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## Overview

### Useful For

Rapid testing for Powassan virus RNA (lineage 1 and lineage 2) using whole blood specimens

An adjunctive test to serology for detection of early Powassan virus infection (ie, first few days after symptom onset)

This assay **should not be used for** screening asymptomatic individuals and should only be used to test patients with signs and symptoms of Powassan virus disease.

### Testing Algorithm

For information see the following:

[-Meningitis/Encephalitis Panel Algorithm](#)

[-Acute Tickborne Disease Testing Algorithm](#)

### Special Instructions

- [Acute Tickborne Disease Testing Algorithm](#)
- [Meningitis/Encephalitis Panel Algorithm](#)

### Method Name

Real-Time Polymerase Chain Reaction (PCR)

### NY State Available

Yes

## Specimen

### Specimen Type

Whole Blood EDTA

### Ordering Guidance

The Powassan virus polymerase chain reaction result may remain positive for a longer time in urine than in blood, serum, and cerebrospinal fluid (7 days or more).

### Specimen Required

**Container/Tube:** Lavender top (EDTA)

**Specimen Volume:** 1 mL

**Collection Instructions:** Send whole blood specimen in original tube. **Do not aliquot.**

### Forms

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

**Specimen Minimum Volume**

0.3 mL

**Reject Due To**

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

**Specimen Stability Information**

Specimen Type	Temperature	Time	Special Container
Whole Blood EDTA	Refrigerated (preferred)	7 days	
	Frozen	7 days	

**Clinical & Interpretive****Clinical Information**

Powassan virus (POWV) is an emerging tick-borne virus, harbored by *Ixodes* species ticks, which are the same ticks that transmit Lyme disease (*Borrelia* spp.), *Babesia* spp., and *Anaplasma phagocytophilum*, among other pathogens. POWV is a member of the *Flavivirus* genus, which includes other arthropod-borne viruses (arboviruses) such as West Nile virus (WNV) and St. Louis encephalitis virus. Two lineages of POWV have been identified, sharing approximately 94% amino acid sequence identity, including lineage 1, which is the prototypical POWV lineage transmitted by *Ixodes marxi* and *Ixodes cookei*, and lineage 2, which includes deer tick virus and is transmitted by *Ixodes scapularis*. POWV is maintained in the environment in groundhogs, skunks, squirrels, and white footed mice. Unlike other tick-borne pathogens, following tick attachment to a host, POWV can be transmitted in as little as 15 minutes.

Following infection, the incubation period can last anywhere from 4 to 14 days, after which approximately 66% of patients will remain asymptomatic. Symptomatic patients may present with a nonspecific influenza-like illness, including high fever, fatigue, malaise, and myalgia. Approximately 30% of symptomatic patients will progress to develop neurologic manifestations, most commonly encephalitis. While some patients may recover, over 50% of individuals will have persistent neurologic sequelae. POWV has been associated with an overall mortality rate of 10%. Although there is no targeted antiviral therapy and treatment is entirely supportive care, diagnosis is important for a number of reasons, including the ability to discontinue empiric antibiotics and to provide prognostic information for patients and families.

While limited data exist for POWV real-time reverse transcription polymerase chain reaction (RT-PCR) detection, data from testing for the related flavivirus, WNV, suggests that blood may be a more sensitive source than serum for detecting viral RNA. Similarly, data from WNV and other flaviviruses suggest that viral RNA may be detected in urine for a longer period of time than in blood or serum. POWV RNA may be detected from cerebrospinal fluid in cases of neuroinvasive disease.

Powassan infections are often diagnosed based on a patient's symptoms and exposure risk in conjunction with molecular and serologic testing. The use of RT-PCR can provide a rapid laboratory confirmation of POWV RNA early in infection, particularly during the first 7 days of illness when serologic testing is typically negative. After 7 days, POWV

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RT-PCR is less sensitive and serologic testing is the preferred diagnostic method. It is common for RT-PCR and serology to be used together for diagnosis of early infection, as they are complementary methods.

**Reference Values**

Negative

**Interpretation**

A positive result indicates the presence of Powassan virus RNA and is consistent with early Powassan virus infection.

**Cautions**

The sensitivity of the assay is dependent upon the time of illness onset in which the specimen is collected. Polymerase chain reaction testing has the greatest utility when used within the first few days of symptom onset.

A negative test does not exclude infection with Powassan virus. Therefore, the results obtained should be used in conjunction with clinical findings and serologic test results to make an accurate diagnosis.

This assay detects both viable and nonviable virus. Test performance depends on viral load in the specimen and may not correlate with cell culture performed on the same specimen.

**Supportive Data**

[The following validation data supports the use of this assay for clinical testing.](#)

**Accuracy/Diagnostic Sensitivity and Specificity:**

Accuracy studies were performed by testing negative clinical specimens with whole viral genomic RNA for lineages 1 and 2 near the limit of detection (LOD) and yielded greater than or equal to 97% sensitivity and specificity.

**Analytical Sensitivity/Limit of Detection:**

The lower LOD of this assay is 1 to 52 target copies/mcL of RNA extract for EDTA whole blood and serum, 0.1 to 5.2 copies/mcL for urine, and 0.1 to 52 copies/mcL for cerebrospinal fluid.

**Precision:**

Interassay and intra-assay precisions are 100%.

**Specificity:**

A panel of 15 organisms that can be found in the specimen types acceptable for this assay, as well as closely-related viruses (eg, dengue types 1-4, Japanese encephalitis virus, hepatitis E virus, Murray Valley encephalitis virus, St. Louis encephalitis virus, tick-borne encephalitis virus, yellow fever virus, Zika virus) and those that can cause a similar clinical syndrome were tested by this assay. No cross-reacting positive results were noted.

**Reportable Range:**

This is a qualitative assay, and the results are reported as either negative or positive for targeted Powassan virus.

**Clinical Reference**

1. Centers for Disease Control and Prevention (CDC), National Center for Emerging and Zoonotic Infectious Diseases, Division of Vector-Borne Diseases: Powassan Virus. For Healthcare Providers: Diagnostic Testing. CDC; Updated January

26, 2023. Accessed March 28, 2023. Available at [www.cdc.gov/powassan/diagnostic-testing.html](http://www.cdc.gov/powassan/diagnostic-testing.html)

2. Piantadosi A, Rubin DB, McQuillen DP, et al. Emerging cases of Powassan virus encephalitis in New England: Clinical presentation, imaging, and review of the literature. *Clin Infect Dis*. 2016 Mar 15;62(6):707-713. doi: 10.1093/cid/civ1005

## Performance

### Method Description

For this real-time reverse-transcription laboratory-developed polymerase chain reaction (PCR) assay, viral nucleic acid is extracted from specimens, followed by amplification and detection on the Roche LightCycler 480 instrument. This PCR assay has been optimized to detect a target sequence in the nonstructural protein. Primers amplify a 179 base pair product. Powassan virus genomic RNA is first transcribed to complementary DNA (cDNA) by reverse transcriptase, followed by amplification of the cDNA product. The LightCycler 480 is an automated instrument that amplifies and monitors the development of target nucleic acid (amplicon) after each cycle of PCR. The detection of amplicon is based on fluorescence resonance energy transfer, which utilizes a 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 Powassan virus lineage 1 and lineage 2. (Unpublished Mayo method)

### PDF Report

No

### Day(s) Performed

Monday through Friday

### Report Available

Same day/1 to 5 days

### Specimen Retention Time

7 days

### Performing Laboratory Location

Rochester

## 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**

87798

**LOINC® Information**

Test ID	Test Order Name	Order LOINC® Value
POWVB	Powassan Virus, PCR, Blood	34456-4

Result ID	Test Result Name	Result LOINC® Value
617488	Lineage 1	34456-4
618144	Lineage 2 (Deer Tick Virus)	34456-4
618148	Powassan Virus PCR Comment	77202-0