Case Report

Successful Diagnosis and Treatment of Borrelia miyamotoi in a Patient With Joint and Muscle Pains, ME/CFS, and Cognitive Dysfunction Following Tick Bites: A Case Report

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Case Report

Successful Diagnosis and Treatment of *Borrelia miyamotoi* in a Patient with Joint and Muscle Pains, ME/CFS, and Cognitive Dysfunction following Tick Bites: A Case Report

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Abstract: Introduction: Diagnosing and managing *Borrelia miyamotoi* disease (BMD) is challenging due to its symptom overlap with Lyme disease (LD) and the lack of reliable laboratory diagnostics for BMD. This case study demonstrates the use of phage-based PCR (phb-PCR) to identify *B. miyamotoi* in a patient who had been bitten by ticks. **Case presentation**: A female patient aged 46 presented with joint and muscle pain, myalgic encephalomyelitis/chronic fatigue syndrome, and cognitive impairment after multiple tick bites in Europe. The patient was assessed for LD at the National Reference Centre in Strasbourg using Enzyme-linked immunosorbent assay (ELISA) and immunoblot. Additional diagnostics involved screening for *Bartonella* and *Anaplasma* antibodies using indirect immunofluorescence, and tests for antinuclear antibodies to check for autoimmune conditions. All serological and immunological tests yielded negative results. However, the phb-PCR identified *B. miyamotoi*. Consequently, the patient received treatment for BMD with a regimen of intravenous ceftriaxone, oral azithromycin, and additional intravenous vitamin and mineral therapy. Post-treatment evaluations showed significant improvements in pain levels, cognitive abilities, and fatigue. **Conclusion**: This case highlights the importance of direct diagnostic methods like phb-PCR for identifying BMD when serological tests fail. Clinicians should consider *B. miyamotoi* testing for accurate and timely management of complex TBDs.

Keywords: *Borrelia miyamotoi*; tick-borne diseases (TBDs); phage-based PCR (phb-PCR); myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS); cognitive dysfunction; case report

Introduction

Diagnosing tick-borne diseases (TBDs) like Lyme disease (LD) and tick-borne relapsing fever (TBRF) is challenging because they share overlapping symptoms, such as fever, chills, headaches, muscle and joint pain, fatigue, arthritis, and neurological manifestations [1]. Although both LD and TBRF diagnoses are difficult, the presence of the erythema migrans (EM) rash in approximately 60-80% of LD patients provides a slight advantage in identifying LD [2,3]. In contrast, the absence of this characteristic rash in TBRF complicates its diagnosis [4].

The geographic distribution of LD-causing *Borrelia* species varies greatly. For instance, *Borrelia burgdorferi* sensu stricto (s.s.) is the most common LD-causing species in the United States, whereas *B. garinii* and *B. afzelii* are predominant in Europe and Asia. Additionally, other *Borrelia* species such as *B. mayonii*, *B. spielmanii*, *B. bissettii*, and *B. valaisiana* are also linked to LD, although they are encountered less frequently [5]. This geographic specificity begs the question of the need for regionally tailored diagnostic and treatment strategies.

The increasing complexity of relapsing fever (RF)-causing *Borrelia* species has significantly complicated their diagnosis and management. *Borrelia* species commonly causing TBRF include *B.*

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coriaceae, *B. lonestari*, *B. duttonii*, *B. crocidurae*, *B. hispanica*, *B. parkeri*, *B. turicatae*, and *B. hermsii*, all of which are transmitted by soft-bodied ticks [6]. In contrast, *B. recurrentis*, which also causes RF, is transmitted by the human body louse [7]. Additionally, *B. miyamotoi*, discovered in Japan in 1995, is an exception among TBRF *Borrelia* species because it is transmitted by hard-bodied ticks, the same vectors responsible for LD [8]. Consequently, a single bite from a hard-bodied tick could potentially transmit both LD and TBRF. This co-transmission of LD and TBRF has overwhelmed diagnostic resources, highlighting the need for improved detection methods and more adaptable testing protocols that are practical for clinical use.

Diagnosing *B. miyamotoi* disease (BMD), a type of TBRF, is challenging because it shares symptoms with LD but lacks the characteristic EM rash [9,10]. Due to these diagnostic challenges, laboratory detection of *B. miyamotoi* is crucial. However, current serological detection methods are unsuitable due to cross-reactivity [3,11]. The CDC (Centers for Disease Control and Prevention) recommends PCR testing to detect *B. miyamotoi*, but bacteria-based PCR methods often have low sensitivity [12,13].

To enhance PCR sensitivity, phage-based PCR (phb-PCR) has emerged as a promising solution for the sensitive detection of bacterial species. Unlike traditional PCR, which targets bacterial DNA directly, phb-PCR detects multiple-copy phage sequences associated with the bacteria, providing higher sensitivity [14,15]. This approach leverages the intrinsic specificity of phages for their bacterial hosts, ensuring that phage detection strongly indicates the presence of the target bacteria [16,17]. Researchers have successfully used phb-PCR to detect tuberculosis, *Borrelia burgdorferi* s.l., and *Borrelia miyamotoi* [15,18,19].

In this case report, we showcased the positive impact of phb-PCR in diagnosing BMD and developing an effective treatment plan. After encountering multiple negative serological tests and struggling to identify the causative agent, we turned to phb-PCR. Recognising the increasing cases of *B. miyamotoi*, we specifically tested for this pathogen [20–22]. Our approach exemplifies the potential of phb-PCR in identifying *B. miyamotoi* infections and guiding appropriate treatment strategies. Testing for *B. miyamotoi* when serological tests yield negative results in complicated tick-borne diseases could be a wise decision when coupled with clinical judgement. Clinicians, scientists, and patient charity workers should collaborate to raise awareness of tick-borne diseases and BMD, ensuring patients receive timely and accurate diagnostics and treatment.

Methods

We explored the complex medical history of a 46-year-old female patient who experienced three distinct tick exposures. The first exposure occurred in July 2006, the second in April 2016, and the third in June 2016. Following these exposures, the patient presented with symptoms including joint and muscle pains, fever, chill, myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS), and cognitive dysfunction ('brain fog'), but did not exhibit the hallmark EM rash of LD.

The various diagnostic tests conducted for the patient are summarised in Table 1. The patient underwent diagnostic evaluation at the National Reference Centre for Lyme Diseases in Strasbourg, France. Initial screening used Enzygnost® *Borrelia* Lyme IgM and Enzygnost® *Borrelia* Lyme link VlsE/IgG Enzyme-linked immunosorbent assay (ELISA) by Siemens to detect the patient's immune response to *B. burgdorferi* s.l. species [23]. Immunoblot testing employed the *Borrelia* Europe plus TpN17 LINE immunoblot by Sekisui Virotech to analyse the detailed immune response to specific *Borrelia* antigens [24].

Subsequent diagnostic efforts included multiple tests to identify the causative agent. Vircell's *Bartonella* indirect immunofluorescence assay (IFA) kits were used to screen for IgG antibodies against *B. henselae* and *B. quintana* [25], while Focus Diagnostics IFA kits were employed to detect IgG and IgM antibodies against *Anaplasma phagocytophilum* [26].

To explore potential autoimmune disorders, an antinuclear antibodies (ANA) test was conducted using the indirect immunofluorescence (IIF) technique on HEp-2 cells [27]. Additionally, specific antibodies against extractable nuclear antigens (ENAs) and other nuclear and cytoplasmic components were tested using ELISA for specific autoantibodies included anti-Ro/SS-A, anti-La/SS-

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B, anti-Sm, anti-RNP, anti-Scl-70, and anti-dsDNA antibodies. Finally, anti-double stranded DNA (anti-dsDNA) were assessed using ELISA for IgG and IgM [28].

A proprietary phb-PCR test designed to detect *B. miyamotoi* was employed [29]. This test, offered by RED Laboratories, was performed in compliance with Good Laboratory Practice (GLP) and conducted according to the MIQE guidelines [30]. The phb-PCR test has been evaluated using analytical, clinical and tick samples, and its accuracy and reliability have been validated and peer-reviewed, with results published in scientific literature [15,19]. The PCR product was sequenced using the Sanger method.

To evaluate the patient's progress post-treatment, a series of detailed follow-up assessments were conducted:

1. **Quality of life measurements**: Quality of life was assessed using the Short Form Health Survey (SF-36) [31]. The SF-36 evaluates multiple dimensions of health, including physical functioning, bodily pain, general health perceptions, vitality, social functioning, emotional role functioning, and mental health.
2. **Symptom severity tracking**: Symptom severity was tracked using the Fatigue Severity Scale (FSS) [32] and the Visual Analogue Scale (VAS) [33] for pain assessment. These scales provide objective data on symptom progression and treatment response, which is essential for effectively managing and adjusting therapeutic strategies.
3. **Cognitive function assessment**: Cognitive function in patients was evaluated using the Montreal Cognitive Assessment (MoCA) [34]. This assessment is crucial for detecting cognitive impairments commonly seen in LD and TBDs and tracking improvements with treatment.
4. **Laboratory testing**: We conducted phb-PCR on the patient's sample after completing the treatment to confirm the absence of *B. miyamotoi*.

Results

1. Diagnostic results:

For clarity and ease of access, detailed diagnostic results and interpretations are summarised in Table 1. Each individual test and its results are explained in the text below.

Table 1. Summary of diagnostic test results for the patient.

Test	Method	Result	Interpretation
B. burgdorferi sensu lato - ELISA	Enzygnost® *Borrelia* Lyme link VlsE/IgG and IgM ELISA	IgG: <4 U/mL, IgM: Negative	No evidence of current or past LD infection
B. burgdorferi sensu lato - Immunoblot	*Borrelia* Europe plus TpN17 LINE by Sekisui Virotech	No reactive bands detected	No evidence of current or past LD infection
B. henselae - IFA	Vircell IFA Kit	IgG: <1:64	Negative, no detectable immune response
B. quintana - IFA	Vircell IFA Kit	IgG: <1:64	Negative, no detectable immune response
A. phagocytophilum - IFA	Focus Diagnostics IFA Kits	IgM: 1:20 (Inconclusive), IgG: <1:64	Inconclusive IgM result, negative IgG result
Antinuclear antibodies (ANA)	Indirect Immunofluorescence (IIF) on HEp-2 cells	Positive at 1/320, homogeneous irregular/speckled	Suggestive of an autoimmune aetiology
Anti-Ro/SS-A antibodies	ELISA	Negative	No evidence of specific autoimmune disease
Anti-La/SS-B antibodies	ELISA	Negative	No evidence of specific autoimmune disease
Anti-Sm antibodies	ELISA	Negative	No evidence of specific autoimmune disease
Anti-RNP antibodies	ELISA	Negative	No evidence of specific autoimmune disease
Anti-Scl-70 antibodies	ELISA	Negative	No evidence of specific autoimmune disease
Anti-dsDNA antibodies	ELISA IgG+IgM	<30 U/mL (Reference: <50 U/mL)	No significant presence of anti-dsDNA antibodies

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phb-PCR	PCR targeting *B. miyamotoi* phage genes	Positive	Confirmed presence of *B. miyamotoi*, validated by sequencing

The patient's diagnostic evaluation for LD yielded negative results. The IgG and IgM ELISA showed IgG levels below the diagnostic threshold (<4 U/mL) and a negative IgM result. Additionally, the immunoblot detected no reactive bands. These findings collectively suggest that the patient has not been exposed to *B. burgdorferi sensu lato* and does not have an active or previous infection.

Subsequent diagnostic efforts using IFA tests revealed no evidence of exposure to *B. henselae* or *B. quintana*, as indicated by negative IgG results (<1:64) for both pathogens. For *A. phagocytophilum*, the IFA tests showed an inconclusive IgM result at 1:20 and a negative IgG result (<1:64). These findings jointly suggest that there is no detectable immune response indicative of infection by *B. henselae*, *B. quintana*, or *A. phagocytophilum* in the patient.

The ANA test for autoimmune markers showed a positive result at a dilution of 1:320 with a homogeneous irregular/speckled pattern. This positive ANA suggests a possible autoimmune aetiology. However, specific autoantibodies, including anti-Ro/SS-A, anti-La/SS-B, anti-Sm, anti-RNP, and anti-Scl-70 (detailed in Table 2), were all negative, indicating no evidence of specific autoimmune diseases commonly associated with these antibodies. Additionally, anti-dsDNA antibodies tested negative, with levels measured at <30 U/mL, which is below the reference value of <50 U/mL.

Table 2. Autoantibodies, their target antigens, associated diseases in autoimmune conditions.

Autoantibody	**Target antigen**	**Associated diseases**
Anti-Ro/SS-A	Ro/SS-A antigens are ribonucleoproteins found in the nucleus and cytoplasm.	Sjögren's syndrome, Systemic lupus erythematosus (SLE)
Anti-La/SS-B	La/SS-B antigens are ribonucleoproteins that interact with nascent RNA transcripts.	Sjögren's syndrome, SLE
Anti-Sm (Anti-Smith)	Sm antigens are nuclear ribonucleoproteins that are major components of the eukaryotic spliceosome.	SLE
Anti-RNP (Anti-Ribonucleoprotein)	U1-RNP antigens are components of small nuclear ribonucleoproteins (snRNPs) involved in pre-mRNA splicing.	Mixed connective tissue disease (MCTD), SLE, systemic sclerosis
Anti-Scl-70 (Anti-Topoisomerase I)	Scl-70 antigen is the topoisomerase I enzyme involved in DNA replication and repair.	Systemic sclerosis, particularly the diffuse cutaneous form of the disease
Anti-dsDNA	Double stranded DNA	SLE

In summary, while the positive ANA result suggests a potential autoimmune condition, the absence of specific autoantibodies does not support the initial ANA finding, thereby excluding certain autoimmune diseases.

Given the negative results from the serological tests, we decided to switch from antibody-based methods to a direct test for detecting *B. miyamotoi*. The phb-PCR assay detected *B. miyamotoi*, and sequencing of the PCR product confirmed its identity. All controls (positive, negative, internal, and extraction) in the PCR functioned correctly.

2. **Therapeutic interventions**:

Following the detection of *B. miyamotoi* from the patient's blood, a targeted treatment regimen was established, focusing on antibiotic therapy against *B. miyamotoi*, complemented by comprehensive nutritional support [35]. The treatment regimen involved:

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Antibiotic Therapy

- Intravenous Ceftriaxone: Administered in 12-day cycles.
- Oral Azithromycin: Administered concurrently with the intravenous ceftriaxone cycles.
- Rest Periods: Each antibiotic cycle was followed by a three-week rest period.

Nutritional Support

During the antibiotic treatment phase, the patient received intravenous nutrient infusions for 18 days. This nutritional support was critical in maintaining the patient's overall health and aiding recovery [36]. The infusions included:

- Vitamin C
- B complex vitamins
- Essential minerals
- Calcium gluconate
- Neurobion (Vitamin B1)
- Spasmag (Magnesium Sulfate)
- Cyanocobalamin

Symptom Improvement

Table 3 illustrates the significant clinical improvements observed with the treatment regimen. The SF-36 scores showed an overall enhancement in quality of life. The patient experienced a marked reduction in fatigue and pain severity scores, along with improved cognitive function and enhanced general well-being. Fatigue and pain scores dropped from high and sever to low and moderate, while cognitive function improved from low to acceptable, indicating substantial recovery. Additionally, the negative phb-PCR result post-treatment confirms the clearance of *B. miyamotoi* infection. This result provides strong microbiological evidence of the treatment's effectiveness.

Table 3. Improvement in symptoms before and after treatment.

Parameter	Pre-treatment	Post-treatment	Interpretation
General health (SF-36)	40%	60%	Significant improvement in overall well-being and physical health.
Energy (SF-36)	25%	70%	Substantial increase in energy levels indicates enhanced physical and mental stamina.
Social (SF-36)	60%	75%	Improved social functioning reflects better social interactions.
Health Change (SF-36)	25%	100%	Improvement in perceived health change shows a positive perception of health progress.
Fatigue (FSS Score)	40	26	Fatigue severity decreased from high to low, demonstrating effective alleviation of fatigue.
Pain (VAS Score)	75 mm	45 mm	Pain severity reduction from severe to moderate demonstrates the efficacy of the treatment.
Cognitive Function (MoCA)	22	26	Cognitive function improved from low to acceptable range.
Phb-PCR	Positive	Negative	The absence of *B. miyamotoi* proves the treatment's effectiveness.

Following the completion of the treatment regimen, the patient underwent regular follow-up assessments to monitor for potential recurrence of the infection and to evaluate overall health and symptomatology. The therapeutic effects have been sustained for eight months, during which the patient has not experienced any relapse of symptoms. After this period, the patient experienced some flare-ups, which were successfully managed with non-antibiotic therapies, including non-corticosteroid anti-inflammatory medications and painkillers.

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Discussion

Given the complexities and challenges associated with diagnosing TBDs, and in light of the guidance provided by the CDC, IDSA (Infectious Diseases Society of America) and ECDC (European Centre for Disease Prevention and Control), our decision to use phb-PCR for diagnosing *B. miyamotoi* was justified by several factors. Initially, the patient underwent standard serological testing for LD and other common co-infections such as *Bartonella* and *A. phagocytophilum*, which all returned negative results. These tests are typically prioritised due to the higher prevalence of LD and its co-infections, as well as the more established diagnostic protocols available for these diseases. Additionally, comprehensive autoimmune screenings were conducted to rule out conditions that could mimic TBD symptoms.

Despite these negative serological results, the clinical presentation of the patient continued to suggest a tick-borne aetiology, warranting further investigation. The CDC and IDSA both acknowledge that *B. miyamotoi* is an emerging pathogen and recommend considering it in differential diagnoses, particularly when standard tests do not yield conclusive results, but clinical suspicion remains high [37–40]. Phb-PCR offers enhanced sensitivity and specificity by targeting the phage genes associated with *B. miyamotoi*, which are part of the pathogen's epigenetic makeup and present in multiple copies within the bacterial cells.

Therefore, the use of phb-PCR was a logical next step to accurately identify *B. miyamotoi*, allowing for the timely initiation of an effective treatment regimen. This approach underscores the importance of utilising advanced diagnostic techniques when standard methods fail to provide definitive answers, ultimately enhancing patient care and outcomes.

After detecting *B. miyamotoi*, we initiated a treatment regimen involving intravenous ceftriaxone and oral azithromycin, supplemented with nutrient infusions. This approach leverages the distinct mechanisms of action of two antibiotics, thereby reducing the risk of antibiotic resistance development. The inclusion of nutritional supplementation was not merely an adjunct but a pivotal component that mitigates the side effects commonly associated with prolonged antibiotic use, such as gastrointestinal disturbances and nutrient depletion. This strategy supports the body’s natural defences and facilitates a more effective recovery.

We recognise the complexities in TBDs. This case report advocates for clinicians to recognise the rising incidence of *B. miyamotoi* in ticks and patients when making differential diagnoses of TBDs. Utilising sensitive direct diagnostic tests, such as phage-based PCR, enhances diagnostic accuracy. This prevents delayed diagnoses and misdiagnoses, reduces patient suffering, and addresses antibiotic resistance issues. This case exemplifies the synergy between laboratory diagnostics and clinical judgment, ultimately improving patient care. By integrating advanced diagnostics with clinical expertise, we can refine the diagnostic algorithm for TBRF and significantly enhance patient outcomes.

Conclusions

This case report highlights the critical importance of direct diagnostic methods for accurately identifying BMD. Diagnosing BMD is challenging due to its symptom overlap with LD and unreliable serological tests. In this case, multiple negative serological tests prompted the use of phb-PCR, which successfully detected *B. miyamotoi* in the patient’s blood. This detection facilitated a tailored treatment plan that included intravenous ceftriaxone, oral azithromycin, and nutritional support, resulting in significant symptom improvement.

The success of phb-PCR in diagnosing BMD underscores its value in managing complex TBDs. Clinicians should consider *B. miyamotoi* testing when serological tests for LD and other common TBDs yield negative results, yet the clinical presentation suggests a tick-borne aetiology. Incorporating BMD testing into diagnostic protocols will ensure timely and accurate treatment.

Author Contributions: Louis Teulières and Jinyu Shan co-conceived the idea for this paper. Louis Teulières handled the clinical aspects of the work and provided the outline for the diagnostic and treatment approaches.

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Jinyu Shan, with the assistance of Ying Jia, was responsible for the meticulous drafting and critical writing of the manuscript with constant conversation with Louis Teulières. Martha Clokie proofread the manuscript.

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Data Availability Statement: The findings of this case report are detailed within the article. Due to privacy and ethical considerations, individual patient data is not publicly accessible. For inquiries about the study's methodologies and anonymized data, please contact the corresponding author, Jinyu Shan, at the provided email address. Data requests will be considered in line with ethical standards and patient confidentiality.

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Conflicts of Interest: The authors declare no conflict of interest.

Ethics Statement: The research conducted and presented in this paper was carried out with strict adherence to ethical principles, respecting the dignity, privacy, and rights of the patient involved. Prior to conducting this study, informed consent was obtained from the patient, ensuring she was fully aware of the research's nature, objectives, potential benefits, and risks. The study was designed and executed with a commitment to maintaining confidentiality and safeguarding the patient's personal and medical information, in accordance with the Declaration of Helsinki and relevant local regulations. The authors have taken meticulous care to ensure that the research methodologies employed were ethically sound, scientifically justified, and conducted under the appropriate oversight. Furthermore, this paper does not contain any data that could lead to the identification of the patient, and all personal information has been anonymised to protect her privacy. The collaboration between the authors from PhelixRD Charity and the University of Leicester was founded on mutual respect, integrity, and a shared goal of advancing medical knowledge for the benefit of patients suffering from tick-borne diseases.

Connections References: *(Please reference all the papers connected to this one, for example a methods paper or supplementary material.).* Lawrence, N., & Brailey, A. (2023). What is the current and expected evolution of prevalence, geographical spread and impact of ticks and tick-borne diseases, and what strategies are needed to improve management, testing, diagnosis, and treatment of these diseases amongst patients and animal populations? *Research Directions: One Health*, 1-4. https://doi.org/10.1017/one.2023.11

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