

Overview

Useful For

Quantifying plasma HIV-1 RNA levels (viral load) in individuals living with HIV, including children, followed by identification of genotypic mutations associated with viral resistance to inhibitors of HIV-1 reverse transcriptase and protease

Guiding initiation or change of combination antiretroviral therapy in individuals, including children, living with HIV

Testing Algorithm

If HIV-1 RNA level is 500 copies/mL or higher, then HIV-1 genotypic drug resistance mutation testing will be performed at an additional charge.

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 Treatment Monitoring Algorithm](#)

Special Instructions

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

Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
HIVPR	HIV-1 Genotypic PR-RT Resistance, P	Yes	No

Method Name

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

NY State Available

Yes

Specimen

Specimen Type

Plasma EDTA

Shipping Instructions

1. Ship specimen frozen on dry ice.
2. If shipment will be delayed for >24 hours, freeze plasma specimen at -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:** 3.6 mL**Collection Instructions:**

1. 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).
2. Freeze aliquoted plasma for shipment.

Reject Due To

Gross hemolysis OK

Gross lipemia OK

Specimen Minimum Volume

2 mL

Specimen Stability Information

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

Clinical & Interpretive**Clinical Information**

HIV-1 is an RNA virus that infects human host cells and is then converted to complementary DNA (cDNA) by the action of viral reverse transcriptase. HIV-1 is the causative agent of AIDS, a severe, life-threatening condition.

Currently, 2 types of HIV: HIV type 1 (HIV-1) and HIV type 2 (HIV-2), are known to infect humans. HIV-1 has been isolated from patients with AIDS, AIDS-related complex, and asymptomatic infected individuals at high-risk for AIDS. Accounting for over 99% of HIV infections in the world, HIV-1 is transmitted by sexual contact, by exposure to infected blood or blood products, from an infected pregnant woman to fetus in utero or during birth, or from an infected mother to infant via breast feeding. HIV-2 has been isolated from infected patients in West Africa and it appears to be endemic only in that region. However, HIV-2 also has been identified in individuals who have lived in West Africa or had sexual relations with individuals from that geographic region. HIV-2 is similar to HIV-1 in its morphology, overall genomic structure, and ability to cause AIDS.

Multiple clinical studies of plasma HIV-1 viral load (expressed as HIV-1 RNA copies/mL of plasma) have shown a clear relationship of HIV-1 RNA copy number to stage of HIV-1 disease and efficacy of anti-HIV-1 therapy. Quantitative HIV-1 RNA level in plasma (ie, HIV-1 viral load) is an important surrogate marker in assessing the risk of disease progression and monitoring response to anti-HIV-1 drug therapy in the routine medical care of HIV-1-infected patients.

Studies have identified a number of mutations associated with antiviral resistance. Genotypic analysis allows identification of nucleotide changes associated with HIV drug resistance. When combination therapy fails, genotyping for drug resistance mutations may help direct appropriate changes in antiretroviral therapy and may result in at least a short-term benefit, as evidenced by viral load reduction.

Reference Values

Undetected

Interpretation

This assay has a plasma HIV-1 RNA quantification result range of 20 to 10,000,000 copies/mL (1.30-7.00 log copies/mL).

An "Undetected" result indicates that the assay was unable to detect HIV-1 RNA within the plasma specimen.

A result of "<20 IU/mL" indicates that HIV-1 RNA is detected, but the level present is less than the lower quantification limit of this assay. Due to the increased sensitivity of this assay, patients with previously low or undetectable HIV-1 viral load may show increased or detectable viral load with this assay. However, the clinical implications of a viral load below 20 copies/mL remain unclear. Possible causes of such a result include very low plasma HIV-1 viral load present (eg, in the range of 1-19 copies/mL), very early HIV-1 infection (ie, less than 3 weeks from time of infection), or absence of HIV-1 infection (ie, false-positive).

A result of ">10,000,000" with the result comment of "HIV-1 RNA level is >10,000,000 copies/mL (>7.00 log copies/mL). This assay cannot accurately quantify HIV-1 RNA above this level" indicates that HIV-1 RNA is detected, but the level present is above the upper quantification limit of this assay.

For the purpose of monitoring patient's response to antiretroviral therapy, the US Health and Human Services Panel on Antiretroviral Guidelines for Adults and Adolescents defines virologic failure as a confirmed viral load of greater than 200 copies/mL, which eliminates most cases of viremia resulting from isolated blips or assay variability. Confirmed viral load rebound (ie, >200 copies/mL) on 2 separate tests obtained at least 2 to 4 weeks apart should prompt a careful evaluation of patient's tolerance of current drug therapy, drug-to-drug interactions, and patient adherence.

If the viral load is greater than or equal to 500 copies/mL, genotypic antiviral drug resistance mutation analysis is performed automatically at an additional charge.

Genotype for drug resistance:

Codon sequences of the patient's HIV-1 reverse transcriptase and protease genes are compared with those in a database of known antiretroviral drug resistance mutations provided in the assay manufacturer's software application. Results are provided that highlight those codon changes associated with specific drug resistance. These mutations are categorized and reported.

"Susceptible (SUSC)" indicates that the genotypic mutations present in patient's HIV-1 strain have not been associated with resistance to the specific drug in question.

"Resistant (RESIST)" indicates that genotypic mutations detected have been associated with maximum reduction in susceptibility to the specific drug.

"Possible resistance (PR)" indicates that genotypic mutations detected have been associated with 1 or both of the following outcomes:

-Diminished virologic response in some, but not all, patients having virus with these mutations

-Intermediate decrease in susceptibility of the virus to the specific drug

"Unable to genotype" result indicates that the sequence data obtained are of poor quality to determine the presence or absence of genotypic resistant mutations in the patient's HIV strain. Probable causes of such poor sequence data include polymorphism in the region of the sequencing primers interfering with primer binding and subsequent sequencing reaction, or low viral load (ie, <500 copies/mL).

"Inconclusive" result indicates inability of the assay to reliably determine antiviral resistance because of the presence of polymerase chain reaction inhibitors or ambiguous or incomplete viral target sequences generated from the assay.

Cautions

The HIV-1 RNA detection and quantification assay is not licensed by the FDA as a screening test for HIV-1 infection in donors of blood, human cells, tissues, or tissue products.

Although this quantitative HIV-1 RNA test is not FDA approved for diagnostic purposes, the US Department of Health and Human Services Panel on Antiretroviral Therapy and Medical Management of HIV-Infected Children recommends the use of molecular-based assays to detect HIV-1 RNA or proviral DNA for the diagnosis of HIV infection in infants younger than 18 months of age and born to HIV-infected mothers.

A single HIV-1 viral load test result should not be used as the sole criterion in guiding therapeutic decisions and intervention in the clinical care of HIV-1-infected patients. Viral load results should be correlated with patient symptoms, clinical presentation, and CD4 cell count. Due to the inherent variability in the assay, physiologic variation and concurrent illnesses in the infected patients, changes of less than 100-fold (<2 log) in plasma HIV-1 viral load should not be considered to be significant changes.

Viral load results below 20 copies/mL do not necessarily indicate absence of HIV-1 viral replication. Inhibitory substances may be present in the plasma specimen, leading to negative or falsely low HIV-1 RNA results. Improper specimen collection or storage may lead to negative or falsely lower plasma viral load results.

Although this commercial HIV-1 viral load assay is optimized for quantification of plasma viral load in HIV-1 infection due to HIV-1 groups M (subtypes A to H) and O strains, results generated from HIV-1 group O strains may be discordant (> or =0.5 log copies/mL) with those obtained from other commercially available HIV-1 viral load assays. The assay is not reliable for quantifying plasma viral loads in infection caused by HIV-1 group N and HIV-2 strains.

ACD plasma specimens are not optimal for HIV-1 viral load testing because such plasma specimens show HIV-1 RNA levels that are approximately 15% lower than those collected in tubes containing EDTA. Plasma specimens stored frozen

in plasma preparation tubes (PPT) are not suitable for HIV-1 viral load testing due to falsely high viral load results from release of intracellular HIV-1 nucleic acids (DNA and RNA) during the freezing process.

Clinical Reference

1. Centers for Disease Control and Prevention and Association of Public Health Laboratories. Laboratory testing for the diagnosis of HIV infection: Updated recommendations. June 27, 2014. Available at <http://stacks.cdc.gov/view/cdc/23447>
2. Gunthard HF, Saag MS, Benson CA, et al: Antiretroviral drugs for treatment and prevention of HIV infection in adults: 2016 recommendations of the International Antiviral Society-USA Panel. JAMA. 2016 Jul 12;316(2):191-210
3. Panel on Antiretroviral Guidelines for Adults and Adolescents: Guidelines for the use of antiretroviral agents in adults and adolescents living with HIV. US Department of Health and Human Services. October 17, 2017. Updated December 18, 2019. Accessed May 13, 2021. Available at <https://aidsinfo.nih.gov/guidelines/html/1/adult-and-adolescent-arv/3/tests-for-initial-assessment-and-follow-up>
4. Wensing AM, Calvez V, Gunthard HF, et al: 2017 Update of the drug resistance mutations in HIV-1. Top Antivir Med. 2017;24(4):132-141

Performance**Method Description**

The cobas HIV-1 assay is an FDA-approved, in vitro nucleic acid amplification test for the quantification of HIV-1 RNA in human plasma using the cobas 6800 System or cobas 8800 System for fully automated viral nucleic acid extraction (generic silica-based capture technique) and automated amplification and detection of the viral nucleic acid sequence. This PCR assay amplifies sequences within the *gag* gene and LTR region of the HIV-1 genome and generates amplification products that are detected and quantified in real-time with 2 sequence-specific TaqMan probes. A non-HIV armored RNA quantitation standard (RNA-QS) is introduced into each specimen during sample preparation to serve as internal control for nucleic acid extraction and PCR amplification / detection processes. Fluorescent reporter dye-labeled TaqMan probes hybridized to the complementary HIV-1 target sequences and RNA-QS sequence undergo hydrolysis during PCR amplification step to generate fluorescent signal detected in 2 different dye channels. Concentration of the HIV-1 RNA in a patient's plasma sample is determined by a ratio of the intensity of the fluorescent dye from the cleaved HIV-1 target sequence probes and that from the RNA-QS target probe detected throughout the PCR process. (Package insert: cobas HIV-1 - Quantitative nucleic acid test for use on the cobas 6800/8800 Systems; Roche Molecular Systems, Inc., Branchburg, NJ; Doc rev. 1.0, 01/2016)

PDF Report

No

Specimen Retention Time

60 days

Performing Laboratory Location

Rochester

Fees & Codes**Test Classification**

This test has been modified from the manufacturer's instructions. Its performance characteristics were 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

87536-HIV-1 Quantification

87901-HIV-1 genotypic drug resistance (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC Value
HIRGT	HIV-1 RNA Reflex Geno PR-RT Resist, P	70241-5

Result ID	Reporting Name	LOINC®
65713	HIV-1 RNA Detect/Quant, P	70241-5