

## Overview

### Useful For

Virologic detection of HIV-1 infection in infants younger than 2 years of age (an age group for which serologic tests are unreliable) born to HIV-1-infected mothers

Early detection of acute HIV-1 infection in children and adults who may be receiving combination antiretroviral prophylaxis or preemptive treatment

Determining eradication of HIV-1 in individuals receiving combination highly active antiretroviral therapies

### Testing Algorithm

The following algorithms are available in Special Instructions:

[-HIV Testing Algorithm \(Fourth-Generation Screening Assay\), Including Follow-up of Reactive Rapid Serologic Test Results](#)

[-HIV Prenatal Testing Algorithm, Including Follow-up of Reactive Rapid Serologic Test Results](#)

### Special Instructions

- [HIV Testing Algorithm \(Fourth-Generation Screening Assay\), Including Follow-up of Reactive Rapid Serologic Test Results](#)
- [HIV Prenatal Testing Algorithm, Including Follow-up of Reactive Rapid Serologic Test Results](#)

### Method Name

Real-Time Polymerase Chain Reaction (PCR)

### NY State Available

No

## Specimen

### Specimen Type

Plasma EDTA

### Ordering Guidance

This assay should not be used as a screening test or primary diagnostic test for HIV-1 infection, except in infants younger than 2 years of age born to HIV-1-infected mothers. For other age groups, order HVCOP / HIV-1 and HIV-2 Antigen and

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Antibody Routine Screen, Plasma as the screening test.

**Shipping Instructions**

1. Ship plasma on ice packs if arrival at Mayo Clinic Laboratories is expected within 5 days of plasma separation.
2. If shipment will be delayed for more than 5 days, freeze plasma specimen -20 to -80 degrees C until shipment on dry ice.

**Specimen Required**

**Supplies:** Aliquot Tube, 5 mL (T465)

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

**Submission Container/Tube:** Plastic vial

**Specimen Volume:** 1 mL

**Collection Instructions:** Centrifuge blood collection tube and aliquot plasma into plastic vial per collection tube manufacturer's instructions (eg, centrifuge and aliquot within 2 hours of collection for BD Vacutainer tubes).

**Additional Information:** This test can be used for detection and diagnosis of HIV-1 infections, including in children younger than 2 years of age when serologic tests are not useful (due to presence of maternal HIV antibodies).

**Reject Due To**

Gross hemolysis OK  
Gross lipemia OK

**Specimen Minimum Volume**

0.5 mL

**Specimen Stability Information**

Specimen Type	Temperature	Time	Special Container
Plasma EDTA	Frozen (preferred)	35 days	
	Refrigerated	5 days	

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

Human immunodeficiency virus (HIV)-1 infection is usually confirmed by detection of HIV-1-specific antibodies in serum. However, serologic testing may not reliably identify HIV-1 infection in neonates with passively acquired maternal HIV-1

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antibodies or with incompletely developed immune systems, individuals with early HIV-1 infection (<30 days from infection), or individuals with "indeterminate" HIV-1 antibody results by supplemental serologic assays. In these situations, detection of HIV-1 nucleic acids (RNA or proviral DNA) by PCR can provide definitive, early evidence of HIV-1 infection (approximately 10 to 14 days after infection), when results of routine diagnostic assays may be inconclusive.

Upon entry into human cells (including peripheral blood mononuclear cells), the HIV-1 RNA is converted into complementary DNA (cDNA) by reverse transcription. These linear cDNA strands are then integrated into the host cell genome, thus representing the proviral form of HIV-1. mRNA, transcribed from the proviral DNA, is used to synthesize the proteins required to make new viral particles. These proteins and viral RNA are packaged in the host's cytoplasm and released from the cell, completing the life cycle of the virus.

For infants born to HIV-1-infected mothers, HIV-1 DNA or RNA tests are recommended at 0 to 2 days, 14 days, 1 to 2 months, and 4 to 6 months after birth. Two consecutive positive HIV-1 virologic test results (HIV-1 DNA and/or RNA) are necessary for confirming the diagnosis of HIV-1 infection in infants younger than 2 years of age.

### Reference Values

Undetected

### Interpretation

A "Detected" result is consistent with HIV infection (see Cautions). Per CDC and US Public Health Services recommendations, a second specimen should be collected from any patient with first-time detectable HIV-1 DNA or RNA result and tested to verify the diagnosis of HIV-1 infection.

An "Undetected" result indicates that neither HIV-1 DNA nor RNA is detected in the specimen (see Cautions). Repeat testing is recommended at 0 to 2 days, 14 days, 1 to 2 months, and 4 to 6 months after birth in infants born to HIV-1 infected mothers. For at-risk individuals older than 2 years of age, repeat testing in 1 to 2 months is recommended. The lower limits of detection (based on 95% detection rate) of this assay in plasma are 311 copies/mL for HIV-1 DNA and 75 copies/mL for HIV-1 RNA.

An "Inconclusive" result indicates that the absence or presence of HIV-1 DNA or RNA could not be determined with certainty after repeat testing of the clinical specimens in the laboratory, possibly due to PCR inhibition. Submission of a new specimen for testing is recommended.

### Cautions

This assay is optimized for the detection of HIV-1 groups M (subtypes A to H), N and O, but it may not detect all HIV-1 group N or O strains.

Diagnosis of HIV-1 infection should not rely solely upon a "Detected" result for HIV-1 DNA and/or RNA. Such a result should be considered in conjunction with patient's clinical presentation, physical findings, and other diagnostic laboratory tests prior to establishing the diagnosis. An "Undetected" result indicates only the absence of HIV-1 DNA and RNA in the specimen tested and does not exclude the diagnosis of HIV-1 infection. "Undetected" results should be interpreted with caution, considering the patient's risk factors for HIV-1 infection, the analytical sensitivity of the assay, and the group of the infecting HIV-1 strain. Follow-up testing is recommended for high-risk patients with initially "Undetected" test results. Individuals on prophylactic or therapeutic antiretroviral therapy may have "Undetected" results.

"Undetected" results together with repeatedly positive HIV-1 antibody supplemental test results may be observed in HIV-2-infected individuals. For such patients with risk factors for HIV-2 infection, specific testing for HIV-2 antibodies (serologic) and HIV-2 DNA and/or RNA is recommended.

### Clinical Reference

1. Huang S, Erickson B, Mak WB, et al: A novel real-time HIV-1 qualitative assay for the detection of HIV-1 nucleic acids in dried blood spots and plasma. *J Virol Meth* 2011;178:216-224
2. Burgard M, Blanche S, Jasseron C, et al: Performance of HIV-1 DNA or HIV-1 RNA tests for early diagnosis of perinatal HIV-1 infection during anti-retroviral prophylaxis. *J Pediatr* 2012;160:60-66
3. Nelson JAE, Hawkins JT, Schanz M, et al: Comparison of the Gen-Probe Aptima HIV-1 and Abbott HIV-1 qualitative assays with the Roche Amplicor HIV-1 DNA assay for early infant diagnosis using dried blood spots. *J Clin Virol* 2014;60:418-421
4. HHS Panel on Treatment of HIV-Infected Pregnant Women and Prevention of Perinatal Transmission: Recommendations for the use of antiretroviral drugs in pregnant women with HIV infection and interventions to reduce perinatal HIV transmission in the United States. Accessed January 5, 2018. Available at <https://aidsinfo.nih.gov/guidelines/html/3/perinatal/0>
5. HHS Panel on Antiretroviral Therapy and Medical Management of HIV-Infected Children: Guidelines for the use of antiretroviral agents in pediatric HIV infection. Accessed January 5, 2018. Available at <https://aidsinfo.nih.gov/guidelines/html/2/pediatric-arv/0>

### Performance

#### Method Description

The Abbott *mSample* Preparation System kit is used with the automated Abbott *m2000sp* sample preparation system to extract and purify viral nucleic acids from human plasma specimens, based on magnetic particle technology. An internal control (IC) is incorporated in the nucleic acid extraction and purification procedure for processing the assay controls

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and clinical specimens. After capture of nucleic acids on magnetic microparticles, the microparticles are washed to remove unbound sample components. Then, the bound nucleic acids are eluted from the microparticles and the eluates are transferred to a 96-well microtiter plate containing PCR master mix reagents for amplification and detection.

During the amplification reaction on the Abbott *m2000rt* system, target DNA is amplified by the DNA polymerase activity of the thermostable rTth DNA polymerase. The target RNA present is first converted to cDNA by the reverse transcriptase activity of the enzyme and subsequently amplified. Amplification of HIV-1 and IC targets takes place simultaneously in the same reaction. The target sequence for this assay is in the *pol* integrase region of the HIV-1 genome and it is highly conserved. The IC sequence is derived from the hydroxypyruvate reductase gene of the pumpkin plant, *Cucurbita pepo*, and is delivered in an Armored RNA particle that has been diluted in negative human plasma.

During the read cycles of amplification on the Abbott *m2000rt*, the temperature is lowered to allow fluorescent detection of amplification products as the HIV-1 and IC probes anneal to their respective targets (real-time fluorescent detection). The HIV-1 probe has a fluorescent moiety that is covalently linked to the 5' end. A short quencher oligonucleotide is complementary to the 5' end of the HIV-1 probe and has a quencher moiety at its 3' end. In the absence of HIV-1 target, the HIV-1 probe fluorescence is quenched through hybridization to the quencher oligonucleotide. In the presence of the HIV-1 target sequence, the HIV-1 probe preferentially hybridizes to the target sequence, dissociating from the quencher oligonucleotide, allowing fluorescent detection. The IC probe is a single stranded DNA oligonucleotide with a fluorophore at the 5' end and a quencher at the 3' end. In the absence of IC sequences, the IC probe adopts a series of random conformations, some of which bring the quencher close enough to the excited fluorophore to absorb its energy before it can be fluorescently emitted. When the IC probe binds to its complementary sequence in the target, the fluorophore and the quencher are held apart, allowing fluorescent emission and detection by the Abbott *m2000rt*. The HIV-1 and IC specific probes are each labeled with a different fluorophore, thus allowing for simultaneous detection of both amplified products. (Package insert: Abbott RealTime HIV-1 Qual; Abbott Molecular, Inc., Des Plaines, IL. 10/2014)

**PDF Report**

No

**Specimen Retention Time**

2 months

**Performing Laboratory Location**

Rochester

**Fees & Codes**

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**Test Classification**

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

**CPT Code Information**

87535

**LOINC® Information**

Test ID	Test Order Name	Order LOINC Value
HIVP	HIV-1 DNA / RNA Qualitative, P	79379-4

Result ID	Reporting Name	LOINC®
64693	HIV-1 DNA / RNA Qualitative, P	79379-4