Test Definition: LNORO

Norovirus PCR, Molecular Detection, Feces

Overview

Useful For

Diagnosing gastrointestinal disease (diarrhea or vomiting) caused by norovirus genogroups 1 and 2

This test **should not be used** as a test-of-cure.

Method Name

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

NY State Available

Yes

Specimen

Specimen Type

Fecal

Shipping Instructions

Place vial in a sealed plastic bag and **ship ambient**. **Specimens received at other temperatures will be rejected, and testing will be canceled**.

Specimen Required

Supplies: Culture and Sensitivity Stool Transport Vial (T058)
Container/Tube: Commercially available transport system specific for recovery of enteric pathogens from fecal specimens (15 mL of non-nutritive transport medium - either Cary-Blair, Para-Pak Culture and Sensitivity Media)
Specimen Volume: Representative portion of diarrheal fecal sample, 1 gram or 5 mL
Collection Instructions:
1. Collect fresh feces and place in preservative within 1 hour of collection.
2. Visibly formed feces are not consistent with Norovirus gastrointestinal disease and should not be submitted for testing.

Reject Due To

Nonpreserved feces Transport media other than Cary-Blair or C and S Modified	Reject

Cary-Blair Visibly formed feces	

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Fecal	Ambient	7 days	

Clinical & Interpretive

Clinical Information

Noroviruses, previously known as Norwalk-like viruses, are highly contagious RNA viruses that cause acute gastroenteritis (diarrhea, vomiting). Although 6 genogroups of norovirus have been identified, only 3 genogroups (genogroup: G1, G2, and G4) cause disease in humans. Furthermore, the majority of outbreaks have been associated with G1 and G2, with G2 being most common.(1)

Noroviruses are transmitted through close, personal contact with an infected individual or via the fecal-oral route in which the virus becomes ingested in contaminated food or water. These viruses are extremely contagious, with fewer than 20 virions being able to cause disease.(1)

Once infected, the incubation period is typically short, between 24 and 72 hours. The onset of symptoms is abrupt, with vomiting and watery nonbloody diarrhea being common. Patients may also experience a low-grade fever, as well as headache and mild body aches.(1)

The diagnosis of norovirus infection can often be made on clinical grounds, and symptoms generally resolve in 24 to 48 hours. However, in certain patients, especially those who are immunocompromised or hospitalized, laboratory testing may be indicated for infection control purposes and to limit the use of antibiotics. Testing of diarrheal feces by real-time polymerase chain reaction allows for a rapid and sensitive means of detecting and differentiating norovirus G1 and G2 in clinical stool samples.

Reference Values

Negative

Interpretation

A positive result indicates that RNA from norovirus genogroups 1 and/or 2 was present in the clinical specimen.

A negative result suggests that RNA from norovirus genogroups 1 and 2 was absent in the clinical specimen.

Cautions

A positive result suggests that norovirus is the cause of gastrointestinal disease (diarrhea or vomiting); however, in certain patients (eg, immunocompromised hosts), norovirus may be shed for weeks to months in the absence of symptoms.(2)

This test should not be used as a test-of-cure, due to the fact that norovirus nucleic acid may be detected in patients for weeks to months following the resolution of symptoms.(2)

A negative result suggests that norovirus is not the cause of gastrointestinal disease (diarrhea or vomiting); however, viral nucleic acid may be present at a level that is below the limit of detection for this test. The results should be interpreted in the context of the patient's clinical presentation and other laboratory findings.

Supportive Data

Accuracy:
A total of 100 clinical stool specimens submitted to Mayo between 11/2015 and 03/2016 for testing by a commercial multiplex gastrointestinal panel (that includes norovirus) were aliquoted, blinded, and tested within 24 to 48 hours of receipt using the norovirus G1/G2 real-time polymerase chain reaction (PCR) assays. Specimens yielding discordant results between the multiplex panel and the real-time PCR were submitted to an outside reference lab and the Minnesota Department of Health (MDH) for norovirus molecular testing.

Table 1. Comparison of results following discordant analysis at an outside reference laboratory and MDH

	Number of samples following discordant resolution with a result of: (a,b)	
Norovirus lab-developed test (LDT)	**Positive**	**Negative**
Positive	63(c)	0
Negative	2(d)	35
Total	65	35

Adjusted Sensitivity (95% confidence interval [CI])=96.9% (88.8, 99.8)
Adjusted Specificity (95% CI)=100% (88.2, 100)
Adjusted Agreement (95% CI)=98% (92.6, 99.9)

a. Samples showing discordant results between the norovirus real-time PCR and FilmArray were tested by a molecular method at Focus Diagnostics and MDH.
b. Samples that showed agreement between the norovirus real-time PCR and FilmArray were not tested further.
c. Four of these 63 samples were identified as G1 by the LDT; the remaining 59 were identified as norovirus G2.
d. One of these 2 samples was positive for norovirus at Focus Diagnostics and MDH, and upon repeat testing in triplicate by the LDT, was also positive. The second sample was positive for norovirus at MDH, but negative by Focus and the LDT upon repeat testing.

Testing of clinical stool samples yielded 59 specimens that were positive for norovirus G2 by the LDT; however, only 4 samples were determined to be positive for norovirus G1. In order to supplement the clinical data and increase the number of samples positive for norovirus G1, spiking studies were performed. Analyte-negative stool samples (n=26) were spiked with norovirus G1 RNA (ATCC) at 1 dilution above the defined limit of detection (LOD) (Table 2).

Table 2. Results of spiking studies for norovirus GI in stool specimens.

Specimen	**Species**	**Concentration of target in spiked sample**	**Number positive/ number tested**	**% Positive**	**Mean crossing**

					point
Stool	Norovirus G1	10 copies/mcL	26/26	100	33.2

Analytical Sensitivity:
The limit of detection of both the G1 and G2 assays was determined to be 5 copies/mcL (5000 copies/mL) of stool.

Analytical Specificity:
A comprehensive specificity panel, consisting of bacteria (n=16), parasites (n=5), or viruses (n=5) known to cause gastrointestinal disease was tested by the norovirus G1/G2 assays. In addition, human DNA was tested. All members of the specificity panel were negative by both the norovirus G1 and G2 assays. In addition, a BLAST (basic local alignment search tool) analysis was performed on the primer and probe sequences and did not reveal significant cross-reactivity with organisms that may be present in stool samples.

Clinical Reference

1. National Center for Immunization and Respiratory Diseases, Division of Viral Diseases: Norovirus. Center for Disease Control and Prevention; 2013. Updated February 23, 2023. Accessed March 28, 2023. Available at www.cdc.gov/norovirus/index.html
2. Echenique IA, Stosor V, Gallon L, Kaufman D, Qi C, Zembower TR: Prolonged norovirus infection after pancreas transplantation: a case report and review of chronic norovirus. Transpl Infect Dis. 2016 Feb;18(1):98-104

Performance

Method Description

This assay utilizes real-time, TaqMan-based polymerase chain reaction (PCR) technology to target the nonstructural polyprotein gene of norovirus G1 and G2. Nucleic acid is extracted from diarrheal stool samples in Cary-Blair media. Following extraction of viral RNA, reverse transcription is performed to convert norovirus genomic RNA to complementary DNA. Real-time amplification and detection are then performed. Two separate real-time PCR reactions are performed for each sample, one specific for norovirus G1 and the second targeting norovirus G2. The assay is able to accurately detect and differentiate these genogroups in clinical stool samples.(Unpublished Mayo method)

PDF Report

No

Day(s) Performed

Monday through Sunday

Report Available

2 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.

CPT Code Information

87798 x 2

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
LNORO	Norovirus PCR, F	88701-8

Result ID	Test Result Name	Result LOINC® Value
65170	Norovirus G1 PCR	54905-5
47553	Norovirus G2 PCR	54906-3