

Overview

Useful For

Rapid testing for West Nile virus (WNV) RNA (lineage 1 and lineage 2) using serum specimens

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

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

Testing Algorithm

The following algorithms are available:

- [Meningitis/Encephalitis Panel Algorithm](#)
- [Mosquito-borne Disease Laboratory Testing](#)

Special Instructions

- [Meningitis/Encephalitis Panel Algorithm](#)
- [Mosquito-borne Disease Laboratory Testing](#)

Method Name

Real-Time Reverse Transcription Polymerase Chain Reaction (RT-PCR)

NY State Available

Yes

Specimen

Specimen Type

Serum

Specimen Required

Collection Container/Tube:

Preferred: Serum gel

Acceptable: Red top

Submission Container: Sterile container

Specimen Volume: 0.5 mL

Collection Instructions:

1. Within 2 hours of collection centrifuge and aliquot serum into a sterile container.
2. Serum specimens not aliquoted from the serum gel collection tube into a sterile container will be rejected.

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

Gross hemolysis	Reject
Heat-inactivated specimen	Reject

Specimen Stability Information

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

Clinical & Interpretive

Clinical Information

West Nile virus (WNV) is a mosquito-borne flavivirus (single-stranded RNA virus) that primarily infects birds but occasionally infects horses and humans (1,2,3). Until the virus was recognized in 1999 in infected birds in New York City, WNV had been detected only in the Eastern hemisphere with a wide distribution in Africa, Asia, the Middle East, and Europe. There are 2 distinct lineages of WNV: lineage 1 has the broadest distribution worldwide, including North America and Europe, whereas lineage 2 is found only in Africa and parts of Europe.

Most people who are infected with WNV do not experience symptoms. It is estimated that about 20% of those who become infected will develop West Nile fever with mild symptoms, including headache, myalgia, and, occasionally, a skin rash on the trunk of the body. About 1 of 150 WNV infections (<1%) results in meningitis or encephalitis. Fatality rates among patients hospitalized during recent outbreaks have ranged from 4% to 14%. Advanced age is the most important risk factor for death, and patients older than 70 years are at particularly high risk.

Laboratory diagnosis is best achieved by demonstration of specific IgG- and IgM-class antibodies in serum specimens. However, polymerase chain reaction (PCR) testing can be used to detect WNV RNA in serum, whole blood, and urine specimens from patients with recent WNV infection (ie, 3-5 days following infection) when specific antibodies to the virus are not yet present. It may also be useful for patients who are immunocompromised when an antibody response is minimal or absent. Finally, PCR can be useful for supporting a serologic diagnosis, given the known cross-reactivity of WNV serology with other flaviviruses.

Studies indicate that whole blood testing by PCR may provide higher sensitivity when testing patients with acute WNV disease (up to 87%) compared to serum, plasma, urine, and cerebrospinal fluid testing.(4) However, viral RNA may be

detected for a longer period of time (> or =10 days after symptom onset) in urine than in other sources.(5) Serum testing offers lower sensitivity (26%) but may be used when it is the only specimen type available.

Reference Values

Negative

Reference values apply to all ages.

Interpretation

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

Cautions

The sensitivity of the assay is very 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.

Whole blood rather than serum has been shown to provide increased sensitivity for detecting West Nile virus (WNV) RNA.

A negative test does not exclude infection with WNV. 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/LOD:

The lower LOD of this assay is 1 to 5 target copies/mcL of RNA extract for serum, urine, and spinal fluid and 27 to 60 copies/mcL for EDTA whole blood.

Precision:

Inter-assay and intra-assay precisions are 100%.

Specificity:

A panel of 42 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 West Nile virus.

Clinical Reference

1. Petersen LR, Brault AC, Nasci RS. West Nile virus: review of the literature. JAMA. 2013;310(3):308-315
2. Colpitts TM, Conway MJ, Montgomery RR, Fikrig E. West Nile virus: Biology, transmission, and human infection. Clin Microbiol Rev. 2012;25(4):635-648
3. Centers for Disease Control and Prevention (CDC), National Center for Emerging and Zoonotic Infectious Disease (NCEZID), Division of Vector-Borne Diseases (DVBD): West Nile Virus. CDC; Accessed January 21, 2025. Available at <https://www.cdc.gov/west-nile-virus/>
4. Lustig Y, Mannasse B, Koren R, et al. Superiority of West Nile virus RNA detection in whole blood for diagnosis of acute infection. J Clin Microbiol. 2016;54(9):2294-2297
5. Barzon L, Pacenti M, Franchin E, et al. Excretion of West Nile virus in urine during acute Infection. J Infect Dis. 2013;208(7):1086-1092

Performance**Method Description**

This real-time reverse transcription polymerase chain reaction (RT-PCR) assay provides qualitative detection of lineages 1 and 2 of West Nile virus. Viral RNA is first extracted from serum on an automated platform. The extract is then used for subsequent RT-PCR. An initial reverse transcription step is employed to convert viral RNA to complementary DNA (cDNA). The viral cDNA target is then amplified on the Roche LightCycler 480 using specific primers and detected via Taqman probe technology. An internal control is also included with each reaction. Analysis of the PCR amplification is accomplished through the use of LightCycler software.(Package insert: RealStar WNV RT-PCR Kit 2.0. Altona Diagnostics; 03/2020)

PDF Report

No

Day(s) Performed

Monday through Friday

Report Available

Same day/1 to 5 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

87798

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
WNVS	West Nile Virus RNA, PCR, Serum	32361-8

Result ID	Test Result Name	Result LOINC® Value
608436	West Nile Virus RNA, PCR, Serum	32361-8