# Test Definition: HRTVC

Heartland Virus, RNA, Molecular Detection, PCR, Spinal Fluid

## Overview

### Useful For
Aiding in the diagnosis of central nervous system infection caused by Heartland virus using spinal fluid specimens

### Testing Algorithm
For more information see Meningitis/Encephalitis Panel Algorithm.

### Special Instructions
- Meningitis/Encephalitis Panel Algorithm

### Method Name
Real-Time Polymerase Chain Reaction (PCR)

### NY State Available
Yes

## Specimen

### Specimen Type
CSF

### Ordering Guidance
Patients with a history of symptoms for more than 1 week may be negative by molecular tests (ie, real-time polymerase chain reaction) and may require serologic testing, which is available through the Centers for Disease Control and Prevention.

### Specimen Required
**Container/Tube:** Sterile vial
**Specimen Volume:** 1 mL
**Collection Instructions:**
1. Send specimen from collection vial 2.
**2. Do not centrifuge or heat inactivate.**

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

### Reject Due To

| | |
|---|---|
| Heat-inactivate | Reject |

| | |
|---|---|
| d specimen | |

### Specimen Stability Information

| Specimen Type | Temperature | Time | Special Container |
|---|---|---|---|
| CSF | Refrigerated (preferred) | 14 days | |
| | Ambient | 24 hours | |
| | Frozen | 14 days | |

## Clinical & Interpretive

### Clinical Information

Heartland virus (HRTV) disease is an emerging zoonosis, transmitted to humans through the bite of infected *Amblyomma americanum* (Lone Star) ticks. HRTV possesses a single-stranded negative-sense RNA genome encoded on small, medium, and large segments. HRTV is a member of the Bandavirus genus, which includes other arthropod-borne viruses (arboviruses), such as severe fever with thrombocytopenia syndrome virus (SFTSV). Reports of human HRTV disease are relatively rare with fewer than 100 cases reported to date, most from the Central, Southern, and Northeastern United States. Symptoms generally occur within 2 weeks of a tick bite and may include non-specific symptoms such as headache, fever, fatigue, anorexia, nausea, diarrhea, and muscle or joint pain. Leukopenia, thrombocytopenia, and elevation of liver transaminases are also common laboratory findings. Rarely, multisystem organ failure and death occur. While there is no targeted antiviral therapy and treatment is entirely supportive care, diagnosis is important for several reasons, including the ability to discontinue empiric antibiotics and to provide prognostic information for patients and families.

Detection of HRTV nucleic acid in cerebrospinal fluid (CSF) is a marker for central nervous system infection caused by this virus. Importantly, the period of time that the virus can be detected in serum and CSF is brief. Therefore, molecular testing should be performed within the first week following onset of symptoms. After this time, serologic testing is the preferred method for diagnosis of HRTV infection. Serologic testing is currently only available through the Centers for Disease Control and Prevention.

### Reference Values

Negative

### Interpretation

Positive:
The detection of Heartland virus (HRTV) nucleic acid in cerebrospinal fluid (CSF) is consistent with acute-phase infection of the central nervous system. HRTV nucleic acid may be detectable during the first week following the onset of symptoms.

Negative:
The absence of HRTV nucleic acid in CSF is consistent with the lack of acute-phase infection but does not rule out infection. Additional serologic testing may be indicated.

## Cautions

Results should be used in conjunction with clinical presentation and exposure history.

Negative Heartland virus (HRTV) polymerase chain reaction results may occur if the specimen was collected more than 7 days following symptom onset. Serologic testing for the presence of antibodies to HRTV may be recommended in such cases.

## Supportive Data

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

Accuracy/Diagnostic Sensitivity and Specificity:
Heartland virus (HRTV) accuracy results produced greater than or equal to 90% positive, negative, and overall agreement among 70 total contrived samples for each matrix (cerebrospinal fluid [CSF] and serum).

Analytical Sensitivity/Limit of Detection:
The HRTV reverse transcription polymerase chain reaction (RT-PCR) assay analytical sensitivity was established and confirmed at 2.5 genome copies/microliter of RNA extract (2500 genome copies/mL) in both CSF and serum.

Precision:
Greater or equal to 95% qualitative concordance among test replicates for HRTV inter-assay and intra-assay reproducibility.

Specificity:
Panel of 13 organisms with similar phylogenetic association, clinical presentation, vector/transmission mode and/or sample type were tested by the HRTV RT-PCR assay and did not show any amplification. Additionally, NCBI BLAST searches of HRTV assay forward/reverse primers and PCR amplicon sequences against a panel of 82 organisms causing similar diseases or commonly found in CSF or serum were also used to determine potential cross-reactivity of notable organisms. The data indicate that HRTV RT-PCR assay will not detect any organisms other than Heartland virus.

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

## Clinical Reference

1. Savage HM, Godsey MS, Lambert A, et al. First detection of heartland virus (Bunyaviridae: Phlebovirus) from field collected arthropods. Am J Trop Med Hyg. 2013;89(3):445-452. doi:10.4269/ajtmh.13-0209
2. Brault AC, Savage HM, Duggal NK, Eisen RJ, Staples JE. Heartland virus epidemiology, vector association, and disease potential. Viruses. 2018;10(9):498. doi:10.3390/v10090498
3. Staples JE, Pastula DM, Panella AJ, et al. Investigation of Heartland virus disease throughout the United States, 2013-2017. Open Forum Infect Dis. 2020;7(5);ofaa125. Published 2020 Apr 11. doi:10.1093/ofid/ofaa125
4. Centers for Disease Control and Prevention (CDC), National Center for Emerging and Zoonotic Infectious Diseases (NCEZID) , Division of Vector-Borne Diseases (DVBD). Heartland virus disease. CDC; Updated January 26, 2023. Accessed April 25, 2024. Available at www.cdc.gov/heartland-virus/healthcare-providers/index.html

## Performance

### Method Description

This Heartland virus (HRTV) real-time reverse transcription polymerase chain reaction (RT-PCR) laboratory-developed test is based on primer and probe sequences reported by Savage et al. (CDC assay, 1). It provides qualitative detection of HRTV RNA from serum and CSF of patients with suspected infection. HRTV RNA in clinical specimens is first extracted using the Roche MagNA Pure 96 instrument. RT-PCR testing is then performed on the Roche LightCycler 480 II (LC480).

This RT-PCR assay employs oligonucleotide forward and reverse primers and a TaqMan hydrolysis probe specific to the small segment of the non-structural protein region of Heartland virus. A reverse transcription step is first performed to convert RNA to complementary DNA (cDNA). The primers then bind to the target cDNA sequence and facilitate amplification of an 86 base pair amplicon product during PCR. Using the LC480 software, analysis of the resultant amplification curves is performed to allow for detection of HRTV RNA. Generation of an amplification curve and a crossing point (Cp) value indicates the presence of HRTV RNA in the specimen and may be used to support the diagnosis of acute HRTV infection in the appropriate clinical and epidemiologic setting.(Unpublished Mayo method.)

### PDF Report

No

### Day(s) Performed

Monday through Sunday

### Report Available

Same day/1 to 5 days

### Specimen Retention Time

7 days

### 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.

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## CPT Code Information

87798

## LOINC® Information

| Test ID | Test Order Name | Order LOINC® Value |
|---|---|---|
| HRTVC | Heartland Virus, PCR, CSF | 94183-1 |

| Result ID | Test Result Name | Result LOINC® Value |
|---|---|---|
| 620056 | Heartland Virus, PCR, CSF | 94183-1 |