (Table 2). The ESGBOR guidelines specify that the diagnostic sensitivity of the intrathecal synthesis is about 80% in patients with shorter duration (< 6–8 weeks) of clinical disease and nearly 100% with longer disease duration [16]. The characteristic spectrum of bands, particularly in the IgG immunoblot, also provides evidence to divide the immune response into an early and a late stage. Antibodies against early phase antigens (e.g., VlsE, OspC, p41) are typically compatible with an early presentation (e.g., facial palsy) or a brief latent infection, whereas late phase antigens (e.g., p100, p17/p18) fit well with late presentations (e.g., arthritis, acrodermatitis chronica atrophicans) [16]. Some guidelines point out that the index of antibody production in CSF at the early stage of disease may be negative. Then, a criterion of inflammatory process, such as pleocytosis in CSF, can be useful [17].

Regarding serum antibody detection, most recommendations suggest that, in case of a negative serology in serum and persisting suspicion of neuroborreliosis, antibody detection in serum should be newly performed (2–4 weeks later) to detect a potential seroconversion after a recent infection [9,10,15,16,22]. The NICE guidelines also recommend to repeat the ELISA test and to perform an immunoblot test for patients with a negative ELISA test who have had symptoms for 12 weeks or more and for whom Lyme disease is still suspected [12]. The Canadian guidelines only recommend an enzyme immunoassay (EIA) (with an approved-in-Canada kit and Western immunoblot confirmation) and then recommend to consider polymerase chain reaction of spinal fluid [19]. Of note, Lyme disease incidence is still low or absent in most parts of central Canada and in certain parts of western Canada [19].

As for PCR, the EFNS reminds that the sensitivity in cerebrospinal fluid is around 40% [16]. Therefore, most guidelines specify that PCR should not be used routinely to diagnose LNB, except in complex cases. The Polish guidelines suggest performing a CSF PCR test up to six weeks after the infection during the period where serological tests are still negative or in patients with immunosuppression who cannot have a positive serology [17,22]. Similarly, the culture of *B. burgdorferi sensu lato* is limited to specific indications such as atypical clinical presentations or patients presenting with immune deficiencies [22]. The NICE guidelines recommend a discussion with or referral to an infectious disease specialist in case of a suspicion of LNB [12]. The German Borreliosis Society guidelines recommend a lymphocyte transformation test for "chronic Lyme borreliosis" and the issue of LNB is not detailed.

Consensual recommendation: to perform a cerebrospinal fluid examination (cell count and protein) and to search for intrathecal antibody synthesis (12/14 guidelines).

A CSF PCR test may be useful in some cases (10/14 guidelines).

Discordant recommendation: a PCR test should be performed on all puncture specimens (1/14 guidelines); a lymphocyte transformation test should be performed in case of chronic Lyme disease (1/14 guideline).

5.4. Joint presentations

5.4.1. Clinical description

Lyme arthritis (LA) is a monoarticular or oligoarticular presentation of arthritis that typically involves the knees, usually over a period of several months or years, without prominent systemic presentations [8,9]. Lyme arthritis is the most common feature of disseminated *B. burgdorferi* infection in the United States [9]. In Europe, where Lyme disease is more frequently caused by *B. garinii* and *B. afzelii* than *B. burgdorferi sensu stricto*, LA is observed in only 3-25% of patients [8,17]. The clinical presentations are too unspecific to confirm a purely clinical diagnosis of Lyme arthritis.

5.4.2. Diagnosis

Serological testing is the mainstay of diagnosis. Contrary to early infection, where some patients may be seronegative, patients presenting with LA—a late manifestation—almost always have positive serological results for IgG and low-titer for IgM antibodies to *B. burgdorferi sensu lato* [16]. Thus, most guidelines recommend a serological test and cell count of synovial fluid in the first-line setting. When an articular puncture is performed, the synovial fluid usually shows mild-to-moderate inflammation, and a predominance of granulocytes [9].

A positive PCR test from the synovial fluid increases the diagnostic certainty [16]. The ESGBOR guidelines indicate that the sensitivity and specificity of synovial fluid PCR are 36% and 100%, respectively [16]. The rate of correct positive results by PCR may be increased by synovial biopsy [11]. However, the suspicion of LA is not a sufficient justification for performing a synovial biopsy, and laboratory confirmation of the diagnosis primarily relies on serum antibody determination. Positive PCR results for a joint fluid specimen from a seronegative patient should be interpreted with caution [9]. As a consequence, a synovial fluid PCR test can be occasionally performed for the detection of *B. burgdorferi sensu lato* as a supplementary diagnostic method.

The German Borreliosis Society does not specifically address the diagnosis of LA. However, for chronic Lyme borreliosis (late stage), they recommend serological tests and a lymphocyte transformation test, and suggest performing a PCR culture and immunofluorescence microscopy to search for *B. burgdorferi sensu lato*.

Consensual recommendation: to perform a two-tier serology (12/13 guidelines); a synovial fluid PCR test may be useful (7/13 guidelines).

Discordant recommendation: systematic synovial fluid PCR test (3/13 guidelines), German Borreliosis Society: to perform a one-tier serology and a lymphocyte transformation test (1/13 guidelines).

6. Diagnosis of cardiac and ocular presentations

6.1. Carditis

6.1.1. Clinical description

Lyme carditis is one of the rarer organic presentations of Lyme disease and occurs in 4% to 10% of untreated patients presenting with Lyme disease in the United States [9]. In the absence of concomitant EM (observed in up to 85% of cases), the clinical presentations of Lyme carditis are too nonspecific to confirm a purely clinical diagnosis [9]. Patients presenting with symptomatic cardiac involvement associated with Lyme disease usually present with varying degrees of intermittent atrioventricular heart block, sometimes in association with clinical evidence of myopericarditis [9].

6.1.2. Diagnosis

The diagnosis requires the presence of anti-*B. burgdorferi sensu lato* antibodies in serum. Most patients with cardiac manifestations of Lyme disease are seropositive at the time of presentation [9]. A positive serology alone is not sufficient to diagnose Lyme carditis and must be associated with newly developed auriculo-ventricular conduction disorder, additional history of existing/previous EM or tick bite, and exclusion of other differential diagnoses [14]. To conclude, almost all guidelines only suggested serological tests, e.g., two-tier tests (Table 2). The German Borreliosis Society is the only one to recommend performing a lymphocyte transformation test [20]. A myocarditis biopsy is only recommended optionally in the guidelines of the Swiss Infectious Diseases Society in case of an uncertain diagnosis [11].

Consensual recommendation: to perform a two-tier serology (11/12 guidelines).

Discordant recommendation: German Borreliosis Society: to perform a one-tier serology and a lymphocyte transformation test (1/12 guidelines).

6.2. Ocular presentations

6.2.1. Clinical description

Ocular presentations of Lyme disease include conjunctivitis, episcleritis, keratitis, uveitis, neuroretinitis, retinal vasculitis, and cranial nerve palsies. Even though possible at every stage of the disease, ocular involvement in Lyme disease is most frequently observed at the late stages [8].

6.2.2. Diagnosis

Due to a lack of data and to the rarity of these presentations, few guidelines specify the diagnosis for ocular presentations of Lyme disease [8,16]. ESGBOR suggests detecting serum IgG antibodies to *B. burgdorferi*, even if no positive and negative predictive value can be given in this presentation [16]. Similarly, the SPILF guidelines specify that the serology is usually positive, but the diagnosis is established on a case-by-case basis with the help of a specialist [8]. The German Borreliosis Society recommends performing a lymphocyte transformation test for *Borrelia*.

Consensual recommendation: to perform a two-tier serology (3/4 guidelines).

Discordant recommendation: German Borreliosis Society: to perform a one-tier serology and a lymphocyte transformation test (1/4 guidelines).

7. Diagnosis of late disseminated Lyme borreliosis

7.1. Acrodermatitis chronica atrophicans

7.1.1. Clinical description

The guidelines of the German Dermatological Society offer the most precise definition of acrodermatitis chronica atrophicans (ACA) [13]. It initially manifests as an infiltrative edematous lesion with a pink reticular, then increasingly purple, edematous infiltrated cushion-like erythema, mostly on one extremity without any pain. Then, an atrophic stage of the disease is described as a purple to brown coloring of the skin, with skin atrophy, loss of body hair, connective and fatty tissues, emergence of veins, fibrous nodules adjacent to the joints and joint involvement, often associated with peripheral neuropathy (50% of cases) and hyperesthesia (50%) [13]. It mainly affects women and is very rare in children. It is primarily due to *B. afzelii* and is therefore more common in Europe than in the United States [9,10,13,16].

7.1.2. Diagnosis

All guidelines remind that ACA is a clinical diagnosis first, that must be confirmed by a two-tier serological test with high sensitivity and specificity [8,10,11,13–17,19]. High IgG titer in a screening test combined with a broad-spectrum borrelial-specific bands in IgG immunoblot confirm the diagnosis [8,10,11,13–15,17]. In the guidelines of the German Dermatological Society, these borrelial-specific bands are mentioned as follows: p83/100, p58, p43, p41, p39, p17/18, and Vlse [13]. A negative IgG serology rules out ACA with high certainty in immunocompetent patients [8,10,11,13,14,17]. Of note, the guidelines of the German Borreliosis Society are the only ones to recommend a one-tier serology in immunoblot [20].

In ambiguous cases, skin biopsies should be performed for histopathological analysis, culture, and *Borrelia* PCR [8,10,11,13,14]. A histopathological confirmation is recommended in the first-line setting by the IDSA guidelines and the

guidelines of the German Dermatological Society. The Polish guidelines recommend a *Borrelia* PCR from a skin biopsy to confirm the diagnosis, but they mention that the absence of standardization of the PCR represents an important limitation to the systematic use of the PCR [17].

Consensual recommendation: clinical diagnosis of ACA and a two-tier serological test (14/16 guidelines).

Discordant recommendation: one-tier serology in immunoblot (German Borreliosis Society) (1/16 guidelines).

7.2. Late neuroborreliosis

7.2.1. Clinical description

Late neuroborreliosis can manifest as a chronic encephalomyelitis (spastic syndrome involving the four limbs, spastic-ataxic gait disorder, and disturbed micturition, cranial neuropathy, cognitive impairment, etc.), radiculoneuritis, meningitis, and stroke-like signs (occlusive vasculitis, cerebral infarction) [8–10,14–17,19,22]. A late peripheral neuropathy is also described in association with ACA and presents as a mild, diffuse "stocking glove", with limb paresthesia and sometimes radicular pain [8,9,22]. A mild, late encephalopathy is also described but still controversial [9,16,19,22]. Belgian and NICE guidelines precise that tiredness and isolated pain are not considered as late neuroborreliosis [12,14]. In children, symptoms can include headache, lethargy, irritability, and focal neurological signs [15] but late neuroborreliosis is very rare in this population [15,22].

7.3. Diagnosis

A two-tier serology in blood and CSF is recommended in all guidelines, to demonstrate intrathecal antibody production [8–10,14–17,19,22,23]. A serological test in blood and CSF and an intrathecal antibody production are almost always positive in late neuroborreliosis, and in case of peripheral neuropathy associated with ACA [8–10,14–17,22]. Tests can remain positive for months after a well-conducted treatment. Most guidelines therefore do not recommend these tests to assess healing [8–10,14–17,22].

A lymphocytic pleocytosis in CSF, a moderately elevated level of protein, and a normal glucose level are often observed [8–10,14,15,17,19,22]. Radiological abnormalities in white matter are described in late neuroborreliosis: typical areas of inflammation with increased signal in T2 and FLAIR MRI and enhancement following contrast product administration [9,14,17].

A CSF PCR test is not recommended by the EFNS guidelines at this stage because of poor sensitivity and specificity [9,14,22]. However, it is suggested in Canadian, Polish, and French guidelines in the second-line setting [8,17,19]. The EFNS and Polish guidelines conclude that neuroborreliosis may be confirmed if the following criteria are met: neurological symptoms indicative of neuroborreliosis, pleocytosis in CSF, and intrathecal *B. burgdorferi sensu lato* antibody production [17,22]. Neuroborreliosis is possible if at least two of these following criteria are met: peripheral polyneuritis, ACA, and a positive serological blood test [17,22].

Of note, the guidelines of the German Borreliosis Society are the only ones to recommend a one-tier test, eventually associated with a lymphocyte transformation test in:

- seronegative patients with a strong suspicion of Lyme borreliosis;
- seropositive patients presenting with ambiguous symptoms;
- clinical suspicion of recurrence of Lyme borreliosis;
- suspicion of reinfection [20]. They do not recommend the analysis of CSF in late Lyme neuroborreliosis [20].

Consensual recommendation: intrathecal synthesis or *Borrelia* antibodies (11 guidelines).

Discordant recommendation: lymphocyte transformation test for *Borrelia* (German Borreliosis Society).

8. The Post-Treatment Lyme Disease Syndrome (PTLDS)

The PTLDS is defined in eight guidelines as the persistence of subjective symptoms for six months (fatigue, cognitive complaints, and musculoskeletal pain) beginning within six months after diagnosis and recommended treatment initiation of an objective Lyme borreliosis [4–7,9,10,13,17]. The SPILF guidelines specify that PTLDS is inappropriately named "chronic Lyme disease" [8]. The main exclusion criteria are a proven active infection with *B. burgdorferi sensu lato*, or another ongoing disease that could explain the symptoms [9]. This entity is not present in the German Borreliosis Society guidelines. These guidelines are the only ones to define a "chronic stage" of Lyme disease occurring six months after the start of the infection and composed of a myriad of clinical presentations: fatigue, encephalopathy, muscular and skeletal symptoms, neurological symptoms, gastrointestinal symptoms, urogenital symptoms, ocular symptoms, cutaneous symptoms, and heart diseases [20].

No guidelines recommend a serological test for Lyme borreliosis in case of PTLDS suspicion. The EFNS guidelines mention the following preliminary tests to rule out other diagnoses: physical examination, clinical and laboratory assessment for prior Lyme borreliosis, complete blood count, blood chemistry, antinuclear antibodies, thyroid stimulating hormone, chest X-ray, psychiatric consultation, computed tomography or magnetic resonance imaging if chronic headaches, lumbar puncture if neurological symptoms, imaging and histopathological evaluation if focal signs [22]. If all results of these tests are negative, PTLDS can be evoked.

9. Insufficiently assessed tests

A number of alternative diagnostic tools for Lyme disease have been proposed in recent years, including various PCR systems and antigen detection in urine or blood, lymphocyte transformation tests, numeration of CD57 cells, positive natural killer cells, enzyme-linked immuno-spot assays (ELISPOT),

xenodiagnosis, and commercially available *B. burgdorferi* rapid diagnostic tests (RDT).

However, these methods have been insufficiently evaluated. As a consequence, immunohistochemical detection of *Borrelia* from tissues, lymphocyte transformation tests, detection of specific cytokines (CXCL13) or circulating immune-complex, CD 57 cells, *Borrelia* antigens from patients' samples, and detection of *Borrelia* in samples by light microscopy are not recommended in most guidelines [9,10,13–15]. The German Borreliosis Society guidelines are the only ones to recommend lymphocyte transformation tests in almost all stages of Lyme disease but do not specify any sensitivity or specificity values for this test [20].

10. Conclusion

Our quality analysis of guidelines showed that most national guidelines obtained elevated quality scores, demonstrating their high quality. The lowest score (1 point) was obtained by the German Borreliosis Society guidelines, which is an organization currently not recognized by the German Association of Scientific Medical Societies.

Concerning the contents of the guidelines, our synthesis shows that the recommendations from Europe and North America are quite homogeneous regarding clinical features of the various stages of Lyme diseases and their diagnostic methods (Table 2). The only guidelines with major discordant recommendations for each stage of the disease are the ones of the German Borreliosis Society. Particularly, these guidelines are the only ones to recommend performing lymphocyte transformation tests for *Borrelia* (Table 2), a test that all other guidelines do not recommend because of insufficient evaluation. These guidelines are also the only ones to define a "chronic stage" of Lyme disease.

As a conclusion, our analysis of existing European and American guidelines shows that, contrary to the intense debate that is taking place on the Internet and in the media of European and American countries, most medical scientific guidelines of good quality agree on the clinical presentations and diagnostic methods of Lyme disease. The only guidelines with discordant recommendations are promoted by the German Borreliosis Society, showing a very low level of evidence.

Disclosure of interest

The authors declare that they have no competing interest.

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