Test Definition: LYMPV

Lyme Disease, Molecular Detection, PCR, Varies

Overview

Useful For

Supporting the diagnosis of Lyme disease in conjunction with serologic testing

Specific indications including testing skin biopsies when a rash lesion is not characteristic of erythema migrans and testing synovial fluid or synovium to support the diagnosis of Lyme arthritis

This test **should not be used** to screen asymptomatic patients.

Testing Algorithm

The following algorithms are available:
-Acute Tick-borne Disease Testing Algorithm
-Meningitis/Encephalitis Panel Algorithm

Special Instructions

- Acute Tick-borne Disease Testing Algorithm
- Meningitis/Encephalitis Panel Algorithm

Method Name

Real-Time Polymerase Chain Reaction (PCR)/DNA Probe Hybridization

NY State Available

Yes

Specimen

Specimen Type

Varies

Ordering Guidance

This assay does not detect *Borrelia miyamotoi.* If infection with this organism is suspected, order BMIPB / *Borrelia miyamotoi* Detection, PCR, Blood or BMIYC / *Borrelia miyamotoi* Detection, PCR, Spinal Fluid.

Necessary Information

Specimen source is required.

Specimen Required

Submit only 1 of the following specimens:

Specimen Type: Spinal fluid
Container/Tube: Sterile vial
Specimen Volume: 1 mL
Collection Instructions: Label specimen as spinal fluid.

Specimen Type: Synovial fluid
Container/Tube: Sterile vial
Specimen Volume: 1 mL
Collection Instructions: Label specimen as synovial fluid.

Specimen Type: Tissue (fresh only)
Sources: Skin or synovial biopsy
Container/Tube: Sterile container with normal saline
Specimen Volume: Approximately 4 mm(3)
Collection Instructions:
1. Submit only fresh tissue.
2. Skin biopsies:
a. Wash biopsy site with an antiseptic soap. Thoroughly rinse area with sterile water. Do not use alcohol or iodine preparations. A local anesthetic may be used.
b. Biopsy specimens are best taken by punch biopsy to include full thickness of dermis.
3. Label specimen with source of tissue.

Forms

If not ordering electronically, complete, print, and send a Microbiology Test Request (T244) with the specimen.

Specimen Minimum Volume

Spinal fluid: 0.3 mL; Synovial fluid: 0.5 mL; Tissue: See Specimen Required

Reject Due To

All specimens will be evaluated at Mayo Clinic Laboratories for test suitability.

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Refrigerated (preferred)	7 days	
	Frozen	7 days	

Clinical & Interpretive

Clinical Information

Lyme disease is a multisystem and multistage tick-transmitted infection caused by spirochetal bacteria in the *Borrelia burgdorferi* sensu lato (Bbsl) complex.(1) Nearly all human infections are caused by 3 Bbsl species; *B burgdorferi* sensu stricto (hereafter referred to as *B burgdorferi*) is the primary cause of Lyme disease in North America, while *Borrelia*

afzelii and *Borrelia garinii* are the primary causes of Lyme disease in Europe. In 2012, *Borrelia mayonii* was identified as a less common cause of Lyme disease in the upper Midwestern United States.(2,3) This organism has only been detected in patients with exposure to ticks in Minnesota and Wisconsin and has not been detected in over 10,000 specimens from patients in other states, including regions of northeast where Lyme disease is endemic.

Lyme disease is the most commonly reported tick-borne infection in Europe and North America, causing an estimated 300,000 cases in the United States each year and 85,000 cases in Europe.(4,5) The clinical features of Lyme disease are broad and may be confused with various immune and inflammatory disorders. The classic presenting sign of early localized Lyme disease caused by *B burgdorferi* is erythema migrans (EM), which occurs in approximately 80% of individuals. Other early signs and symptoms include malaise, headache, fever, lymphadenopathy, and myalgia. Arthritis, neurological disease, and cardiac disease may be later stage manifestations. EM has also been seen in patients with *B mayonii* infection, but diffuse rashes are more commonly reported.(2) The chronic skin condition, acrodermatitis chronicum atrophicans, is also associated with *B afzelii* infection.

The presence of EM in the appropriate clinical setting is considered diagnostic for Lyme disease; no confirmatory laboratory testing is needed. In the absence of a characteristic EM lesion, serologic testing is the diagnostic method of choice for Lyme disease.(6) However, serology may not be positive until 1 to 2 weeks after onset of symptoms and may show decreased sensitivity for detection of infection with *B mayonii.* Therefore, detection of Bbsl DNA using polymerase chain reaction (PCR) may be a useful adjunct to serologic testing for detection of acute disease. PCR has shown utility for detection of *Borrelia* DNA from skin biopsies of Lyme-associated rashes and can be used to detect *Borrelia* DNA from synovial fluid and synovium biopsies. Less commonly, *Borrelia* DNA can be detected in cerebrospinal fluid.(7) Lyme PCR should always be performed in conjunction with US Food and Drug Administration-approved serologic tests, and the results should be correlated with serologic and epidemiologic data and clinical presentation of the patient.(8) The Mayo Clinic Lyme PCR test detects and differentiates the main causes of Lyme disease in North America (*B burgdorferi* and *B mayonii*) and Europe (*B afzelii* and *B garinii*).(2,7)

Reference Values

Negative
Reference values apply to all ages.

Interpretation

A positive result indicates the presence of DNA from *Borrelia burgdorferi*, *Borrelia mayonii*, *Borrelia afzelii,* or *Borrelia garinii,* the main agents of Lyme disease.

A negative result indicates the absence of detectable target DNA in the specimen. Due to the clinical sensitivity limitations of the polymerase chain reaction assay, a negative result does not preclude the presence of the organism or active Lyme disease.

Cautions

Serologic tests are recommended for diagnosis of Lyme disease. Polymerase chain reaction (PCR) may play an adjunctive role but may not detect *Borrelia burgdorferi* DNA from cerebrospinal fluid (CSF) in cases of active or chronic disease. The presence of inhibitory substances may also cause a false-negative result. If clinical features of illness are highly indicative of Lyme neuroborreliosis, serologic testing on CSF is warranted. PCR test results should be used as an aid in diagnosis and not considered diagnostic by themselves. These results should be correlated with serologic and epidemiologic data and clinical presentation of the patient.

Testing of CSF by PCR in patients with suspected Lyme neuroborreliosis should be requested only on patients with positive *B burgdorferi* antibody in serum confirmed by Western blot assay (LYWB / Lyme Disease Antibody, Immunoblot, Serum) and with abnormal CSF findings (elevated protein and WBC >10 cells/high-power field).

Concurrent infections with multiple tick-borne pathogens, including *Ehrlichia muris eauclairensis, Anaplasma phagocytophilum, Babesia microti,* and *Borrelia miyamotoi* (a relapsing fever *Borrelia*) have been reported in the United States, and consideration should be given to testing for other pathogens, if clinically indicated.

This assay detects most members of the *Borrelia burgdorferi* sensu lato (Bbsl) complex, including *Borrelia andersonii, Borrelia americana,* and *Borrelia bissettii,* which have been rarely detected in humans. Detection of DNA from these organisms would be reported as an atypical result and prompt additional laboratory testing to further identify the DNA present. The sensitivity of this assay for detecting these organisms has not been determined.

This assay also detects some members of the Bbsl complex that are not considered to be human pathogens but may be found in ticks and other animals. Therefore, this assay should not be used to test nonhuman specimens.

Supportive Data

The following validation data supports the use of this assay for clinical testing.

Analytical Sensitivity/Limit of Detection:
The lower limit of detection (LOD) is approximately 300 to 1000 genomic copies/mL in cerebrospinal fluid (CSF), tissue, blood, and synovial fluid.

Accuracy/Diagnostic Sensitivity and Specificity:
Spiking studies of whole organism in fresh tissue, synovial fluid, and CSF (spiked near the approximate LOD) showed 100% recovery.

Analytical Specificity:
No polymerase chain reaction signal was obtained from the extracts of 22 bacterial, viral, parasitic, and fungal isolates that can cause symptoms similar to Lyme disease, including *Rickettsia rickettsii, Rickettsia typhi, Ehrlichia canis, Babesia microti, Plasmodium falciparum, Plasmodium vivax, Bartonella henselae, Bartonella quintana,* herpes simplex virus, and *Toxoplasma gondii.* Relapsing fever borreliae (including *Borrelia miyamotoi*) are also not detected with this assay.

Precision:
Interassay precision was 100%, and intra-assay precision was 100%.

Reference Range:
The reference range for this assay is negative. This assay is only to be used for patients with a clinical history and symptoms consistent with Lyme disease and must be interpreted in the context of serologic tests, which are the gold standard for diagnosis of Lyme disease.

Reportable Range:
This is a qualitative assay, and the results are reported as negative or positive for targeted *Borrelia burgdorferi*, *Borrelia*

afzelii, Borrelia garinii, or *Borrelia mayonii*.

Clinical Reference

1. Stanek G, Wormser GP, Gray J, Strle F. Lyme borreliosis. Lancet. 2012;379(9814):461-473
2. Pritt BS, Mead PS, Johnson DKH, et al. Identification of a novel pathogenic *Borrelia* species causing Lyme borreliosis with unusually high levels of spirochetaemia: a descriptive study. Lancet Infect Dis. 2016;16(5):556-564
3. Pritt BS, Respicio-Kingry LB, Sloan LM, et al. *Borrelia mayonii* sp. nov., a member of the *Borrelia burgdorferi* sensu lato complex, detected in patients and ticks in the upper midwestern United States. Int J Sys Evol Microbiol. 2016;66(11):4878-4880
4. Hinckley AF, Connally NP, Meek JI, et al. Lyme disease testing by large commercial laboratories in the United States. Clin Infect Dis. 2014;59(5):676-681
5. Lindgren E, Jaenson TGT. Lyme borreliosis in Europe: influences of climate and climate change, epidemiology, ecology and adaptation measures. World Health Organization; 2006
6. Centers for Disease Control and Prevention (CDC). Recommendations for test performance and interpretation from the Second National Conference on Serologic Diagnosis of Lyme Disease. MMWR Morb Mortal Wkly Rep. 1995;44(31):590-591
7. Babady NE, Sloan LM, Vetter EA, Patel R, Binnicker MJ. Percent positive rate of Lyme real-time polymerase chain reaction in blood, cerebrospinal fluid, synovial fluid, and tissue. Diagn Microbiol Infect Dis. 2008;62(4):464-466
8. Centers for Disease Control and Prevention (CDC). Lyme disease--United States, 1995. MMWR Morb Mortal Wkly Rep. 1996;45(23):481-484

Performance

Method Description

Nucleic acid is extracted from clinical specimens using the automated MagNA Pure LC instrument system. The extract is then transferred to individual wells of a 96-well plate for amplification. The LightCycler is an automated instrument that amplifies and monitors the development of target nucleic acid (amplicon) after each cycle of polymerase chain reaction (PCR). The DNA target for PCR assay is the 283-base pairs plasminogen-binding protein gene (*OppA2*), which is present at a frequency of 1 copy per organism in all 4 confirmed pathogenic species of the *Borrelia burgdorferi* sensu lato genogroup (*B burgdorferi* sensu stricto, *Borrelia afzelii*, *Borrelia garinii*, and *Borrelia mayonii*). A specific base pair DNA target sequence is amplified by PCR. The detection of amplicon is based on fluorescence resonance energy transfer, which utilizes 1 hybridization probe with a donor fluorophore, fluorescein, at the 3' end, and a second hybridization probe with an acceptor fluorophore, LC-Red 610, at the 5' end. When the target amplicon is present, the LC-Red 610 emits a measurable and quantifiable light signal at a specific wavelength. Presence of the specific organism nucleic acid may be confirmed by performing a melting curve analysis of the amplicon. Using features of the melting curve analysis, the assay primers and specific hybridization probes are able to detect and differentiate *B burgdorferi* sensu stricto from *B mayonii, B afzelii*, and *B garinii,* although the melting curve analysis cannot differentiate between *B afzelii* and *B garinii*. Each assay run can be completed within 60 minutes.(Unpublished Mayo method)

PDF Report

No

Day(s) Performed

Monday through Sunday

Report Available
Same day/1 to 4 days

Specimen Retention Time
1 week

Performing Laboratory Location
Mayo Clinic Laboratories - Rochester Main Campus

Fees & Codes

Fees
- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their account representative. For assistance, contact Customer Service.

Test Classification
This test was developed and its performance characteristics determined by Mayo Clinic in a manner consistent with CLIA requirements. It has not been cleared or approved by the US Food and Drug Administration.

CPT Code Information
87476
87798 x 2
87801 (if appropriate for government payers)

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
LYMPV	Lyme Disease, PCR, Varies	94253-2

Result ID	Test Result Name	Result LOINC® Value
LYMS	Specimen Source	31208-2
618333	B. burgdorferi PCR	94250-8
618334	B. mayonii PCR	94251-6
618335	B. garinii/B. afzelii PCR	94252-4
618336	Lyme CSF Comment	59464-8