GeneXpert HIV-1 Quantitative assay

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INTRODUCTION

• Cepheid HIV-1 Quant Assay is an automated single cartridge-based quantitative nucleic acid amplification test that is designed to detect Human Immunodeficiency Virus Type 1 (HIV-1) total nucleic acids, using human plasma.

• The GeneXpert Dx System consist of an instrument, personal computer, barcode scanner, and preloaded software for running tests on collected samples and viewing the results.
Types of HIV-1 RNA PCR POC CARTRIDGES
TYPES OF GENEXPERT INSTRUMENTS
GENEXPERT INSTRUMENT

- Loading of instrument
Fig. 3: GeneXpert Gx-IV mode of operation.
CARTRIDGE
• The GeneXpert Dx System integrates and automates sample purification, nucleic acid amplification, and detection of the target sequence in simple or complex samples using real-time reverse transcriptase PCR and results are obtained in 90 minutes.

• This system requires the use of single-use disposable GeneXpert cartridge that holds the RT-PCR reagents and hosts the RT-PCR processes. Cross-contamination between samples is minimized due to the cartridges being self-contained.
• The HIV-1 Quant Assay includes reagents for the detection of HIV-1 RNA in specimens as well as 2 controls used for quantitation of HIV-1 RNA.

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PROCEDURE

• Blood collected in EDTA tube, spin down for 10 minutes at $\leq 1300$ rpm.
• Inspect the test cartridge for damage, if damaged do not use.
• Label the HIV-1 Quant cartridge with the sample ID, taking care not to write on the lid or the 2D barcode on the front of the cartridge.
• Open the lid of the cartridge.
• Transfer 1000uL of the plasma, using the 1 mL transfer pipette provided.
• Close the cartridge lid firmly.
• Starting test
• Before starting test ensure all modules status are functioning and have passed self-test after logging onto the GeneXpert Dx system.

• In the GeneXpert Dx System window, click Create Test. The Scan Sample ID dialog box appears.
• In the **Sample ID box**, scan or type the sample ID. The scan Cartridge Barcode dialog box will then appear.

• Scan the barcode on the HIV-1 Qual cartridge. The Create Test window appears displaying the barcode information of the cartridge automatically.

• Click **Start Test**. Enter your user name and password.

• A green flashing light will indicate will module is going to perform the test. Open the instrument module door with blinking light and load the cartridge.

• Close the module door until you hear the click and the green light stops flashing, but remains green. Indicating that the test has started.
• Once the test is complete after 1 hour 30 minutes, the green light on the door turns off and the system will release the door lock, opening the module door. You can remove the used cartridge and discard the cartridge appropriately.

• The printer will automatically print the result once the test is complete.

• LOD Whole blood- 200-300 cp/ml
• DBS(70ul) - 500 cp/ml
1. Transfer of 1ml plasma into HIV-1 viral load cartridge

2. Scan, insert cartridge and start test

3. Time to result 90 minutes for HIV-1 viral load, GeneXpert®
## Validation

Template #2: one laboratory only interested in validating one 4-module instrument over 5 days.

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<th>GeneXpert Instrument #1</th>
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<td></td>
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Diagnostic Accuracy of the Point-of-care Xpert® HIV-1 Viral Load
Assay in a South African HIV clinic

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2Departments of Global Health, Medicine, and Epidemiology, Schools of Medicine and Public Health, University of Washington, Seattle
3Department of Epidemiology, Columbia University, New York City, USA
• Min of 1000ul of plasma is required for the assay

• EDTA tube may be stored at 15-35 degrees celsius for up to 6 hours or at 2-8 degrees Celsius for up to 72 hours prior to preparation and testing.

• Specimens are stable frozen and can be used for the assay. Need to ensure sample is completely thawed, spin to remove any clots and then introduce into the GeneXpert cartridge.
• Ensure sample is introduced into the cartridge properly with no air bubbles being introduced as well as that will affect the assay.
• Note that most GeneXpert kits currently have short shelf life, will improve as further tests indicating stability of reagents progress.
• Clean and service the GeneXpert according to manufacturers instructions.
• Do not use reagents or cartridges post expiry date.
• Do not open a cartridge lid until ready to perform testing.
• Cartridge should be used within 30 minutes after opening from pouch.
• The assay is very easy to perform.

• A report prints out once the test is complete that includes the primary result.

• The beauty of the GeneXpert POC VL assay in addition to the short turn around time is that
  • you can start different patient tests at different times and
  • also you can run different assays like CT/NG or TV or MTB Rif on the different modules for patients at once.
USERS
Acknowledgements

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Example of Acknowledgements

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