



BIDS RECEIVED



09/06/2013

San Diego County

Roche Diagnostics would like to provide the following information to support your request for bid for an HIV-1 viral load assay system (RFB No. 6130).

Roche HIV-1 viral load tests have been the gold standard for over 17 years. With >75% market share, more than 280 U.S. laboratories, including all major reference laboratories, and 15 million tests performed, the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0 is the most clinically validated viral load assay worldwide.

Currently, 43 US Federal Government institutions, including 32 VAs, utilize Roche systems and assays for viral load monitoring. Within the State of California, there are 5 Public Health labs that utilize the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0.

The following features are unique to the Roche COBAS® AmpliPrep / COBAS® TaqMan® HIV-1 Test versions 2.0:

- The HIV-1 v2.0 Test is Roche's 7<sup>th</sup> generation HIV-1 assay
- The Roche HIV-1 v2.0 Test is designed with dual target amplification – two regions are targeted, the *gag* and LTR regions which are highly conserved and not current drug targets.
- The Roche HIV-1 v2.0 Test is the most sensitive HIV-1 viral load test available with a linear range of 20 copies/mL - 10,000,000 copies/mL

The following features are unique to the COBAS® AmpliPrep / COBAS® TaqMan® system:

- Each lot of reagents is calibrated within the manufacturing process to ensure traceability to the WHO standard. No user calibration is required.
- Reagents are packaged in ready-to-use barcoded cassettes. System software automates tracking of reagent use and stability. No manual reagent preparation is required.
- Physical contamination control measures include closed-tube processing, spatial separation of sample and reagent pipetting and dedicated consumables per sample.
- QS (Quantitation Standard) that acts as an internal control and is co-amplified with the target for the purpose of titer calculations within each reaction.
- Proprietary AmpErase enzyme is included in each reaction to ensure degradation of carry-over amplicon from prior reactions.
- Approved for single room operation.
- The COBAS® AmpliPrep / COBAS® TaqMan® system has sample in and result out capability, including overnight unattended operation.

We appreciate your consideration of this proposal and look forward to your response.

Kind Regards,

Joni Zurawinski,  
Group Marketing Manager, Virology  
Office: 317-521-1839  
Email: [joni.zurawinski@roche.com](mailto:joni.zurawinski@roche.com)

**Roche Diagnostics  
Corporation**

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Indianapolis, IN 46250-0457  
USA

Phone: 800-428-5074



**Table of Contents**

RFB No. 6130.....  
    Roche Response to Statement of Work.....  
    Roche Response to Pricing.....  
    Roche Response to Terms and Conditions.....  
    **Supporting Documents**.....  
        HIV Dual Target Brochure.....  
        HIV-1 Version 2 Package Insert.....



TABLE OF CONTENTS

**SIGN AND RETURN ALL SECTIONS**

**SECTION A REQUEST FOR BID AND GENERAL INFORMATION**

1. REQUEST FOR BID .....A-1  
2. TABLE OF CONTENTS .....A-2&3  
3. REPRESENTATIONS AND CERTIFICATIONS .....A-4  
4. STATEMENT OF WORK .....A-5  
5. PRICE SCHEDULE .....A-6  
6. PUBLIC AGENCY/RENEWAL .....A-7  
7. COUNTY CONTRACTOR PARTICIPATION (JULY 2008) .....A-7  
8. AUTOMATIC CONTRACT RENEWAL .....A-7  
9. CALIFORNIA REVENUE AND TAXATION CODE SECTION .....A-7  
10. FRANCHISE TAX BOARD WEBSITES .....A-7

**SECTION B INSTRUCTIONS FOR COMPLETING REQUEST FOR BIDS**

1. PRICING YOUR BID .....B-1  
2. SUBMITTING YOUR BID .....B-1  
3. EVALUATION AND AWARD .....B-2  
4. PROTEST PROCEDURES .....B-2  
5. LOCAL BUSINESS PREFERENCE .....B-2

**SECTION C STANDARD TERMS AND CONDITIONS**

1. DEFINITIONS .....C-1  
2. DISABLED VETERANS BUSINESS ENTERPRISE (DVBE) PARTICIPATION ENCOURAGED .....C-1  
3. ASSIGNMENT OF RIGHTS, TITLE AND INTEREST .....C-1  
4. CAL OSHA .....C-1  
5. FORMAL BIDS .....C-1  
6. DELIVERY .....C-1  
7. INSPECTION .....C-1  
8. TERMINATION FOR DEFAULT .....C-1&2  
9. TERMINATION FOR CONVENIENCE .....C-2  
10. TITLE .....C-2  
11. VARIATIONS IN SPECIFICATIONS .....C-2  
12. HAZARDOUS SUBSTANCES .....C-2  
13. PROHIBITED CONTRACTS .....C-2&3  
14. ESTIMATED QUANTITIES .....C-3  
15. AVAILABILITY OF FUNDING .....C-3  
16. INSPECTION OF SERVICE/MATERIALS/SUPPLIES .....C-3  
17. DISPUTES .....C-3&4  
18. CHANGES .....C-4  
19. ASSIGNABILITY .....C-4  
20. INDEMNITY .....C-4  
21. CONDUCT OF CONTRACTOR .....C-4  
22. DISALLOWANCE .....C-4  
23. GOVERNING LAW .....C-5  
24. AUDIT AND INSPECTION OF RECORDS .....C-5  
25. PATENT AND COPYRIGHT INFRINGEMENT .....C-5  
26. CONTRACTOR REPRESENTATION .....C-5  
27. WARRANTY .....C-5  
28. INSURANCE .....C-5&6  
29. PERMITS, NOTICES, FEES AND LAWS .....C-6  
30. AIR, WATER POLLUTION CONTROL, SAFETY AND HEALTH .....C-6  
31. FINDINGS CONFIDENTIAL .....C-6  
32. PUBLICATION, REPRODUCTION AND USE OF MATERIAL .....C-6

33. <u>NOTICE</u> .....	C-6
34. <u>PRODUCT IDENTIFICATION AND LABELING</u> .....	C-6
35. <u>DRUG &amp; ALCOHOL FREE WORKPLACE</u> .....	C-7
36. <u>ORDERING WITH BLANKET PURCHASE AGREEMENT</u> .....	C-7
37. <u>INVOICES</u> .....	C-7
38. <u>PAYMENTS AND INVOICES</u> .....	C-7
39. <u>ACCEPTANCE OF COUNTY CREDIT CARD FOR PAYMENT</u> .....	C-7&8
40. <u>FLAMMABILITY AND TOXICITY</u> .....	C-8
41. <u>BRAND NAME OR EQUAL</u> .....	C-8
42. <u>CONTRACT EXTENSION OPTION</u> .....	C-8
43. <u>SEVERABILITY</u> .....	C-8

**County of San Diego  
Department of Purchasing and Contracting  
REPRESENTATIONS AND CERTIFICATIONS**

The following representations and certifications are to be completed, signed and returned with the offer.

**NOT-FOR-PROFIT ORGANIZATIONS**

Attach proof of status and omit Paragraph 3.

**2. INTERLOCKING DIRECTORATE**

In accordance with Board of Supervisors Policy A-79, if Offeror is a non-profit as indicated in paragraph 1 above, Offeror is required to identify any related for-profit subcontractors in which an interlocking directorate, management or ownership relationship exists. By submission of this offer, Offeror certifies it will not enter into a subcontract relationship with a related for-profit entity if Offeror is a non-profit entity. If Offeror is a non-profit and will be subcontracting with a related for-profit entity, Offeror must list the entity(ies) on an attached separate sheet listing them all and the contract must be approved by the Board of Supervisors

**3. BUSINESS REPRESENTATION**

**3.1. REPRESENTATION AS DISABLED VETERANS BUSINESS ENTERPRISE**

"Disabled Veterans Business Enterprise" means a business which is at least fifty-one (51%) owned and operated by one or more veterans with a service related disability as certified by Equal Opportunity Management Office (EOMO), California Department of General Services, Office of Small Business and members of Joint Agencies Contracting Opportunities (JACO), (California Military and Veterans code, Article 6, section 999).

This Offeror represents as a part of this offer that the ownership, operation and control of the business are in accordance with the specific definition in 3.1. I am currently certified by:

Certifying Government Agency: \_\_\_\_\_

Certification #: \_\_\_\_\_

**4. CERTIFICATE REGARDING DEBARMENT, SUSPENSION AND RELATED MATTERS**

Offeror hereby certifies to the best of its knowledge that neither it nor any of its officers:

- 4.1. Are presently debarred, suspended, proposed for debarment, declared ineligible, or voluntarily excluded from covered transactions by any Federal department or agency; and
- 4.2. Have within a three (3) year period preceding this agreement been convicted of or had a civil judgment rendered against them for commission of fraud or criminal offense in connection with obtaining, attempting to obtain, or performing a public (Federal, State, or local) transaction or contract under a public transaction; violation of Federal or State antitrust statutes or commission of embezzlement, theft, forgery, bribery, falsification or destruction of records, making false statements, or receiving stolen property; and
- 4.3. Are presently indicted for or otherwise criminally or civilly charged by a government entity (Federal, State, or local) with the commission of any of the offenses enumerated in paragraph 4.2 of this certification; and

- 4.4. Have within a three (3) year period preceding this agreement had one or more public transactions (Federal, State or local) terminated for cause or default.
- 4.5. Are presently the target or subject of any investigation, accusation or charges by any Federal, State or local law enforcement, licensing or certification body and if they are, the appropriate information is included in the proposal, as requested in the Submittal Requirements.
- 4.6. Contractor will report in writing to the County Department of Purchasing and Contracting within five business days of knowing or have any reason to know any change in status as certified in the preceding paragraphs 4.1 through 4.5, and that occur prior to award (in the case of bids) and contract execution (in the case of negotiated procurements).
- 4.7. Offeror and its proposed subcontractors, agents and consultants have not previously contracted with the County to perform work on this project (e.g. preparing components of the statement of work or plans and specifications for this project). If Offeror or any of its subcontractors, agents or consultants, have previously contracted with the County to perform work on this project, Offeror shall identify those previous agreement(s) and submit that list along with the proposal.

**5. CERTIFICATE OF CURRENT COST OR PRICING**

This is to certify that, to the best of my knowledge and belief, cost and/or pricing data submitted with this offer, or specifically identified by reference if actual submission of the data is impracticable, is/are accurate, complete, and current as of the date signed below.

**6. CERTIFICATE OF INDEPENDENT PRICING**

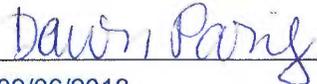
By submission of this offer, each Offeror certifies, and in the case of a joint offers, each party thereto certifies as to its own organization, that in relation to this procurement:

- 6.1. The prices in this offer have been arrived at independently, without consultation, communication, or agreement, for the purpose of restricting competition, as to any matter relating to such prices with other Offeror; with any competitor; or with any County employee(s) or consultant(s) involved in this or related procurements; and
  - 6.2. Unless otherwise required by law, the prices which have been quoted in this offer have not been knowingly disclosed by the Offeror and will not knowingly be disclosed by the Offeror prior to opening, in the case of a bid, or prior to award, in the case of a proposal, directly or indirectly to any other Offeror or to any competitor; and
  - 6.3. No attempt has been made or will be made by the Offeror to induce any other person or firm to submit or not to submit an offer for the purpose of restricting competition.
7. The Offeror understands that prior to receiving a contract award from the County, the Offeror must submit a completed IRS W-9 form to provide a Federal Tax ID number, or if not available, to provide a Social Security Number (SSN).

**CERTIFICATION**

The information furnished in Paragraphs 1 through 7 is certified to be factual and correct as of the date submitted and this certification is made under penalty of perjury under the laws of the State of California.

Name: Dawn Paris

Signature: 

Title: Manager, Contracting

Date: 09/06/2013

Company/Organization: Roche Diagnostics Corporation

**SUBMIT THIS FORM AS DIRECTED IN THE REQUEST FOR SOLICITATION DOCUMENTS OR WITH THE OFFER**

**RFB 6130 - COUNTY OF SAN DIEGO  
HEALTH AND HUMAN SERVICES AGENCY  
PUBLIC HEALTH LABORATORY  
HIV-1 VIRAL LOAD TESTING SYSTEM  
SCOPE OF WORK/PURPOSE**

THE VENDOR SHALL PROVIDE AN INCLUSIVE SYSTEM FOR THE FULLY AUTOMATED TESTING FOR THE HIV-1 VIRAL LOAD IN PLASMA, INCLUDING THE REAGENTS, CONSUMABLES, EQUIPMENT, ANNUAL PREVENTATIVE MAINTENANCE AND REPAIRS (I.E., LABOR AND PARTS). THE PRICE PER TEST FOR AN ESTIMATED 600 TESTS PER MONTH IS TO BE INCLUDED IN THE PRICE OF THE TEST REAGENTS.

**REQUIRED SPECIFICATIONS FOR THE HIV-1 VIRAL LOAD ASSAY SYSTEM**

THE HIV-1 VIRAL LOAD ASSAY SYSTEM MUST: BE A FULLY-AUTOMATED REAL-TIME RT-PCR (IN-VITRO REVERSE TRANSCRIPTION-POLYMERASE CHAIN REACTION (RT-PCR) ASSAY) SYSTEM FOR DETECTION AND QUANTIFICATION OF HUMAN IMMUNODEFICIENCY VIRUS TYPE 1 (HIV-1) WITH A QUANTIFICATION RANGE IN HUMAN PLASMA BETWEEN 40 TO 10,000,000 COPIES/ML. FULL AUTOMATION MEANS THAT THE PLASMA WILL BE SAMPLED DIRECTLY FROM A VACUTAINER-TYPE SAMPLE TUBE WITHOUT TRANSFER TO A SECOND TUBE OR ASSAY CUVETTE. THE SYSTEM MUST BE CAPABLE OF READING SD PHL LIMS BAR-CODE LABELS (CODE 128) DIRECTLY OFF THE SAMPLE TUBE. ONCE THE SAMPLE TUBES ARE LOADED INTO THE SYSTEM, ALL PIPETTING, SAMPLE EXTRACTION, NUCLEIC ACID PURIFICATION AND ASSAY REAGENT AND TEMPLATE HANDLING IS PERFORMED BY THE SYSTEM WITHOUT OPERATOR INTERVENTION.

**BACKGROUND INFORMATION**

THE SAN DIEGO COUNTY PUBLIC HEALTH LABORATORY (PHL), LOCATED AT 3851 ROSECRANS STREET, SUITE 716, SAN DIEGO, CA 92110, IS A COMPREHENSIVE PUBLIC HEALTH REFERENCE LABORATORY FOR THE COUNTY OF SAN DIEGO. THE PHL REQUIRES AN AUTOMATED SYSTEM FOR THE QUANTIFICATION OF HUMAN IMMUNODEFICIENCY VIRUS TYPE 1 (HIV-1) IN HUMAN PLASMA FROM HIV-1 INFECTED INDIVIDUALS OVER THE RANGE OF 40 TO 10,000,000 COPIES/ML. THE APPROXIMATE VOLUME FOR VIRAL LOAD TESTING WOULD BE 600 PATIENT SPECIMENS PER MONTH. REAGENTS WILL BE ORDERED BY THE LABORATORY ON AN AS NEEDED BASIS.

**OBJECTIVES**

- VENDOR SHALL PROVIDE A FULLY-AUTOMATED, FDA-APPROVED, REAL-TIME PCR BASED SYSTEM FOR HIV-1 VIRAL LOAD TESTING AND ALL INSTRUMENTS AND REQUIRED EQUIPMENT UNDER A LEASE/RENTAL AGREEMENT TO INCLUDE ROUTINE SERVICE AND MAINTENANCE FOR THE CONTRACT PERIOD. THE COST OF THIS WILL BE INCLUDED AND QUOTED AS THE COST PER TEST.
- VENDOR SHALL PROVIDE QUOTATION FOR FDA APPROVED REAGENTS INCLUDING CONSUMABLES (TIPS, TRAYS, REAGENT/ASSAY PLATES AND TUBES. ETC.) FOR THE EXPECTED TESTING VOLUME OF 600 SAMPLES PER MONTH.

**SPECIFIC REQUIREMENTS OF THE SYSTEM**

1. EQUIPMENT SHALL BE LOANED OR RENTED TO THE COUNTY OF SAN DIEGO. THE FULL COST OF THE EQUIPMENT, SCHEDULED MAINTENANCE AND REPAIRS (I.E., LABOR AND PARTS) ARE TO BE INCLUDED IN THE PRICE OF THE REAGENTS (SOMETIMES DESCRIBED AS A REAGENT RENTAL AGREEMENT).
2. THE CONTRACT BASE PERIOD WILL BE FOR TWO (2) YEARS WITH OPTIONS TO RENEW FOR YEARS 3 THROUGH 5.
3. ANY AND ALL PARTS, ADDITIONAL ITEMS, OR ACCESSORIES NOT USED ROUTINELY AS PART OF THE ASSAY, OR PROVIDED UNDER THE SERVICE CONTRACT (E.G. SAMPLE TUBE RACKS OR OTHER DURABLE PARTS), FOR THE SYSTEM SHOULD BE PROVIDED AS AN ITEMIZED LIST BY THE VENDOR AS A PERCENT OF DISCOUNT FROM LIST PRICE, SUBMIT LIST ALONG WITH PRICE SHEETS, PAGE A-6 & 6A.
4. THE SYSTEM MUST BE CAPABLE OF FULLY AUTOMATED READING OF BAR-CODE LABELS LABORATORY AND PATIENT BLOOD TUBES (SYMBOLGY CODE 128).
5. FULLY AUTOMATED PRECISION PIPETTING TO ELIMINATE MANUAL MIXING OR MANIPULATION OF SAMPLES OR REAGENTS.
6. SYSTEM MUST BE CAPABLE OF DIRECT SAMPLING OF 5 ML VACUTAINER-TYPE BLOOD COLLECTION TUBES.
7. FLEXIBLE THROUGHPUT OPTIONS OF 24 TO 96 SAMPLES IN ONE 8-HOUR SHIFT.
8. HAVE FDA APPROVAL: THE SYSTEM MUST HAVE FDA APPROVAL FOR HIV-1 VIRAL LOAD TESTING.
9. REGULARLY SCHEDULED PREVENTATIVE MAINTENANCE WILL BE PROVIDED ONCE ANNUALLY DURING REGULAR BUSINESS HOURS OF MONDAY THROUGH FRIDAY, 8:00 A.M. TO 5:00 P.M.
10. REPAIR SERVICE WILL BE AVAILABLE MONDAY THROUGH FRIDAY, 8:00 A.M. TO 5:00 P.M.



**RFB 6130 - COUNTY OF SAN DIEGO  
HEALTH AND HUMAN SERVICES AGENCY PUBLIC HEALTH LABORATORY  
HIV-1 VIRAL LOAD TESTING SYSTEM**

**SCOPE OF WORK/PURPOSE**

THE VENDOR SHALL PROVIDE AN INCLUSIVE SYSTEM FOR THE FULLY AUTOMATED TESTING FOR THE HIV-1 VIRAL LOAD IN PLASMA, INCLUDING THE REAGENTS, CONSUMABLES, EQUIPMENT, ANNUAL PREVENTATIVE MAINTENANCE AND REPAIRS (I.E., LABOR AND PARTS). THE PRICE PER TEST FOR AN ESTIMATED 600 TESTS PER MONTH IS TO BE INCLUDED IN THE PRICE OF THE TEST REAGENTS.

**REQUIRED SPECIFICATIONS FOR THE HIV-1 VIRAL LOAD ASSAY SYSTEM**

THE HIV-1 VIRAL LOAD ASSAY SYSTEM MUST: BE A FULLY-AUTOMATED REAL-TIME RT-PCR (IN-VITRO REVERSE TRANSCRIPTION-POLYMERASE CHAIN REACTION (RT-PCR) ASSAY) SYSTEM FOR DETECTION AND QUANTIFICATION OF HUMAN IMMUNODEFICIENCY VIRUS TYPE 1 (HIV-1) WITH A QUANTIFICATION RANGE IN HUMAN PLASMA BETWEEN 40 TO 10,000,000 COPIES/ML.

**Roche complies with these specifications.**

The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, version 2.0 (HIV-1, v2.0 Test) is an *in vitro* (FDA approved) nucleic acid amplification test (real-time PCR test) for the quantitation of human immunodeficiency virus type 1 (HIV-1) RNA in human plasma.

The HIV-1, v2.0 Test can quantitate HIV-1 RNA over the range of 20 - 10,000,000 copies (cp)/mL. This assay is the only real-time PCR IVD that is calibrated to the WHO International Standard for HIV-1 RNA for Nucleic Acid-Based Techniques (NAT) (NIBSC 97/656).

With a lower limit of quantitation (LOQ) and lower limit of detection (LOD) of 20 copies/mL, the HIV-1, v2.0 test is the most sensitive HIV-1 viral load monitoring test on the market today.

The HIV-1, v2.0 Test is fully automated with sample in, results out capabilities, requiring minimal user interaction. This is only system on the market for HIV-1 viral load monitoring that is FDA approved for use in one room, and in a docked configuration can run unattended overnight.

Primary or secondary specimen tube handling, as well as pipetting of test controls is fully automated on the **cobas** p 630 instrument. Sample extraction is fully automated on the COBAS<sup>®</sup> AmpliPrep Instrument. Amplification and detection are fully automated on the COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer or COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer. Together these instruments form the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> system.

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The HIV-1, v2.0 Test has a unique dual target design, targeting two distinct, highly conserved areas of the HIV-1 genome, the gag and LTR regions. These regions are not currently drug targets and therefore are not under selective drug pressure, which is not the case for other HIV-1 tests on the market today.

The gag region has been a target for Roche HIV-1 assays for 17 years and is therefore well studied and understood in the context of assay design. Similarly, the LTR region has been used in Roche MPx multiplex assays in blood screening applications where accuracy is paramount. Targeting two regions with the HIV-1 genome provides built in redundancy to minimize the impact of mutations that can limit the ability of any real-time PCR assay to accurately quantify the HIV-1 virus.

FULL AUTOMATION MEANS THAT THE PLASMA WILL BE SAMPLED DIRECTLY FROM A VACUTAINER-TYPE SAMPLE TUBE WITHOUT TRANSFER TO A SECOND TUBE OR ASSAY CUVETTE.

**Roche complies with this specification.**

The **cobas p 630** component of the COBAS® AmpliPrep/COBAS® TaqMan® system conducts preanalytical handling of patient samples. An aliquot of plasma required for processing is transferred directly from a vacutainer type sample (primary tube) to racks used in the nucleic acid extraction process.

THE SYSTEM MUST BE CAPABLE OF READING SD PHL LIMS BAR-CODE LABELS (CODE 128) DIRECTLY OFF THE SAMPLE TUBE.

**Roche complies with this specification.**

The COBAS® AmpliPrep/COBAS® TaqMan® System barcode reader is capable of reading and decoding the following barcode formats: Code 2 of 5 interleaved; Code39 (3 of 9 interleaved), Codabar, and Code 128.

ONCE THE SAMPLE TUBES ARE LOADED INTO THE SYSTEM, ALL PIPETTING, SAMPLE EXTRACTION, NUCLEIC ACID PURIFICATION AND ASSAY REAGENT AND TEMPLATE HANDLING IS PERFORMED BY THE SYSTEM WITHOUT OPERATOR INTERVENTION.

**Roche complies with these specifications.**

Samples tubes (racks) are loaded on the COBAS AmpliPrep instrument for sample extraction, nucleic acid purification and assay reagent and template handling (i.e.: PCR reaction set up) without user intervention. Prepared PCR plates can be automatically transferred by the instrument to the COBAS TaqMan analyzer for amplification and detection without user intervention when in the docked configuration. The COBAS AmpliPrep/COBAS TaqMan System is the only viral load monitoring system on the market that has this capability.



In addition, the COBAS AmpliPrep/COBAS TaqMan system can be continuously loaded while in operation to further streamline workflow and turnaround time.

## BACKGROUND INFORMATION

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## OBJECTIVES

- VENDOR SHALL PROVIDE A FULLY-AUTOMATED, FDA-APPROVED, REAL-TIME PCR BASED SYSTEM FOR HIV-1 VIRAL LOAD TESTING AND ALL INSTRUMENTS AND REQUIRED EQUIPMENT UNDER A LEASE/RENTAL AGREEMENT TO INCLUDE ROUTINE SERVICE AND MAINTENANCE FOR THE CONTRACT PERIOD. THE COST OF THIS WILL BE INCLUDED AND QUOTED AS THE COST PER TEST.
- VENDOR SHALL PROVIDE QUOTATION FOR FDA APPROVED REAGENTS INCLUDING CONSUMABLES (TIPS, TRAYS, REAGENT/ASSAY PLATES AND TUBES. ETC.) FOR THE EXPECTED TESTING VOLUME OF 600 SAMPLES PER MONTH.

## SPECIFIC REQUIREMENTS OF THE SYSTEM

1. EQUIPMENT SHALL BE LOANED OR RENTED TO THE COUNTY OF SAN DIEGO. THE FULL COST OF THE EQUIPMENT, SCHEDULED MAINTENANCE AND REPAIRS (I.E., LABOR AND PARTS) ARE TO BE INCLUDED IN THE PRICE OF THE REAGENTS (SOMETIMES DESCRIBED AS A REAGENT RENTAL AGREEMENT).

Roche complies. Please refer to the attached Pricing for additional information.

2. THE CONTRACT BASE PERIOD WILL BE FOR TWO (2) YEARS WITH OPTIONS TO RENEW FOR YEARS 3 THROUGH 5.

Roche complies. Please refer to the attached Pricing for additional information.

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3. ANY AND ALL PARTS, ADDITIONAL ITEMS, OR ACCESSORIES NOT USED ROUTINELY AS PART OF THE ASSAY, OR PROVIDED UNDER THE SERVICE CONTRACT (E.G. SAMPLE TUBE RACKS OR OTHER DURABLE PARTS), FOR THE SYSTEM SHOULD BE PROVIDED AS AN ITEMIZED LIST BY THE VENDOR AS A PERCENT OF DISCOUNT FROM LIST PRICE, SUBMIT LIST ALONG WITH PRICE SHEETS, PAGE A-6 & 6A.

Roche complies. Please refer to the attached Pricing for additional information.

4. THE SYSTEM MUST BE CAPABLE OF FULLY AUTOMATED READING OF BAR-CODE LABELS LABORATORY AND PATIENT BLOOD TUBES (SYMBOLGY CODE 128).

**Roche complies with this specification.**

Bar-code reading is fully automated as patient blood tube labels are read as the rack is loaded into the system. The system software provides full traceability (positive sample ID tracking) from the scanned patient blood tubes through final result reporting.

The COBAS® AmpliPrep/COBAS® TaqMan® System barcode reader is capable of reading and decoding the following barcode formats: Code 2 of 5 interleaved; Code39 (3 of 9 interleaved), Codabar, and Code 128.

5. FULLY AUTOMATED PRECISION PI PETTING TO ELIMINATE MANUAL MIXING OR MANIPULATION OF SAMPLES OR REAGENTS.

**Roche complies with this specification.**

The COBAS® AmpliPrep/COBAS® TaqMan® System is the only system that does not require thawing, mixing, pouring, labeling or any other manual manipulation of reagents or samples prior to loading on the system.

Reagents are provided in pre-filled, ready-to-use, pre-calibrated barcoded cassettes. These are stored at 4°C not frozen. Cassettes are removed from the refrigerator, loaded on racks, and then loaded on the instrument.

The COBAS® AmpliPrep/COBAS® TaqMan® System is the only system that has independent precision pipetting systems, one dedicated to patient samples and processing steps and one that only handles reagents. The reagent pipetting system is monitored by capacitive or electrical level detection. The sample pipetting system has a sensor to detect insufficient volume as well as pressure monitoring of the process for the presence of clots in the patient samples.

6. SYSTEM MUST BE CAPABLE OF DIRECT SAMPLING OF 5 ML VACUTAINER-TYPE BLOOD COLLECTION TUBES.

**Roche complies with this specification.**



Not only is the system capable of direct sampling of 5 mL vacutainer type blood collection tubes, there are a broad range of tube types that are compatible (see Table). This allows for greater flexibility for the laboratory should the type of tube change in the future.

Manufacturer	Primary tube sizes and vacuum volume						
<b>Sarstedt S-Monovette<sup>®</sup></b>	16 x 92 mm 9 ml	15 x 92 mm 7.5 ml	15 x 75 mm 5.5 ml	13 – 90 mm 4.9 ml	13 – 65 mm 2.6 ml	11 – 92 mm 4.0 ml	11 – 66 mm 2.7 ml
<b>Greiner Bio one Vacuette<sup>®</sup></b>	16 x 100 mm 7 – 9 ml	13 x 100 mm 3.5 – 6 ml	13 x 75 mm 1 – 4.5 ml	-	-	-	-
<b>BD Vacutainer<sup>®</sup></b>	16 x 100 mm 10 – 6 ml	13 x 100 mm 7 – 4 ml	13 x 75 mm 5 – 1.8 ml	-	-	-	-

7. FLEXIBLE THROUGHPUT OPTIONS OF 24 TO 96 SAMPLES IN ONE 8-HOUR SHIFT.

**Roche complies with this specification.**

The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> system is the only system that has no minimal batch size requirements and routine runs as low as 9 patient samples per run can be conducted with no reagent waste. In the docked configuration, the system can process up to 147 patient samples per 8 hour shift.

This processing design allows for greater flexibility in workflow for the laboratory and in turnaround time. Even lower volume laboratories have the possibility to report out HIV-1 results daily.

8. HAVE FDA APPROVAL: THE SYSTEM MUST HAVE FDA APPROVAL FOR HIV -I VIRAL LOAD TESTING.

**Roche complies with this specification.**

Please see above for additional details regarding the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1, v2.0 Test.

9. REGULARLY SCHEDULED PREVENTATIVE MAINTENANCE WILL BE PROVIDED ONCE ANNUALLY DURING REGULAR BUSINESS HOURS OF MONDAY THROUGH FRIDAY, 8:00A.M. TO 5:00P.M.

**Roche complies with this specification.**

10. REPAIR SERVICE WILL BE AVAILABLE MONDAY THROUGH FRIDAY, 8:00A.M. TO 5:00P.M.

**Roche complies with this specification.**

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MD

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In addition, Roche has a 24/7/365 Technical Support Hotline to address questions for the laboratory at any time. With optional remote diagnostics capability, Roche technical support specialists can assist laboratory technicians at any time over a secured connection.

RFB 6130  
COUNTY OF SAN DIEGO  
HEALTH AND HUMAN SERVICES AGENCY  
PUBLIC HEALTH LABORATORY  
HIV-1 VIRAL LOAD TESTING SYSTEM  
SECTION A  
PRICING SCHEDULE

**PAGE 6 IS AN EXCEL SPREAD SHEET AND ALL VENDORS MUST COMPLETE. ALL THE VENDOR NEEDS TO DO IS FILL IN THE PRICE PER UNIT FOR THE BASE TERM, FIRST, SECOND AND THIRD OPTION PERIODS AND THE EXCEL SPREADSHEET WILL CALCULATE THE TOTALS. ONCE YOU HAVE THE TOTAL YOU CAN PRINT AND SEND WITH THE OTHER DOCUMENTATION. ALL BIDS MUST BE RECEIVED BY 11 AM ON WEDNESDAY, SEPTEMBER 4, 2013. ALL DOCUMENTS MUST BE SUBMITTED WITH RFB 6130 FOR THE BID TO BE CONSIDERED.**

**AWARD SHALL CONSIST OF MONTHLY RENTAL OF EQUIPMENT, REAGENTS, YEARLY MAINTENANCE AND REPAIR SERVICE AS NEEDED BASED ON A MONTHLY USAGE OF PATIENT SPECIMANS.**

RF8 6130  
HIV-1 VIRAL LOAD  
PRICING SCHEDULE

Item #	Item Description	UOM	Base Term Period: Date of Award thru 31-Aug-2015			First Option Period: 01-Sep-2015 thru 31-Aug-2016			Second Option Period: 01-Sep-2016 thru 31-Aug-2017			Third Option Period: 01-Sep-2017 thru 31-Aug-2018			BASIS OF AWARD	NOTES	
			Est. Monthly Qty	Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier			Est. Extended Price
1	HIV-1 Viral Load Test	EA	600		24	\$0.00		12	\$0.00		12	\$0.00		12	\$0.00	\$0.00	
BASIS OF AWARD TOTAL OF ALL PERIODS - BASE, FIRST OPTION, SECOND OPTION AND THIRD OPTION PERIOD															\$0.00		

SAMPLE

REPLACE THIS SAMPLE WORKSHEET WITH YOUR COMPLETED WORKSHEET

F '0  
HIV-1 . LOAD  
PRICING SCHEDULE

Item #	Item Description	UOM	Est. Monthly Qty	Base Term Period: Date of Award thru 31-Aug-2015			First Option Period: 01-Sep-2015 thru 31-Aug-2016			Second Option Period: 01-Sep-2016 thru 31-Aug-2017			Third Option Period: 01-Sep-2017 thru 31-Aug-2018			BASIS OF AWARD	NOTES
				Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier	Est. Extended Price		
1	HIV-1 Viral Load Test	EA	600	\$35.70	24	\$514,080.00	\$35.70	12	\$257,040.00	\$35.70	12	\$257,040.00	\$35.70	12	\$257,040.00	\$1,285,200.00	
<b>BASIS OF AWARD TOTAL OF ALL PERIODS - BASE, FIRST OPTION, SECOND OPTION AND THIRD OPTION PERIOD</b>															<b>\$1,285,200.00</b>		



San Diego County Public Health/FSS

San Diego, CA 92123

Material #	Description	Package Contents	Contract Price
<b>HIV-1 MONITOR Specific Reagents</b>			
05212308190	COBAS® AmpliPrep/COBAS TaqMan® HIV-1 Test v2.0	1	\$ 1,646.54 *

**Specific Reagents TOTAL**

\* Contract price includes instrument costs

<b>COBAS® AmpliPrep GPRs</b>			
03587797190	System Wash Reagent	5.1 L	\$ 10.83

**Committed GPRs TOTAL**

<b>COBAS® AmpliPrep Consumables</b>			
03137040001	Input Tubes with Barcode Clips	12 x 24	\$ 37.53
03287343001	K-tips	12 x 36	\$ 38.98
03755525001	SPUs	12 x 24	\$ 90.56

<b>COBAS® TaqMan® Consumables</b>			
03137082001	K-tubes, rack	12 x 96	\$ 864.00

**Consumables TOTAL**

Used pricing is for customer consideration and evaluation and remains in effect for 30 days from the date of this exhibit.

All pricing contingent on customer execution of final contract agreeable to Roche Diagnostics.

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San Diego County Public Health/FSS

San Diego, CA 92123

Material #	Description	Package Contents	Contract Price
<b>COBAS® AmpliPrep Ancillary Items</b>			
03286436001	K-carrier Rack	1	\$ 68.00
03286886001	Cassette Opener Tool	1	\$ 0.03
03287335001	K-carrier Rack Labels, 1-8	8	\$ 106.00
04916140001	Sample Rack Barcode Labels, 21-40	20	\$ 42.00
04916166001	Sample Rack Barcode Labels, 41-60	20	\$ 42.00
05165261001	Sample Rack Labels, 101-200	60	\$ 58.00
05165334001	Sample Rack Labels, 61-100	40	\$ 27.00
05166365001	Reagent Rack Labels, 41-100	60	\$ 46.00
05166373001	Reagent Rack Labels, 101-200	100	\$ 58.00
05167221001	Reagent Rack Barcode Labels, 41-100	60	\$ 80.00
05168732001	Reagent Rack Barcode Labels, 101-200	100	\$ 130.00
05182794001	DNA Away, 8 oz	1	\$ 20.00
05325005001	UPS Power Manager	1	\$ 2,200.00
05471664001	SPU Rack w/Cones	1	\$ 20.66
28048355001	Sample Rack Labels, 1-20	20	\$ 25.00
28048398001	Reagent Rack Barcode Labels, 1-20	20	\$ 56.69
28055203001	Sample Rack Labels, 21-40	20	\$ 5.00
28070687001	Reagent Rack Barcode Labels, 21-40	20	\$ 56.69
28073112001	Reagent Rack Labels, 1-20	20	\$ 20.10
28073465001	Reagent Rack Labels, 21-40	20	\$ 22.90
28090262001	Sample Rack Labels, 41-60	20	\$ 5.00
28122172001	Sample Rack	1	\$ 127.00
28122199001	Reagent Rack	1	\$ 58.75
22911001	Syringe, 2.5 mL	1	\$ 277.15
28127328001	UV Tube Light	1	\$ 80.75
28136289001	Sample Rack Barcode Labels, 1-20	20	\$ 55.11
28136815001	Seal Cap Syringes	5	\$ 32.67
28154104001	O-ring Grippers	10	\$ 14.52
28173362001	Reagent Tip	1	\$ 276.51

<b>COBAS® TaqMan® Ancillary Items</b>			
03127575001	Waste Bags	25	\$ 68.15
03132307001	Power Supply Filter	1	\$ 8.48
03279995001	K-carrier Labels, 1-25	25	\$ 92.00
03287696001	K-carrier Holders	2	\$ 58.73
03339874001	K-tube Capper, manual	1	\$ 49.30
03517519001	K-carrier Transporter	1	\$ 82.05
05070856001	Halogen Lamp	1	\$ 270.00
28035792001	Fan Filter	1	\$ 6.86
28150397001	K-carrier	1	\$ 187.64
28171815001	Plate Rack	1	\$ 3,400.00

Proposed pricing is for customer consideration and evaluation and remains in effect for 30 days from the date of this exhibit.

All pricing contingent on customer execution of final contract agreeable to Roche Diagnostics.

Roche Diagnostics Corporation: Confidential information - Do not copy or distribute.



San Diego County Public Health/FSS

San Diego, CA 92123

Material #	Description	Package Contents	Contract Price
<b>COBAS® AMPLICOR® Ancillary Items</b>			
28108285001	Waste Reservoir, 10L	1	\$ 114.08
<b>cobas® p 630 Ancillary Items</b>			
05547431001	Rack Adapter	6	\$ 1,582.47
05547440001	Rack Control	2	\$ 2,156.74
<b>Ancillary Items TOTAL</b>			

AMPLICOR, AMPLIPREP, COBAS and TAQMAN are trademarks of Roche. All other names and trademarks are the property of their respective owners.

**Proposed pricing is for customer consideration and evaluation and remains in effect for 30 days from the date of this exhibit.**

**All pricing contingent on customer execution of final contract agreeable to Roche Diagnostics.**

**Roche Diagnostics Corporation: Confidential information - Do not copy or distribute.**

**PUBLIC AGENCY PARTICIPATION (July 2008)**

It is intended that any other public agency (i.e., city, district, public authority, public agency, municipality and other political sub-division or public corporation of California) located in San Diego County shall have the option to participate in any award made as a result of this solicitation. Any agency located outside of San Diego County shall have the option to participate, but shall incur all freight charges from location of awarded vendor to delivery point. The County of San Diego shall incur no financial responsibility in connection with orders issued under the authority of this provision or in making payments to the vendor.

**COUNTY CONTRACTOR PARTICIPATION (July 2008)**

It is intended that any educational institution or non profit organization that is currently under contract with the County of San Diego to provide direct support to the County with reimbursement for such support coming directly from the County shall have the option to participate in any award made as a result of this solicitation. The contractor agrees to provide the items called for in the schedule of this contract to educational institutions or non profit organization under the authority of this provision. The contractor is responsible for confirming that any educational institution or non profit organization has a current contract with the County of San Diego. The County shall incur no financial responsibility in connection with orders issued under the authority of this provision. The ordering organization shall be solely responsible for verifying they are currently under contract with the County, placing orders, and making payments to the contractor.

**AUTOMATIC CONTRACT RENEWAL (July 2008)**

Unless County notifies Contractor in writing, not less than 30 days prior to the expiration date that they do not intend to renew the Agreement, the Agreement will be automatically renewed for another year. Term not to exceed August 31, 2018.

**WINNING AWARD WILL BE REQUIRED TO COMPLY WITH THE FOLLOWING:**

**CALIFORNIA REVENUE AND TAXATION CODE SECTION 18662.**

In compliance with California Revenue and Taxation code section 18662, if you are a non resident of California (out-of-state invoices) who receives California source income, the County will pay California Use Tax directly to the State of California per permit no. SR FH 25-632384. Fifteen (15) business days prior to the first payment, new suppliers or suppliers with expired forms or forms with incorrect information, must submit new forms to the County (forms are available from the Franchise Tax Board website listed below).

Under certain circumstances you may be eligible for reduced or waived nonresident withholding. If you have already received a waiver or a reduced withholding response from the State of California and the response is still valid, submit the response to the County in lieu of the forms. Failure to submit the required forms will result in withholding of payments. Refer to the Franchise Tax Board websites (listed below) for tax forms and information on nonresident withholding, including waivers or reductions. The County will not give you any tax advice. It is recommended you speak with your tax adviser and/or the State of California for guidance.

**FRANCHISE TAX BOARD WEBSITES:**

<http://www.ftb.ca.gov>

[http://www.ftb.ca.gov/individuals/Withholding\\_Definitions.shtml](http://www.ftb.ca.gov/individuals/Withholding_Definitions.shtml)

[http://www.ftb.ca.gov/individuals/wsc/Processing\\_Changes\\_for\\_2010.shtml](http://www.ftb.ca.gov/individuals/wsc/Processing_Changes_for_2010.shtml)

[http://www.ftb.ca.gov/individuals/wsc/forms\\_and\\_publications.shtml](http://www.ftb.ca.gov/individuals/wsc/forms_and_publications.shtml)

[http://www.ftb.ca.gov/individuals/wsc/decision\\_chart.shtml](http://www.ftb.ca.gov/individuals/wsc/decision_chart.shtml)

**RFB 6003**  
**SECTION B**  
**COUNTY OF SAN DIEGO'S**  
**INSTRUCTIONS FOR COMPLETING REQUEST FOR BID**  
**AND PRE-AWARD REQUIREMENTS**

Rev 01/04

**1. PRICING YOUR BID**

- 1.1 Bid on each item separately. Prices should be stated per unit(s) specified herein.
- 1.2 Unless otherwise specified, all prices shall be F.O.B. destination. Bids other than F.O.B. destination shall be considered non-responsive and will be rejected. Prices shall include all freight charges.
- 1.3 Unless otherwise specified, prices bid herein should **NOT** include California sales/use tax or Federal excise tax. The County generally is required to pay California sales/use tax, and it should be shown as a **separate item** on invoices. The County is exempt from payment of Federal excise tax. It must **NOT** be included in invoices.
- 1.4 All prices and notations must be in ink or typewritten. No erasures permitted. Mistakes may be crossed out and corrected and must be initialed in ink by person(s) signing the bid.
- 1.5 Discounts of less than thirty- (30) days will not be considered in evaluation of bids to determine overall apparent low bidder.
- 1.6 Net terms of less than 30 days will not be accepted.

**2. SUBMITTING YOUR BID**

- 2.1 Each bid must be in a separate sealed envelope **WITH BID NUMBER ON THE OUTSIDE** and must be delivered to the County Purchasing and Contracting Department, Front Desk (where it will be time stamped to indicate time of receipt), 5560 Overland Avenue, Suite 270, San Diego, California 92123, by 11:00 a.m. on the day specified. Bids will be publicly opened at that time.
- 2.2 Failure to bid on authorized County form may be cause for rejection of bid.
- 2.3 Any bid received at the County Purchasing and Contracting Department after the exact time for receipt will not be considered and will be rejected as a late bid.
- 2.4 Late bids will be returned to the bidder unopened unless it is determined that the late receipt was due solely to mishandling by the Purchasing and Contracting Department and such determination is made prior to award.
- 2.5 The County's primary means of providing bids and addenda is the County BuyNet Internet website:
- 2.6 No oral interpretation shall be made to modify any provisions of any bid specifications. Requests for an interpretation shall be made in writing to the County Director of Purchasing and Contracting prior to bid opening and a written response will be posted on the County BuyNet website.
- 2.7 Any vendor desiring to withdraw its bid must do so before County bid opening. If there are any questions or comments relative to technicalities of the bid, they must be submitted in writing to County of San Diego, Director of Purchasing and Contracting, within 24 hours after bid opening.
- 2.8 Bids submitted in response to this Request for Bid must be in full conformance with the terms and conditions set forth herein. Further, all specification requirements must be met unless the language of the Request for Bid specifically indicates alternate specifications will be considered.
- 2.9 Samples of items, when required, must be furnished free of expense to the County, and if not destroyed by tests will, upon request, be returned at the bidder's expense.
- 2.10 All bids must be signed with the firm name and by an authorized officer or employee. Obligations assumed by such signature must be fulfilled.

3. **EVALUATION AND AWARD**

- 3.1 Bids are subject to acceptance at any time within 30 days after opening of same, unless otherwise stipulated by the County.
- 3.2 In determining the lowest bid, discounts of 30 days or greater will be considered. Discounts will be calculated from receipt and acceptance of merchandise or invoice, whichever is later.
- 3.3 Award will be made by the Department of Purchasing and Contracting as stated on the cover/pricing page to the lowest responsive, responsible bidder.
- 3.4 The County reserves the right to waive a variation in specification if, in the opinion of the County, such variation does not materially change the item or its performance within parameters acceptable to the County.
- 3.5 The County reserves the right to reject any or all bids and to accept or reject any item(s) thereon, or waive any informality in the bid.
- 3.6 In the event of a conflict between unit price bid and bidder's extended price, the unit price will prevail unless price is so obviously unreasonable as to indicate an error. In that event, the bid will be rejected as non-responsive for the reason of inability to determine the intended bid.
- 3.7 The County reserves the right to perform a pre-award survey of the bidder to determine capability to perform, including but not limited to facilities, financial responsibility, materials/supplies, and past performance. The determination of the County as to the bidder's prospective ability to perform the contract shall be conclusive.

4. **PROTEST PROCEDURES**

Any protest resulting from this procurement is to be processed as prescribed in Board of Supervisors' Policy A-97, Protest Procedures for Award of Contracts. All protests shall be in writing, be made **prior** to Award, and be made only by an offeror. Such protests shall clearly state the ground for the protest and the relief sought. Protests shall be filed with the County's contracting office identified in the solicitation package.

Whenever a contract is contemplated to be awarded to other than the low bidder in a formally advertised procurement, the low bidder shall be so notified five working days prior to award, in addition to the posting of the proposed award in a public place in the Contracting Office for the same period of time. Copies of Policy A-97 are available upon request from the Clerk of the Board, 1600 Pacific Highway, San Diego, CA 92101 or via the County of San Diego's Internet website: <http://www.co.san-diego.ca.us/cob/policy/index.html>

5. **LOCAL BUSINESS PREFERENCE**

Responsive bids from responsible local San Diego County businesses shall be given preference for award over bids received from non-local businesses. "Local Business" is defined as a business with a valid license issued by a city within the County, employing San Diego residents, and with a verifiable address within the County, or a business employing San Diego residents and with a verifiable address in an unincorporated area of the county. Post Office Boxes do not qualify as verifiable local business addresses. If a tie bid occurs between a local business and a non-local business, award shall be made to the local business.

If the lowest responsive, responsible bid is submitted by a non-local business, one percent (1%) shall be subtracted from the lowest responsive, responsible bid submitted by a local business in evaluating the bids for award. If application of the one percent (1%) factor results in the local business bid being equal to or lower than the non-local business bid, contract award will be made to the local business at the local business bid price, except for public works and construction bids, or if prohibited by State or Federal law.

**RFB 6003 - SECTION C  
TERMS & CONDITIONS OF REQUEST FOR BIDS  
AND RESULTANT CONTRACT OR PURCHASE ORDER**

**1. DEFINITIONS**

"County" shall mean The County of San Diego, California

"Offeror" shall mean any person, firm, partnership, or corporation submitting a proposal to County in response to this solicitation.

"Contractor" shall mean the offeror whose proposal is accepted by County and who has entered into an agreement with County to provide the equipment and services described herein.

"Vendor" shall mean the same as contractor.

**2. DISABLED VETERANS BUSINESS ENTERPRISE PARTICIPATION ENCOURAGED (Rev. 11/97)**

County Board of Supervisor's policies B-53 and B-39 A encourages the participation of small and Disabled Veterans Business Enterprises (DVBE) in County procurement. Section A of this solicitation (Representations & Certifications) contains a description of the County's requirements to qualify as an (DVBE). Perspective (DVBE) bidders/offerors are encouraged to contact the Contracting Office representative listed on the face of this Request for Bid (RFB) or Request for Proposal (RFP) for information concerning the County's procurement procedures.

**3. ASSIGNMENT OF RIGHTS, TITLE AND INTEREST**

In submitting a bid to a public purchasing body, the vendor offers and agrees that if the bid is accepted, it will assign to the purchasing body all rights, title and interest in and to all causes of action it may have under Section 4 of the Clayton Act (15 U.S.C. Sec. 15) or under the Cartwright Act (Chapter 1 (commencing with Section 16700) of Part 2 of Division 7 of the Business and Professions Code), arising from purchases of goods, materials, or services by the bidder for sale to the purchasing body pursuant to the bid. Such assignment shall be made and become effective at the time the purchasing body tenders final payment to the vendor.

**4. CAL OSHA**

As applicable, all items furnished under this bid shall meet or exceed the standards established by the California Occupational Safety and Health Act of 1973 and current amendments thereto, provided the end use of the item is for the purpose for which the item is intended.

**5. FORMAL BIDS**

In the event this bid results in a purchase order, terms and conditions of this bid are incorporated herein and from a part of the purchase order. In the event of any conflict or inconsistency between the terms of the formal bid or award, the terms of this formal bid shall control.

**6. DELIVERY**

Time is of the essence, and the purchase order is subject to termination for failure to deliver on time. The acceptance by buyer of late performance with or without objection or reservation shall not waive the right to claim damage for such breach nor constitute a waiver of the requirements for the timely performance of any obligation remaining to be performed by the vendor.

**7. INSPECTION**

All items or services are subject to final inspection and acceptance at designation by the County. Such final inspection shall be made within a reasonable time after delivery.

**8. TERMINATION FOR DEFAULT**

The County may, by written notice of default to the vendor, terminate any resulting order in whole or in part should the vendor fail to make satisfactory progress, fail to deliver within time specified therein or fail to deliver in strict conformance to

specifications and requirements set forth therein. In the event of such termination, the County reserves the right to purchase or obtain the supplies or services elsewhere, and the defaulting vendor shall be liable for the difference between the prices set forth in the terminated order and the actual cost thereof to the County. The prevailing market price shall be considered the fair repurchase price.

- 8.1. If, after notice of termination of this contract under the provisions of this clause, it is determined for any reason that the Contractor was not in default under this provisions of this clause, the rights and obligations of the parties shall, if the contract contains a clause providing for termination for convenience of the County, be the same as if the notice of termination had been issued pursuant to such clause.
- 8.2. The rights and remedies of County provided in this article shall not be exclusive and are in addition to any other rights and remedies provided by law or under resulting order.

#### 9. TERMINATION FOR CONVENIENCE

The County may, by written notice stating the extent and effective date, terminate any resulting order for convenience in whole or in part, at any time. The County shall pay the vendor as full compensation for performance until such termination:

- 9.1. The unit or pro rata price for the delivered and accepted portion.
- 9.2. A reasonable amount, as costs of termination, not otherwise recoverable from other sources by the vendor as approved by the County, with respect to the undelivered or unaccepted portion of the order, provided compensation hereunder shall in no event exceed the total price.
- 9.3. In no event shall the County be liable for any loss of profits on the resulting order or portion thereof so terminated.
- 9.4. The rights and remedies of County provided in this article shall not be exclusive and are in addition to any other rights and remedies provided by law or under resulting order.

#### 10. TITLE

Title to the material and supplies purchased shall pass directly from vendor to County at the F.O.B. point shown, subject to the right of County to reject upon inspection.

#### 11. VARIATIONS IN SPECIFICATIONS

The County reserves the right to waive a variation in specification if, in the opinion of the County, such variation does not materially change the item or its performance within parameters acceptable to the County.

#### 12. HAZARDOUS SUBSTANCES (July 2008)

If any product being delivered or supplied to the County under this contract/purchase order is listed in the Hazardous Substances List of the Regulations of the Director of Industrial Relations with the California Occupational Safety and Health Standards Board, or if the product presents a physical or health hazard as defined in the California Code of Regulations, General Industry Safety Order, Section 5194 (T8CCR), Hazard Communication, then the contractor must include a Material Safety Data Sheet (MSDS) with delivery, or shipment. Each MSDS must reference the contract/purchase order number, and identify the "Ship To Address". All shipments and containers must comply with the labeling requirements of Title 49, Code of Federal Regulations by identifying the hazardous substance, name and address of manufacturer, and appropriate hazard warning regarding potential physical safety and health hazard. (County of San Diego Administrative Manual, 0300-02, Hazard Communication Program).

No product which is manufactured with fully halogenated chlorofluorocarbons (CFC) shall be delivered or supplied, or used on a job site in performance of this contract/purchase order unless specifically described in the stated requirements of this contract/purchase order or otherwise explicitly authorized by the County Director, Purchasing and Contracting.

#### 13. PROHIBITED CONTRACTS

Section 67 of the San Diego County Administrative Code provides that the County shall not contract with, and shall reject any bid or proposal submitted by the person or entities specified below, unless the Board of Supervisors finds that special circumstances exist which justify the approval of such contract:

- 13.1. Persons employed by the County or of public agencies for which the Board of Supervisors is the governing body;
- 13.2. Profit-making firms or businesses in which employees described in sub-section (a) of code serve as officers, principals, partners, or major shareholders;
- 13.3. Persons who, within the immediately preceding twelve (12) months came within the provisions of the above sub-section and who (1) were employed in positions of substantial responsibility in the area of service to be performed by the contract, or (2) participated in any way in developing the contract or its service specifications; and
- 13.4. Profit-making firms or businesses in which the former employees described in sub-section 16.3 of code serve as officers, principals, partners, or major shareholders.

With the affixing of a signature to your response to this solicitation, offeror certifies that the above provisions of the Code have been complied with, and that any exception will cause any ensuing contract to be invalid.

14. **ESTIMATED QUANTITIES** (March 1993)

The Estimated Quantities in Section "A", Pricing Schedule, are provided solely for evaluation of bids. They represent approximate anticipated use based on historical consumption. If the County's actual requirements do not result in orders in the quantities described as "estimated" in the Schedule, that fact shall not constitute the basis for price adjustment.

15. **AVAILABILITY OF FUNDING**

The County's obligation for payment of any contract beyond the current fiscal year end is contingent upon the availability of funding from which payment can be made. No legal liability on the part of the County shall arise for payment beyond June 30 of the calendar year unless funds are made available for such performance.

16. **INSPECTION OF SERVICE/MATERIALS/SUPPLIES**

- 16.1. All performance (which includes services, materials, supplies and equipment furnished or utilized in the performance of this contract, and workmanship in the performance of services) shall be subject to inspection and test by the County at all times during the term of the contract. The Contractor shall provide adequate cooperation to any inspector assigned by the County to permit the inspector to determine the Contractor's conformity with these specifications and the adequacy of the services being contractually provided. All inspection by the County shall be made in such a manner as not to unduly interfere with Contractor performance.
- 16.2. If any services performed hereunder are not in conformity with the specifications and requirements of this contract, the County shall have the right to require the Contractor to perform the services in conformity with said specifications and requirements at no additional increase in total contract amount. When the services to be performed are of such nature that the difference cannot be corrected, the County shall have the right to (1) require the Contractor immediately to take all necessary steps to ensure future performance of the services in conformity with requirements of the contract, and (2) reduce the contract price to reflect the reduced value of the services performed. In the event the Contractor fails to perform the services promptly or to take necessary steps to ensure future performance of the service in conformity with the specifications and requirements of the contract, the County shall have the right to either (1) by contract or to otherwise have the services performed in conformity with the contract specifications and charge to the Contractor any cost occasioned to the County that is directly related to the performance of such services, or (2) terminate this contract for default as provided in the Termination clause.

17. **DISPUTES**

- 17.1. Except as otherwise provided in this contract, any dispute concerning a question of fact arising under this contract which is not disposed of by agreement shall be decided by the Contracting Officer who shall furnish the decision to the Contractor in writing. The decision of the Contracting Officer shall be final and conclusive unless determined by the court of competent jurisdiction to have been fraudulent or capricious, or arbitrary, or so grossly erroneous as necessarily to imply bad faith. The Contractor shall proceed diligently with the performance of the contract pending the Contracting Officer's decision.
- 17.2. The "Disputes" clause does not preclude consideration of legal questions in connection with decisions provided

for in paragraph (A) above. Nothing in this contract shall be construed as making final the decision of any administrative official, representative, or board on a question of law.

#### **18. CHANGES**

The Contracting Officer may at any time, by written order, make changes within the general scope of this contract, in the definition of services to be performed, and the time (i.e., hours of the day, days of the week, etc.) and place of performance thereof. If any such change causes an increase or decrease in the cost of, or the time required for the performance of any part of the work under this contract, whether changed or not changed by any such order, an equitable adjustment shall be made in the contract price or delivery schedule, or both, and the contract shall be modified in writing accordingly. Any claim by the Contractor for adjustment under this clause must be asserted within 30 days from the date of receipt by the Contractor of the notification of change; provided however, that the Contracting Officer, if he decides that the facts justify such action, may receive and act upon any such claim asserted at any time prior to final payment under this contract. Where the cost of property made obsolete or excess as a result of a change is included in the Contractor's claim for adjustment, the Contracting Officer shall have the right to prescribe the manner of disposition of such property. Failure to agree to any adjustment shall be a dispute concerning a question of fact within the meaning of the clause of this contract entitled "Disputes". However, nothing in this clause shall excuse the Contractor from proceeding with the contract as changed.

#### **19. ASSIGNABILITY**

The Contractor shall not assign any interest in this contract, and shall not transfer any interest in the same (whether by assignment or novation), without the prior written consent of the County thereto; provided however, that claims for money due or to become due to the Contractor from the County under this contract may be assigned without such approval. Notice of any such assignment or transfer shall be furnished promptly to the County.

#### **20. INDEMNITY**

County shall not be liable for, and Contractor shall defend and indemnify County and the employees and agents of County collectively, "County Parties") against any and all claims, demands, liability, judgments, awards, fines, mechanics' liens or other liens, labor disputes, losses, damages, expenses, changes or costs of any kind or character, including attorneys' fees and court costs (hereinafter collectively referred to as "Claims"), related to or arising out of this contract, and arising either directly or indirectly from any act, error, omission or negligence of Contractor or its subcontractors, licensees, agents, servants or employees, including Claims caused by the concurrent negligent act, error or omission, whether active or passive, of County Parties. However, Contractor shall have no obligation to defend or indemnify County Parties from a Claim if it is determined by a court of competent jurisdiction that such Claim was caused by the sole negligence or willful misconduct of County Parties.

#### **21. CONDUCT OF CONTRACTOR**

- 21.1. The Contractor agrees to inform the County of all the Contractor's interests, if any, which are or which the Contractor believes to be incompatible with any interests of the County.
- 21.2. The Contractor shall not, under circumstances which might reasonably be interpreted as an attempt to influence the recipient in the conduct of his duties, accept any gratuity or special favor from individuals or organizations with whom the Contractor is doing business or proposing to do business, in accomplishing the work under the contract.
- 21.3. The Contractor shall not use for personal gain or make other improper use of privileged information which is acquired in connection with his employment. In this connection, the term "privileged information" includes, but is not limited to, unpublished information relating to technological and scientific development; medical, personnel, or security records of the individuals; anticipated materials requirements or pricing actions; and knowledge of selections of contractors or subcontractors in advance of official announcement.
- 21.4. The Contractor or employees thereof shall not offer gifts, gratuity, favors, entertainment directly or indirectly to County employees.

#### **22. DISALLOWANCE**

In the event the Contractor receives payment for services under this contract which is later disallowed by the County, the Contractor shall promptly refund the disallowed amount to the County on request, or at its option, the County may offset the amount disallowed from any payment due to the Contractor under any contract with the County.

**23. GOVERNING LAW**

This contract shall be construed and interpreted according to the laws of the State of California.

**24. AUDIT AND INSPECTION OF RECORDS**

- 24.1. General. The County shall have the audit and inspection rights described in this section.
- 24.2. Cost or pricing data. If the Contractor submitted cost or pricing data in connection with the pricing of this contract or any change or modification thereto, unless such pricing was based on adequate price competition, established catalog or market prices of commercial items sold in substantial quantities of the general public, or prices set by law or regulation, the Contracting Officer or his representatives who are employees of the County or its agent shall have the right to examine all books, records, documents and other data of the Contractor related to the negotiation pricing or performance of such contract, change or modification, for the purpose of evaluating the accuracy, completeness and currency of the cost or pricing data submitted.
- 24.3. Availability. The materials described above shall be made available at the office of the Contractor, at all reasonable times, for inspection, audit or reproduction, until the expiration of 3 years from the date of final payment under this contract, or by (1) and (2) below:
  - 24.3.1. If this contract is completely or partially terminated, the records relating to the work terminated shall be made available for a period of three years from the date of any resulting final settlement.
  - 24.3.2. Records which relate to appeals under the "Disputes" clause of this contract, or litigation or the settlement of claims arising out of the performance of this contract, shall be made available until such appeals, litigation, or claims have been disposed of, or three years after contract completion, whichever is longer.
- 24.4. The Contractor shall insert a clause containing all the provisions of this entire clause in all subcontracts hereunder except altered as necessary for proper identification of the contracting parties and the contracting officer under the County's prime contract.

**25. PATENT AND COPYRIGHT INFRINGEMENT**

The contractor shall report to the contracting officer, promptly and in reasonable written detail, each notice or claim of patent or copyright infringement based on the performance of this contract of which the contractor has knowledge.

**26. CONTRACTOR REPRESENTATION**

Unless the contractor expressly states otherwise in his proposal, where functional requirements are expressly stated as part of the requirements of this solicitation, the contractor, by responding, represents that in its opinion the system proposed is capable of meeting those requirements. In the event of any inconsistency between the functional specifications and the detailed specifications contained in the solicitation, the former will control.

**27. WARRANTY**

Contractor agrees that the equipment, supplies or services to be furnished shall be covered by the most favorable commercial warranties the contractor gives to any customer for the same or substantially similar equipment, supplies or services and that the rights and remedies so provided are in addition to and do not limit any rights afforded to County.

**28. INSURANCE**

Within 10 working days prior to the inception of the contract Contractor shall submit to County certificates of insurance and appropriate separate endorsements to the actual insurance policy, evidencing that the Contractor has obtained for the period of the Contract, at its sole expense, insurance in the following forms of coverage and minimum amounts specified from insurance carriers with a Best's Rating of not less than A-, VII or a company of equal financial stability approved in writing by County's Risk Management Division.

- a. An occurrence policy of Commercial General Liability insurance insuring Contractor against liability for bodily injury, personal injury or property damage arising out of or in connection with the Contractor's performance of work or service under this Contract of not less than \$1,000,000 per occurrence and \$2,000,000 general aggregate. The County of San Diego, its officers, agents, employees, and volunteers shall be added as Additional Insured by separate endorsement to the policy.
- b. Statutory Workers' Compensation, as required by State of California and Employer's Liability at \$1,000,000 each accident for bodily injury or disease. Coverage shall include waiver of subrogation endorsement in favor of County of San Diego.
- c. Comprehensive Automobile Liability covering all owned, non-owned and hired vehicles for bodily injury and property damage of not less than \$1,000,000 each accident.
- d. Certificates of insurance provided by Contractor must evidence that the insurer providing the policy will provide notice of any cancellation, lapse, reduction or other adverse change respecting such insurance in accordance with policy provisions.

The County of San Diego shall retain the right to review the coverage, form and amount of insurance required herein and may require Contractor to obtain insurance reasonably sufficient in coverage, form and amount to provide adequate protection against the kind and extent of risk which exists at the time a change in insurance is required. County requirements shall be reasonable. County retains the right to demand a certified copy of any insurance policy required herein after 15 days notice.

#### **29. PERMITS, NOTICES, FEES AND LAWS**

The contractor shall, at contractor's expense, obtain all necessary permits and licenses, give all necessary notices, pay all fees required by law, and comply with all laws, ordinances, rules and regulations relating to work and to the preservation of the public health and safety.

#### **30. AIR, WATER POLLUTION CONTROL, SAFETY AND HEALTH**

Contractor shall comply with all air pollution control, water pollution, Safety and Health Ordinances and statutes which apply to the work performed pursuant to this contract, including any requirements specified in state government codes.

#### **31. FINDINGS CONFIDENTIAL**

Any reports, information, data, etc., given to or prepared or assembled by the Contractor under this Agreement which the County requests to be kept as confidential shall not be made available to any individual or organization by the Contractor without the prior written approval of the County.

#### **32. PUBLICATION, REPRODUCTION AND USE OF MATERIAL**

No material produced, in whole or in part, under this Agreement shall be subject to copyright in the United States or in any other country. The County shall have unrestricted authority to publish, disclose, distribute and otherwise use, in whole or in part, any reports, data or other materials prepared under this Agreement. All reports, data and other materials prepared under this Agreement shall be the property of the County upon completion of this Agreement.

#### **33. NOTICE**

Any notice or notices required or permitted to be given pursuant to this Agreement may be personally served on the other party by the party giving such notice, or may be served by certified mail, return receipt requested, to the addresses set forth herein.

#### **34. PRODUCT IDENTIFICATION AND LABELING**

Each package shall be identified with manufacturer's label, which shall conform to the requirements of the Fair Packaging and Labeling Act and Section 12604 of the California Business and Professions Code.

### **35. DRUG & ALCOHOL FREE WORKPLACE**

The County of San Diego, in recognition of individual rights to work in a safe, healthful and productive work place, has adopted a requirement for a drug and alcohol free work place, County of San Diego Drug and Alcohol Use Policy C-25. This policy provides that all County employed Contractors and Contractor employees shall assist in meeting this requirement.

- 35.1. As a material condition of this agreement, the Contractor agrees that the Contractor and the Contractor employees, while performing service for the County, on County property, or while using County equipment:
  - 35.1.1. Shall not be in any way impaired because of being under the influence of alcohol or a drug.
- 35.2. Shall not possess an open container of alcohol or consume alcohol or possess or be under the influence of an illegal drug.
  - 35.2.1. Shall not sell, offer, or provide alcohol or a drug to another person.
  - 35.2.2. Shall not be applicable to a Contractor or Contractor employee who, as part of the performance of normal job duties and responsibilities prescribes or administers medically prescribed drugs.
- 35.3. The Contractor shall inform all employees that are performing service for the County on County property or using County equipment, of the County objective of a safe, healthful and productive work place and the prohibition of drug or alcohol use or impairment from same while performing such service for the County.
- 35.4. The County may Terminate for Default or Breach this Agreement and any other Agreement the Contractor has with the County, if the Contractor, or Contractor employees are determined by the Contracting Officer not to be in compliance with the above.

### **36. ORDERING WITH BLANKET PURCHASE AGREEMENT**

A blanket purchase agreement for the estimated requirements will be sent to the successful bidder. This will authorize the acceptance of releases from designated County departments for their requirements. The vendor shall complete delivery of items ordered to destinations set forth in the release. Each release shipment shall be accompanied by a priced invoice itemizing all material. Partial shipments are not acceptable when ordered by release.

### **37. INVOICES**

All deliveries must be accompanied by invoices or delivery tickets. A copy of each invoice or delivery ticket must be signed by the individual accepting delivery. Invoices shall include item, description, quantity, delivery point, price, terms, purchase order number, release number (if applicable to a blanket purchase agreement) and any data relative to the shipment. Original invoices shall be mailed to the County address as specified in the purchase order or blanket purchase agreement release. Discounts will be calculated from receipt of merchandise or invoice, whichever is later.

### **38. PAYMENTS AND INVOICES**

The County is precluded from making payments prior to receipt of service or supplies (advance payments). The County will accept invoice for progress payments and if approved reimburse the Contractor up to 90% of the value of goods received. Invoice subject to following:

Original invoices will be submitted monthly, or at the completion of each phase or task, or at contract completion to the address specified in the purchase order or blanket purchase agreement release.

- 38.1. A copy of the invoice shall be submitted to the Contracting Officer's Technical Representative (COTR).
- 38.2. The invoice must specify items and deliverables for all items described in the "Statement of Work".
- 38.3. Payment shall be Net 30 Days from receipt and approval of invoice unless otherwise stated. Discounts will be calculated from receipt of merchandise or invoice, whichever is later.

### **39. ACCEPTANCE OF COUNTY CREDIT CARD FOR PAYMENT**

- 39.1. Orders may be paid using the County of San Diego credit card (VISA).

39.2. For your bid to be considered responsive, you must accept County of San Diego credit card for payment.

39.3. Pricing remains the same whether paid by credit card or check.

40. **FLAMMABILITY AND TOXICITY**

Materials furnished under this order must meet or exceed minimum California State Fire Marshal's standard for flammability and toxicity for institutional fabrics. Vendor shall provide evidence of California Marshal's test results and approval number.

41. **BRAND NAME OR EQUAL**

41.1. If items called for in this Request for Bids have been identified in the schedule by a "brand name or equal" description; such identification is intended to be descriptive, but not restrictive, and is to indicate the quality and characteristics of products (including products of the brand name manufacturer other than the one described by the brand name) will be considered for award if such products are determined by the County to meet fully the salient characteristic requirements listed in the request.

41.2. Unless the bidder clearly indicates in the bid that an "or equal" product is being offered, bid shall be considered as offering the brand name product specified.

41.3. If the bidder proposes to furnish an "equal product, the brand name, if any, of the product to be furnished shall be inserted in the space provided in the Request for Bids, or such product shall be clearly identified in the bid. The evaluation of the bids and the determination as to equality of the product offered shall be the responsibility of the County and will be based upon the information furnished by the bidder, or identified in the bid as well as other information reasonably available to the Purchasing Activity. CAUTION TO BIDDERS: The Purchasing Activity is not responsible for locating or securing any information which is not identified in the bid and reasonably available to the Purchasing Activity. Accordingly, to insure that sufficient information is available, the bidder must furnish, as part of the bid, all descriptive material (such as cuts, illustrations, drawings, or other information) necessary for the Purchasing Activity to (I) determine whether the product offered meets the salient characteristic requirements of the Request for Bids, and (II) establish exactly what the bidder proposes to furnish and what the County is binding itself to purchase by making an award. The information furnished may include specific references to information previously furnished or to information otherwise available to the Purchasing Activity.

41.4. If the bidder proposes to modify a product so as to make it conform to the requirements of the Request for Bids, he shall (I) include in the bid a clear description of such modifications and (II) clearly mark any descriptive to show the proposed modifications.

41.5. Modifications proposed after bid opening to make a product conform to a brand name product referenced in the Request for Bids will not be considered.

42. **CONTRACT EXTENSION OPTION**

42.1. One to six months - end of contract period

The providing of goods and/or services described in Section A or B may be extended in one or more increments for a total of no less than one (1) nor more than three (3) calendar months at the discretion of the County Purchasing Director. Each extension shall be affected by written contract modification delivered to the Contractor no less than fifteen (15) calendar days prior to expiration of the contract. The rates set forth in the pricing section shall apply to any extension made pursuant to this option provision unless provision for appropriate price adjustment has been made elsewhere in this contract. All payments are subject to General Terms and Conditions, Clause titled "AVAILABILITY OF FUNDING".

43. **SEVERABILITY**

Should any part of this agreement be held to be invalid by a court of competent jurisdiction, the remainder of the agreement shall be considered as the whole agreement and be binding on the contracting parties.



## **CONTRACTOR'S MODIFICATIONS TO COUNTY TERMS AND CONDITIONS**

Contractor submits this bid subject to the following revisions to the resulting contract between County of San Diego and Roche Diagnostics Corporation. So that Roche may offer the County the competitive pricing outlined in this bid, the County will need to make a minimum volume commitment during the Base Term Period. The revisions to Sections 9, 14 and 15 below reflect this requirement.

**Section 9 "Termination for Convenience"** is revised to provide that County may not exercise its right to terminate any resulting contract for its convenience under Section 9 (or Section 8.1) at any time during the Base Term Period which expires August 31, 2015.

**Section 14 "Estimated Quantities."** The provisions of Section 14 shall not apply during the Base Term Period which expires August 31, 2015. The County agrees to purchase a minimum annual volume of 172 kits (48) of the COBAS® AmpliPrep / COBAS® TaqMan® HIV-1 Test, v2.0, during each 12-month period of the Base Term Period ("Annual Commitment"). If the County fails to purchase the Annual Commitment during the Base Term Period, the County is required to pay Contractor an amount equal to 25% of the Annual Commitment.

**Section 15 "Availability of Funding."** The words "such performance" shall be replaced with "the type of testing described under this contract."

**Section 17.1 "Disputes"** is modified in its entirety to read: Any dispute concerning a question of fact arising under this contract shall be decided by agreement between the parties or a mutually acceptable dispute resolution process."

**Section 22 "Disallowance"** is modified by removing the text "disallowed by the County" and replacing it with "mutually determined to have been erroneously paid to Contractor".

**Section 37 "Invoices"** this is removed in its entirety "Discounts will be calculated from receipt of merchandise or invoice, whichever is later" Roche Diagnostics does not provide discounts.

**Section 38 "Payments and Invoices"** this is removed "The County will accept invoice for progress payments and if approved reimburse the Contractor up to 90% of the value of goods received" and replaced in its entirety to read as follows: Roche will provide You with payment terms of net thirty (30) days from the date of invoice. Payment by credit card is acceptable at point of sale only.

**Section 38.3** this is removed in its entirety "Discounts will be calculated from receipt of merchandise or invoice, whichever is later" Roche Diagnostics does not provide discounts.

**Supplemental Term:** The following language will be added to the resulting contract between County of San Diego and Roche Diagnostics Corporation.

**Opportunity to Cure.** Prior to a party's exercise of any of the remedies set forth in this contract for a default by the other party, the non-defaulting party shall provide the defaulting party written notice describing the default and a 30-day opportunity to cure the default.

# It takes more than just a single target

BY THE TIME YOU HAVE READ THIS, HIV - I WILL HAVE MUTATED.  
BY THH TIME YOU HAVE READ THIS, HSV - I WILL HAVE MUTATED.  
BY THH TIME YOUYHAVE READ THVS, HSV - I WILL HAVE MUTATED.  
BYETHH TIME YOUYHAVE READ THVS, HSO - I WILL HAVE MUTATED.  
TYETHH LLMEGESUYHA F CE EVTHVE, HSO STWYLONH VTEMUTHTAD.  
TYETHHALLMEGESUYHA FACE EVTHVE, HSO STWYLONH STEMUTHTAD.  
TYE HHALLMEGESUYHU FACE EVTHVE, HSO STWYLONH STEM THTAD.  
THE HHALLMEGESUYHU FACE EVOHVE, HSO STWYLONH STEM AHTAD.  
THE HHALLMEGES YHU FACE EVOHVE, HSO STWYLONH STEP AHTAD.  
THE HHALLMNGES YHU FACE EVOLVE, HSO STWYLONE STEP AHTAD.  
THE CHALLMNGES YHU FACE EVOLVE, HSO STWYLONE STEP AHTAD.  
THE CHALLENGES YHU FACE EVOLVE, SO STAYLONE STEP AHTAD.  
THE CHALLENGES YHU FACE EVOLVE, SO STAY ONE STEP AHTAD.  
THE CHALLENGES YOU FACE EVOLVE, SO STAY ONE STEP AHEAD.



## As the challenges you face evolve...

### HIV mutates

*“No HIV-1 mutation can be considered to be neutral”<sup>1</sup>*

- Growing evidence indicates all HIV subtypes may be prone to errors; posing enormous challenges to viral load monitoring.<sup>2</sup>
- HIV-1 diversity is increasing and recombinants of greater complexity are being created.<sup>1,3</sup>
  - Produces  $10^{10}$  virions / day.<sup>4</sup>
  - Creates a polymorphism every 2,000–5,000 nucleotides.<sup>4</sup>
- Drug pressure and polymorphisms can lead to RT-PCR inefficiencies.<sup>2,3,5-7</sup>
- Mismatches and mutations unseen by single target assays can lead to underquantification.<sup>3,6</sup>

***Underquantification can have major clinical repercussions; delaying the detection of drug resistance<sup>2,3</sup>***

## Treatments evolve

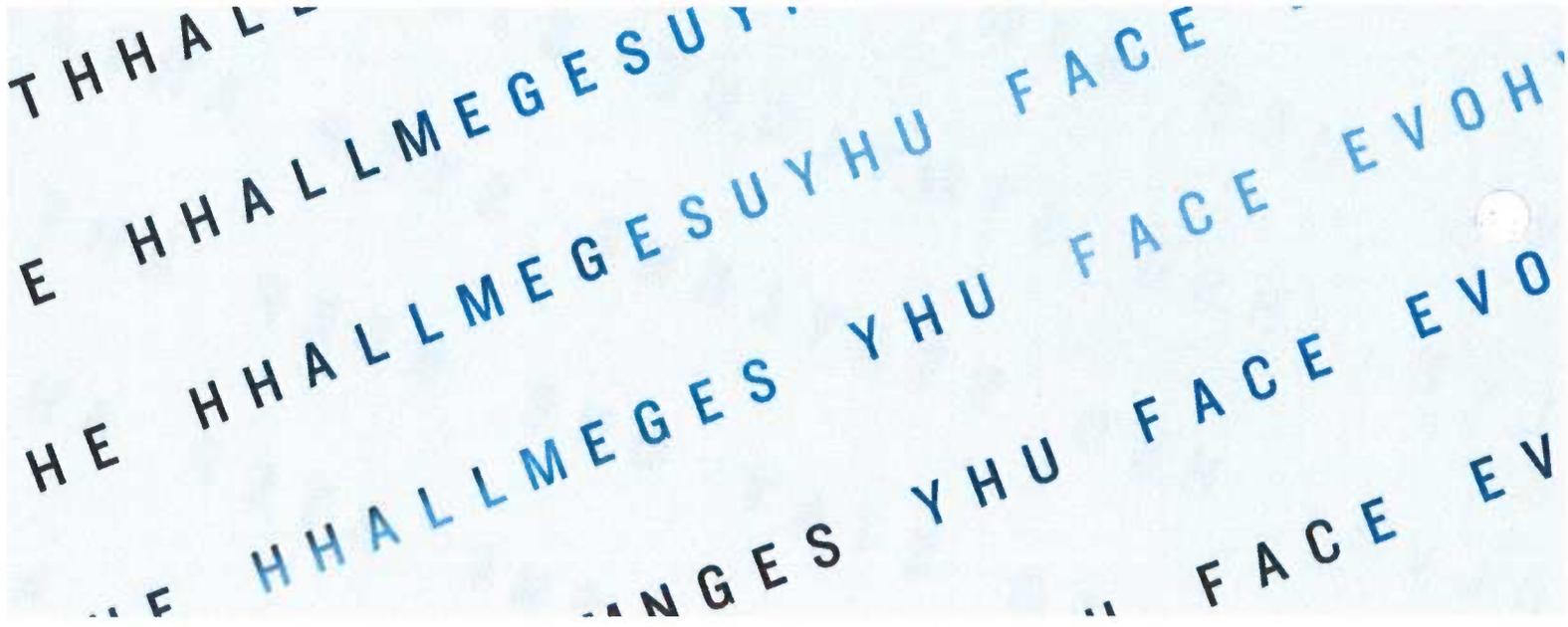
### *Newer classes of medications change treatment regimens*

- Updated IAS and DHHS guidelines recommend INSTIs for 1<sup>st</sup> line therapy.<sup>8,9</sup>
- In 2012, the use of raltegravir increased 25%.<sup>10</sup>
- The integrase gene is an attractive target for drug development.<sup>11</sup>
  - Raltegravir and elvitegravir are FDA approved.
  - Dolutegravir under FDA review.
  - Additional compounds are in development.

### *Drug resistance remains a central problem*

- Associated with all antiretrovirals, including INSTIs.<sup>12-15</sup>
- Over 42 mutations are associated with resistance to raltegravir.<sup>16,17</sup>

***Selective pressure on a drug target has the potential to compromise treatment efficacy<sup>11</sup>***



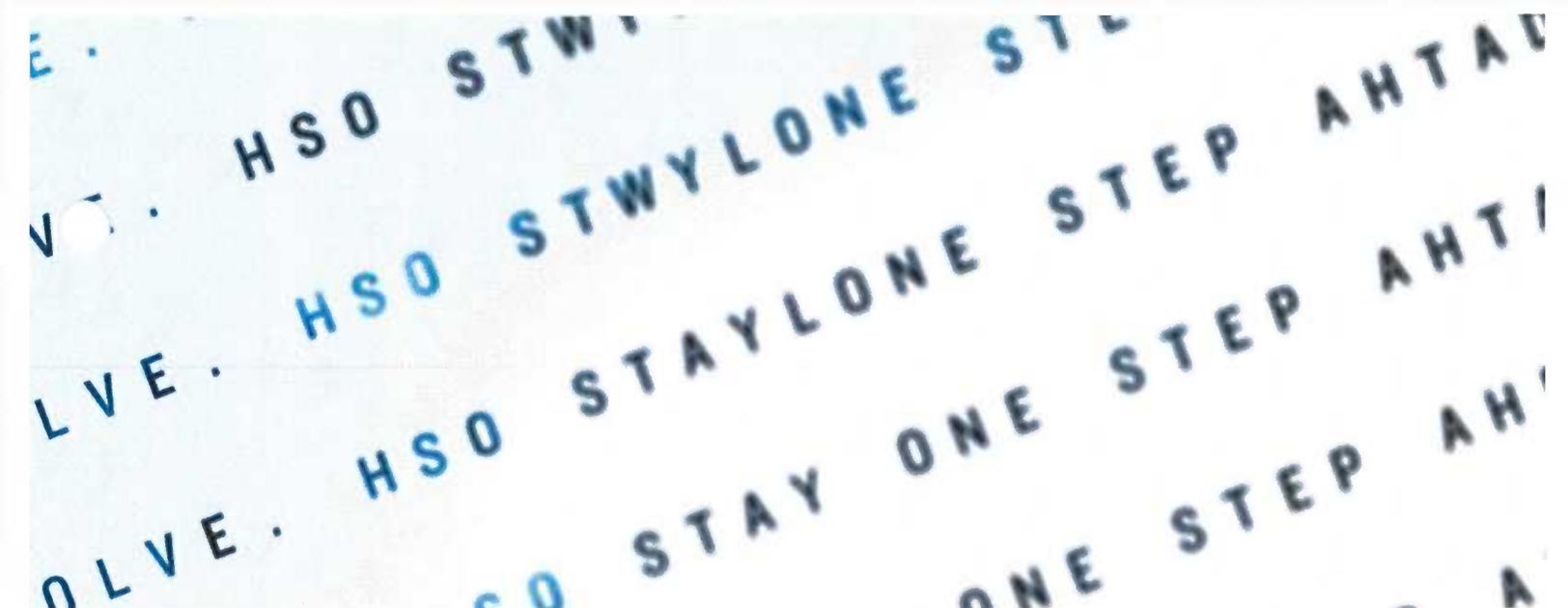
## So does Roche and the support we provide.

### Two targets

*“Represents an important step forward”<sup>5</sup>*

- Targeting two regions improves genotype inclusivity, detects HIV-1 variants and potentially avoids underquantification.<sup>5,6,18</sup>
  - 30 samples not quantified by the single target assay were quantified by the Roche dual target HIV-1 assay.<sup>5</sup>
  - The single target comparator assay quantified 19% of samples significantly lower than the Roche dual target HIV-1 assay.<sup>3</sup>
- Amplification of a less ideal target region might explain discrepancies already observed in the literature.<sup>3,5,6,18</sup>

***Accurately quantifying HIV-1 RNA with a dual target assay contributes to optimal treatment decisions for patient management<sup>2,5,18,19</sup>***



## Superior sensitivity

*“Evolution of viral resistance can occur in the setting of low-level viremia”<sup>8,11</sup>*

- Two clinical trials and a cohort analysis detected new resistance mutations in 37% and 65% respectively of patients who had developed persistent low-level viremia.<sup>8,20,21</sup>
- Viremia between 20-49 RNA copies/mL have been associated with higher baseline viral load and less time on ART.<sup>22,23</sup>
- Quantifying HIV-1 viremia between 20-49 copies/mL may have value.<sup>19,22</sup>

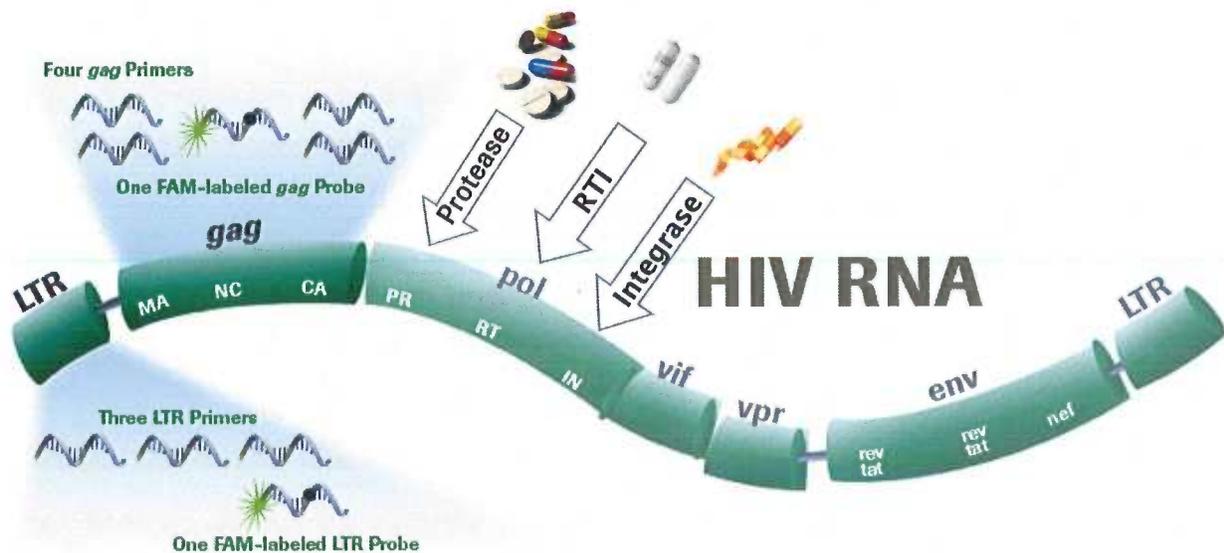
***Sensitive assays provide insight into disease awareness, assist in research eradication efforts, and may lead to improvements in disease management for patients living with the HIV-1 virus<sup>3,18</sup>***

## Stay one step ahead

With the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test v2.0\*

*Performance for today; prepared for tomorrow*

It takes more than just a single target to stay ahead of HIV-1. A diversified approach includes multiple safeguards, such as a dual target and increased sensitivity, providing confidence in test results for patients living with the HIV-1 virus<sup>2,5,22-24</sup>



\*This test is intended for use in conjunction with clinical presentation and other laboratory markers of disease progress for the clinical management of HIV-1 infected patients. The test can be used to assess patient prognosis by measuring the baseline HIV-1 RNA level or to monitor the effects of antiretroviral therapy by measuring changes in EDTA plasma HIV-1 RNA levels during the course of antiretroviral treatment.

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Indianapolis, IN 46256  
MyLabOnline.com

**COBAS® AmpliPrep/COBAS® TaqMan®  
HIV-1 Test, version 2.0**



**FOR IN VITRO DIAGNOSTIC USE.**

COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, version 2.0	<b>HI2CAP</b>	48 Tests	P/N: 05212308 190
COBAS® AmpliPrep/COBAS® TaqMan® Wash Reagent	<b>PG WR</b>	5.1 Liters	P/N: 03587797 190

**TABLE OF CONTENTS**

Table of Contents.....	7
INTENDED USE.....	7
SUMMARY AND EXPLANATION OF THE TEST.....	2
PRINCIPLES OF THE PROCEDURE.....	3
Target Selection.....	3
Specimen Preparation.....	4
Reverse Transcription and PCR Amplification.....	4
Detection of PCR Products in a COBAS® TaqMan® Test.....	5
Fundamentals of COBAS® TaqMan® Test Quantitation.....	5
HIV-1 RNA Quantitation.....	7
REAGENTS.....	8
WARNINGS AND PRECAUTIONS.....	11
STORAGE AND HANDLING REQUIREMENTS.....	12
MATERIALS PROVIDED.....	12
MATERIALS REQUIRED BUT NOT PROVIDED.....	13
Instrumentation and Software.....	13
Disposables.....	13
OTHER MATERIALS REQUIRED BUT NOT PROVIDED.....	13
SPECIMEN COLLECTION, TRANSPORT, AND STORAGE.....	14
A. Specimen Collection.....	14
B. Specimen Transport.....	14
C. Specimen Storage.....	14
INSTRUCTIONS FOR USE.....	15
Specimen and Control Preparation.....	16
Amplification and Detection.....	19
RESULTS.....	19
AMPLILINK Software.....	20
Batch Validation.....	20
Interpretation of Results.....	21
QUALITY CONTROL.....	22
Negative Control.....	22
Positive Controls.....	22
PROCEDURAL PRECAUTIONS.....	23
PROCEDURAL LIMITATIONS.....	23
INTERFERING SUBSTANCES.....	23

NON-CLINICAL PERFORMANCE EVALUATION.....	24
A. Limit of Detection.....	24
B. Precision.....	26
C. Linear Range.....	26
D. Inclusivity of HIV-1 Group M.....	27
E. HIV-1 Group O Detection.....	28
F. Specificity.....	29
G. Analytical Specificity.....	29
H. Method Correlation.....	30
CLINICAL PERFORMANCE EVALUATION.....	32
Reproducibility.....	32
Clinical Sensitivity, Specificity and Method Comparison.....	34
Clinical Method Comparison.....	37
Conclusion.....	40
REFERENCES.....	41

#### INTENDED USE

The COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, version 2.0 (v2.0) is an *in vitro* nucleic acid amplification test for the quantitation of human immunodeficiency virus type 1 (HIV-1) RNA in human plasma using the COBAS® AmpliPrep Instrument for automated specimen processing and the COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer for automated amplification and detection. The test can quantitate HIV-1 RNA over the range of 20 - 10,000,000 copies (cp)/mL. One copy of HIV-1 RNA is equivalent to 1.7 ± 0.1 International Units (IU) based on the WHO 1<sup>st</sup> International Standard for HIV-1 RNA for Nucleic Acid-Based Techniques (NAT) (NIBSC 97/656)<sup>1</sup>.

This test is intended for use in conjunction with clinical presentation and other laboratory markers of disease progress for the clinical management of HIV-1 infected patients. The test can be used to assess patient prognosis by measuring the baseline HIV-1 RNA level or to monitor the effects of antiretroviral therapy by measuring changes in EDTA plasma HIV-1 RNA levels during the course of antiretroviral treatment.

**The COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0 is not intended for use as a screening test for the presence of HIV-1 in blood or blood products or as a diagnostic test to confirm the presence of HIV-1 infection.**

**NOTE: The COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0 exhibits a higher level of sensitivity compared with the COBAS® AMPLICOR HIV-1 MONITOR Test, v1.5 and the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, reporting values ≥ 20 cp/mL that were undetectable or < Lower Limit of Quantitation (LLOQ) in these methods.**

#### SUMMARY AND EXPLANATION OF THE TEST

The human immunodeficiency virus (HIV) is the etiologic agent of acquired immunodeficiency syndrome (AIDS) and is primarily transmitted by sexual contact, exposure to infected blood or blood products, or by an infected mother to her fetus<sup>2-4</sup>. Once patients are infected with the virus, quantitative HIV-1 RNA measurement is a critical tool for disease management as well as the monitoring of antiretroviral therapy. Several studies have established the correlation of higher virus levels with increased risk of clinical progression of HIV disease, and that reductions in plasma virus levels can decrease this risk<sup>5-7</sup>. The clinical utility of viral load monitoring during antiretroviral therapy has been determined through multiple clinical trials for drugs<sup>8-9</sup>. The key viral endpoints for successful treatment include the magnitude of viral load reduction over time (log<sub>10</sub> units), percentage of patients demonstrating viral suppression below the limit of detection (LOD) at key time points, and time-to-loss of virologic response (resistance or non-adherence)<sup>8-9</sup>. There is an expected concordant CD4 cell count increase in relation to the viral suppression in patients responding to therapy. These endpoints have evolved and been incorporated into all of the major United States and global HIV treatment guidelines<sup>10-13</sup>. Specific medical decision points for HIV-1 RNA levels have been established for initiating antiretroviral therapy, determining treatment response, frequency of viral load monitoring, and determining virologic failure<sup>10-13</sup>. In summary, quantitative HIV-1 RNA level monitoring remains the most important surrogate marker for antiretroviral response.

Virus levels in the peripheral blood can be quantified by measurement of the HIV p24 antigen in serum, by quantitative culture of HIV from plasma, or by direct measurement of viral RNA in plasma using nucleic acid amplification or signal amplification technologies<sup>14-19</sup>. The initial direct viral RNA quantitative assays utilized endpoint measurement of either PCR or nucleic acid amplification reactions<sup>15,18</sup>. The detection phase of end-point PCR is executed after amplification and has limitations which include limited linear range, and time consuming process<sup>19</sup>. The introduction of real-time target amplification systems implements simultaneous amplification and detection, and has significantly improved upon the limitations of end point amplification<sup>17-19</sup>. The benefits of real-time PCR include increased sensitivity, broader linear range, and reduced turn-around processing time due to decreased reaction time<sup>19</sup>.

More recently, the recognition of increasing genetic diversity of HIV has highlighted the need to further evolve the design of next-generation real-time amplification tests<sup>20</sup>. In order to address the risk of mutational escape, a novel PCR design (the dual-target approach) was implemented resulting in coamplification of two target regions of HIV-1. The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 is an in vitro nucleic acid amplification test that quantitates all major subtypes of HIV-1 group M and HIV-1 group O. Three primers and one probe from the HIV-1 Long Terminal Repeat (LTR) as well as four primers and one probe in the *gag* region target and amplify the two HIV-1 regions. Both the *gag* and the LTR region are phylogenetically highly conserved to ensure broad subtype coverage.

#### PRINCIPLES OF THE PROCEDURE

The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 is a nucleic acid amplification test for the quantitation of human immunodeficiency virus type 1 (HIV-1) RNA in human plasma. The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 is based on three major processes: (1) specimen preparation to isolate HIV-1 RNA; (2) reverse transcription of the target RNA to generate complementary DNA (cDNA), and (3) simultaneous PCR amplification of target cDNA and detection of cleaved dual-labeled oligonucleotide detection probe specific to the target.

The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 permits automated specimen preparation followed by automated reverse transcription, PCR amplification and detection of HIV-1 target RNA and HIV-1 Quantitation Standard (QS) Armored RNA. The Master Mix reagent contains primers and probes specific for both HIV-1 RNA and HIV-1 QS RNA. The Master Mix has been developed to ensure equivalent quantitation of group M subtypes of HIV-1 and HIV-1 group O. The detection of amplified DNA is performed using target-specific and QS-specific dual-labeled oligonucleotide probes that permit independent identification of HIV-1 amplicon and HIV-1 QS amplicon.

The quantitation of HIV-1 viral RNA is performed using the HIV-1 QS. It compensates for effects of inhibition and controls the preparation and amplification processes, allowing a more accurate quantitation of HIV-1 RNA in each specimen. The HIV-1 QS is a non-infectious Armored RNA construct that contains HIV sequences with identical primer binding sites as the HIV-1 target RNA and a unique probe binding region that allows HIV-1 QS amplicon to be distinguished from HIV-1 target amplicon.

The HIV-1 QS is added to each specimen at a known copy number and is carried through the subsequent steps of specimen preparation, reverse transcription, simultaneous PCR amplification and detection of cleaved dual-labeled oligonucleotide detection probes. The COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer or COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer calculates the HIV-1 RNA concentration in the test specimens by comparing the HIV-1 signal to the HIV-1 QS signal for each specimen and control.

#### Target Selection

Selection of the target RNA sequence for HIV-1 depends on identification of regions within the HIV-1 genome that show maximum sequence conservation among the various HIV-1 group M subtypes and HIV-1 group O specimens. In order to address the high genetic variability of the virus, two regions of HIV genome are simultaneously targeted for amplification and detection by the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0. Two target-specific and one QS-specific dual-labeled oligonucleotide probes permit independent identification of the HIV-1 amplicon and of the HIV-1 QS amplicon. Accordingly, the appropriate selection of the primers and the dual-labeled oligonucleotide probes is critical to the ability of the test to amplify and detect the HIV-1 group M subtypes and HIV-1 group O. The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 uses reverse transcription and PCR amplification primers that define sequences within the highly conserved regions of the HIV-1 *gag* gene and of the HIV-1 LTR region<sup>21</sup>.

### Specimen Preparation

The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 utilizes automated specimen preparation on the COBAS<sup>®</sup> AmpliPrep Instrument by a generic silica-based capture technique. The procedure processes 850 µL of plasma. The HIV-1 virus particles are lysed by incubation at elevated temperature with a protease and chaotropic lysis/binding buffer that releases nucleic acids and protects the released HIV-1 RNA from RNases in plasma. Protease and a known number of HIV-1 QS Armored RNA molecules are introduced into each specimen along with the lysis reagent and magnetic glass particles. Subsequently, the mixture is incubated and the HIV-1 RNA and HIV-1 QS RNA are bound to the surface of the magnetic glass particles. Unbound substances, such as salts, proteins and other cellular impurities, are removed by washing the magnetic glass particles. After separating the magnetic glass particles and completing the washing steps, the adsorbed nucleic acids are eluted at elevated temperature with an aqueous solution. The processed specimen, containing the magnetic glass particles as well as released HIV-1 RNA and HIV-1 QS RNA, is added to the amplification mixture and transferred to the COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer or COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer. The HIV-1 target RNA and the HIV-1 QS RNA are then reverse transcribed, amplified and simultaneously detected by cleavage of two target-specific and one QS-specific dual-labeled oligonucleotide probe.

### Reverse Transcription and PCR Amplification

The reverse transcription and PCR amplification reaction is performed with the thermostable recombinant enzyme *Thermus spec* Z05 DNA Polymerase (Z05). In the presence of manganese (Mn<sup>2+</sup>) and under the appropriate buffer conditions, Z05 has both reverse transcriptase and DNA polymerase activity<sup>22-23</sup>. This allows both reverse transcription and PCR amplification to occur together with real-time detection of the amplicon.

Processed specimens are added to the amplification mixture in amplification tubes (K-tubes) in which both reverse transcription and PCR amplification occur. The reaction mixture is heated to allow the downstream primers to anneal specifically to the HIV-1 target RNA and to the HIV-1 QS RNA. In the presence of Mn<sup>2+</sup> and excess deoxynucleotide triphosphates (dNTPs), including deoxyadenosine, deoxyguanosine, deoxycytidine, deoxyuridine and deoxythymidine triphosphates, Z05 polymerase extends the annealed primers forming DNA strands complementary to the RNA target.

### Target Amplification

Processed specimens are added to the amplification mixture in amplification tubes (K-tubes) in which PCR amplification occurs. Following reverse transcription of the HIV-1 target RNA and the HIV-1 QS RNA, the Thermal Cycler in the COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer or COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer heats the reaction mixture to denature the RNA:cDNA hybrids and to expose the specific primer target sequences. As the mixture cools, the primers anneal to the target DNA. Z05 in the presence of Mn<sup>2+</sup> and excess deoxynucleotide triphosphates (dNTPs), including deoxyadenosine, deoxyguanosine, deoxycytidine, deoxyuridine and deoxythymidine triphosphates, extends the annealed primers along the target template to produce double-stranded DNA molecules termed amplicons. The COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer or COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer automatically repeats this process for a designated number of cycles, with each cycle intended to double the amount of amplicon DNA. The required number of cycles is preprogrammed into the COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer or COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer. Amplification occurs only in the two regions of the HIV-1 genome between the primers; the entire HIV-1 genome is not amplified.

### Selective Amplification

Selective amplification of target nucleic acid from the specimen is achieved in the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 by the use of AmpErase (uracil-N-glycosylase) enzyme and deoxyuridine triphosphate (dUTP). The AmpErase enzyme recognizes and catalyzes the destruction of DNA strands containing deoxyuridine, but not DNA containing deoxythymidine<sup>24</sup>. Deoxyuridine is not present in naturally occurring DNA, but is always present in amplicon due to the use of deoxyuridine triphosphate as one of the dNTPs in the Master Mix reagent; therefore, only amplicon contains deoxyuridine. Deoxyuridine renders contaminating amplicon susceptible to destruction by the AmpErase enzyme prior to amplification of the target DNA. Also, any nonspecific product formed after initial activation of the Master Mix by manganese is destroyed by the AmpErase enzyme. The AmpErase enzyme, which is included in the Master Mix reagent, catalyzes the cleavage of deoxyuridine-containing DNA at the deoxyuridine residues by opening the deoxyribose chain at the C1-position. When heated in the first thermal cycling step, the amplicon DNA chain breaks at the position of the deoxyuridine, thereby rendering the DNA non-amplifiable. The AmpErase enzyme remains inactive for a prolonged period of time once exposed to

temperatures above 55°C, i.e., throughout the thermal cycling steps, and therefore does not destroy target amplicon formed during amplification.

#### **Detection of PCR Products in a COBAS<sup>®</sup> TaqMan<sup>®</sup> Test**

The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 utilizes real-time PCR technology<sup>25,26</sup>. The use of dual-labeled fluorescent probes allows for real-time detection of PCR product accumulation by monitoring of the emission intensity of fluorescent reporter dyes released during the amplification process. The probes consist of HIV-1 and HIV-1 QS-specific oligonucleotide probes with a reporter dye and a quencher dye. In the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 the HIV-1 and HIV-1 QS probes are labeled with different fluorescent reporter dyes. When these probes are intact, the fluorescence of the reporter dye is suppressed by the proximity of the quencher dye due to Förster-type energy transfer effects. During PCR, the probe hybridizes to a target sequence and is cleaved by the 5' → 3' nuclease activity of the thermostable Z05 DNA polymerase. Once the reporter and quencher dyes are released and separated, quenching no longer occurs, and the fluorescent activity of the reporter dye is increased. The amplification of HIV-1 RNA and HIV-1 QS RNA are measured independently at different wavelengths. This process is repeated for a designated number of cycles, each cycle effectively increasing the emission intensity of the individual reporter dyes, permitting independent identification of HIV-1 RNA and HIV-1 QS RNA. The PCR cycle where a growth curve starts exponential growth is related to the amount of starting material at the beginning of the PCR.

#### **Fundamentals of COBAS<sup>®</sup> TaqMan<sup>®</sup> Test Quantitation**

The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 is inherently quantitative over a very wide dynamic range since the monitoring of amplicon is performed during the exponential phase of amplification. The higher the HIV-1 titer of a specimen, the earlier the fluorescence of the reporter dye of the HIV-1 probes rises above the baseline fluorescence level (see Figure 1). Since the amount of HIV-1 QS RNA is constant between all specimens, the fluorescence of the reporter dye of the HIV-1 QS probe should appear at a similar cycle for all specimens (see Figure 2). In specimens where the QS fluorescence is affected, the concentration is adjusted accordingly. The appearance of the specific fluorescent signals is reported as a critical threshold value (Ct). The Ct is defined as the fractional cycle number where reporter dye fluorescence exceeds a predetermined threshold (the Assigned Fluorescence Level), and starts the exponential growth phase of this signal (see Figure 3). A higher Ct value indicates a lower titer of initial HIV-1 target material. A 2-fold increase in titer correlates with a decrease of 1 Ct for target HIV-1 RNA, while a 10-fold increase in titer correlates with a decrease of 3.3 Ct.

Figure 1 shows the target growth curves for a dilution series spanning a 5- $\log_{10}$  range. As the concentration of the virus increases, the growth curves shift to earlier cycles. Therefore, the leftmost growth curve corresponds to the highest viral titer level, whereas, the rightmost growth curve corresponds to the lowest viral titer level.

**Figure 1**  
**Target Growth Curves for a Dilution Series Spanning a 5-Log<sub>10</sub> Range**

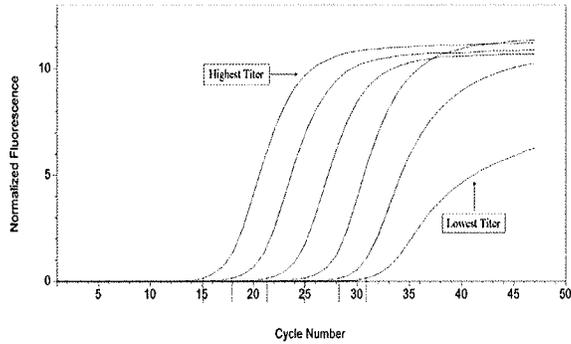


Figure 2 shows the Quantitation Standard growth curves for specimens from a viral dilution series that spans a 5- $\log_{10}$  range. The amount of Quantitation Standard added to each specimen is constant for each reaction. The Ct value of the Quantitation Standard is similar regardless of the viral titer.

**Figure 2**  
**Quantitation Standard Growth Curves for a Dilution Series of Virus Spanning a 5-Log<sub>10</sub> Range**

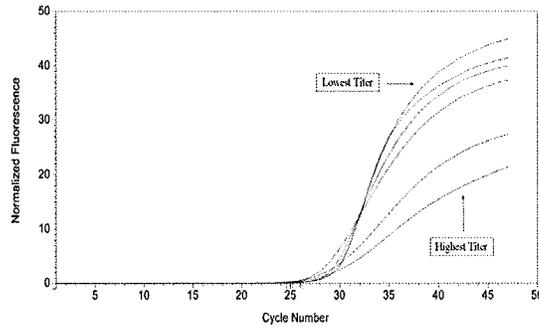
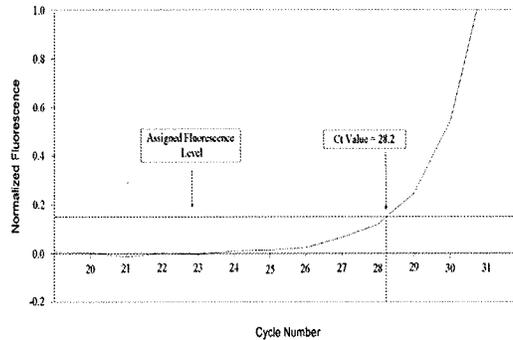


Figure 3 provides an example of how the fluorescence values at every cycle are normalized for each growth curve. The Ct value is calculated where the fluorescence signal crosses the Assigned Fluorescence Level.

**Figure 3**  
**Fluorescence Values at Every Cycle are Normalized for Each Growth Curve**



#### HIV-1 RNA Quantitation

The COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0 quantitates HIV-1 viral RNA by utilizing a second target sequence (HIV-1 Quantitation Standard) that is added to each test specimen at a known concentration. The HIV-1 QS is a non-infectious Armored RNA construct, containing fragments of HIV-1 sequences with primer binding regions identical to those of the HIV-1 *gag* target sequence. The HIV-1 QS contains HIV-1 primer binding regions and generates an amplification product of the same length and base composition as the HIV-1 *gag* target RNA. The detection probe binding region of the HIV-1 QS has been modified to differentiate HIV-1 QS amplicon from HIV-1 *gag* target amplicon.

During the annealing phase of the PCR in the COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer, the specimens are illuminated and excited by filtered light, and filtered emission fluorescence data are collected for each specimen. The readings from each specimen are then corrected for instrumental fluctuations. These fluorescence readings are sent by the instrument to the AMPLILINK software and stored in a database. Pre-Checks are used to determine if the HIV-1 RNA and HIV-1 QS RNA data represent sets that are valid, and flags are generated when the data lie outside the preset limits. After all Pre-Checks are completed and passed, the fluorescence readings are processed to generate Ct values for the HIV-1 RNA and the HIV-1 QS RNA. The lot-specific calibration constants provided with the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0 are used to calculate the titer value for the specimens and controls based upon the HIV-1 RNA and HIV-1 QS RNA Ct values. The COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0 is standardized against the WHO 1<sup>st</sup> International Standard for HIV-1 RNA for Nucleic Acid-Based Techniques (NAT) (NIBSC 97/656)<sup>1</sup>. Titer results are reported in cp/mL. The conversion factor between reported HIV-1 RNA cp/mL and HIV-1 IU/mL has been determined by Roche Molecular Systems, Inc. to be 0.6 cp/IU (1.7 IU/cp).

**REAGENTS**

**COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0** HI2CAP **48 Tests**  
(P/N: 05212308 190)

**HIV-1 v2.0 CS1** 1 x 48 Tests

(HIV-1 Magnetic Glass Particles Reagent Cassette)

Magnetic glass particles

93% Isopropanol

Xi 93% (w/w) Isopropanol



Irritant

F 93% (w/w) Isopropanol



Highly  
Flammable

**HIV-1 v2.0 CS2** 1 x 48 Tests

(HIV-1 Lysis Reagent Cassette)

Sodium citrate dihydrate

42.5% Guanidine thiocyanate

< 14% Polydocanol

0.9% Dithiothreitol

Xn 42.5% (w/w) Guanidine thiocyanate



Harmful

**HIV-1 v2.0 CS3** 1 x 48 Tests

HIV-1 Multi-Reagent Cassette containing:

**Pase** 1 x 3.8 mL

(Proteinase Solution)

Tris buffer

< 0.05% EDTA

Calcium chloride

Calcium acetate

≤ 7.8% Proteinase

Glycerol

Xn ≤ 7.8% (w/w) Proteinase



Harmful

**EB** 1 x 7.0 mL

(Elution Buffer)

Tris-base buffer

0.2% Methylparaben

**HIV-1 v2.0 CS4** 1 x 48 Tests

HIV-1 Test-Specific Reagent Cassette containing:

<p><b>HIV-1 QS</b> (HIV-1 Quantitation Standard) Tris-HCl buffer EDTA &lt; 0.005% Poly rA RNA (synthetic) &lt; 0.001% Armored HIV-1 RNA construct containing HIV-1 primer binding sequences and a unique probe binding region (non-infectious RNA in MS2 bacteriophage) 0.05% Sodium azide</p>	<p>1 x 3.6 mL</p>
<p><b>HIV-1 MMX</b> (HIV-1 Master Mix) Tricine buffer Potassium acetate Potassium hydroxide 20% Dimethylsulfoxide Glycerol &lt; 0.04% dATP, dCTP, dGTP, dUTP, dTTP &lt; 0.003% Upstream and downstream primers to the <i>gag</i> and the LTR region of HIV-1 &lt; 0.003% Oligonucleotide aptamer &lt; 0.003% Fluorescent-labeled oligonucleotide probes specific for HIV-1 and the HIV-1 Quantitation Standard &lt; 0.05% Z05 DNA Polymerase (microbial) &lt; 0.1% AmpErase (uracil-N-glycosylase) enzyme (microbial) 0.09% Sodium azide</p>	<p>1 x 2.5 mL</p>
<p><b>CAP/CTM Mn<sup>2+</sup></b> (CAP/CTM Manganese Solution) &lt; 0.5% Manganese acetate Glacial acetic acid 0.09% Sodium azide</p>	<p>1 x 19.8 mL</p>
<p><b>HIV-1 H(+ )C, v2.0</b> (HIV-1 High Positive Control, v2.0) &lt; 0.001% Armored HIV-1 RNA construct containing HIV-1 sequences (non-infectious RNA in MS2 bacteriophage). Negative Human Plasma, non-reactive by tests for antibody to HCV, antibody to HIV-1/2, HIV p24 antigen and HBsAg; HIV-1 RNA, HCV RNA and HBV DNA not detectable by PCR methods 0.1% ProClin<sup>®</sup> 300 preservative</p>	<p>4 x 1.0 mL</p>
<p>Xi  (3:1) mixture of 5-Chloro-2-methyl-2H-isothiazol-3-one and 2-Methyl-2H-isothiazol-3-one Irritant R36/38: Irritating to eyes and skin R43: May cause sensitization by skin contact</p>	

<p><b>HIV-1 L(+)<b>C</b>, v2.0</b>  (HIV-1 Low Positive Control, v2.0)  &lt; 0.001% Armored HIV-1 RNA construct containing HIV-1 sequences (non-infectious RNA in MS2 bacteriophage).  Negative Human Plasma, non-reactive by tests for antibody to HCV, antibody to HIV-1/2, HIV p24 antigen and HBsAg; HIV-1 RNA, HCV RNA and HBV DNA not detectable by PCR methods  0.1% ProClin® 300 preservative</p>	<p>4 x 1.0 mL</p>
<p>Xi  (3:1) mixture of 5-Chloro-2-methyl-2H-isothiazol-3-one and 2-Methyl-2H-isothiazol-3-one  Irritant  R36/38: Irritating to eyes and skin  R43: May cause sensitization by skin contact</p>	
<p><b>CTM (-) C</b>  [COBAS® TaqMan® Negative Control (Human Plasma)]  Negative Human Plasma, non-reactive by tests for antibody to HCV, antibody to HIV-1/2, HIV p24 antigen and HBsAg; HIV-1 RNA, HCV RNA and HBV DNA not detectable by PCR methods  0.1% ProClin® 300 preservative</p>	<p>4 x 1.0 mL</p>
<p>Xi  (3:1) mixture of 5-Chloro-2-methyl-2H-isothiazol-3-one and 2-Methyl-2H-isothiazol-3-one  Irritant  R36/38: Irritating to eyes and skin  R43: May cause sensitization by skin contact</p>	
<p><b>HIV-1 H(+)<b>C</b>, v2.0 Clip</b>  (HIV-1 High Positive Control, v2.0 Barcode Clip)</p>	<p>1 x 4 Clips</p>
<p><b>HIV-1 L(+)<b>C</b>, v2.0 Clip</b>  (HIV-1 Low Positive Control, v2.0 Barcode Clip)</p>	<p>1 x 4 Clips</p>
<p><b>HIV-1 (-) C Clip</b>  (HIV-1 Negative Control, v2.0 Barcode Clip)</p>	<p>1 x 4 Clips</p>
<p><b>COBAS® AmpliPrep/COBAS® TaqMan® Wash Reagent</b>  (P/N: 03587797 190)</p>	<p><b>PG WR</b>  1 x 5.1 L</p>
<p><b>PG WR</b>  (COBAS® AmpliPrep/COBAS® TaqMan® Wash Reagent)  Sodium citrate dihydrate  &lt; 0.1% N-Methylisothiazolone-HCl</p>	

#### WARNINGS AND PRECAUTIONS

##### A. FOR IN VITRO DIAGNOSTIC USE.

- B. This test is for use with human plasma collected in the anticoagulant EDTA.
- C. Do not pipette by mouth.
- D. Do not eat, drink, or smoke in laboratory work areas. Wear protective disposable gloves, laboratory coats, and eye protection when handling specimens and kit reagents. Wash hands thoroughly after handling specimens and test reagents.
- E. **Avoid microbial and ribonuclease contamination of reagents when removing aliquots from control vials.**
- F. **The use of sterile disposable pipettes and RNase-free pipette tips is recommended.**
- G. Do not pool controls from different lots or from different bottles of the same lot.
- H. Do not mix reagent cassettes or controls from different kits.
- I. Do not open COBAS<sup>®</sup> AmpliPrep cassettes and exchange, mix, remove or add bottles.
- J. Dispose of unused reagents, waste and specimens in accordance with country, federal, state and local regulations.
- K. Do not use a kit after its expiration date.
- L. Material Safety Data Sheets (MSDS) are available on request from your local Roche office.
- M. Specimens and controls should be handled as if infectious, using safe laboratory procedures such as those outlined in *Biosafety in Microbiological and Biomedical Laboratories* and in the CLSI document M29-A3<sup>27,28</sup>. Thoroughly clean and disinfect all work surfaces with a freshly prepared solution of 0.5% sodium hypochlorite in deionized or distilled water.

**Note: Commercial liquid household bleach typically contains sodium hypochlorite at a concentration of 5.25%. A 1:10 dilution of household bleach will produce a 0.5% sodium hypochlorite solution.**

- N. **CAUTION: CTM (-) C, HIV-1 L(+)C, v2.0 and HIV-1 H(+)C, v2.0** contain human plasma derived from human blood. The source material has been tested and found non-reactive for the presence of hepatitis B surface antigen (HBsAg), antibodies to HIV-1/2 and HCV, and HIV p24 antigen. Testing of negative human plasma by PCR methods showed no detectable HIV-1 RNA, HCV RNA or HBV DNA. No known test methods can offer complete assurance that products derived from human blood will not transmit infectious agents. Therefore, all human sourced material should be considered potentially infectious. **CTM (-) C, HIV-1 L(+)C, v2.0 and HIV-1 H(+)C, v2.0** should be handled as if infectious, using safe laboratory procedures such as those outlined in *Biosafety in Microbiological and Biomedical Laboratories* and in the CLSI Document M29-A3<sup>27,28</sup>. Thoroughly clean and disinfect all work surfaces with a freshly prepared solution of 0.5% sodium hypochlorite in deionized or distilled water.
- O. **HIV-1 QS, CAP/CTM Mn<sup>2+</sup> and HIV-1 MMX** contain sodium azide. Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. While disposing of sodium azide-containing solutions down laboratory sinks, flush the drains with a large volume of water to prevent azide buildup.
- P. Wear eye protection, laboratory coats, and disposable gloves when handling any reagent. Avoid contact of these materials with the skin, eyes, or mucous membranes. If contact does occur, immediately wash with large amounts of water. Burns can occur if left untreated. If spills of these reagents occur, dilute with water before wiping dry.

- Q. Do not allow **HIV-1 v2.0 CS2** and liquid waste from the COBAS<sup>®</sup> AmpliPrep Instrument, which contain guanidine thiocyanate, to contact sodium hypochlorite (bleach) solution. These mixtures can produce a highly toxic gas.
- R. When disposing of used COBAS<sup>®</sup> AmpliPrep Sample Processing Units (SPUs), which contain guanidine thiocyanate, avoid any contact with sodium hypochlorite (bleach) solution. These mixtures can produce a highly toxic gas.

**STORAGE AND HANDLING REQUIREMENTS**

- A. **Do not freeze reagents or controls.**
- B. Store **HIV-1 v2.0 CS1**, **HIV-1 v2.0 CS2**, **HIV-1 v2.0 CS3** and **HIV-1 v2.0 CS4** at 2°C to 8°C. Unused, these reagents are stable until the expiration date indicated. Once used, these reagents are stable for 28 days at 2°C to 8°C or until the expiration date, whichever comes first. **HIV-1 v2.0 CS1**, **HIV-1 v2.0 CS2**, **HIV-1 v2.0 CS3** and **HIV-1 v2.0 CS4** can be used for a maximum of 64 hours cumulative on board the COBAS<sup>®</sup> AmpliPrep Instrument. Reagents must be stored at 2°C to 8°C between instrument cycles.
- C. Store **HIV-1 H(+)**C****, **v2.0**, **HIV-1 L(+)**C****, **v2.0** and **CTM (-) **C**** at 2°C to 8°C. The controls are stable until the expiration date indicated. Once opened, any unused portion must be discarded.
- D. Store Barcode clips [**HIV-1 H(+)**C****, **v2.0 Clip**, **HIV-1 L(+)**C****, **v2.0 Clip** and **HIV-1 (-) **C** Clip**] at 2°C to 30°C.
- E. Store **PG WR** at 2°C to 30°C. **PG WR** is stable until the expiration date indicated. Once opened, this reagent is stable for 28 days at 2°C to 30°C or until the expiration date, whichever comes first.

**MATERIALS PROVIDED**

- A. **COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0** **HI2CAP**  
(P/N: 05212308 190)
  - HIV-1 v2.0 CS1**  
(HIV-1 Magnetic Glass Particles Reagent Cassette)
  - HIV-1 v2.0 CS2**  
(HIV-1 Lysis Reagent Cassette)
  - HIV-1 v2.0 CS3**  
(HIV-1 Multi-Reagent Cassette)
  - HIV-1 v2.0 CS4**  
(HIV-1 Test-Specific Reagent Cassette)
  - HIV-1 H(+)**C****, **v2.0**  
(HIV-1 High Positive Control, v2.0)
  - HIV-1 L(+)**C****, **v2.0**  
(HIV-1 Low Positive Control, v2.0)
  - CTM (-) **C****  
[COBAS<sup>®</sup> TaqMan<sup>®</sup> Negative Control (Human Plasma)]
  - HIV-1 H(+)**C****, **v2.0 Clip**  
(HIV-1 High Positive Control, v2.0 Barcode Clip)
  - HIV-1 L(+)**C****, **v2.0 Clip**  
(HIV-1 Low Positive Control, v2.0 Barcode Clip)
  - HIV-1 (-) **C** Clip**  
(HIV-1 Negative Control Barcode Clip)

B. **COBAS® AmpliPrep/COBAS® TaqMan® Wash Reagent**  
(P/N: 03587797 190)

PG R

**PG WR**  
(COBAS® AmpliPrep/COBAS® TaqMan® Wash Reagent)

**MATERIALS REQUIRED BUT NOT PROVIDED**

**Instrumentation and Software**

- COBAS® AmpliPrep Instrument
- COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer
- Optional: Docking Station
- Optional: **cobas p 630** instrument
- AMPLILINK Software, Version 3.2 Series or 3.3 Series
- Data Station for the AMPLILINK software, with printer
- AMPLILINK Software v3.2 or v3.3 Series Manuals:
  - COBAS® AmpliPrep instrument Instrument Manual For use with the COBAS® TaqMan® analyzer, COBAS® TaqMan® 48 analyzer, COBAS® AMPLICOR analyzer, or **cobas p 630** instrument, and the AMPLILINK software, version 3.2 and 3.3 series
  - COBAS® TaqMan® analyzer (plus optional docking station) Instrument Manual For use with the AMPLILINK software, version 3.2 and 3.3 series Application Manual
  - COBAS® TaqMan® 48 analyzer Instrument Manual For use with the AMPLILINK software, version 3.2 and 3.3 series Application Manual
  - AMPLILINK Software Version 3.2 Series Application Manual For use with the COBAS® AmpliPrep Instrument, COBAS® TaqMan® Analyzer, COBAS® TaqMan® 48 Analyzer, and COBAS® AMPLICOR® Analyzer
  - or
  - AMPLILINK software Version 3.3 Series Application Manual For use with COBAS® AmpliPrep instrument, COBAS® TaqMan® analyzer, COBAS® TaqMan® 48 analyzer, COBAS® AMPLICOR analyzer and **cobas p 630** instrument
  - Optional: **cobas p 630** instrument Operator's Manual Software Version 2.2

**Disposables**

- Sample processing units (SPUs)
- Sample input tubes (S-tubes) with barcode clips
- Racks of K-tips
- K-tube Box of 12 x 96

**OTHER MATERIALS REQUIRED BUT NOT PROVIDED**

- Sample Rack (SK 24 rack)
- Reagent Rack
- SPU rack
- K-tube capper, motorized
- K-tube capper
- K-carrier
- K-carrier Transporter
- K-carrier rack

- Pipettors with aerosol barrier or positive displacement RNase-free tips (capacity 1000 µL)\*
- Disposable gloves, powderless
- Vortex mixer

\* Pipettors should be accurate within 3% of stated volume. Aerosol barrier or positive displacement RNase-free tips must be used where specified to prevent specimen and amplicon cross-contamination.

#### SPECIMEN COLLECTION, TRANSPORT, AND STORAGE

**Note:** Handle all specimens and controls as if they are capable of transmitting infectious agents.

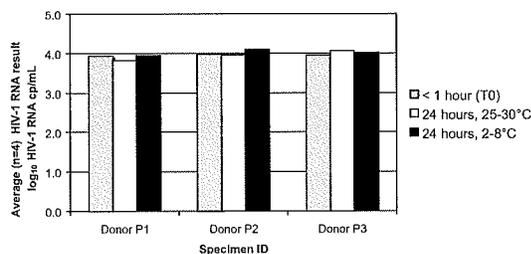
**Note:** This test has been validated for use with only human plasma collected in EDTA anticoagulant. Testing of other specimen types may result in inaccurate results.

#### A. Specimen Collection

The COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0 is for use with plasma specimens. Blood should be collected in sterile tubes using EDTA (lavender top) as the anticoagulant and mixed adequately according to the tube manufacturer's instructions.

Store whole blood at 2°C to 25°C for no longer than 24 hours. Separate plasma from whole blood within 24 hours of collection by centrifugation at 800-1600 x g for 20 minutes at room temperature. Transfer plasma to a sterile polypropylene tube. Figure 4 shows specimen stability data from specimen stability studies performed with the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test.

**Figure 4**  
**HIV-1 Stability in Whole Blood (collected in EDTA-plasma tubes)**



#### B. Specimen Transport

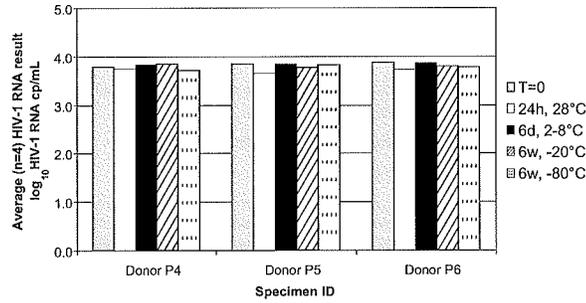
Transportation of whole blood or plasma must comply with country, federal, state and local regulations for the transport of etiologic agents<sup>29</sup>. Whole blood must be transported at 2-25°C and centrifuged within 24 hours of collection. Plasma may be transported at 2-8°C or frozen at -20°C to -80°C.

#### C. Specimen Storage

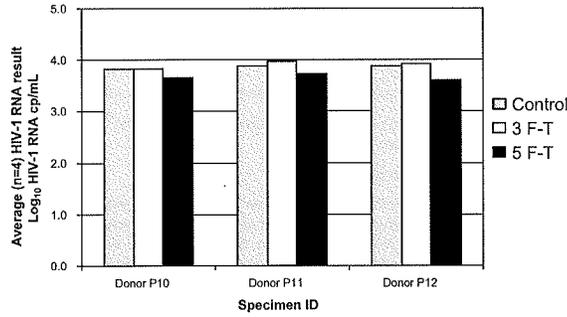
Plasma specimens may be stored at room temperature (25°C to 30°C) for up to 1 day, at 2°C to 8°C for up to 6 days, or frozen at -20°C to -80°C for up to 6 weeks. It is recommended that specimens be stored in 1100-1200 µL aliquots in sterile, 2.0 mL polypropylene screw-cap tubes (such as Sarstedt 72.694.006). Figure 5 shows the specimen stability data from specimen storage studies performed with the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test.

Plasma specimens may be frozen and thawed up to 5 times without a significant loss of HIV-1 RNA. Figure 6 shows the data from a freeze-thaw study performed with the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test.

**Figure 5**  
**HIV-1 Stability in EDTA-Plasma**



**Figure 6**  
**HIV-1 Results after up to Five Freeze-Thaw (F-T) Cycles (EDTA-plasma)**



**INSTRUCTIONS FOR USE**

**Note:** For detailed operating instructions, a detailed description of the possible configurations, printing results and interpreting flags, comments and error messages, refer to (1) the COBAS<sup>®</sup> AmpliPrep Instrument Manual For use with the COBAS<sup>®</sup> TaqMan<sup>®</sup> analyzer, COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 analyzer, COBAS<sup>®</sup> AMPLICOR<sup>®</sup> analyzer, or cobas p 630 instrument, and the AMPLILINK software, version 3.2 and 3.3 series (2) the COBAS<sup>®</sup> TaqMan<sup>®</sup> analyzer (plus optional docking station) Instrument Manual For use with the AMPLILINK software, version 3.2 and 3.3 series Application Manual (3) the COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 analyzer Instrument Manual For use with the AMPLILINK software, version 3.2 and 3.3 series Application Manual (4) the AMPLILINK Software Version 3.2 Series Application Manual For use with the COBAS<sup>®</sup> AmpliPrep Instrument, COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer, COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer, and COBAS<sup>®</sup> AMPLICOR<sup>®</sup> Analyzer or the AMPLILINK software Version 3.3 Series Application Manual For use with COBAS<sup>®</sup> AmpliPrep instrument, COBAS<sup>®</sup> TaqMan<sup>®</sup> analyzer, COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 analyzer, COBAS<sup>®</sup> AMPLICOR<sup>®</sup> analyzer, and cobas p 630 instrument (5) Optional: cobas p 630 instrument Operator's Manual Software Version 2.2

### Batch Size

Each kit contains reagents sufficient for 48 tests which may be performed in batches of 12 to 24 tests. At least one replicate each of **CTM (-) C, HIV-1 L(+)+C, v2.0** and **HIV-1 H(+)+C, v2.0** must be included in each batch (see "Quality Control" section).

### Workflow

The COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer or COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer run must be started within 120 minutes following completion of specimen and control preparation.

**Note: Do not freeze or store processed specimens and controls.**

### Specimen and Control Preparation

**Note: If using frozen specimens, place the specimens at room temperature (15°C to 30°C) until completely thawed and vortex for 3-5 seconds before use. Controls should be removed from 2°C to 8°C storage and equilibrated to room temperature before use.**

### COBAS<sup>®</sup> AmpliPrep Instrument Set-up

#### Part A. Maintenance and Priming

- A1. The COBAS<sup>®</sup> AmpliPrep Instrument is ready for operation in stand-by mode.
- A2. Turn the Data Station for the AMPLILINK software **ON**. Prepare the Data Station as follows:
  - a. Log onto the Windows<sup>®</sup> XP operating system.
  - b. Double click the AMPLILINK software icon.
  - c. Log onto AMPLILINK software by entering the assigned User ID and password.
- A3. Check the supply of **PG WR** using the **Status** Screen and replace if necessary.
- A4. Perform all Maintenance that is listed in the Due Tab. The COBAS<sup>®</sup> AmpliPrep Instrument will automatically prime the system.

#### Part B. Loading of Reagent Cassettes

**Note: All reagent cassettes should be removed from 2-8°C storage, immediately loaded onto the COBAS<sup>®</sup> AmpliPrep Instrument and allowed to equilibrate to ambient temperature on the instrument for at least 30 minutes before the first specimen is to be processed. Do not let reagent cassettes come to ambient temperature outside the instrument as condensation may form on the barcode labels. Do not wipe off condensation if it appears on the barcode labels.**

- B1. Place **HIV-1 v2.0 CS1** onto a reagent rack. Place **HIV-1 v2.0 CS2**, **HIV-1 v2.0 CS3** and **HIV-1 v2.0 CS4** onto a separate reagent rack.
- B2. Load the reagent rack containing **HIV-1 v2.0 CS1** onto rack position **A** of the COBAS<sup>®</sup> AmpliPrep Instrument.
- B3. Load the reagent rack containing **HIV-1 v2.0 CS2**, **HIV-1 v2.0 CS3** and **HIV-1 v2.0 CS4** onto rack position **B, C, D** or **E** of the COBAS<sup>®</sup> AmpliPrep Instrument. (see Table 1 for additional information).

#### Part C. Loading of Disposables

**Note: Determine the number of COBAS<sup>®</sup> AmpliPrep reagent cassettes, Sample Processing Units (SPUs), Input Sample tubes (S-tubes), K-tips and K-tubes needed. One SPU, one Input S-tube, one K-tip and one K-tube are needed for each specimen or control.**

Multiple workflows for use of the COBAS<sup>®</sup> AmpliPrep Instrument with the COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer or COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer are possible. For reference, see Table 1 below. Depending on the workflow used, load the appropriate number of reagent cassette racks, sample racks with Input S-tubes, SPU racks, K-tip racks, K-tube racks and K-carriers on K-carrier racks onto the respective rack positions of the COBAS<sup>®</sup> AmpliPrep Instrument (see Table 1 for additional information).

- C1. Place the SPUs in the SPU rack(s) and load the rack(s) onto rack position **J, K, or L** of the COBAS<sup>®</sup> AmpliPrep Instrument.
- C2. Depending on the workflow used, load full K-tube rack(s) onto rack position **M, N, O, or P** of the COBAS<sup>®</sup> AmpliPrep Instrument.
- C3. Load full K-tip rack(s) onto rack position **M, N, O, or P** of the COBAS<sup>®</sup> AmpliPrep Instrument.
- C4. For workflow 3 using the COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer, load K-carriers on K-carrier rack(s) onto rack positions **M & N, or O & P** of the COBAS<sup>®</sup> AmpliPrep Instrument.

**Table 1**  
Possible Workflows for using the COBAS<sup>®</sup> AmpliPrep Instrument with the COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer or COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer

Workflow	Transfer Mode to COBAS <sup>®</sup> TaqMan <sup>®</sup> Analyzer or COBAS <sup>®</sup> TaqMan <sup>®</sup> 48 Analyzer	Racks, Carriers and Disposables	Position on COBAS <sup>®</sup> AmpliPrep Instrument	
1	COBAS <sup>®</sup> AmpliPrep Instrument plus Docking Station plus COBAS <sup>®</sup> TaqMan <sup>®</sup> Analyzer	Automated transfer of K-carrier	K-tubes in full K-tube racks	M - P
			K-tips in full K-tip racks	M - P
			Input S-tubes containing specimens and controls on sample racks	F - H
			SPUs in SPU racks	J - L
			CS1 on Cassette rack	A
2	COBAS <sup>®</sup> AmpliPrep Instrument plus COBAS <sup>®</sup> TaqMan <sup>®</sup> Analyzer	Manual transfer of K-tubes via sample rack(s) onto COBAS <sup>®</sup> TaqMan <sup>®</sup> Analyzer	CS2, CS3, CS4 on Cassette rack	B - E
			K-tubes in full K-tube racks	M - P
			K-tips in full K-tip racks	M - P
			Input S-tubes containing specimens and controls on sample racks	F - H
			SPUs in SPU racks	J - L
			CS1 on Cassette rack	A
			CS2, CS3, CS4 on Cassette rack	B - E
3	COBAS <sup>®</sup> AmpliPrep Instrument plus COBAS <sup>®</sup> TaqMan <sup>®</sup> 48 Analyzer(s)	Manual transfer of K-carrier via K-carrier rack(s) onto COBAS <sup>®</sup> TaqMan <sup>®</sup> 48 Analyzer	<u>After specimen processing is finished:</u> K-tubes on sample racks (ready for manual transfer)	F - H
			K-tubes on sample racks	F - H
			K-tips in full K-tip racks	M - P
			Input S-tubes containing specimens and controls on sample racks	F - H
			SPUs in SPU racks	J - L
			CS1 on Cassette rack	A
			CS2, CS3, CS4 on Cassette rack	B - E
			Empty barcoded K-carrier on K-carrier rack	M - P
<u>After specimen processing is finished:</u> K-tubes in K-carrier on K-carrier rack	M - P			

#### Part D. Ordering and Loading of Specimens

**Note:** If using the cobas p 630 instrument for preparation of specimens, refer to the cobas p 630 instrument Operators Manual.

- D1. Prepare sample racks as follows: Attach a barcode label clip to each sample rack position where a specimen (S-tube) is to be placed. Attach one of the specific barcode label clips for the controls [CTM (-) C, HIV-1 L(+)C, v2.0 and HIV-1 H(+)C, v2.0] to each sample rack position where the controls (S-tube) are to be placed. The barcode label clips for controls should have the same control lot number as the lot number on the control vials in the kit. Make sure to assign the right control to the position with the appropriate control barcode clip. Place one Input S-tube into each position containing a barcode label clip.
- D2. Using the AMPLILINK software, create specimen orders for each specimen and control in the **Orders** window **Sample** folder. Select the appropriate test file and complete by saving.
- D3. Assign specimen and control orders to sample rack positions in the **Orders** window **Sample Rack** folder. The sample rack number must be for the rack prepared in Step D1.
- D4. Print the **Sample Rack Order** report to use as a worksheet.
- D5. Prepare specimen and control racks in the designated area for specimen and control addition as follows: Vortex each specimen and control [CTM (-) C, HIV-1 L(+)C, v2.0 and HIV-1 H(+)C, v2.0] for 3 to 5 seconds. Avoid contaminating gloves when manipulating the specimens and controls.
- D6. Transfer 1000 to 1050 µL of each specimen and control [CTM (-) C, HIV-1 L(+)C, v2.0 and HIV-1 H(+)C, v2.0] to the appropriate barcode labeled Input S-tube using a micropipettor with an aerosol barrier or positive displacement RNase-free tip. **Avoid transferring particulates and/or fibrin clots from the original specimen to the Input S-tube.** Specimens and controls should be transferred to tube positions as assigned and recorded on the worksheet in Step D4. The barcode label clips for controls should have the same control lot number as the lot number on the control vials in the kit. Assign the right control to the position with the appropriate control barcode clip. **Avoid contaminating the upper part of the S-tubes with specimens or controls.**
- D7. For workflows 1 and 2 (see Table 1 on page 17), load the sample rack(s) filled with Input S-tubes onto rack positions **F, G, or H** of the COBAS® AmpliPrep Instrument.
- D8. For workflow 3 (see Table 1 on page 17) using the COBAS® TaqMan® 48 Analyzer, load sample rack(s) with Input S-tubes and K-tubes (one for each Input S-tube, loaded in the right position adjacent to Input S-tubes) onto rack position **F, G, or H** of the COBAS® AmpliPrep Instrument.

#### Part E. Start of COBAS® AmpliPrep Instrument Run

- E1. Start the COBAS® AmpliPrep Instrument using the AMPLILINK software.

#### Part F. End of COBAS® AmpliPrep Instrument Run and Transfer to COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer (for workflows 2 and 3 only)

- F1. Check for flags or error messages.
- F2. Remove processed specimens and controls from the COBAS® AmpliPrep Instrument on either sample racks (for COBAS® TaqMan® Analyzer without Docking Station) or K-carrier racks (for COBAS® TaqMan® 48 Analyzer), depending on the workflow (for further details see Part G).
- F3. Remove waste from the COBAS® AmpliPrep Instrument.

**Note: Do not expose processed specimens and controls to light after completion of specimen and control preparation.**

#### **Amplification and Detection**

##### **COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer Set-up**

The COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer run must be started within 120 minutes following completion of specimen and control preparation.

**Note: Do not freeze or store processed specimens and controls.**

#### **Part G. Loading Processed Specimens**

- G1. Depending on which workflow is used (see Table 1 on page 17), perform the appropriate steps listed below to transfer the K-tubes to the COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer:

*Workflow 1:* Automated transfer of K-carrier via docking station to the COBAS® TaqMan® Analyzer. Manual intervention is unnecessary.

*Workflow 2:* Manual transfer of K-tubes in sample rack(s) to the COBAS® TaqMan® Analyzer.

*Workflow 3:* Manual transfer of K-carrier on K-carrier rack(s) to the COBAS® TaqMan® 48 Analyzer. Manual transfer of K-carriers into COBAS® TaqMan® 48 Analyzer using the K-carrier Transporter.

#### **Part H. Start of the COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer Run**

- H1. Start the COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer by one of the options below depending on the workflow used:

*Workflow 1:* No intervention necessary.

*Workflow 2:* Automatic start of the COBAS® TaqMan® Analyzer after insertion of sample rack(s).

*Workflow 3:* Fill K-carrier with empty K-tubes if there are fewer than 6 K-tubes on the K-carrier. Filling is guided by the AMPLILINK software. Open thermal cycler cover, load K-carrier into thermal cycler, and close lid. Start the COBAS® TaqMan® 48 Analyzer run.

#### **Part I. End of COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer Run**

- I1. At the completion of the COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer run, print the Results Report. Check for flags or error messages in the Results Report. Specimens with flags and comments are interpreted as described in the Results section below. After acceptance of results, store data in archive.
- I2. Remove used K-tubes from the COBAS® TaqMan® Analyzer or COBAS® TaqMan® 48 Analyzer.

#### **RESULTS**

The COBAS® TaqMan® Analyzer or the COBAS® TaqMan® 48 Analyzer automatically determines the HIV-1 RNA concentration for the specimens and controls. The HIV-1 RNA concentration is expressed in cp/mL. The conversion factor for the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0 is 0.6 cp/IU, using the WHO 1<sup>st</sup> International Standard for HIV-1 RNA for Nucleic Acid-Based Techniques (NAT) (NIBSC 97/656)<sup>1</sup>.

### AMPLILINK Software

The AMPLILINK software determines

- the Ct for the HIV-1 RNA and the HIV-1 QS RNA.
- the HIV-1 RNA concentration based upon the Ct values for the HIV-1 RNA and HIV-1 QS RNA and the lot-specific calibration coefficients provided on the cassette barcodes.
- that the calculated cp/mL for **HIV-1 L(+)**C**, v2.0** and **HIV-1 H(+)**C**, v2.0** fall within the assigned ranges.

### Batch Validation-- AMPLILINK version 3.2 Series

Check the AMPLILINK software results window or printout for flags and comments to ensure that the batch is valid. For control orders, the AMPLILINK software performs a check to determine if the cp/mL value for the control is within its specified range. If the cp/mL value for the control lies outside of its range, a FLAG (error message) is generated to show the control has failed.

The batch is valid if no flags appear for any of the controls **[HIV-1 L(+)**C**, v2.0; HIV-1 H(+)**C**, v2.0 and CTM (-) **C**]**.

The batch is not valid if any of the following flags appear for the HIV-1 Controls:

#### Negative Control

Flag	Result	Interpretation
_N_NC_INVALID	Invalid	An invalid result or a valid result that was not negative for the HIV-1 target

#### HIV-1 Low Positive Control, v2.0

Flag	Result	Interpretation
_L_LPCINVALID	< 2.00E+01 cp/mL	Control below range
_L_LPCINVALID	Target Not Detected	Control below range
_L_LPCINVALID	A numeric titer X.XXE+XX cp/mL	Control out of range
_L_LPCINVALID	> 1.00E+07 cp/mL	Control above range
_L_LPCINVALID	Invalid	An invalid result

**HIV-1 High Positive Control, v2.0**

Flag	Result	Interpretation
_H_HPCINVALID	< 2.00E+01 cp/mL	Control below range
_H_HPCINVALID	Target Not Detected	Control below range
_H_HPCINVALID	A numeric titer X.XXE+XX cp/mL	Control out of range
_H_HPCINVALID	> 1.00E+07 cp/mL	Control above range
_H_HPCINVALID	Invalid	An invalid result

If the batch is invalid, repeat the entire batch including specimen and control preparation, amplification and detection.

**Batch Validation – AMPLILINK version 3.3 Series**

Check AMPLILINK software results window or printout for flags and comments to ensure that the batch is valid. For control orders, a check is made to determine if the cp/mL value for the control is within its specified range. If the cp/mL value for the control lies outside of its range, a FLAG is generated to show the control has failed.

The batch is valid if no flags appear for any of the controls **[HIV-1 L(+)]C, v2.0; HIV-1 H(+)]C, v2.0 and CTM (-) C]**.

The batch is not valid if any of the following flags appear for the HIV-1 Controls:

**Negative Control:**

Flag	Result	Interpretation
NC_INVALID	Invalid	An invalid result or a "valid" result that was not negative for HIV-1 target

**Low Positive Control:**

Flag	Result	Interpretation
LPCINVALID	Invalid	An invalid result or a control out of range

**High Positive Control:**

Flag	Result	Interpretation
HPCINVALID	Invalid	An invalid result or a control out of range

If the batch is invalid, repeat the entire batch including specimen and control preparation, reverse transcription, amplification and detection.

**Interpretation of Results**

For a valid batch, check each individual specimen for flags or comments on the results printout. A valid batch may include both valid and invalid specimen results depending on whether flags and/or comments are obtained for the individual specimens. Interpret the results as follows:

Specimen results are interpreted as follows:

Titer Result	Interpretation
Target Not Detected	Ct value for HIV-1 above the limit for the assay or no Ct value for HIV-1 obtained. Report results as "HIV-1 RNA not detected".
< 2.00E+01 cp/mL	Calculated cp/mL are below the Limit of Detection of the assay. Report results as "HIV-1 RNA detected, less than 20 HIV-1 RNA cp/mL".
≥ 2.00E+01 cp/mL and ≤ 1.00E+07 cp/mL	Calculated results greater than or equal to 20 cp/mL and less than or equal to 1.00E+07 cp/mL are within the Linear Range of the assay..
> 1.00E+07 cp/mL	Calculated cp/mL are above the range of the assay. Report results as "greater than 1.00E+07 HIV-1 RNA cp/mL ". If quantitative results are desired, the original specimen should be diluted 1:100 with HIV-1-negative human EDTA-plasma and the test repeated. Multiply the reported result by the dilution factor.

**Note:** Specimens above the range of the assay may also produce an invalid result with a flag "QS\_INVALID". If quantitative results are desired, the original specimen should be diluted 1:100 with HIV-1-negative human EDTA-plasma and the test repeated. Multiply the reported result by the dilution factor.

#### QUALITY CONTROL

One replicate each of the COBAS® TaqMan® Negative Control, the HIV-1 Low Positive Control, v2.0 and the HIV-1 High Positive Control, v2.0 must be included in each test batch. The batch is valid if no flags appear for any of the controls [HIV-1 L(+)**C**, v2.0, HIV-1 H(+)**C**, v2.0 and CTM (-) **C**].

Check the batch printout for flags and comments to ensure that the batch is valid.

#### Negative Control

The CTM (-) **C** must yield a "Target Not Detected" result. If the CTM (-) **C** is flagged as invalid, then the entire batch is invalid. Repeat the entire process (specimen and control preparation, amplification and detection). If CTM (-) **C** is consistently invalid in multiple batches, contact your local Roche office for technical assistance.

#### Positive Controls

The assigned titer range for HIV-1 L(+)**C**, v2.0 and HIV-1 H(+)**C**, v2.0 is specific for each lot of reagents, and is provided on the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0 reagent cassette barcodes.

The HIV-1 RNA cp/mL for HIV-1 L(+)**C**, v2.0 and HIV-1 H(+)**C**, v2.0 should fall within their assigned titer ranges. If one or both of the positive controls are flagged as invalid, then the entire batch is invalid. Repeat the entire process (specimen and control preparation, amplification and detection). If the HIV-1 RNA titer of one or both of the positive controls is consistently outside the ranges in multiple batches, contact your local Roche office for technical assistance.

## PROCEDURAL PRECAUTIONS

As with any test procedure, good laboratory technique is essential to the proper performance of this assay.

## PROCEDURAL LIMITATIONS

1. This test has been validated for use with only human plasma collected in EDTA anticoagulant. Testing of other specimen types may result in inaccurate results.
2. The performance of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 has neither been evaluated with specimens containing HIV-1 group N, nor with specimens containing HIV-2.
3. Reliable results are dependent on adequate specimen collection, transport, storage and processing procedures.
4. The presence of AmpErase enzyme in the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 Master Mix reduces the risk of amplicon contamination. However, contamination from HIV-1 positive controls and clinical specimens can be avoided only by good laboratory practices and careful adherence to the procedures specified in this Package Insert.
5. Use of this product should be limited to personnel trained in the techniques of PCR.
6. This product can only be used with the COBAS<sup>®</sup> AmpliPrep Instrument and the COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer or COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer.
7. The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 demonstrated a linear range from 20 cp/mL to 1.0E+07 cp/mL for HIV-1 group M specimen in EDTA plasma and a linear range from 2.0E+02 cp/mL to 2.0E+05 cp/mL for HIV-1 group O specimen in EDTA plasma.
8. Though rare, mutations within the highly conserved regions of the viral genome covered by the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 primers and/or probes may result in the under-quantitation of or failure to detect the virus.
9. Detection of HIV-1 RNA is dependent on the number of virus particles present in the specimen and may be affected by specimen collection methods and patient factors, (i.e., age, presence of symptoms, and/or stage of the infection).
10. Due to inherent differences between technologies, it is recommended that prior to switching from one technology to the next, users perform method correlation studies in their laboratory to quantify technology differences.
11. This product exhibits a higher level of sensitivity compared to its predecessors, reporting values  $\geq$  20 cp/mL that were undetectable or  $<$  LLoQ in prior methods. In addition, the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 returns higher titers than the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, except at both the higher range ( $>$  5 log<sub>10</sub> cp/mL) and the lower range ( $<$  2 log<sub>10</sub> cp/mL) where it returns titers that are lower.

## INTERFERING SUBSTANCES

Elevated levels of triglycerides (up to 3500 mg/dL), bilirubin (up to 28 mg/dL), albumin (up to 8900 mg/dL), hemoglobin (up to 900 mg/dL) and human DNA (up to 0.4 mg/dL) in specimens as well as the presence of autoimmune diseases or respective markers such as Systemic lupus erythematosus (SLE), rheumatoid arthritis (RA) and antinuclear antibody (ANA) were shown not to interfere with the quantitation of HIV-1 RNA or impact the specificity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0. The evaluation was performed according to CLSI Guideline EP7-A2 using one lot of COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 reagents.

The following drug compounds tested at 3 times the Peak Plasma Level (Cmax) have been shown not to interfere with the quantitation of HIV-1 RNA or impact the specificity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0:

<b>Nucleotide DNA Polymerase Inhibitors</b>  Adefovir dipivoxil	<b>Nucleoside Reverse Transcriptase and DNA Polymerase Inhibitors</b>  Lamivudine, 3TC Zidovudine Stavudine, 4dT Abacavir sulfate Didanosine, ddl Entecavir Tenofovir DF Telbivudine Emtricitabine
<b>HIV Protease Inhibitors</b> Saquinavir Ritonavir Lopinavir/Ritonavir Atazanavir Nelfinavir mesylate Darunavir Tipranavir Fosamprenavir	<b>Non-nucleoside HIV Reverse Transcriptase Inhibitors</b> Nevirapine Efavirenz  <b>HIV Fusion Inhibitors</b> Enfurvitide
<b>HIV Integrase Inhibitor</b> Raltegravir	<b>HIV Entry Inhibitor</b> Maraviroc
<b>Immune Modulators</b> Ribavirin Peginterferon alfa-2a Peginterferon alfa-2b	<b>Compounds for Treatment of Herpes Viruses</b> Ganciclovir Valganciclovir HCl Acyclovir

## NON-CLINICAL PERFORMANCE EVALUATION

### A. Limit of Detection

The limit of detection of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 was determined by testing the 2<sup>nd</sup> International HIV-1 RNA WHO Standard, NIBSC Code 97/650<sup>30</sup>, HIV-1 subtype B, diluted in HIV-1-negative human EDTA plasma. The limit of detection was determined for 3 reagent lots. Three dilution series were analyzed for each reagent lot. A total of approximately 126 replicates per concentration level were tested. The evaluation was performed according to CLSI Guideline EP17-A.

The concentration of HIV-1 RNA that can be detected with a positivity rate of greater than 95% as determined by PROBIT Analysis, is 20 cp/mL or 33 IU/mL. The results for the individual lots were 17.7 cp/mL (95% confidence interval: 13.7 – 26.9 cp/mL) for lot 1, 17.0 cp/mL (95% confidence interval: 14.0 – 22.6 cp/mL) for lot 2 and 14.2 cp/mL (95% confidence interval: 11.2 – 22.1 cp/mL) for lot 3. The combined results for all three reagent lots are shown in Table 2. The conversion factor is 0.6 cp/IU for the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0.

**Table 2**  
**Limit of Detection of the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0 Using the WHO International Standard and PROBIT Analysis**

Nominal Input (HIV-1 RNA IU/mL)	Nominal Input (HIV-1 RNA cp/mL)	No. Replicates	No. Positives	Positivity Rate
<b>100</b>	<b>60</b>	<b>126</b>	<b>126</b>	<b>100%</b>
67	40	186	185	99%
50	30	126	125	99%
<b>33</b>	<b>20</b>	<b>126</b>	<b>124</b>	<b>98%</b>
25	15	59	53	90%
17	10	126	108	86%
8	5	125	66	53%
0	0	126	0	0%
<b>PROBIT 95% Hit Rate</b>		<b>27.5 IU/mL</b> (95% confidence interval: 23.8 – 33.0 IU/mL) <b>16.5 cp/mL</b> (95% confidence interval: 14.3 – 19.8 cp/mL)		

In addition, dilutions of cell culture supernatants representing HIV-1 group M subtypes A-H in HIV-1-negative human EDTA plasma were analyzed with 2 reagent lots. Concentrations above, at and below the LOD of 20 cp/mL were tested in replicates of n = 24 per reagent lot. The assignment of nominal concentrations to the cell culture stock materials was performed by averaging the titers of the COBAS® AMPLICOR® HIV-1 MONITOR Test, v1.5; the VERSANT® HIV-1 RNA 3.0 assay (bDNA); and the Abbott RealTime HIV-1 assay. Hit rate analysis shows a positivity rate of > 95% for all subtypes at ≤ 20 cp/mL. The combined results for the 2 reagent lots are shown in Table 3.

**Table 3**  
**Limit of Detection Verification for the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0 Using HIV-1 group M subtypes A-H 95% Hit Rate Analysis**

Subtype	Isolate Designation	Lowest Concentration Level 95% Hit Rate (cp/mL)
<b>A</b>	92UG029	10
<b>A</b>	4237A/98	20
<b>B</b>	92TH026	20
<b>B</b>	8E5/LAV	20
<b>C</b>	92BR025	20
<b>C</b>	3777A/97	11
<b>D</b>	92UG021	20
<b>D</b>	92UG035	11
<b>E</b>	92TH022	12
<b>E</b>	92TH009	14
<b>F</b>	93BR020	20
<b>G</b>	ARP173/RU570	13
<b>H</b>	HIV V1557	16

**B. Precision**

The precision of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 was determined by analysis of serial dilutions of a HIV-1 cell culture supernatant specimen (HIV-1 subtype B) in HIV-1-negative human EDTA plasma. The titer assignment of the cell culture supernatant (stock concentration) was performed by a method that ensures traceability to the 1<sup>st</sup> International HIV-1 RNA WHO Standard, NIBSC Code 97/656<sup>†</sup>. Three reagent lots were analyzed and 15 runs per reagent lot were performed, each consisting of 6 dilution levels and 3 replicates at each level. Each specimen was taken through the entire COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 procedure, including specimen preparation, amplification and detection. Therefore, the precision reported here represents all aspects of the test procedure. The results for each reagent lot and for the 3 reagent lots combined are shown in Table 4.

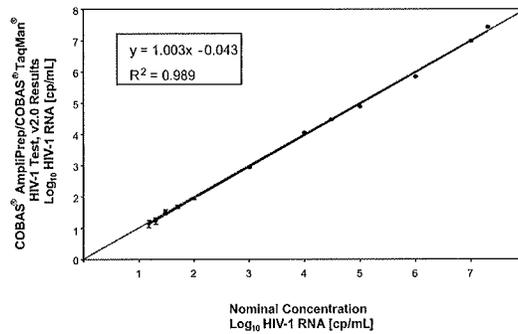
**Table 4**  
**Precision of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0**

Titer (cp/mL)	Lot 1	Lot 2	Lot 3	All 3 lots combined	
	Total SD in log	Total Log normal CV (%)			
1.0E+02	0.19	0.16	0.17	0.17	41
1.0E+03	0.07	0.09	0.07	0.08	20
1.0E+04	0.07	0.07	0.06	0.07	16
1.0E+05	0.04	0.05	0.07	0.06	15
1.0E+06	0.10	0.09	0.10	0.10	25
1.0E+07	0.11	0.12	0.14	0.13	33

**C. Linear Range**

The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 was found to give a linear response from 20 (Log<sub>10</sub> = 1.30) HIV-1 RNA cp/mL to 1.0E+07 (Log<sub>10</sub> = 7.00) HIV-1 RNA cp/mL. The evaluation was performed according to CLSI Guideline EP6-A using 2 lots of COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 reagents and serial dilutions of a high titer HIV-1 RNA (+) cell culture supernatant specimen<sup>33</sup>. Two reagent lots were analyzed and 15 runs per reagent lot were performed, each consisting of 12 dilution levels and 3 replicates at each level. The results for 1 reagent lot are shown in Figure 7.

**Figure 7**  
**Linearity for the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0**



#### D. Inclusivity of HIV-1 Group M

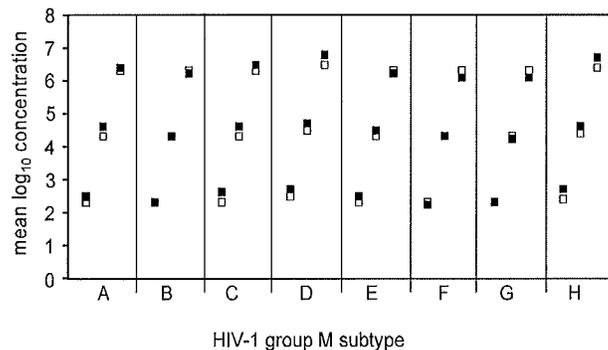
Eight subtype categories have been proposed for HIV-1 group M based on nucleotide divergence. These subtypes are designated with capital alphabetical letters from A through H<sup>31</sup>.

The performance of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 on HIV-1 subtypes was evaluated by analysis of cell culture stock material of representatives for each HIV-1 group M subtypes A through H. The assignment of nominal concentrations to the cell culture stock materials was performed by averaging the titers of the COBAS<sup>®</sup> AMPLICOR<sup>®</sup> HIV-1 MONITOR Test, v1.5; the VERSANT<sup>®</sup> HIV-1 RNA 3.0 assay (bDNA); and the Abbott RealTime HIV-1 assay. Each cell culture stock material was diluted to nominal concentrations of approximately 2.00E+02, 2.00E+04 and 2.00E+06 cp/mL in EDTA plasma. The concentrations were then tested in 10 replicates by the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 using 1 reagent lot. The mean log<sub>10</sub> titers of all concentrations and subtypes were compared to the respective log<sub>10</sub> nominal titers.

The evaluation of the 8 HIV-1 subtype isolates by the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 demonstrates equivalent results for all tested representatives of the HIV-1 group M subtypes (see Figure 8). Mean log<sub>10</sub> concentration results for all subtypes were within ±0.3 log<sub>10</sub> of the assigned input concentration.

**Figure 8**  
**COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0**  
**Inclusivity Testing – Cell Culture Supernatants**

□ nominal concentration ■ COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0

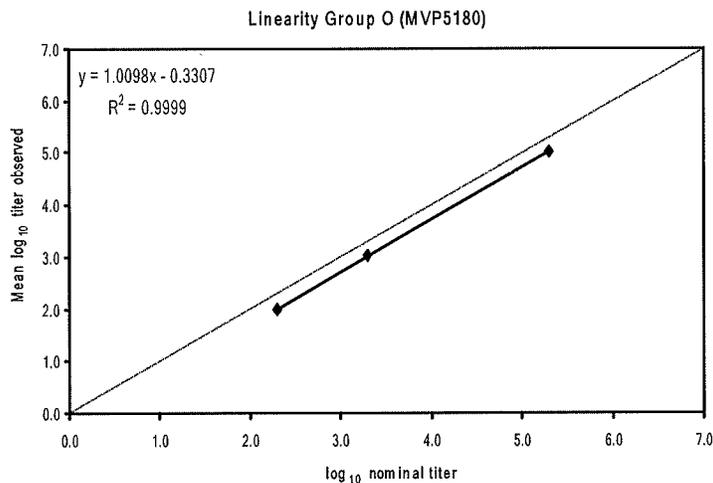


**E. HIV-1 Group O Detection**

Dilutions of a HIV-1 group O cell culture supernatant (isolate MVP5180) in human EDTA plasma were analyzed with 2 reagent lots. Five concentration levels at approximately 10, 20, 30, 50 and 75 cp/mL were tested in 24 replicates per reagent lot. Assignment of the nominal concentration to the cell culture stock material was performed by an FDA-approved assay. Hit rate analysis shows a positivity rate of greater than 95% at 20 cp/mL.

The HIV-1 group O cell culture stock material was diluted to nominal concentrations of approximately 2.00E+02, 2.00E+03, and 2.00E+05 cp/mL in EDTA plasma. The concentrations were then tested in 10 replicates by the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 using 1 reagent lot. The mean log<sub>10</sub> titers of all concentrations were linear and within ± 0.3 log<sub>10</sub> of the respective log<sub>10</sub> nominal titer (see Figure 9).

**Figure 9**  
**COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0**  
**Inclusivity Testing – HIV-1 Group O**



In addition, 10 cell culture materials and one diluted patient specimen (11613) representing HIV-1 group O were tested in parallel in the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 and in the Abbott RealTime HIV-1 assay. All 11 specimens were found positive with the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 (see Table 5). Both tests returned a mean log<sub>10</sub> titer for the 11 specimens within 0.1 log<sub>10</sub>.

**Table 5**  
**Recognition of HIV-1 Group O Isolates by the**  
**COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0**

<b>Isolate Designation</b>	<b>Log<sub>10</sub> Titer COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0</b>	<b>Log<sub>10</sub> Titer Abbott RealTime HIV-1 Assay</b>
BBI PRD 301, BV-5050	3.09	2.42
BBI PRD 301, BV-5051	2.86	3.35
BBI PRD 301, BV-5003	3.00	2.71
BBI PRD 301, BV-5024	2.87	2.69
MVP5180	2.78	3.25
HIV-1 CA-9	3.31	3.08
BCF01	5.71	5.61
BCF02	5.16	5.39
BCF07	4.27	4.81
BCF011	5.57	5.26
11613	2.97	2.05
<b>Mean Log<sub>10</sub> Titer</b>	<b>3.78</b>	<b>3.69</b>

**F. Specificity**

The specificity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 was determined with 2 reagent lots by analysis of HIV-1-negative EDTA plasma specimens from blood donors. A total of 660 individual EDTA plasma specimens showed valid results and all were negative for HIV-1 RNA in the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0. Based on these results, the specificity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 is 100% (one-sided lower 95% confidence limit: 99.6%).

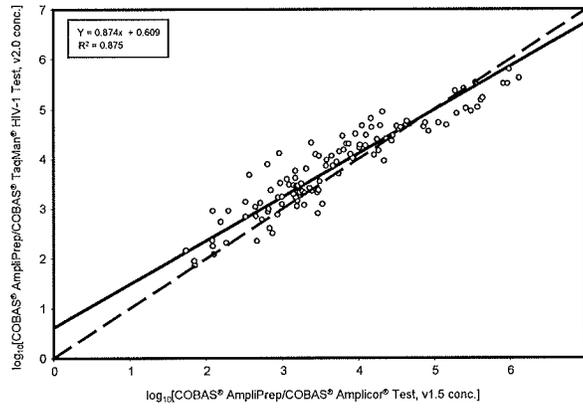
**G. Analytical Specificity**

The analytical specificity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 was evaluated by adding cultured organisms (viruses, bacteria, yeast) or DNA (HTLV-II) at 5E+04 particles/mL input concentration into HIV-1-negative human EDTA plasma and into HIV-1-positive EDTA plasma at 1.5E+02 cp/mL HIV-1 (see Table 6).

None of the organisms tested showed cross reactivity with the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0. HIV-1-positive specimens returned titer results that were within  $\pm 0.5 \log_{10}$  from a HIV-1-positive control.

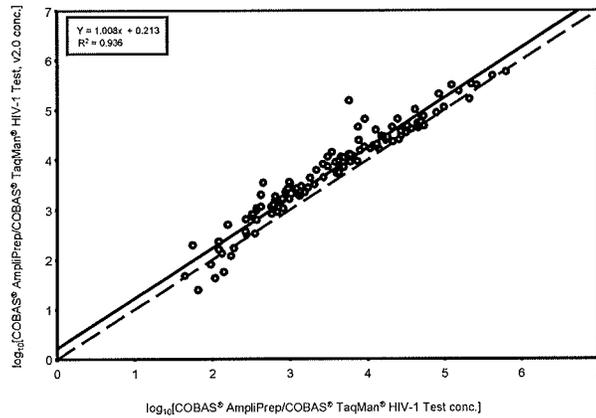


**Figure 10**  
**Correlation of the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0**  
**and the COBAS® AmpliPrep/COBAS® AMPLICOR® HIV-1 MONITOR Test, v1.5**



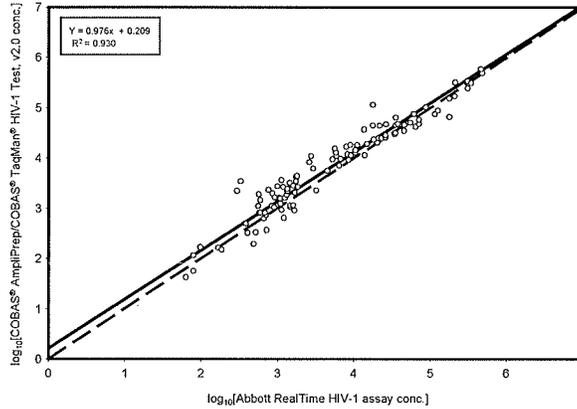
**Note:** The dashed line indicates perfect agreement between any two test methods, i.e.,  $y = x$ .

**Figure 11**  
**Correlation of the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test, v2.0**  
**and the COBAS® AmpliPrep/COBAS® TaqMan® HIV-1 Test**



**Note:** The dashed line indicates perfect agreement between any two test methods, i.e.,  $y = x$ .

**Figure 12**  
**Correlation of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0**  
**and the Abbott RealTime HIV-1 assay**



**Note:** The dashed line indicates perfect agreement between any two test methods, i.e.,  $y = x$ .

#### CLINICAL PERFORMANCE EVALUATION

##### Reproducibility

Reproducibility of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 Test was evaluated in EDTA plasma using 2 different workflows (COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer System and COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer System). The study was performed using panels constructed from well-characterized HIV-1 group M, subtype B cultured virus stock and from EDTA plasma that was negative for HIV-1 RNA and HIV-1/2 antibodies. The panel covered the dynamic range of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 as well as the key medical decision points for the intended use and supported by the 2008 Department of Health and Human Services Guidelines for the Use of Antiretroviral Agents in HIV-1-Infected Adults and Adolescents<sup>15</sup>. The study was designed to evaluate key variables contributing to total precision variance, including lot, site/instrument, operator, day/run, and within-run. Additional analysis were conducted to compare the performance characteristics and comparative precision variability between the two workflows. Two operators at each of 3 sites performed 5 days of testing with each of 3 reagent kit lots using each workflow. Each run consisted of one set of controls (1 high positive, 1 low positive, and 1 negative) and a 7-member panel tested in triplicate (21 sample) on the COBAS<sup>®</sup> AmpliPrep Instrument. The prepared samples and controls were amplified and detected on the COBAS<sup>®</sup> TaqMan<sup>®</sup> Analyzer or on COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 Analyzer.

Reproducibility was evaluated by using a random effects model with terms for (a) lot, (b) site/instrument, (c) operator nested within site/instrument, (d) day/run nested within lot, site/instrument, and operator, and (e) aliquots within-run components by using PROC MIXED and  $\log_{10}$  transformed results. The percentage of variability due to each component and coefficient of variation of the  $\log_{10}$  transformed HIV-1 RNA concentration were calculated. Only the Within Assay Range (2.00E+1 to 1.00E+7 cp/mL) data were investigated.

Table 7 shows the total precision variance and total precision standard deviation obtained from the COBAS® AmpliPrep/COBAS® TaqMan® Analyzer System as determined by analysis of variance. In general, the within-run component contributed more variability than other components.

**Table 7**  
**Attributable Percentage of Total Variance, Total Precision Standard Deviation, and Lognormal CV of HIV-1 RNA Concentration ( $\log_{10}$  cp/mL) from Tests Within Assay Range**

HIV-1 RNA Concentration ( $\log_{10}$ cp/mL)			Contribution to Total Variance (%)					Total Precision
Expected	Observed (Average)	No. of Valid Tests <sup>1</sup>	Lot	Site/Instrument	Operator	Day/Run	Within-Run	Standard Deviation (Lognormal %CV)
1.699	1.832	270	5%	2%	0%	8%	85%	0.20 (48%)
2.602	2.676	275	6%	1%	0%	17%	77%	0.11 (25%)
3.000	3.067	274	16%	0%	4%	12%	69%	0.10 (24%)
3.699	3.822	273	20%	6%	0%	17%	57%	0.10 (23%)
4.699	4.746	273	27%	0%	0%	14%	59%	0.07 (17%)
5.699	5.644	274	33%	10%	0%	19%	38%	0.10 (23%)
6.699	6.751	259	27%	14%	0%	20%	39%	0.12 (27%)

Note: Within assay range results are from 20 cp/mL to 1.00E+7 cp/mL (1.30  $\log_{10}$  cp/mL to 7.00  $\log_{10}$  cp/mL), inclusive.

<sup>1</sup> Number of tests within assay range.

Results obtained from the COBAS® AmpliPrep/COBAS® TaqMan® 48 System Workflow are summarized in Table 8. In general, the within-run component contributed more variability than other components with the exception of the highest titer panel member.

**Table 8**  
**Attributable Percentage of Total Variance, Total Precision Standard Deviation, and Lognormal CV of HIV-1 RNA Concentration ( $\log_{10}$  cp/mL) from Tests Within Assay Range**

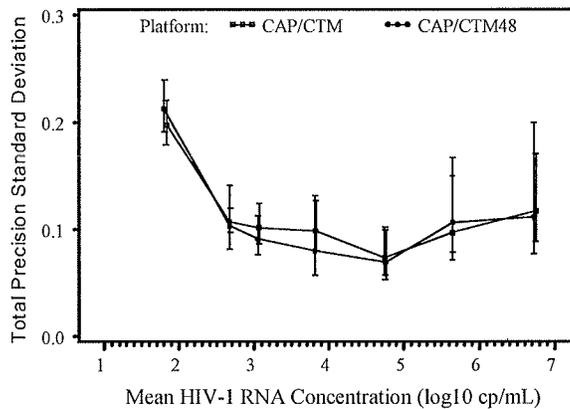
HIV-1 RNA Concentration ( $\log_{10}$ cp/mL)			Contribution to Total Variance (%)					Total Precision
Expected	Observed (Average)	No. of Valid Tests <sup>1</sup>	Lot	Site/Instrument	Operator	Day/Run	Within-Run	Standard Deviation (Lognormal %CV)
1.699	1.804	266	7%	2%	0%	2%	89%	0.21 (52%)
2.602	2.672	273	26%	0%	2%	5%	68%	0.10 (24%)
3.000	3.048	272	17%	0%	0%	6%	77%	0.09 (21%)
3.699	3.814	271	39%	0%	2%	13%	46%	0.08 (19%)
4.699	4.756	272	30%	0%	0%	10%	61%	0.07 (16%)
5.699	5.647	272	35%	0%	6%	16%	43%	0.11 (25%)
6.699	6.727	269	45%	0%	4%	13%	38%	0.11 (26%)

Note: Within assay range results are from 20 cp/mL to 1.00E+7 cp/mL (1.30  $\log_{10}$  cp/mL to 7.00  $\log_{10}$  cp/mL), inclusive.

<sup>1</sup> Number of tests within assay range.

The results shown in Figure 13 display the plot of the total precision standard deviation with the corresponding approximate 95% Confidence Intervals against the mean  $\log_{10}$  HIV-1 RNA concentrations. These results indicate a comparable precision performance between the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> (CAP/CTM) System and the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> 48 (CAP/CTM48) System configurations.

**Figure 13**  
**Total Precision Standard Deviation (approximate 95% CI)**  
**versus Mean HIV-1 RNA Concentration**



Note: The approximate 95% CI for the total precision standard deviation was calculated by taking the square root of the 95% CI bounds of the total precision variance.

### Clinical Sensitivity, Specificity and Method Comparison

#### Methodology

The primary objective of this study was to evaluate the clinical specificity and sensitivity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 in specimens from HIV-negative and HIV-1-positive subjects. Both fresh (never frozen) and frozen EDTA plasma samples were tested in each of the evaluations. The secondary objectives were to compare results and evaluate the positive percent agreement and negative percent agreement of COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 results to those obtained with the FDA-approved tests, COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test and the COBAS<sup>®</sup> AMPLICOR HIV-1 MONITOR Test, v1.5.

Clinical specificity was evaluated with the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 by testing 148 fresh (never frozen) samples and 418 frozen samples collected from blood donors who were negative for HIV-1/2 antibodies. Clinical sensitivity of the test was evaluated with the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 by testing 117 fresh samples and 301 frozen samples in EDTA plasma collected from HIV-1-infected subjects (frozen samples were randomly distributed across test sites by CD4 cell count category). Test results from the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 were compared to those obtained with the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test and COBAS<sup>®</sup> AMPLICOR HIV-1 MONITOR Test, v1.5. Testing was conducted at 3 test sites, with 1 COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> System per site. Three COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 reagent lots were used.

### Statistical Methods

Fresh and frozen samples from HIV-negative and HIV-1-positive subjects were tested with the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0, the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, and the COBAS<sup>®</sup> AMPLICOR<sup>®</sup> HIV-1 MONITOR Test, v1.5. HIV-negative subjects were evaluable for statistical analyses of the specificity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 if they generated valid COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 results. HIV-1-positive subjects were evaluable for statistical analyses of the sensitivity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 if they generated valid COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 results and had valid COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test results within the linear range of the assay.

The clinical specificity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 was calculated as the percentage of evaluable HIV-negative subjects who had Target Not Detected COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 results. The associated 95% exact confidence interval (CI) was also provided. The clinical sensitivity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 was calculated as the percentage of evaluable HIV-1-positive subjects who had detectable HIV-1 viral load on the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0. The associated 95% exact confidence interval (CI) was also provided. The method comparison evaluated COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 results separately with both comparative platforms (COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test and the COBAS<sup>®</sup> AMPLICOR<sup>®</sup> HIV-1 MONITOR Test, v1.5). Positive and negative percent agreements were calculated between the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 and each comparative platform. Paired samples from HIV-1-positive subjects contributing within linear range results for both the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 and the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test were compared using scatter plots and analyzed using the Deming regression.

### Results

A total of 566 evaluable HIV-negative and 418 HIV-1-positive patient specimens were included in clinical specificity and sensitivity analyses. Approximately 75% of the patient specimens were frozen and 25% were fresh. The specific distribution of each platform is summarized in Table 9.

**Table 9**  
**Evaluable HIV-1 Negative and Positive Subjects by Sample Type**

Sample Type	HIV-Negative Specimens	HIV-1-Positive Specimens
Fresh	148 ( 26.1%)	117 ( 28.0% )
Frozen	418 ( 73.9%)	301 ( 72.0% )
Total	566	418

The demographic characteristics of the 418 evaluable HIV-1-positive specimens are summarized in Table 10. The CD4 cell counts of the subjects distributed approximately evenly across CD4 cell count categories (<200, 200-500, >500 cells/uL). Most of the subjects were male (74.2%) and between 30 to 49 years of age (72.5%). The ethnic distribution is comparable to that observed in the HIV-1 population of the United States<sup>32</sup>.

**Table 10**  
**Demographic Characteristics of Evaluable HIV-1-Positive Subjects**

Demographic Characteristic	Category	HIV-1-Positive Subjects
<b>Overall</b>	Total	418
<b>CD4 Cell Count (cells/uL)</b>	< 200	130 (31.1%)
	200 - 500	152 (36.4%)
	> 500	136 (32.5%)
<b>Sample Type</b>	Fresh	117 (28.0%)
	Frozen	301 (72.0%)
<b>Sex</b>	Male	310 (74.2%)
	Female	108 (25.8%)
<b>Age (Years)</b>	18-29	23 (5.5%)
	30-39	100 (23.9%)
	40-49	203 (48.6%)
	50-59	74 (17.7%)
	≥ 60	18 (4.3%)
<b>Ethnicity</b>	Caucasian	129 (30.9%)
	Hispanic	46 (11.0%)
	Black	223 (53.3%)
	Asian / Pacific Islander	3 (0.7%)
	Other	17 (4.1%)
<b>On Antiretroviral Medication</b>	Yes	240 (57.4%)
	No	178 (42.6%)

The clinical specificity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 (Table 11) was 99.3% (562/566; 95% CI = 98.2% to 99.8%), with 4 specimens classified as false positives. Three of these specimens were reported at < 20 cp/mL, below the LLoQ of the assay. The remaining single specimen out of the 566 tested was within the linear range but at a very low titer (28.8 cp/mL). The clinical specificity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 was similar for both fresh specimens (99.3% [147/148; 95% CI = 96.3% to 100%]) and frozen specimens (99.3% [415/418; 95% CI = 97.9% to 99.9%]).

**Table 11**  
**Clinical Specificity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0**

Subject Group	CAP/CTM HIV-1 Test, v2.0		Total N	Clinical Specificity (95% exact CI)
	Positive	Negative		
<b>HIV-Negative</b>	4 ( 0.7% )	562 ( 99.3% )	566	99.3% (98.2%, 99.8%)

The clinical sensitivity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 was defined as the percentage of evaluable HIV-1-positive subjects who had a positive COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 result and is summarized in Table 12. The clinical sensitivity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 was 100% (418/418; 95% CI = 99.1% to 100%). There were no subjects that had false negative COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 results. The clinical sensitivity was tested in an HIV-patient population reflective of that in the United States with regards to gender, age, ethnicity and exposure to antiretroviral therapy<sup>20</sup>. The test demonstrated 100% clinical sensitivity independent of the above listed demographics, CD4 cell count, or sample type (fresh versus frozen).

**Table 12**  
**Clinical Sensitivity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0**

Subject Group	CAP/CTM HIV-1 Test, v2.0		Total N	Clinical Sensitivity (95% exact CI)
	Positive	Negative		
<b>HIV-1-Positive</b>	418 ( 100.0% )	0 ( 0.0% )	418	100.0% (99.1%, 100.0%)

#### Clinical Method Comparison

##### COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 versus the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test

The comparison of COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 and COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test results for the 950 subjects eligible for the analysis is summarized in Table 13. The positive percent agreement of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 with respect to the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test was 99.5% (427/429; 95% CI = 98.3% to 99.9%). The negative percent agreement of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 with respect to the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test was 98.1% (511/521; 95% CI = 96.5% to 99.1%). There were 10 samples with positive COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 results and negative COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test results. Three samples were at titers below the LLoQ of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, most likely a reflection of the increased sensitivity of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0. Three samples were false positive COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test v2.0 results from HIV-negative subjects identified in the clinical specificity analysis that again were below the LLoQ. Four samples had titers ranging from 24.9 cp/mL to 156 cp/mL and are likely reflective of the known variability associated with low titer quantitation.

**Table 13**  
**Comparison of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 versus the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test**

CAP/CTM HIV-1 Test, v2.0	CAP/CTM HIV-1 Test		Total
	Positive	Negative	
Positive	427	10	437
Negative	2	511	513
Total	429	521	950
Positive Percent Agreement (95% exact CI)	99.5% (98.3%, 99.9%)		
Negative Percent Agreement (95% exact CI)		98.1% (96.5%, 99.1%)	

CI = confidence interval; CAP/CTM HIV-1 Test = COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan HIV-1 Test; CAP/CTM HIV-1 Test, v2.0 = COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan HIV-1 Test, v2.0.

Note: HIV-negative and HIV-1-positive subjects contributing both valid CAP/CTM HIV-1 Test, v2.0 and CAP/CTM HIV-1 Test results were included in this summary table.

A total of 417 paired HIV-1-positive samples had results within the linear range of both assays and were evaluable for the method comparison analysis. Table 14 shows the mean paired difference and 95% CI for the bias between the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 and the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test. The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 returns higher titers than the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, except at both the higher range (> 5 log<sub>10</sub> cp/mL) and the lower range (< 2 log<sub>10</sub> cp/mL) where it returns titers that are lower (see Figure 14). The overall systematic bias is estimated as 0.2591 log<sub>10</sub> cp/mL.

**Table 14**  
**Mean Paired Difference and 95% CI for the bias between the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 and the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test**

Number of Paired HIV-1-Positive Samples Within Linear Range of Both Assays = 417		
Mean Difference (log <sub>10</sub> cp/mL)	Standard Error	95% CI
0.2591	0.0122	( 0.235, 0.283 )

CI = confidence interval; CAP/CTM HIV-1 Test = COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan HIV-1 Test; CAP/CTM HIV-1 Test, v2.0 = COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan HIV-1 Test, v2.0.

Note: HIV-1-positive subjects contributing both valid CAP/CTM HIV-1 Test and CAP/CTM HIV-1 Test, v2.0 results within the linear range of each assay were included in this summary table.

The results of the Deming regression analysis between COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 and COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test results for paired HIV-1-positive specimens within the linear range of both assays are tabulated in Table 15 and displayed graphically in Figure 14 (in this figure, the dashed line indicates perfect agreement between the two test methods, i.e., y = x).

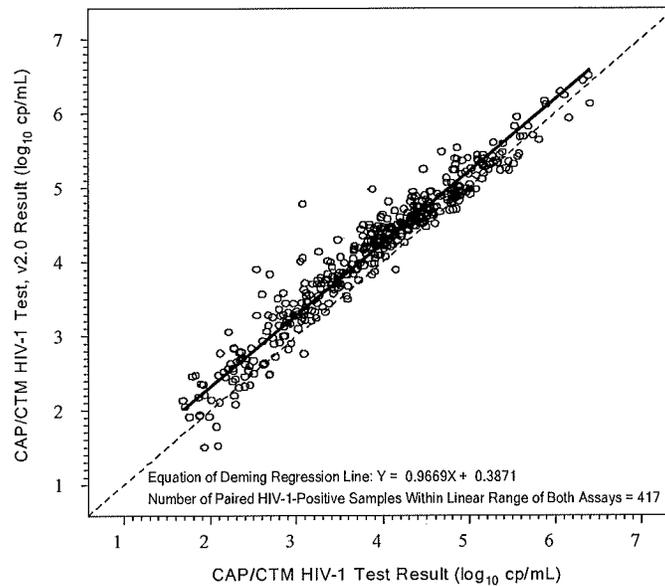
**Table 15**  
**Parameter Estimates from Deming Regression Analysis Between the**  
**CAP/CTM HIV-1 Test, v2.0 and the CAP/CTM HIV-1 Test**

Number of Paired HIV-1-Positive Samples Within Linear Range of Both Assays = 417				
Parameter	Parameter Estimate log <sub>10</sub> cp/mL	Standard Error	95% CI	r <sup>2</sup>
Intercept	0.3871	0.0488	( 0.291, 0.483)	0.9375
Slope	0.9669	0.0122	( 0.943, 0.991)	

CI = confidence interval; CAP/CTM HIV-1 Test = COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan HIV-1 Test; CAP/CTM HIV-1 Test, v2.0 = COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan HIV-1 Test, v2.0.

Note: HIV-1-positive subjects contributing both valid CAP/CTM HIV-1 Test and CAP/CTM HIV-1 Test, v2.0 results within the linear range of each assay were included in this summary table.

**Figure 14**  
**Deming Regression Analysis Between the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0**  
**and the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test**



COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 versus COBAS<sup>®</sup> AMPLICOR<sup>®</sup> HIV-1 MONITOR Test, v1.5

Table 16 shows the comparison of COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 and CA HIV-1 MONITOR Test, v1.5 results for 991 subjects eligible for the analysis. The positive percent agreement of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 with respect to the COBAS<sup>®</sup> AMPLICOR<sup>®</sup> (CA) HIV-1 MONITOR Test, v1.5 was 100% (419/419; 95% CI = 99.1% to 100%). The negative percent agreement of the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 with respect to the COBAS<sup>®</sup> AMPLICOR<sup>®</sup> HIV-1 MONITOR Test, v1.5 was 97.4% (557/572; 95% CI = 95.7% to 98.5%). Of the 15 subjects with positive COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test v2.0 results and negative COBAS<sup>®</sup> AMPLICOR<sup>®</sup> HIV-1 MONITOR Test, v1.5 results, 4 were false positive COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test v2.0 results from HIV-negative subjects identified in the clinical specificity analysis that again were below the LLoQ. Eleven were from HIV-1-positive subjects with COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test v2.0 results ranging from below the LLoQ to 223 cp/mL and negative COBAS<sup>®</sup> AMPLICOR<sup>®</sup> HIV-1 MONITOR Test, v1.5 results.

**Table 16**  
**Comparison of the CAP/CTM HIV-1 Test, v2.0 With the Cobas Amplicor HIV-1 MONITOR Test, v1.5**

CAP/CTM HIV-1 Test, v2.0	CA HIV-1 MONITOR Test, v1.5		Total
	Positive	Negative	
Positive	419	15	434
Negative	0	557	557
Total	419	572	991
Positive Percent Agreement (95% exact CI)	100.0% (99.1%, 100.0%)		
Negative Percent Agreement (95% exact CI)		97.4% (95.7%, 98.5%)	

CI = confidence interval; CAP/CTM HIV-1 Test = COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test; CAP/CTM HIV-1 Test, v2.0 = COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0.

Note: HIV-negative and HIV-1-positive subjects contributing both valid CAP/CTM HIV-1 Test, v2.0 and CA HIV-1 MONITOR Test, v1.5 results were included in this summary table.

**Conclusion**

The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 exhibits high levels of agreement with the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test in quantitative analyses ( $r^2 = 0.9375$ ) and in concordance analyses (positive percent agreement = 99.5%; negative percent agreement = 98.1%). It quantifies clinical specimens 0.2591  $\log_{10}$  cp/mL higher overall than the COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, with lower quantitation at the higher range ( $> 5 \log_{10}$  cp/mL) and the lower range ( $< 2 \log_{10}$  cp/mL).

The COBAS<sup>®</sup> AmpliPrep/COBAS<sup>®</sup> TaqMan<sup>®</sup> HIV-1 Test, v2.0 also shows high levels of agreement with the COBAS<sup>®</sup> AMPLICOR<sup>®</sup> HIV-1 MONITOR Test, v1.5 in concordance analyses (positive percent agreement = 100.0%; negative percent agreement = 97.4%).

These test results support the utility of the test for the intended use of assessing disease progression and monitoring antiretroviral therapy in HIV-1 infected patients.

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Document Revision Information	
Doc Rev. 2.0 09/2011	<p>Update of section "<b>MATERIALS REQUIRED BUT NOT PROVIDED</b>" to reflect the introduction of AL 3.3 and <b>cobas p 630</b></p> <p>AL 3.3 and <b>cobas p 630</b> manual reference in section "<b>INSTRUCTIONS FOR USE</b>" needs to be added.</p> <p>Add of a note in section "<b>Part D: Loading and Ordering of Specimen</b>" to refer to the <b>cobas p 630</b> manual</p> <p>Section "<b>Batch Validation</b>" is renamed to "<b>Batch Validation – AMPLILINK version 3.2 Series</b>".</p> <p>Section "<b>Batch Validation – AMPLILINK version 3.3 Series</b>" added.</p> <p>Update chemical name for ProClin in the <b>REAGENTS</b> section.</p> <p>Please contact your local Roche Representative if you have any questions.</p>



Roche Molecular Systems, Inc., Branchburg, NJ 08876 USA  
A Member of the Roche Group



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09/2011

05328276001-02

Doc Rev. 2.0

05328276001-02EN

43

Doc Rev. 2.0

The following symbols are now used in labeling for Roche PCR diagnostic products.

	Ancillary Software		For <i>in vitro</i> diagnostic use
	Authorized representative		For IVD Performance Evaluation only
	Barcode Data Sheet		Lower Limit of Assigned Range
	Batch code		Manufactured by
	Biological Risk (Potentially biohazardous material)		Store in the dark
	Catalogue Number		Temperature limitation (Store At)
	Consult instructions for use		Test Definition File
	Contains sufficient for < n > tests		Upper Limit of Assigned Range
	Contents of kit		Use by (last day of month)
	Distributed by		

US Customer Technical Support 1-800-428-2336

[SP1EN-01]  
05328276001-02  
(Variable IFU M/N Rev)

NEXT BID

September 4, 2013

Abbott Molecular, a division of Abbott Laboratories, specializes in the development and commercialization of molecular diagnostics testing products for the Clinical Diagnostic Market. In June, 2013, we celebrated our eighth year anniversary of successful commercialization of RealTime PCR products.

Abbott Molecular combines our highly sensitive assays with the advanced technology of the *m2000* System, a fully automated RealTime PCR platform. The *m2000* System combines automated precision pipetting, primary tube sampling and RealTime PCR technology to create a platform that helps laboratories maximize workflow efficiency, minimize hands-on time and improve laboratory throughput.

With our *m2000* system, we have a versatile platform that enables menu consolidation through advanced automation and helps increase confidence in your clinical results. By continuing to perform your HIV-1 viral load testing on the *m2000* platform that already exists in your laboratory, you eliminate any costs and additional labor required to train on a new system. Additional labor savings can be realized by eliminating the need to re-validate HIV-1 on a new platform which can be labor intensive. Furthermore, potential for future consolidation allows for better return on footprint and a reduction on service and inventory costs associated with multiple platforms.

Abbott's investment in research and development of novel assays in the area of HIV, spans over four decades. It is this experience that allows us to deliver best in class assays and a fully automated testing solution for the San Diego Public Health Laboratory. We are confident we can continue to demonstrate that our testing solution will provide the best in patient care by delivering superior overall value to help drive improved performance and outcomes, and assist with cost containment through improved workflow efficiencies and informatics.

Our system not only meets the requirements outlined in this solicitation, but also includes the following unique specifications not found in competitive systems:

- A single molecular platform capable of performing FDA approved tests which include HIV-1, HCV, HBV, HCV Genotype, CT/NG, VRE, and Flu A, Flu B, RSV
- The *m2000sp* is an open platform that offers flexible protocols for various sample type and volumes, including RNA, DNA and Total Nucleic Acid options
- The *m2000rt* allows for complete flexibility in defining laboratory-based real-time PCR applications. Abbott FDA approved assays, ASR's and non-Abbott reagents can be run on the system
- 2-point external calibration to generate a stored calibration curve which lends to higher precision assays
- The Abbott Molecular *m2000* system is the only PCR based system that uses a non-competing internal control to validate assay performance
- The *m2000rt* is the only system that uses the maxRatio Data Analysis which incorporates multiple validity checks for improved confidence in patient results. This validates the growth curves and performance of each reaction
- A control strategy where only one set of controls is required regardless of batch size, and may be placed anywhere within the run

Enclosed is Abbott Molecular's response to San Diego County Public Health Laboratory RFB for HIV-1 Viral Load testing that includes equipment (Reagent Rental) and reagents to quantify nucleic acid in patient plasma specimens.



1300 E. Touhy Avenue  
Des Plaines, IL 60018

Our proposal is based upon continuing to provide the most comprehensive program as defined by the detailed discussions with San Diego County Public Health Lab regarding the technical characteristics required by the site. We believe that the proposal offered by Abbott represents the best technological option and best value at the best price for the San Diego County testing program.

Abbott has worked diligently to contain costs within this offer and yet include the robust requirements to meet every technical, service, and support specification of the RFB.

We are proud to work with the County of San Diego to extend the finest healthcare to its patients through the California State Therapeutic Monitoring Program (TMP).

Thank you for allowing Abbott Molecular to propose one *m2000* automated RealTime PCR System as a solution for your facility. We appreciate your business and look forward to the opportunity to continue our partnership.

If there are any questions, please feel free to contact me.

Dan Delany  
Government Enterprise Manager  
Cell: 224-637-0182  
Email: [daniel.delany@abbott.com](mailto:daniel.delany@abbott.com)

**RFB 6130 - COUNTY OF SAN DIEGO  
HEALTH AND HUMAN SERVICES AGENCY  
PUBLIC HEALTH LABORATORY  
HIV-1 VIRAL LOAD TESTING SYSTEM  
SCOPE OF WORK/PURPOSE**

THE VENDOR SHALL PROVIDE AN INCLUSIVE SYSTEM FOR THE FULLY AUTOMATED TESTING FOR THE HIV-1 VIRAL LOAD IN PLASMA, INCLUDING THE REAGENTS, CONSUMABLES, EQUIPMENT, ANNUAL PREVENTATIVE MAINTENANCE AND REPAIRS (I.E., LABOR AND PARTS). THE PRICE PER TEST FOR AN ESTIMATED 600 TESTS PER MONTH IS TO BE INCLUDED IN THE PRICE OF THE TEST REAGENTS.

Abbott agrees to provide an inclusive system for fully automated testing for HIV-1 viral load in plasma which includes reagents, consumables, equipment, annual preventative maintenance and repairs (labor, travel, and parts) of the equipment in price of test reagents. **Please see Appendix A (PRICING SCHEDULE).**

**REQUIRED SPECIFICATIONS FOR THE HIV-1 VIRAL LOAD ASSAY SYSTEM**

THE HIV-1 VIRAL LOAD ASSAY SYSTEM MUST: BE A FULLY-AUTOMATED REAL-TIME RT-PCR (IN-VITRO REVERSE TRANSCRIPTION-POLYMERASE CHAIN REACTION (RT-PCR) ASSAY) SYSTEM FOR DETECTION AND QUANTIFICATION OF HUMAN IMMUNODEFICIENCY VIRUS TYPE 1 (HIV-1) WITH A QUANTIFICATION RANGE IN HUMAN PLASMA BETWEEN 40 TO 10,000,000 COPIES/ML. FULL AUTOMATION MEANS THAT THE PLASMA WILL BE SAMPLED DIRECTLY FROM A VACUTAINER-TYPE SAMPLE TUBE WITHOUT TRANSFER TO A SECOND TUBE OR ASSAY CUVETTE. THE SYSTEM MUST BE CAPABLE OF READING SD PHL LIMS BAR-CODE LABELS (CODE 128) DIRECTLY OFF THE SAMPLE TUBE. ONCE THE SAMPLE TUBES ARE LOADED INTO THE SYSTEM, ALL PIPETTING, SAMPLE EXTRACTION, NUCLEIC ACID PURIFICATION AND ASSAY REAGENT AND TEMPLATE HANDLING IS PERFORMED BY THE SYSTEM WITHOUT OPERATOR INTERVENTION.

The *m2000* System leverages advanced automation to maximize workflow efficiency and minimize Operator hands-on time. According to Argent Global Services, a third party, process engineering consulting firm, total hands-on time is less than 20 minutes. In addition, the Abbott RealTime assay(s) process includes large blocks of walk-away time during the specimen extraction process and PCR thermal cycling steps. The Abbott Molecular RealTime HIV-1 assay is an in-vitro, reverse-transcription polymerase chain reaction (RT-PCR) test that measures the amount of HIV-1 virus circulating in a positive patient's blood. Abbott's HIV-1 RealTime viral load test detects and quantitates all known HIV-1 groups and subtypes with high precision. RealTime viral load test detects and quantitates all known HIV-1 groups and subtypes with high precision. The Abbott Molecular RealTime HIV-1 Assay is the only FDA approved Real Time PCR HIV viral load assay that has a similar quantitation range of 40 copies/ml to 10 million copies/ml for all M, N, O groups and subtypes.

For the last four years, the Abbott *m2000* has been the fastest growing molecular platform with 28% growth last year. The *m2000* System™ is a fully automated, RealTime PCR platform comprised of two main components:

The *m2000sp*™ – Sample Preparation Platform

An automated sample preparation platform for the extraction of DNA, RNA or Total Nucleic Acid, preparation of RealTime PCR mastermix and preparation of the PCR plate.

2. THE CONTRACT BASE PERIOD WILL BE FOR TWO (2) YEARS WITH OPTIONS TO RENEW FOR YEARS 3 THROUGH 5.

Abbott agrees. Please see Appendix A-PRICING SCHEDULE.

3. ANY AND ALL PARTS, ADDITIONAL ITEMS, OR ACCESSORIES NOT USED ROUTINELY AS PART OF THE ASSAY, OR PROVIDED UNDER THE SERVICE CONTRACT (E.G. SAMPLE TUBE RACKS OR OTHER DURABLE PARTS), FOR THE SYSTEM SHOULD BE PROVIDED AS AN ITEMIZED LIST BY THE VENDOR AS A PERCENT OF DISCOUNT FROM LIST PRICE, SUBMIT LIST ALONG WITH PRICE SHEETS, PAGE A-6 & 6A.

Abbott agrees. Please see Appendix B-ANCILLARY PRODUCTS LIST.

4. THE SYSTEM MUST BE CAPABLE OF FULLY AUTOMATED READING OF BAR-CODE LABELS LABORATORY AND PATIENT BLOOD TUBES (SYMBOLGY CODE 128).

The m2000 is capable of reading all common barcode symbologies including Code 128.

5. FULLY AUTOMATED PRECISION PIPETTING TO ELIMINATE MANUAL MIXING OR MANIPULATION OF SAMPLES OR REAGENTS.

Abbott's design of the *m2000* system provides improved efficiency. Abbott *m2000* HIV-1 Viral Load laboratory barcoded test tubes can be loaded directly on the *m2000* system allowing for a leaner process. This feature eliminates the need for additional non-value added requirements to set up manufacturer specific commodities (such as tubes) and subsequent pipetting of samples and controls into those specific commodities that would be required with some competitive systems. The *m2000* system reads the laboratory barcoded test tube providing confidence in managing patient identification or chain of custody. This eliminates the need to create traymaps or worklists that link any manufacturer specific commodities to patient sample ID. Removing these non-value added steps will allow laboratories to take advantage of increased efficiency gains.

Primary specimen tubes or primary pour-off tubes received from accessioning can be placed directly into the *m2000sp* Sample Racks, so no pipetting of patient sample is required by the operator.

All Sample Preparation and Assay Amplification reagents come fully prepared and ready for placement on the *m2000* system. Assay calibration and control materials come prepared in individual, single use barcoded tubes, ready for placement on the *m2000*.

The *m2000* System improves sample and result quality through:

#### Automated Precision Pipetting

- *m2000* System automates the manual pipetting and mixing steps involved in NAAT technology
  - Automated Sample Preparation Extraction
    - automated extraction of RNA, DNA or Total Nucleic Acid
    - concentrates the target DNA or RNA
    - removes potential inhibitors of amplification
  - Automated generation of Master Mix
  - Automated pipetting and set up of optical reaction plate
- Automated precision pipetting increases result precision
- Enables Primary Tube Sampling

**6. SYSTEM MUST BE CAPABLE OF DIRECT SAMPLING OF 5 ML VACUTAINER-TYPE BLOOD COLLECTION TUBES.**

A number of studies have been conducted to determine the effect of specimen handling parameters on the measurement of HIV viral load. A recent Clinical Virology Symposium poster evaluated the effect of freezing plasma *in situ* in PPT on the accuracy of HIV-1 viral load results using the Abbott RealTime HIV-1 assay, and the effect of whole blood storage in the PPT for six hours at room temperature prior to centrifugation (PPT6H) instead of two hours as specified in the PPT product insert (PPT control). The author's conclusions were as follows:

**CONCLUSIONS**

We conclude that using the Abbott RealTime HIV-1 Viral Load Assay, specimens can be collected in BD Vacutainer® Plasma Preparation Tubes and:

- Centrifuged within 6 hours of phlebotomy with no effect on viral load results.
- Plasma separated in PPTs can be stored frozen *in situ* until the time of testing with no effect on viral load results.
- There is no misclassification of HIV viral load status when using PPT in combination with the Abbott RealTime HIV-1 Viral Load Assay for any of the handling conditions tested.

Adapted from Fernandes H. et al, Evaluation of Specimen Handling Conditions in BD Vacutainer® Plasma Preparation Tube HIV-1 Viral Load as Measured by the Abbott RealTime HIV-1 Assay, *Clinical Virology Symposium April 2009, Florida*

**7. FLEXIBLE THROUGHPUT OPTIONS OF 24 TO 96 SAMPLES IN ONE 8-HOUR SHIFT.**

The Abbott m2000 has the capability of flexible throughput of 24 to 96 samples in one 8-hour shift.

**8. HAVE FDA APPROVAL: THE SYSTEM MUST HAVE FDA APPROVAL FOR HIV-1 VIRAL LOAD TESTING.**

HIV-1 is characterized by exceptional genetic diversity which has significant implications

for diagnosis, monitoring and clinical management of patients. The Abbott Molecular RealTime HIV-1 assay is an in-vitro, reverse-transcription polymerase chain reaction (RT-PCR) test that measures the amount of HIV-1 virus circulating in a positive patient's blood. Abbott's HIV-1 RealTime viral load test detects and quantitates all known HIV-1 groups and subtypes with high precision. RealTime viral load test detects and quantitates all known HIV-1 groups and subtypes with high precision. The Abbott Molecular RealTime HIV-1 Assay is the only FDA approved Real Time PCR HIV viral load assay that has a similar quantitation range of 40 copies/ml to 10 million copies/ml for all M, N, O groups and subtypes.

The foundation for the RealTime HIV-1 assay was based on three primary elements:

- selection of primer and probe sequences from highly conserved regions within the HIV-1 genome
- utilization of a probe technology with unprecedented tolerance to nucleotide mismatches (partially double-stranded linear probe) coupled with low temperature annealing/read-out
- optimization of assay and cycling conditions to enhance amplification/detection of target sequences harboring mismatches

The Abbott HIV Global Surveillance Program was leveraged to aid in selection of the optimal location for the primer and probe sites. This program was initiated in 1995 to pro-actively address the challenge of HIV diversity and its impact on diagnostic, screening and monitoring assays. The primary goals of this comprehensive program are to:

- monitor global diversification of HIV and emergence of new strains of epidemiological significance
- establish a well-characterized large-volume panel of genetically diverse specimens (group/subtype/CRF)
- create databases of sequence information from diagnostically relevant regions of the genome
- evaluate comparative performance of serological and molecular assays
- generate assay development tools

### Assay Design:

The Abbott RealTime HIV-1 Assay comprises:

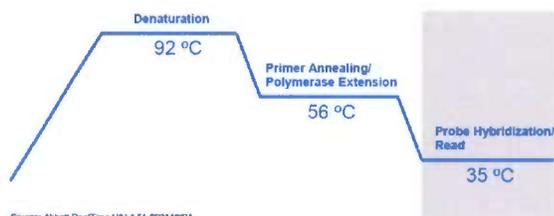
- **Target Region:** Primers and probes targets the pol/INT portion of the HIV-genome to minimize primer, probe mismatches
- **Probe Design:** HIV Probe uses a unique extended length, partially double-stranded probe to increase tolerability for mismatches
- **Cycling Conditions:** Low probe read temperature (35° C) is more thermodynamically favorable
- **Reagent Reuse-** with the recent launch of our new mPLUS, our system has the ability to extend the use of the *m2000* amplification packs for HIV-1 which will allow the users to customize workflow to sample arrival patterns. This new feature will help reduce waste, costs, and turnaround time. The updated software automatically tracks number of reuses available and total test remaining by amplification pack
- **Specimen Types:** Plasma (ACD-A and EDTA)
- **Specimen Input:** Multiple sample input volumes of 1.0 mL, 0.6 mL, 0.5 mL and 0.2 mL
- **Assay Performance:** Assay delivers exceptional precision at critical decision points.
  - Precision – Inter-assay standard deviation (SD) of  $\leq 0.25$  log copies/mL across entire range (40 copies/mL – 10 million copies/mL)
  - Sensitivity: 1.0 mL = 25 copies/ml; 0.6 mL = 40 copies/ml; 0.5 mL = 75 copies/ml; 0.2 mL = 150 copies/ml
  - Specificity: 100%

- HIV-1 Subtype Detection - Group M subtypes A-H, Group O and Group N in plasma (ACD-A and EDTA). Publication demonstrates detection of group P.
- **Controls:** Requires only 3 controls per batch of 21 to 93 patient specimens, allowing more specimens to be completed with each run
- **Calibrators:** Uses a 2-point external calibration to generate a stored calibration curve which lends to higher precision assay.

### RealTime HIV-1 Cycling Conditions

#### Low Temperature Read Cycles

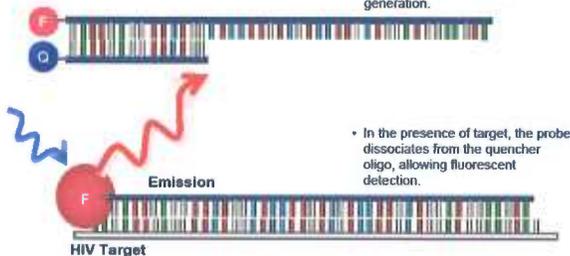
- The cycling conditions for the Abbott RealTime HIV-1 encompass a low temperature read cycle which allows the probe to tolerate mismatches more effectively.



### RealTime HIV-1 Probe Design

#### Partially Double-Stranded Probe

- In the absence of target, the probe hybridizes to the quencher oligo preventing fluorescent signal generation.



The limit of detection (LOD) is defined as the HIV-1 RNA concentration detected with a probability of 95% or greater.

The LOD claim for the Abbott RealTime HIV-1 assay is 40 copies/mL with the 0.6 mL sample volume procedure. The quantitative linear range: 40 copies/mL – 10 million copies/ml. Probit analysis of the data determined that the concentration of HIV-1 RNA detected with 95% probability was 39 copies/mL (95% CI 33-49).

The LOD claim for the Abbott RealTime HIV-1 assay is 75 copies/mL with the 0.5 mL sample volume procedure. The quantitative linear range: 75 copies/mL – 10 million copies/ml. Probit analysis of the data determined that the concentration of HIV-1 RNA detected with 95% probability was 65 copies/mL (95% CI 51-88).

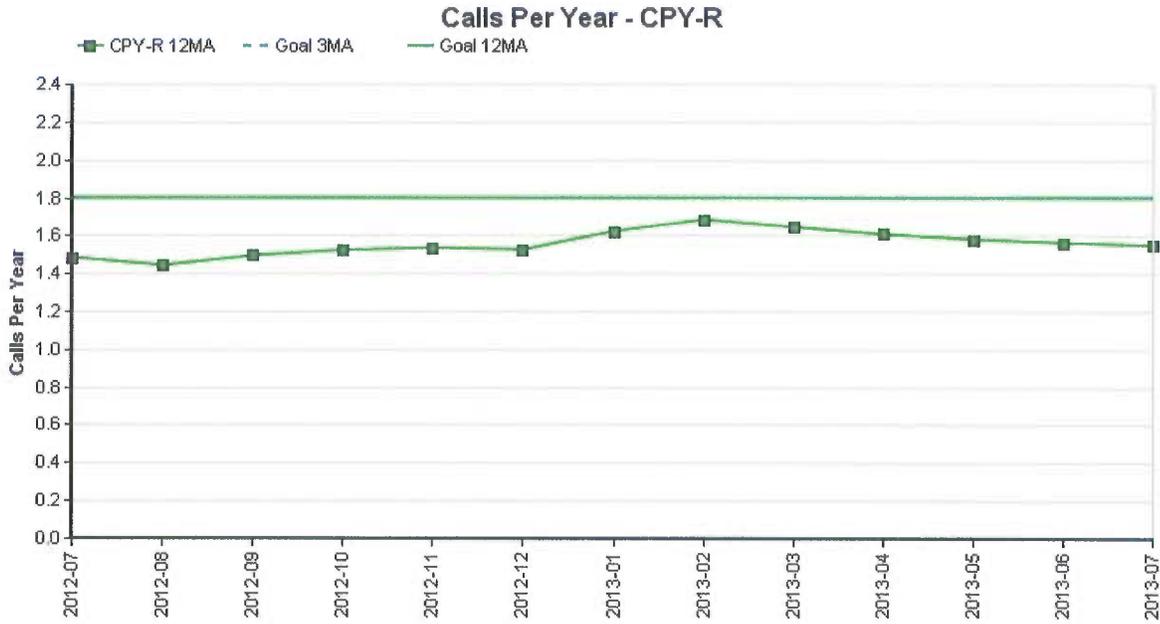
The LOD claim for the Abbott RealTime HIV-1 assay is 150 copies/mL with the 0.2 mL sample volume procedure. The quantitative linear range: 150 copies/mL – 10 million copies/ml. Probit analysis of the



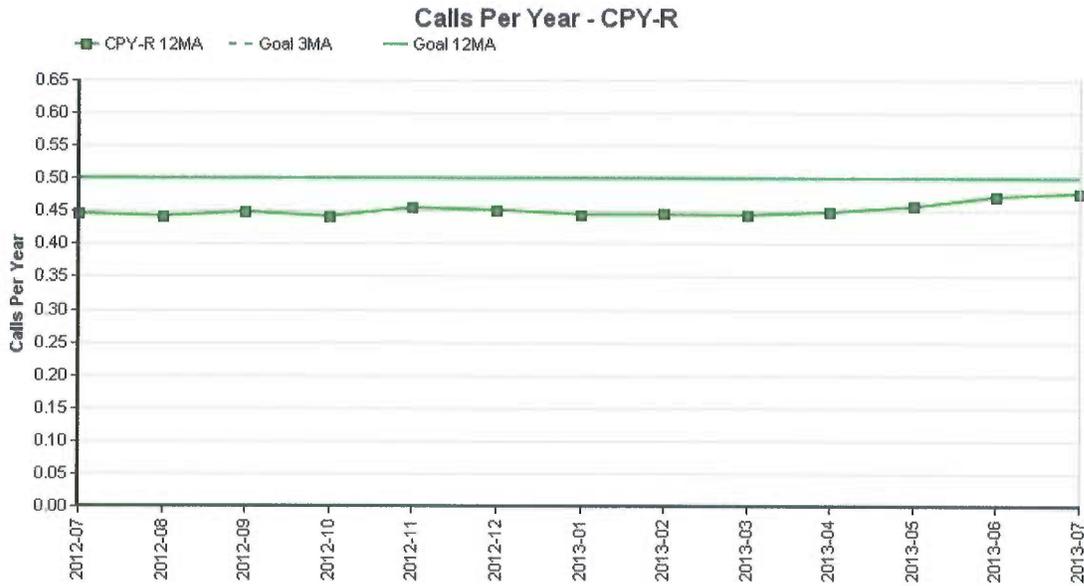
Reliability:

The charts below represent the Calls Per Year for the *m2000sp* and *m2000rt* systems for the last 12 months:

**M2000SP (605) - WorldWide**



**M2000RT (610) - WorldWide**



Abbott Molecular promotes the use of continuous remote monitoring via AbbottLink, to expedite resolution and maximize uptime, while maintaining the privacy of your patient population. To ensure we exceed our customer's expectations, multiple AbbottLink features are available in our One Care Service Package:

#### AbbottLink Remote Diagnostics

- Remote diagnostics allows technical support and field service to collaborate with your facility over a secure connection and access your system to understand and assess current performance.

#### AbbottLink Instant Virtual Presence

- Instant Virtual Presence (IVP) is a secure and efficient tool to share your screen with service and support, live! IVP can be utilized to assist with training, troubleshooting, or to simply receive clarification on instrument messages, alerts, or other information presented through the user interface.

#### Abbott Link Proactive Monitoring

- Proactive monitoring services provide real time notification of errors to trained service and support staff, while maintaining the privacy of your patient population. Immediate investigation and escalation of certain tracked error codes may prevent later more serious errors from occurring. This active notification service can ensure that your lab runs at peak efficiency.
- Please note that this system still requires follow up by an Engineer or Technical Support to assess the notification, troubleshoot, and schedule on-site service as needed.

AbbottLink uses secure, encrypted technology to transfer key logs, report generation, and remote services developments, including software revisions and upgrades. AbbottLink does not allow any visibility to patient identification information.

Please see [Appendix I-ABBOTTLINK BROCHURE](#)



John M. Pellegrino  
Director

*County of San Diego*  
**Department of Purchasing and Contracting**  
5560 Overland Avenue, Suite 270, San Diego, California 92123-1204

TELEPHONE (858) 505-6367  
FAX (858) 715-6452

September 3, 2013

**ADDENDUM No. 1**

**REAGENTS FOR HIV-1 VIRAL LOAD ASSAY WITH OPTION  
FOR CT/NG VITRO PCR ASSAY TESTING - RFB 6130**

Addendum No. 1. forms a part of the contract documents and changes the original documents only in the manner and to the extent stated.

**BID DUE DATE TIME HAS BEEN CHANGED:**

BID OPENING DATE AND TIME: September 11, 2013 at 11 AM

**QUESTIONS AND ANSWERS:**

1. Question: Given that each vendor's viral load assay requires number of components, the inclusion of which is required to produce an accurate and true total cost per test, how will you verify each vendor has included all of the costs necessary to produce a cost per test which is comparable between the vendors? We suggest each vendor be required to submit a list of the cost components used in the calculation of the total cost per test, so a comparable, accurate cost per test is verifiable by all participants in the RFB.

Answer: Yes, we would like each vendor to submit a list of cost components used in the calculation of the total cost per test.

2. Question: Per our discussion today, I wanted to ask for clarification on how you would like the HIV cost per test broken out on the HIV viral load pricing schedule. Per the pricing schedule, we're being asking for reagent costs for 600 HIV tests per month. The total number of tests required to run 600 tests per month is dependent on how often the test is run and how many tests are included in each batch. Without having this information, the cost per test could vary significantly depending on what assumptions are made.

Answer: The pricing should be based on two runs per week with 75 patient samples in each run (batch).

3. Question: Can you clarify whether you'd like us to calculate the test price entered on the pricing schedule to include all consumables?

Answer: Yes

4. If so, we'll need to know the number of batches per week that HIV testing will be performed. The other option would be to list the HIV viral load reagent only price on the pricing schedule and list all consumables on a separate exhibit.

Answer: Two batches per week with 75 patient samples run in each batch.

If you have any questions, please contact Ronald Higley, Procurement Specialist, at (858) 505-6359, or by email at [ronald.higley@sdcounty.ca.gov](mailto:ronald.higley@sdcounty.ca.gov).

*Ronald Higley for*  
John M. Pellegrino, Director  
Department of Purchasing and Contracting

JMP:rsh



TABLE OF CONTENTS

**SIGN AND RETURN ALL SECTIONS**

**SECTION A REQUEST FOR BID AND GENERAL INFORMATION**

1. REQUEST FOR BID .....A-1  
2. TABLE OF CONTENTS ..... A-2&3  
3. REPRESENTATIONS AND CERTIFICATIONS .....A-4  
4. STATEMENT OF WORK .....A-5  
5. PRICE SCHEDULE .....A-6  
6. PUBLIC AGENCY/RENEWAL .....A-7  
7. COUNTY CONTRACTOR PARTICIPATION (JULY 2008) .....A-7  
8. AUTOMATIC CONTRACT RENEWAL .....A-7  
9. CALIFORNIA REVENUE AND TAXATION CODE SECTION .....A-7  
10. FRANCHISE TAX BOARD WEBSITES .....A-7

**SECTION B INSTRUCTIONS FOR COMPLETING REQUEST FOR BIDS**

1. PRICING YOUR BID ..... B-1  
2. SUBMITTING YOUR BID ..... B-1  
3. EVALUATION AND AWARD ..... B-2  
4. PROTEST PROCEDURES ..... B-2  
5. LOCAL BUSINESS PREFERENCE ..... B-2

**SECTION C STANDARD TERMS AND CONDITIONS**

1. DEFINITIONS ..... C-1  
2. DISABLED VETERANS BUSINESS ENTERPRISE (DVBE) PARTICIPATION ENCOURAGED ..... C-1  
3. ASSIGNMENT OF RIGHTS, TITLE AND INTEREST ..... C-1  
4. CAL OSHA ..... C-1  
5. FORMAL BIDS ..... C-1  
6. DELIVERY ..... C-1  
7. INSPECTION ..... C-1  
8. TERMINATION FOR DEFAULT ..... C-1&2  
9. TERMINATION FOR CONVENIENCE ..... C-2  
10. TITLE ..... C-2  
11. VARIATIONS IN SPECIFICATIONS ..... C-2  
12. HAZARDOUS SUBSTANCES ..... C-2  
13. PROHIBITED CONTRACTS ..... C-2&3  
14. ESTIMATED QUANTITIES ..... C-3  
15. AVAILABILITY OF FUNDING ..... C-3  
16. INSPECTION OF SERVICE/MATERIALS/SUPPLIES ..... C-3  
17. DISPUTES ..... C-3&4  
18. CHANGES ..... C-4  
19. ASSIGNABILITY ..... C-4  
20. INDEMNITY ..... C-4  
21. CONDUCT OF CONTRACTOR ..... C-4  
22. DISALLOWANCE ..... C-4  
23. GOVERNING LAW ..... C-5  
24. AUDIT AND INSPECTION OF RECORDS ..... C-5  
25. PATENT AND COPYRIGHT INFRINGEMENT ..... C-5  
26. CONTRACTOR REPRESENTATION ..... C-5  
27. WARRANTY ..... C-5  
28. INSURANCE ..... C-5&6  
29. PERMITS, NOTICES, FEES AND LAWS ..... C-6  
30. AIR, WATER POLLUTION CONTROL, SAFETY AND HEALTH ..... C-6  
31. FINDINGS CONFIDENTIAL ..... C-6  
32. PUBLICATION, REPRODUCTION AND USE OF MATERIAL ..... C-6

33. NOTICE ..... C-6  
34. PRODUCT IDENTIFICATION AND LABELING ..... C-6  
35. DRUG & ALCOHOL FREE WORKPLACE ..... C-7  
36. ORDERING WITH BLANKET PURCHASE AGREEMENT ..... C-7  
37. INVOICES ..... C-7  
38. PAYMENTS AND INVOICES ..... C-7  
39. ACCEPTANCE OF COUNTY CREDIT CARD FOR PAYMENT ..... C-7&8  
40. FLAMMABILITY AND TOXICITY ..... C-8  
41. BRAND NAME OR EQUAL ..... C-8  
42. CONTRACT EXTENSION OPTION ..... C-8  
43. SEVERABILITY ..... C-8

County of San Diego  
Department of Purchasing and Contracting  
REPRESENTATIONS AND CERTIFICATIONS

The following representations and certifications are to be completed, signed and returned with the offer.

1. NOT-FOR-PROFIT ORGANIZATIONS

Attach proof of status and omit Paragraph 3.

2. INTERLOCKING DIRECTORATE

In accordance with Board of Supervisors Policy A-79, if Offeror is a non-profit as indicated in paragraph 1 above, Offeror is required to identify any related for-profit subcontractors in which an interlocking directorate, management or ownership relationship exists. By submission of this offer, Offeror certifies it will not enter into a subcontract relationship with a related for-profit entity if Offeror is a non-profit entity. If Offeror is a non-profit and will be subcontracting with a related for-profit entity, Offeror must list the entity(ies) on an attached separate sheet listing them all and the contract must be approved by the Board of Supervisors

3. BUSINESS REPRESENTATION

3.1. REPRESENTATION AS DISABLED VETERANS BUSINESS ENTERPRISE

"Disabled Veterans Business Enterprise" means a business which is at least fifty-one (51%) owned and operated by one or more veterans with a service related disability as certified by Equal Opportunity Management Office (EOMO), California Department of General Services, Office of Small Business and members of Joint Agencies Contracting Opportunities (JACO), (California Military and Veterans code, Article 6, section 999):

This Offeror represents as a part of this offer that the ownership, operation and control of the business are in accordance with the specific definition in 3.1. I am currently certified by:

Certifying Government Agency: Not Applicable

Certification #: Not Applicable

4. CERTIFICATE REGARDING DEBARMENT, SUSPENSION AND RELATED MATTERS

Offeror hereby certifies to the best of its knowledge that neither it nor any of its officers:

- 4.1. Are presently debarred, suspended, proposed for debarment, declared ineligible, or voluntarily excluded from covered transactions by any Federal department or agency; and
- 4.2. Have within a three (3) year period preceding this agreement been convicted of or had a civil judgment rendered against them for commission of fraud or criminal offense in connection with obtaining, attempting to obtain, or performing a public (Federal, State, or local) transaction or contract under a public transaction; violation of Federal or State antitrust statutes or commission of embezzlement, theft, forgery, bribery, falsification or destruction of records, making false statements, or receiving stolen property; and
- 4.3. Are presently indicted for or otherwise criminally or civilly charged by a government entity (Federal, State, or local) with the commission of any of the offenses enumerated in paragraph 4.2 of this certification; and

4.4. Have within a three (3) year period preceding this agreement had one or more public transactions (Federal, State or local) terminated for cause or default.

4.5. Are presently the target or subject of any investigation, accusation or charges by any Federal, State or local law enforcement, licensing or certification body and if they are, the appropriate information is included in the proposal, as requested in the Submittal Requirements.

4.6. Contractor will report in writing to the County Department of Purchasing and Contracting within five business days of knowing or have any reason to know any change in status as certified in the preceding paragraphs 4.1 through 4.5, and that occur prior to award (in the case of bids) and contract execution (in the case of negotiated procurements).

4.7. Offeror and its proposed subcontractors, agents and consultants have not previously contracted with the County to perform work on this project (e.g. preparing components of the statement of work or plans and specifications for this project). If Offeror or any of its subcontractors, agents or consultants, have previously contracted with the County to perform work on this project, Offeror shall identify those previous agreement(s) and submit that list along with the proposal.

5. CERTIFICATE OF CURRENT COST OR PRICING

This is to certify that, to the best of my knowledge and belief, cost and/or pricing data submitted with this offer, or specifically identified by reference if actual submission of the data is impracticable, is/are accurate, complete, and current as of the date signed below.

6. CERTIFICATE OF INDEPENDENT PRICING

By submission of this offer, each Offeror certifies, and in the case of a joint offers, each party thereto certifies as to its own organization, that in relation to this procurement:

6.1. The prices in this offer have been arrived at independently, without consultation, communication, or agreement, for the purpose of restricting competition, as to any matter relating to such prices with other Offeror; with any competitor; or with any County employee(s) or consultant(s) involved in this or related procurements; and

6.2. Unless otherwise required by law, the prices which have been quoted in this offer have not been knowingly disclosed by the Offeror and will not knowingly be disclosed by the Offeror prior to opening, in the case of a bid, or prior to award, in the case of a proposal, directly or indirectly to any other Offeror or to any competitor; and

6.3. No attempt has been made or will be made by the Offeror to induce any other person or firm to submit or not to submit an offer for the purpose of restricting competition.

7. The Offeror understands that prior to receiving a contract award from the County, the Offeror must submit a completed IRS W-9 form to provide a Federal Tax ID number, or if not available, to provide a Social Security Number (SSN).

CERTIFICATION

The information furnished in Paragraphs 1 through 7 is certified to be factual and correct as of the date submitted and this certification is made under penalty of perjury under the laws of the State of California.

Name: Laurea Prantz Signature: [Signature]

Title: Sr. Business Project Mng. Date: 9/10/13

Company/Organization: Abbott Laboratories Inc.

SUBMIT THIS FORM AS DIRECTED IN THE REQUEST FOR SOLICITATION DOCUMENTS OR WITH THE OFFER

**RFB 6130 - COUNTY OF SAN DIEGO  
HEALTH AND HUMAN SERVICES AGENCY  
PUBLIC HEALTH LABORATORY  
HIV-1 VIRAL LOAD TESTING SYSTEM  
SCOPE OF WORK/PURPOSE**

THE VENDOR SHALL PROVIDE AN INCLUSIVE SYSTEM FOR THE FULLY AUTOMATED TESTING FOR THE HIV-1 VIRAL LOAD IN PLASMA, INCLUDING THE REAGENTS, CONSUMABLES, EQUIPMENT, ANNUAL PREVENTATIVE MAINTENANCE AND REPAIRS (I.E., LABOR AND PARTS). THE PRICE PER TEST FOR AN ESTIMATED 600 TESTS PER MONTH IS TO BE INCLUDED IN THE PRICE OF THE TEST REAGENTS.

**REQUIRED SPECIFICATIONS FOR THE HIV-1 VIRAL LOAD ASSAY SYSTEM**

THE HIV-1 VIRAL LOAD ASSAY SYSTEM MUST: BE A FULLY-AUTOMATED REAL-TIME RT-PCR (IN-VITRO REVERSE TRANSCRIPTION-POLYMERASE CHAIN REACTION (RT-PCR) ASSAY) SYSTEM FOR DETECTION AND QUANTIFICATION OF HUMAN IMMUNODEFICIENCY VIRUS TYPE 1 (HIV-1) WITH A QUANTIFICATION RANGE IN HUMAN PLASMA BETWEEN 40 TO 10,000,000 COPIES/ML. FULL AUTOMATION MEANS THAT THE PLASMA WILL BE SAMPLED DIRECTLY FROM A VACUTAINER-TYPE SAMPLE TUBE WITHOUT TRANSFER TO A SECOND TUBE OR ASSAY CUVETTE. THE SYSTEM MUST BE CAPABLE OF READING SD PHL LIMS BAR-CODE LABELS (CODE 128) DIRECTLY OFF THE SAMPLE TUBE. ONCE THE SAMPLE TUBES ARE LOADED INTO THE SYSTEM, ALL PIPETTING, SAMPLE EXTRACTION, NUCLEIC ACID PURIFICATION AND ASSAY REAGENT AND TEMPLATE HANDLING IS PERFORMED BY THE SYSTEM WITHOUT OPERATOR INTERVENTION.

**BACKGROUND INFORMATION**

THE SAN DIEGO COUNTY PUBLIC HEALTH LABORATORY (PHL), LOCATED AT 3851 ROSECRANS STREET, SUITE 716, SAN DIEGO, CA 92110, IS A COMPREHENSIVE PUBLIC HEALTH REFERENCE LABORATORY FOR THE COUNTY OF SAN DIEGO. THE PHL REQUIRES AN AUTOMATED SYSTEM FOR THE QUANTIFICATION OF HUMAN IMMUNODEFICIENCY VIRUS TYPE 1 (HIV-1) IN HUMAN PLASMA FROM HIV-1 INFECTED INDIVIDUALS OVER THE RANGE OF 40 TO 10,000,000 COPIES/ML. THE APPROXIMATE VOLUME FOR VIRAL LOAD TESTING WOULD BE 600 PATIENT SPECIMENS PER MONTH. REAGENTS WILL BE ORDERED BY THE LABORATORY ON AN AS NEEDED BASIS.

**OBJECTIVES**

- VENDOR SHALL PROVIDE A FULLY-AUTOMATED, FDA-APPROVED, REAL-TIME PCR BASED SYSTEM FOR HIV-1 VIRAL LOAD TESTING AND ALL INSTRUMENTS AND REQUIRED EQUIPMENT UNDER A LEASE/RENTAL AGREEMENT TO INCLUDE ROUTINE SERVICE AND MAINTENANCE FOR THE CONTRACT PERIOD. THE COST OF THIS WILL BE INCLUDED AND QUOTED AS THE COST PER TEST.
- VENDOR SHALL PROVIDE QUOTATION FOR FDA APPROVED REAGENTS INCLUDING CONSUMABLES (TIPS, TRAYS, REAGENT/ASSAY PLATES AND TUBES. ETC.) FOR THE EXPECTED TESTING VOLUME OF 600 SAMPLES PER MONTH.

**SPECIFIC REQUIREMENTS OF THE SYSTEM**

1. EQUIPMENT SHALL BE LOANED OR RENTED TO THE COUNTY OF SAN DIEGO. THE FULL COST OF THE EQUIPMENT, SCHEDULED MAINTENANCE AND REPAIRS (I.E., LABOR AND PARTS) ARE TO BE INCLUDED IN THE PRICE OF THE REAGENTS (SOMETIMES DESCRIBED AS A REAGENT RENTAL AGREEMENT).
2. THE CONTRACT BASE PERIOD WILL BE FOR TWO (2) YEARS WITH OPTIONS TO RENEW FOR YEARS 3 THROUGH 5.
3. ANY AND ALL PARTS, ADDITIONAL ITEMS, OR ACCESSORIES NOT USED ROUTINELY AS PART OF THE ASSAY, OR PROVIDED UNDER THE SERVICE CONTRACT (E.G. SAMPLE TUBE RACKS OR OTHER DURABLE PARTS), FOR THE SYSTEM SHOULD BE PROVIDED AS AN ITEMIZED LIST BY THE VENDOR AS A PERCENT OF DISCOUNT FROM LIST PRICE, SUBMIT LIST ALONG WITH PRICE SHEETS, PAGE A-6 & 6A.
4. THE SYSTEM MUST BE CAPABLE OF FULLY AUTOMATED READING OF BAR-CODE LABELS LABORATORY AND PATIENT BLOOD TUBES (SYMBOLGY CODE 128).
5. FULLY AUTOMATED PRECISION PIPETTING TO ELIMINATE MANUAL MIXING OR MANIPULATION OF SAMPLES OR REAGENTS.
6. SYSTEM MUST BE CAPABLE OF DIRECT SAMPLING OF 5 ML VACUTAINER-TYPE BLOOD COLLECTION TUBES.
7. FLEXIBLE THROUGHPUT OPTIONS OF 24 TO 96 SAMPLES IN ONE 8-HOUR SHIFT.
8. HAVE FDA APPROVAL: THE SYSTEM MUST HAVE FDA APPROVAL FOR HIV-1 VIRAL LOAD TESTING.
9. REGULARLY SCHEDULED PREVENTATIVE MAINTENANCE WILL BE PROVIDED ONCE ANNUALLY DURING REGULAR BUSINESS HOURS OF MONDAY THROUGH FRIDAY, 8:00 A.M. TO 5:00 P.M.
10. REPAIR SERVICE WILL BE AVAILABLE MONDAY THROUGH FRIDAY, 8:00 A.M. TO 5:00 P.M.

RFB 6130  
COUNTY OF SAN DIEGO  
HEALTH AND HUMAN SERVICES AGENCY  
PUBLIC HEALTH LABORATORY  
HIV-1 VIRAL LOAD TESTING SYSTEM  
SECTION A  
PRICING SCHEDULE

**PAGE 6 IS AN EXCEL SPREAD SHEET AND ALL VENDORS MUST COMPLETE. ALL THE VENDOR NEEDS TO DO IS FILL IN THE PRICE PER UNIT FOR THE BASE TERM, FIRST, SECOND AND THIRD OPTION PERIODS AND THE EXCEL SPREADSHEET WILL CALCULATE THE TOTALS. ONCE YOU HAVE THE TOTAL YOU CAN PRINT AND SEND WITH THE OTHER DOCUMENTATION. ALL BIDS MUST BE RECEIVED BY 11 AM ON WEDNESDAY, SEPTEMBER 4, 2013. ALL DOCUMENTS MUST BE SUBMITTED WITH RFB 6130 FOR THE BID TO BE CONSIDERED.**

**AWARD SHALL CONSIST OF MONTHLY RENTAL OF EQUIPMENT, REAGENTS, YEARLY MAINTENANCE AND REPAIR SERVICE AS NEEDED BASED ON A MONTHLY USAGE OF PATIENT SPECIMANS.**

RF8 6130  
HIV-1 VIRAL LOAD  
PRICING SCHEDULE

Item #	Item Description	UOM	Base Term Period: Date of Award thru 31-Aug-2015			First Option Period: 01-Sep-2015 thru 31-Aug-2016			Second Option Period: 01-Sep-2016 thru 31-Aug-2017			Third Option Period: 01-Sep-2017 thru 31-Aug-2018			BASIS OF AWARD	NOTES	
			Est. Monthly Qty	Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier			Est. Extended Price
1	HIV-1 Viral Load Test	EA	600		24	\$0.00		12	\$0.00		12	\$0.00		12	\$0.00	\$0.00	
BASIS OF AWARD TOTAL OF ALL PERIODS - BASE, FIRST OPTION, SECOND OPTION AND THIRD OPTION PERIOD															\$0.00		

SAMPLE

REPLACE THIS SAMPLE WORKSHEET WITH YOUR COMPLETED WORKSHEET

**PUBLIC AGENCY PARTICIPATION (July 2008)**

It is intended that any other public agency (i.e., city, district, public authority, public agency, municipality and other political sub-division or public corporation of California) located in San Diego County shall have the option to participate in any award made as a result of this solicitation. Any agency located outside of San Diego County shall have the option to participate, but shall incur all freight charges from location of awarded vendor to delivery point. The County of San Diego shall incur no financial responsibility in connection with orders issued under the authority of this provision or in making payments to the vendor.

**COUNTY CONTRACTOR PARTICIPATION (July 2008)**

It is intended that any educational institution or non profit organization that is currently under contract with the County of San Diego to provide direct support to the County with reimbursement for such support coming directly from the County shall have the option to participate in any award made as a result of this solicitation. The contractor agrees to provide the items called for in the schedule of this contract to educational institutions or non profit organization under the authority of this provision. The contractor is responsible for confirming that any educational institution or non profit organization has a current contract with the County of San Diego. The County shall incur no financial responsibility in connection with orders issued under the authority of this provision. The ordering organization shall be solely responsible for verifying they are currently under contract with the County, placing orders, and making payments to the contractor.

**AUTOMATIC CONTRACT RENEWAL (July 2008)**

Unless County notifies Contractor in writing, not less than 30 days prior to the expiration date that they do not intend to renew the Agreement, the Agreement will be automatically renewed for another year. Term not to exceed August 31, 2018.

**WINNING AWARD WILL BE REQUIRED TO COMPLY WITH THE FOLLOWING:**

**CALIFORNIA REVENUE AND TAXATION CODE SECTION 18662.**

In compliance with California Revenue and Taxation code section 18662, if you are a non resident of California (out-of-state invoices) who receives California source income, the County will pay California Use Tax directly to the State of California per permit no. SR FH 25-632384. Fifteen (15) business days prior to the first payment, new suppliers or suppliers with expired forms or forms with incorrect information, must submit new forms to the County (forms are available from the Franchise Tax Board website listed below).

Under certain circumstances you may be eligible for reduced or waived nonresident withholding. If you have already received a waiver or a reduced withholding response from the State of California and the response is still valid, submit the response to the County in lieu of the forms. Failure to submit the required forms will result in withholding of payments. Refer to the Franchise Tax Board websites (listed below) for tax forms and information on nonresident withholding, including waivers or reductions. The County will not give you any tax advice. It is recommended you speak with your tax adviser and/or the State of California for guidance.

**FRANCHISE TAX BOARD WEBSITES:**

<http://www.ftb.ca.gov>

[http://www.ftb.ca.gov/individuals/Withholding\\_Definitions.shtml](http://www.ftb.ca.gov/individuals/Withholding_Definitions.shtml)

[http://www.ftb.ca.gov/individuals/wsc/Processing\\_Changes\\_for\\_2010.shtml](http://www.ftb.ca.gov/individuals/wsc/Processing_Changes_for_2010.shtml)

[http://www.ftb.ca.gov/individuals/wsc/forms\\_and\\_publications.shtml](http://www.ftb.ca.gov/individuals/wsc/forms_and_publications.shtml)

[http://www.ftb.ca.gov/individuals/wsc/decision\\_chart.shtml](http://www.ftb.ca.gov/individuals/wsc/decision_chart.shtml)

**RFB 6003  
SECTION B  
COUNTY OF SAN DIEGO'S  
INSTRUCTIONS FOR COMPLETING REQUEST FOR BID  
AND PRE-AWARD REQUIREMENTS**

Rev 01/04

**1. PRICING YOUR BID**

- 1.1 Bid on each item separately. Prices should be stated per unit(s) specified herein.
- 1.2 Unless otherwise specified, all prices shall be F.O.B. destination. Bids other than F.O.B. destination shall be considered non-responsive and will be rejected. Prices shall include all freight charges.
- 1.3 Unless otherwise specified, prices bid herein should **NOT** include California sales/use tax or Federal excise tax. The County generally is required to pay California sales/use tax, and it should be shown as a **separate item** on invoices. The County is exempt from payment of Federal excise tax. It must **NOT** be included in invoices.
- 1.4 All prices and notations must be in ink or typewritten. No erasures permitted. Mistakes may be crossed out and corrected and must be initialed in ink by person(s) signing the bid.
- 1.5 Discounts of less than thirty- (30) days will not be considered in evaluation of bids to determine overall apparent low bidder.
- 1.6 Net terms of less than 30 days will not be accepted.

**2. SUBMITTING YOUR BID**

- 2.1 Each bid must be in a separate sealed envelope **WITH BID NUMBER ON THE OUTSIDE** and must be delivered to the County Purchasing and Contracting Department, Front Desk (where it will be time stamped to indicate time of receipt), 5560 Overland Avenue, Suite 270, San Diego, California 92123, by 11:00 a.m. on the day specified. Bids will be publicly opened at that time.
- 2.2 Failure to bid on authorized County form may be cause for rejection of bid.
- 2.3 Any bid received at the County Purchasing and Contracting Department after the exact time for receipt will not be considered and will be rejected as a late bid.
- 2.4 Late bids will be returned to the bidder unopened unless it is determined that the late receipt was due solely to mishandling by the Purchasing and Contracting Department and such determination is made prior to award.
- 2.5 The County's primary means of providing bids and addenda is the County BuyNet Internet website:
- 2.6 No oral interpretation shall be made to modify any provisions of any bid specifications. Requests for an interpretation shall be made in writing to the County Director of Purchasing and Contracting prior to bid opening and a written response will be posted on the County BuyNet website.
- 2.7 Any vendor desiring to withdraw its bid must do so before County bid opening. If there are any questions or comments relative to technicalities of the bid, they must be submitted in writing to County of San Diego, Director of Purchasing and Contracting, within 24 hours after bid opening.
- 2.8 Bids submitted in response to this Request for Bid must be in full conformance with the terms and conditions set forth herein. Further, all specification requirements must be met unless the language of the Request for Bid specifically indicates alternate specifications will be considered.
- 2.9 Samples of items, when required, must be furnished free of expense to the County, and if not destroyed by tests will, upon request, be returned at the bidder's expense.
- 2.10 All bids must be signed with the firm name and by an authorized officer or employee. Obligations assumed by such signature must be fulfilled.

3. **EVALUATION AND AWARD**

- 3.1 Bids are subject to acceptance at any time within 30 days after opening of same, unless otherwise stipulated by the County.
- 3.2 In determining the lowest bid, discounts of 30 days or greater will be considered. Discounts will be calculated from receipt and acceptance of merchandise or invoice, whichever is later.
- 3.3 Award will be made by the Department of Purchasing and Contracting as stated on the cover/pricing page to the lowest responsive, responsible bidder.
- 3.4 The County reserves the right to waive a variation in specification if, in the opinion of the County, such variation does not materially change the item or its performance within parameters acceptable to the County.
- 3.5 The County reserves the right to reject any or all bids and to accept or reject any item(s) thereon, or waive any informality in the bid.
- 3.6 In the event of a conflict between unit price bid and bidder's extended price, the unit price will prevail unless price is so obviously unreasonable as to indicate an error. In that event, the bid will be rejected as non-responsive for the reason of inability to determine the intended bid.
- 3.7 The County reserves the right to perform a pre-award survey of the bidder to determine capability to perform, including but not limited to facilities, financial responsibility, materials/supplies, and past performance. The determination of the County as to the bidder's prospective ability to perform the contract shall be conclusive.

4. **PROTEST PROCEDURES**

Any protest resulting from this procurement is to be processed as prescribed in Board of Supervisors' Policy A-97, Protest Procedures for Award of Contracts. All protests shall be in writing, be made prior to Award, and be made only by an offeror. Such protests shall clearly state the ground for the protest and the relief sought. Protests shall be filed with the County's contracting office identified in the solicitation package.

Whenever a contract is contemplated to be awarded to other than the low bidder in a formally advertised procurement, the low bidder shall be so notified five working days prior to award, in addition to the posting of the proposed award in a public place in the Contracting Office for the same period of time. Copies of Policy A-97 are available upon request from the Clerk of the Board, 1600 Pacific Highway, San Diego, CA 92101 or via the County of San Diego's Internet website: <http://www.co.san-diego.ca.us/cob/policy/index.html>

5. **LOCAL BUSINESS PREFERENCE**

Responsive bids from responsible local San Diego County businesses shall be given preference for award over bids received from non-local businesses. "Local Business" is defined as a business with a valid license issued by a city within the County, employing San Diego residents, and with a verifiable address within the County, or a business employing San Diego residents and with a verifiable address in an unincorporated area of the county. Post Office Boxes do not qualify as verifiable local business addresses. If a tie bid occurs between a local business and a non-local business, award shall be made to the local business.

If the lowest responsible, responsive bid is submitted by a non-local business, one percent (1%) shall be subtracted from the lowest responsive, responsible bid submitted by a local business in evaluating the bids for award. If application of the one percent (1%) factor results in the local business bid being equal to or lower than the non-local business bid, contract award will be made to the local business at the local business bid price, except for public works and construction bids, or if prohibited by State or Federal law.

**RFB 6003 - SECTION C  
TERMS & CONDITIONS OF REQUEST FOR BIDS  
AND RESULTANT CONTRACT OR PURCHASE ORDER**

**1. DEFINITIONS**

"County" shall mean The County of San Diego, California

"Offeror" shall mean any person, firm, partnership, or corporation submitting a proposal to County in response to this solicitation.

"Contractor" shall mean the offeror whose proposal is accepted by County and who has entered into an agreement with County to provide the equipment and services described herein.

"Vendor" shall mean the same as contractor.

**2. DISABLED VETERANS BUSINESS ENTERPRISE PARTICIPATION ENCOURAGED (Rev. 11/97)**

County Board of Supervisor's policies B-53 and B-39 A encourages the participation of small and Disabled Veterans Business Enterprises (DVBE) in County procurement. Section A of this solicitation (Representations & Certifications) contains a description of the County's requirements to qualify as an (DVBE). Perspective (DVBE) bidders/offerors are encouraged to contact the Contracting Office representative listed on the face of this Request for Bid (RFB) or Request for Proposal (RFP) for information concerning the County's procurement procedures.

**3. ASSIGNMENT OF RIGHTS, TITLE AND INTEREST**

In submitting a bid to a public purchasing body, the vendor offers and agrees that if the bid is accepted, it will assign to the purchasing body all rights, title and interest in and to all causes of action it may have under Section 4 of the Clayton Act (15 U.S.C. Sec. 15) or under the Cartwright Act (Chapter 1 (commencing with Section 16700) of Part 2 of Division 7 of the Business and Professions Code), arising from purchases of goods, materials, or services by the bidder for sale to the purchasing body pursuant to the bid. Such assignment shall be made and become effective at the time the purchasing body tenders final payment to the vendor.

**4. CAL OSHA**

As applicable, all items furnished under this bid shall meet or exceed the standards established by the California Occupational Safety and Health Act of 1973 and current amendments thereto, provided the end use of the item is for the purpose for which the item is intended.

**5. FORMAL BIDS**

In the event this bid results in a purchase order, terms and conditions of this bid are incorporated herein and from a part of the purchase order. In the event of any conflict or inconsistency between the terms of the formal bid or award, the terms of this formal bid shall control.

**6. DELIVERY**

Time is of the essence, and the purchase order is subject to termination for failure to deliver on time. The acceptance by buyer of late performance with or without objection or reservation shall not waive the right to claim damage for such breach nor constitute a waiver of the requirements for the timely performance of any obligation remaining to be performed by the vendor.

**7. INSPECTION**

All items or services are subject to final inspection and acceptance at designation by the County. Such final inspection shall be made within a reasonable time after delivery.

**8. TERMINATION FOR DEFAULT**

The County may, by written notice of default to the vendor, terminate any resulting order in whole or in part should the vendor fail to make satisfactory progress, fail to deliver within time specified therein or fail to deliver in strict conformance to

specifications and requirements set forth therein. In the event of such termination, the County reserves the right to purchase or obtain the supplies or services elsewhere, and the defaulting vendor shall be liable for the difference between the prices set forth in the terminated order and the actual cost thereof to the County. The prevailing market price shall be considered the fair repurchase price.

- 8.1. If, after notice of termination of this contract under the provisions of this clause, it is determined for any reason that the Contractor was not in default under this provisions of this clause, the rights and obligations of the parties shall, if the contract contains a clause providing for termination for convenience of the County, be the same as if the notice of termination had been issued pursuant to such clause.
- 8.2. The rights and remedies of County provided in this article shall not be exclusive and are in addition to any other rights and remedies provided by law or under resulting order.

9. **TERMINATION FOR CONVENIENCE**

The County may, by written notice stating the extent and effective date, terminate any resulting order for convenience in whole or in part, at any time. The County shall pay the vendor as full compensation for performance until such termination:

- 9.1. The unit or pro rata price for the delivered and accepted portion.
- 9.2. A reasonable amount, as costs of termination, not otherwise recoverable from other sources by the vendor as approved by the County, with respect to the undelivered or unaccepted portion of the order, provided compensation hereunder shall in no event exceed the total price.
- 9.3. In no event shall the County be liable for any loss of profits on the resulting order or portion thereof so terminated.
- 9.4. The rights and remedies of County provided in this article shall not be exclusive and are in addition to any other rights and remedies provided by law or under resulting order.

10. **TITLE**

Title to the material and supplies purchased shall pass directly from vendor to County at the F.O.B. point shown, subject to the right of County to reject upon inspection.

11. **VARIATIONS IN SPECIFICATIONS**

The County reserves the right to waive a variation in specification if, in the opinion of the County, such variation does not materially change the item or its performance within parameters acceptable to the County.

12. **HAZARDOUS SUBSTANCES** (July 2008)

If any product being delivered or supplied to the County under this contract/purchase order is listed in the Hazardous Substances List of the Regulations of the Director of Industrial Relations with the California Occupational Safety and Health Standards Board, or if the product presents a physical or health hazard as defined in the California Code of Regulations, General Industry Safety Order, Section 5194 (T8CCR), Hazard Communication, then the contractor must include a Material Safety Data Sheet (MSDS) with delivery, or shipment. Each MSDS must reference the contract/purchase order number, and identify the "Ship To Address". All shipments and containers must comply with the labeling requirements of Title 49, Code of Federal Regulations by identifying the hazardous substance, name and address of manufacturer, and appropriate hazard warning regarding potential physical safety and health hazard. (County of San Diego Administrative Manual, 0300-02, Hazard Communication Program).

No product which is manufactured with fully halogenated chlorofluorocarbons (CFC) shall be delivered or supplied, or used on a job site in performance of this contract/purchase order unless specifically described in the stated requirements of this contract/purchase order or otherwise explicitly authorized by the County Director, Purchasing and Contracting.

13. **PROHIBITED CONTRACTS**

Section 67 of the San Diego County Administrative Code provides that the County shall not contract with, and shall reject any bid or proposal submitted by the person or entities specified below, unless the Board of Supervisors finds that special circumstances exist which justify the approval of such contract:

- 13.1. Persons employed by the County or of public agencies for which the Board of Supervisors is the governing body;
- 13.2. Profit-making firms or businesses in which employees described in sub-section (a) of code serve as officers, principals, partners, or major shareholders;
- 13.3. Persons who, within the immediately preceding twelve (12) months came within the provisions of the above sub-section and who (1) were employed in positions of substantial responsibility in the area of service to be performed by the contract, or (2) participated in any way in developing the contract or its service specifications; and
- 13.4. Profit-making firms or businesses in which the former employees described in sub-section 16.3 of code serve as officers, principals, partners, or major shareholders.

With the affixing of a signature to your response to this solicitation, offeror certifies that the above provisions of the Code have been complied with, and that any exception will cause any ensuing contract to be invalid.

**14. ESTIMATED QUANTITIES (March 1993)**

The Estimated Quantities in Section "A", Pricing Schedule, are provided solely for evaluation of bids. They represent approximate anticipated use based on historical consumption. If the County's actual requirements do not result in orders in the quantities described as "estimated" in the Schedule, that fact shall not constitute the basis for price adjustment.

**15. AVAILABILITY OF FUNDING**

The County's obligation for payment of any contract beyond the current fiscal year end is contingent upon the availability of funding from which payment can be made. No legal liability on the part of the County shall arise for payment beyond June 30 of the calendar year unless funds are made available for such performance.

**16. INSPECTION OF SERVICE/MATERIALS/SUPPLIES**

- 16.1. All performance (which includes services, materials, supplies and equipment furnished or utilized in the performance of this contract, and workmanship in the performance of services) shall be subject to inspection and test by the County at all times during the term of the contract. The Contractor shall provide adequate cooperation to any inspector assigned by the County to permit the inspector to determine the Contractor's conformity with these specifications and the adequacy of the services being contractually provided. All inspection by the County shall be made in such a manner as not to unduly interfere with Contractor performance.
- 16.2. If any services performed hereunder are not in conformity with the specifications and requirements of this contract, the County shall have the right to require the Contractor to perform the services in conformity with said specifications and requirements at no additional increase in total contract amount. When the services to be performed are of such nature that the difference cannot be corrected, the County shall have the right to (1) require the Contractor immediately to take all necessary steps to ensure future performance of the services in conformity with requirements of the contract, and (2) reduce the contract price to reflect the reduced value of the services performed. In the event the Contractor fails to perform the services promptly or to take necessary steps to ensure future performance of the service in conformity with the specifications and requirements of the contract, the County shall have the right to either (1) by contract or to otherwise have the services performed in conformity with the contract specifications and charge to the Contractor any cost occasioned to the County that is directly related to the performance of such services, or (2) terminate this contract for default as provided in the Termination clause.

**17. DISPUTES**

- 17.1. Except as otherwise provided in this contract, any dispute concerning a question of fact arising under this contract which is not disposed of by agreement shall be decided by the Contracting Officer who shall furnish the decision to the Contractor in writing. The decision of the Contracting Officer shall be final and conclusive unless determined by the court of competent jurisdiction to have been fraudulent or capricious, or arbitrary, or so grossly erroneous as necessarily to imply bad faith. The Contractor shall proceed diligently with the performance of the contract pending the Contracting Officer's decision.
- 17.2. The "Disputes" clause does not preclude consideration of legal questions in connection with decisions provided

for in paragraph (A) above. Nothing in this contract shall be construed as making final the decision of any administrative official, representative, or board on a question of law.

#### **18. CHANGES**

The Contracting Officer may at any time, by written order, make changes within the general scope of this contract, in the definition of services to be performed, and the time (i.e., hours of the day, days of the week, etc.) and place of performance thereof. If any such change causes an increase or decrease in the cost of, or the time required for the performance of any part of the work under this contract, whether changed or not changed by any such order, an equitable adjustment shall be made in the contract price or delivery schedule, or both, and the contract shall be modified in writing accordingly. Any claim by the Contractor for adjustment under this clause must be asserted within 30 days from the date of receipt by the Contractor of the notification of change; provided however, that the Contracting Officer, if he decides that the facts justify such action, may receive and act upon any such claim asserted at any time prior to final payment under this contract. Where the cost of property made obsolete or excess as a result of a change is included in the Contractor's claim for adjustment, the Contracting Officer shall have the right to prescribe the manner of disposition of such property. Failure to agree to any adjustment shall be a dispute concerning a question of fact within the meaning of the clause of this contract entitled "Disputes". However, nothing in this clause shall excuse the Contractor from proceeding with the contract as changed.

#### **19. ASSIGNABILITY**

The Contractor shall not assign any interest in this contract, and shall not transfer any interest in the same (whether by assignment or novation), without the prior written consent of the County thereto; provided however, that claims for money due or to become due to the Contractor from the County under this contract may be assigned without such approval. Notice of any such assignment or transfer shall be furnished promptly to the County.

#### **20. INDEMNITY**

County shall not be liable for, and Contractor shall defend and indemnify County and the employees and agents of County collectively, "County Parties") against any and all claims, demands, liability, judgments, awards, fines, mechanics' liens or other liens, labor disputes, losses, damages, expenses, changes or costs of any kind or character, including attorneys' fees and court costs (hereinafter collectively referred to as "Claims"), related to or arising out of this contract, and arising either directly or indirectly from any act, error, omission or negligence of Contractor or its subcontractors, licensees, agents, servants or employees, including Claims caused by the concurrent negligent act, error or omission, whether active or passive, of County Parties. However, Contractor shall have no obligation to defend or indemnify County Parties from a Claim if it is determined by a court of competent jurisdiction that such Claim was caused by the sole negligence or willful misconduct of County Parties.

#### **21. CONDUCT OF CONTRACTOR**

- 21.1. The Contractor agrees to inform the County of all the Contractor's interests, if any, which are or which the Contractor believes to be incompatible with any interests of the County.
- 21.2. The Contractor shall not, under circumstances which might reasonably be interpreted as an attempt to influence the recipient in the conduct of his duties, accept any gratuity or special favor from individuals or organizations with whom the Contractor is doing business or proposing to do business, in accomplishing the work under the contract.
- 21.3. The Contractor shall not use for personal gain or make other improper use of privileged information which is acquired in connection with his employment. In this connection, the term "privileged information" includes, but is not limited to, unpublished information relating to technological and scientific development; medical, personnel, or security records of the individuals; anticipated materials requirements or pricing actions; and knowledge of selections of contractors or subcontractors in advance of official announcement.
- 21.4. The Contractor or employees thereof shall not offer gifts, gratuity, favors, entertainment directly or indirectly to County employees.

#### **22. DISALLOWANCE**

In the event the Contractor receives payment for services under this contract which is later disallowed by the County, the Contractor shall promptly refund the disallowed amount to the County on request, or at its option, the County may offset the amount disallowed from any payment due to the Contractor under any contract with the County.

**23. GOVERNING LAW**

This contract shall be construed and interpreted according to the laws of the State of California.

**24. AUDIT AND INSPECTION OF RECORDS**

- 24.1. General. The County shall have the audit and inspection rights described in this section.
- 24.2. Cost or pricing data. If the Contractor submitted cost or pricing data in connection with the pricing of this contract or any change or modification thereto, unless such pricing was based on adequate price competition, established catalog or market prices of commercial items sold in substantial quantities of the general public, or prices set by law or regulation, the Contracting Officer or his representatives who are employees of the County or its agent shall have the right to examine all books, records, documents and other data of the Contractor related to the negotiation pricing or performance of such contract, change or modification, for the purpose of evaluating the accuracy, completeness and currency of the cost or pricing data submitted.
- 24.3. Availability. The materials described above shall be made available at the office of the Contractor, at all reasonable times, for inspection, audit or reproduction, until the expiration of 3 years from the date of final payment under this contract, or by (1) and (2) below:
  - 24.3.1. If this contract is completely or partially terminated, the records relating to the work terminated shall be made available for a period of three years from the date of any resulting final settlement.
  - 24.3.2. Records which relate to appeals under the "Disputes" clause of this contract, or litigation or the settlement of claims arising out of the performance of this contract, shall be made available until such appeals, litigation, or claims have been disposed of, or three years after contract completion, whichever is longer.
- 24.4. The Contractor shall insert a clause containing all the provisions of this entire clause in all subcontracts hereunder except altered as necessary for proper identification of the contracting parties and the contracting officer under the County's prime contract.

**25. PATENT AND COPYRIGHT INFRINGEMENT**

The contractor shall report to the contracting officer, promptly and in reasonable written detail, each notice or claim of patent or copyright infringement based on the performance of this contract of which the contractor has knowledge.

**26. CONTRACTOR REPRESENTATION**

Unless the contractor expressly states otherwise in his proposal, where functional requirements are expressly stated as part of the requirements of this solicitation, the contractor, by responding, represents that in its opinion the system proposed is capable of meeting those requirements. In the event of any inconsistency between the functional specifications and the detailed specifications contained in the solicitation, the former will control.

**27. WARRANTY**

Contractor agrees that the equipment, supplies or services to be furnished shall be covered by the most favorable commercial warranties the contractor gives to any customer for the same or substantially similar equipment, supplies or services and that the rights and remedies so provided are in addition to and do not limit any rights afforded to County.

**28. INSURANCE**

Within 10 working days prior to the inception of the contract Contractor shall submit to County certificates of insurance and appropriate separate endorsements to the actual insurance policy, evidencing that the Contractor has obtained for the period of the Contract, at its sole expense, insurance in the following forms of coverage and minimum amounts specified from insurance carriers with a Best's Rating of not less than A-, VII or a company of equal financial stability approved in writing by County's Risk Management Division.

- a. An occurrence policy of Commercial General Liability insurance insuring Contractor against liability for bodily injury, personal injury or property damage arising out of or in connection with the Contractor's performance of work or service under this Contract of not less than \$1,000,000 per occurrence and \$2,000,000 general aggregate. The County of San Diego, its officers, agents, employees, and volunteers shall be added as Additional Insured by separate endorsement to the policy.
- b. Statutory Workers' Compensation, as required by State of California and Employer's Liability at \$1,000,000 each accident for bodily injury or disease. Coverage shall include waiver of subrogation endorsement in favor of County of San Diego.
- c. Comprehensive Automobile Liability covering all owned, non-owned and hired vehicles for bodily injury and property damage of not less than \$1,000,000 each accident.
- d. Certificates of insurance provided by Contractor must evidence that the insurer providing the policy will provide notice of any cancellation, lapse, reduction or other adverse change respecting such insurance in accordance with policy provisions.

The County of San Diego shall retain the right to review the coverage, form and amount of insurance required herein and may require Contractor to obtain insurance reasonably sufficient in coverage, form and amount to provide adequate protection against the kind and extent of risk which exists at the time a change in insurance is required. County requirements shall be reasonable. County retains the right to demand a certified copy of any insurance policy required herein after 15 days notice.

**29. PERMITS, NOTICES, FEES AND LAWS**

The contractor shall, at contractor's expense, obtain all necessary permits and licenses, give all necessary notices, pay all fees required by law, and comply with all laws, ordinances, rules and regulations relating to work and to the preservation of the public health and safety.

**30. AIR, WATER POLLUTION CONTROL, SAFETY AND HEALTH**

Contractor shall comply with all air pollution control, water pollution, Safety and Health Ordinances and statutes which apply to the work performed pursuant to this contract, including any requirements specified in state government codes.

**31. FINDINGS CONFIDENTIAL**

Any reports, information, data, etc., given to or prepared or assembled by the Contractor under this Agreement which the County requests to be kept as confidential shall not be made available to any individual or organization by the Contractor without the prior written approval of the County.

**32. PUBLICATION, REPRODUCTION AND USE OF MATERIAL**

No material produced, in whole or in part, under this Agreement shall be subject to copyright in the United States or in any other country. The County shall have unrestricted authority to publish, disclose, distribute and otherwise use, in whole or in part, any reports, data or other materials prepared under this Agreement. All reports, data and other materials prepared under this Agreement shall be the property of the County upon completion of this Agreement.

**33. NOTICE**

Any notice or notices required or permitted to be given pursuant to this Agreement may be personally served on the other party by the party giving such notice, or may be served by certified mail, return receipt requested, to the addresses set forth herein.

**34. PRODUCT IDENTIFICATION AND LABELING**

Each package shall be identified with manufacturer's label, which shall conform to the requirements of the Fair Packaging and Labeling Act and Section 12604 of the California Business and Professions Code.

### **35. DRUG & ALCOHOL FREE WORKPLACE**

The County of San Diego, in recognition of individual rights to work in a safe, healthful and productive work place, has adopted a requirement for a drug and alcohol free work place, County of San Diego Drug and Alcohol Use Policy C-25. This policy provides that all County employed Contractors and Contractor employees shall assist in meeting this requirement.

- 35.1. As a material condition of this agreement, the Contractor agrees that the Contractor and the Contractor employees, while performing service for the County, on County property, or while using County equipment:
  - 35.1.1. Shall not be in any way impaired because of being under the influence of alcohol or a drug.
- 35.2. Shall not possess an open container of alcohol or consume alcohol or possess or be under the influence of an illegal drug.
  - 35.2.1. Shall not sell, offer, or provide alcohol or a drug to another person.
  - 35.2.2. Shall not be applicable to a Contractor or Contractor employee who, as part of the performance of normal job duties and responsibilities prescribes or administers medically prescribed drugs.
- 35.3. The Contractor shall inform all employees that are performing service for the County on County property or using County equipment, of the County objective of a safe, healthful and productive work place and the prohibition of drug or alcohol use or impairment from same while performing such service for the County.
- 35.4. The County may Terminate for Default or Breach this Agreement and any other Agreement the Contractor has with the County, if the Contractor, or Contractor employees are determined by the Contracting Officer not to be in compliance with the above.

### **36. ORDERING WITH BLANKET PURCHASE AGREEMENT**

A blanket purchase agreement for the estimated requirements will be sent to the successful bidder. This will authorize the acceptance of releases from designated County departments for their requirements. The vendor shall complete delivery of items ordered to destinations set forth in the release. Each release shipment shall be accompanied by a priced invoice itemizing all material. Partial shipments are not acceptable when ordered by release.

### **37. INVOICES**

All deliveries must be accompanied by invoices or delivery tickets. A copy of each invoice or delivery ticket must be signed by the individual accepting delivery. Invoices shall include item, description, quantity, delivery point, price, terms, purchase order number, release number (if applicable to a blanket purchase agreement) and any data relative to the shipment. Original invoices shall be mailed to the County address as specified in the purchase order or blanket purchase agreement release. Discounts will be calculated from receipt of merchandise or invoice, whichever is later.

### **38. PAYMENTS AND INVOICES**

The County is precluded from making payments prior to receipt of service or supplies (advance payments). The County will accept invoice for progress payments and if approved reimburse the Contractor up to 90% of the value of goods received. Invoice subject to following:

Original invoices will be submitted monthly, or at the completion of each phase or task, or at contract completion to the address specified in the purchase order or blanket purchase agreement release.

- 38.1. A copy of the invoice shall be submitted to the Contracting Officer's Technical Representative (COTR).
- 38.2. The invoice must specify items and deliverables for all items described in the "Statement of Work".
- 38.3. Payment shall be Net 30 Days from receipt and approval of invoice unless otherwise stated. Discounts will be calculated from receipt of merchandise or invoice, whichever is later.

### **39. ACCEPTANCE OF COUNTY CREDIT CARD FOR PAYMENT**

- 39.1. Orders may be paid using the County of San Diego credit card (VISA).

39.2. For your bid to be considered responsive, you must accept County of San Diego credit card for payment.

39.3. Pricing remains the same whether paid by credit card or check.

**40. FLAMMABILITY AND TOXICITY**

Materials furnished under this order must meet or exceed minimum California State Fire Marshal's standard for flammability and toxicity for institutional fabrics. Vendor shall provide evidence of California Marshal's test results and approval number.

**41. BRAND NAME OR EQUAL**

- 41.1. If items called for in this Request for Bids have been identified in the schedule by a "brand name or equal" description; such identification is intended to be descriptive, but not restrictive, and is to indicate the quality and characteristics of products (including products of the brand name manufacturer other than the one described by the brand name) will be considered for award if such products are determined by the County to meet fully the salient characteristic requirements listed in the request.
- 41.2. Unless the bidder clearly indicates in the bid that an "or equal" product is being offered, bid shall be considered as offering the brand name product specified.
- 41.3. If the bidder proposes to furnish an "equal product, the brand name, if any, of the product to be furnished shall be inserted in the space provided in the Request for Bids, or such product shall be clearly identified in the bid. The evaluation of the bids and the determination as to equality of the product offered shall be the responsibility of the County and will be based upon the information furnished by the bidder, or identified in the bid as well as other information reasonably available to the Purchasing Activity. CAUTION TO BIDDERS: The Purchasing Activity is not responsible for locating or securing any information which is not identified in the bid and reasonably available to the Purchasing Activity. Accordingly, to insure that sufficient information is available, the bidder must furnish, as part of the bid, all descriptive material (such as cuts, illustrations, drawings, or other information) necessary for the Purchasing Activity to (I) determine whether the product offered meets the salient characteristic requirements of the Request for Bids, and (II) establish exactly what the bidder proposes to furnish and what the County is binding itself to purchase by making an award. The information furnished may include specific references to information previously furnished or to information otherwise available to the Purchasing Activity.
- 41.4. If the bidder proposes to modify a product so as to make it conform to the requirements of the Request for Bids, he shall (I) include in the bid a clear description of such modifications and (II) clearly mark any descriptive to show the proposed modifications.
- 41.5. Modifications proposed after bid opening to make a product conform to a brand name product referenced in the Request for Bids will not be considered.

**42. CONTRACT EXTENSION OPTION**

42.1. One to six months - end of contract period

The providing of goods and/or services described in Section A or B may be extended in one or more increments for a total of no less than one (1) nor more than three (3) calendar months at the discretion of the County Purchasing Director. Each extension shall be affected by written contract modification delivered to the Contractor no less than fifteen (15) calendar days prior to expiration of the contract. The rates set forth in the pricing section shall apply to any extension made pursuant to this option provision unless provision for appropriate price adjustment has been made elsewhere in this contract. All payments are subject to General Terms and Conditions, Clause titled "AVAILABILITY OF FUNDING".

**43. SEVERABILITY**

Should any part of this agreement be held to be invalid by a court of competent jurisdiction, the remainder of the agreement shall be considered as the whole agreement and be binding on the contracting parties.



John M. Pellegrino  
Director

*County of San Diego*  
**Department of Purchasing and Contracting**  
5560 Overland Avenue, Suite 270, San Diego, California 92123-1204

TELEPHONE (858) 605-6367  
FAX (858) 715-8452

September 3, 2013

**ADDENDUM No. 1**

**REAGENTS FOR HIV-1 VIRAL LOAD ASSAY WITH OPTION  
FOR CT/NG VITRO PCR ASSAY TESTING - RFB 6130**

Addendum No. 1. forms a part of the contract documents and changes the original documents only in the manner and to the extent stated.

**BID DUE DATE TIME HAS BEEN CHANGED:**

**BID OPENING DATE AND TIME:** September 11, 2013 at 11 AM

**QUESTIONS AND ANSWERS:**

1. Question: Given that each vendor's viral load assay requires number of components, the inclusion of which is required to produce an accurate and true total cost per test, how will you verify each vendor has included all of the costs necessary to produce a cost per test which is comparable between the vendors? We suggest each vendor be required to submit a list of the cost components used in the calculation of the total cost per test, so a comparable, accurate cost per test is verifiable by all participants in the RFB.

Answer: Yes, we would like each vendor to submit a list of cost components used in the calculation of the total cost per test.

2. Question: Per our discussion today, I wanted to ask for clarification on how you would like the HIV cost per test broken out on the HIV viral load pricing schedule. Per the pricing schedule, we're being asking for reagent costs for 600 HIV tests per month. The total number of tests required to run 600 tests per month is dependent on how often the test is run and how many tests are included in each batch. Without having this information, the cost per test could vary significantly depending on what assumptions are made.

Answer: The pricing should be based on two runs per week with 75 patient samples in each run (batch).

3. Question: Can you clarify whether you'd like us to calculate the test price entered on the pricing schedule to include all consumables?

Answer: Yes

4. If so, we'll need to know the number of batches per week that HIV testing will be performed. The other option would be to list the HIV viral load reagent only price on the pricing schedule and list all consumables on a separate exhibit.

Answer: Two batches per week with 75 patient samples run in each batch.

If you have any questions, please contact Ronald Higley, Procurement Specialist, at (858) 505-6359, or by email at [ronald.higley@sdcounty.ca.gov](mailto:ronald.higley@sdcounty.ca.gov).

*Ronald Higley for*

John M. Pellegrino, Director  
Department of Purchasing and Contracting

JMP:rsh

## APPENDIX TABLE OF CONTENTS

APPENDIX A (SECTION A, PRICE SCHEDULE)

APPENDIX B (PRICE EXHIBIT)

APPENDIX C (ANCILLARY PRODUCTS LIST)

APPENDIX D (*m*2000 BROCHURE)

APPENDIX E (*m*PLUS SELL SHEET)

APPENDIX F (*m*2000 HIV-1 VIRAL LOAD PACKAGE INSERT)

APPENDIX G (*m*2000 HIV-1 SELL SHEET)

APPENDIX H (ABBOTT REALTIME HIV-1 ASSAY: MEET WITH HIV CHALLENGE)

APPENDIX I (ABBOTTLINK BROCHURE)

RFb 6130  
HIV-1 VIRAL LOAD  
PRICING SCHEDULE  
APPENDIX A

Item #	Item Description	UOM	Est. Monthly Qty	Base Term Period: Date of Award thru 31-Aug-2015			First Option Period: 01-Sep-2015 thru 31-Aug-2016			Second Option Period: 01-Sep-2016 thru 31-Aug-2017			Third Option Period: 01-Sep-2017 thru 31-Aug-2018			BASIS OF AWARD
				Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier	Est. Extended Price	Unit Price	Multiplier	Est. Extended Price	
1	HIV-1 Viral Load Test	EA	600	\$47.50	24	\$684,000.00	\$47.50	12	\$342,000.00	\$47.50	12	\$342,000.00	\$47.50	12	\$342,000.00	\$1,710,000.00
<b>BASIS OF AWARD TOTAL OF ALL PERIODS - BASE, FIRST OPTION, SECOND OPTION AND THIRD OPTION PERIOD</b>															<b>\$1,710,000.00</b>	
NOTES																
Unit price includes Equipment, Service, Installation, Integration, Reagents, Sample Preparation, Controls, Calibrators and consumables. The unit price of \$47.50 is the burdened Cost Per Reportable cost, based on a total patient volume of 7,200 per year, running 2 batches per week both at batch size of 72. Please refer to Appendix A and Appendix B for complete Product Pricing and Ancillary Items.																

PRODUCT PRICE EXHIBIT						
Batches Per Week	Batch Size	Assay	Annual Test Volume	TERM	5 Year	
2	72	HIV	7,200	TERM	5 Year	
		<b>Total Annual Test Volume:</b>	<b>7,200</b>			
Equipment & Service Pricing						
		01K35-001	m2000 Complete System (1 sp & 1 RT)	Quantity: 1	Placement Type: RAP	
Reagent, Calibration, Controls and Consumable Pricing Based upon an Annual Test Volume of 7,200 patients running two (2) batches of HIV-VL per week in Batch Size of seventy-two (72).						
				Price Per Kit	Estimated Annual Kit Volume	
		06L18-090	HIV-1 Reagent Kit	\$3,671.71	79	
		06L18-080	HIV-1 Control Kit	\$475.00	13	
		06L18-070	HIV-1 Calibrator Kit	\$950.00	1	
					Subtotal: \$ 297,190.09	
		04J70-024	Sample Prep GPR Pack RNA	\$280.80	79	
					Subtotal: \$ 22,183.20	
		04J71-010	1ml, Pipette Tips	\$471.00	35	
		04J71-017	Pipette Tips, 200uL (24x96 tips)	\$244.00	5	
		04J71-020	Reaction Vessels	\$233.00	6	
		04J71-030	96 Deep Well Plates	\$198.00	13	
		04J71-060	200ml, Rigi Vessel	\$29.00	7	
		04J71-070	96 Well, Optical Reaction Plate	\$49.00	6	
		04J71-075	Optical Adhesive Covers	\$225.00	2	
		04J71-080	M2K MSTRMX VL/CP	\$45.00	1	
			Estimated Cost Per Test:		Subtotal: \$ 22,843.00	
<b>ABBOTT is owner of, and retains title to, the Equipment. Equipment, service, installation, implementation and on-site training is included in the Reagent Product Pricing for the length of the commitment. Estimated Total Annual Dollar (including Equipment):</b>					\$ 342,016.29	\$47.50
Ancillary Items						
		m2000sp	Part Number	List Price		
		BioHazard Bags	04J71-045	\$35.00		
		System liquid container, or liquid waste container	04J72-008	\$126.00		
		Solid waste container	04J72-014	\$133.00		
		Sample rack, 13 mm (set of 6 racks)	04J72-082	\$606.00		
		Sample rack, 16 mm (set of 6 racks)	04J72-086	\$606.00		
		1 mL Subsystem carrier	04J97-050	\$1,130.00		
		2 mL Subsystem carrier cover	04J97-051	\$984.00		
		Waste system	04J72-029	\$558.00		
		Solid waste chute	04N22-003	\$485.00		
		200 uL DITI rack	04J72-030	\$166.00		
		1000 uL DITI rack	04J72-032	\$184.00		
		Reagent vessel carrier	01N06-001	\$880.00		
		PCR Plate Foam Insulation	01N08-001	\$81.00		
		mSystems Wrench	01N71-001	\$60.00		
		Worktable labels	01L39-001	\$42.00		
		System Liquid and Liquid Waste Container labels	04J97-062	\$77.00		
		Bar code labels	04J97-052	\$97.00		
		13mm Post Set	01N83-001	\$107.00		
		16mm Post Set	01N84-001	\$107.00		
		m2000rt	Part Number	List Price		
		Abbott m2000rt Optical Calibration plate	04J71-093	\$295.00		
		Abbott Splash free support base	09K31-001	\$70.00		
		Abbott Adhesive cover applicator	09K32-001	\$28.42		
		ABBOTT m2000rt Halogen Lamp, Replacement	09K33-001	\$225.00		
		ABBOTT m2000rt Plate Holder, Replacement	09K34-001	\$59.48		

APPENDIX C

<b>Ancillary Items</b>		
<b>m2000sp</b>	<b>Part Number</b>	<b>List Price</b>
BioHazard Bags	04J71-045	\$35.00
System liquid container, or liquid waste container	04J72-008	\$126.00
Solid waste container	04J72-014	\$133.00
Sample rack, 13 mm (set of 6 racks)	04J72-082	\$606.00
Sample rack, 16 mm (set of 6 racks)	04J72-086	\$606.00
1 mL Subsystem carrier	04J97-050	\$1,130.00
2 mL Subsystem carrier cover	04J97-051	\$984.00
Waste system	04J72-029	\$558.00
Solid waste chute	04N22-003	\$485.00
200 µL DiTi rack	04J72-030	\$166.00
1000 µL DiTi rack	04J72-032	\$184.00
Reagent vessel carrier	01N06-001	\$880.00
PCR Plate Foam Insulation	01N08-001	\$81.00
mSystems Wrench	01N71-001	\$60.00
Worktable labels	01L39-001	\$42.00
System Liquid and Liquid Waste Container labels	04J97-062	\$77.00
Bar code labels	04J97-052	\$97.00
13mm Post Set	01N93-001	\$107.00
16mm Post Set	01N94-001	\$107.00
<b>m2000rt</b>	<b>Part Number</b>	<b>List Price</b>
Abbott m2000rt Optical Calibration plate	04J71-093	\$295.00
Abbott Splash free support base	09K31-001	\$70.00
Abbott Adhesive cover applicator	09K32-001	\$28.42
ABBOTT m2000rt Halogen Lamp, Replacement	09K33-001	\$225.00
ABBOTT m2000rt Plate Holder, Replacement	09K34-001	\$59.48



# Abbott *m2000* RealTime System

## Abbott *m2000* RealTime Automation



### *m2000sp*: Sample Extraction Automation

- Barcoded Primary Tubes
  - Reduces transcription error and provides positive sample ID
- Precision Pipetting
  - Eliminates manual mixing or manipulation
- Open Mode
  - Flexible protocol for various sample types and volumes
- Efficient Sample Extraction
  - Flexible throughput options of 24 to 96 samples

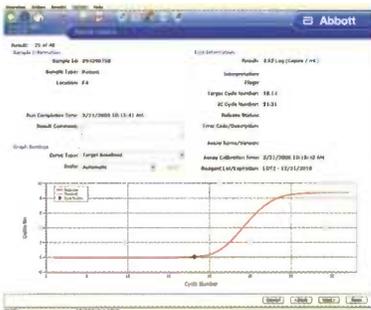


### *m2000rt*: RealTime PCR

- Real-time PCR Amplification and Detection
  - 5 excitation, 5 emission filters
- *maxRatio* Data Analysis
  - Multiple validity checks for improved confidence in patient results
- Minimal Maintenance
  - Halogen bulb replacement

### *m2000* System Software: Simplified Data Management

- Intuitive User Interface
  - Windows XP™
- LIS Capability
  - Standardized interface with LIS
- Data Archiving
  - Calibrator, control, and patient data logs
- Automated Quality Checks and Calibration
  - Provides accurate results
- AbbottLink
  - Remote Instrument Monitoring



# Abbott *m2000* RealTime Performance

The Abbott *m2000sp* and *m2000rt* meet the clinical needs of today's changing molecular laboratory.

Abbott *m2000* RealTime System brings you:

- Walk-Away Automation
- Contamination Control
- Real-Time PCR Technology
- Open Mode Capability



## Abbott *m2000sp*

List Number: 09K14-090

### CHARACTERISTICS

#### Size

Total Length 145 cm (57.1 in)

Total Height 138 cm (53 in)

Total Depth 79.4 cm (31.3 in)

**Weight** 292.7 kg (645.3 lbs)  
instrument and cabinet

**Power Source** 100-240 V

## Abbott *m2000rt*

List Number: 09K15-090

### CHARACTERISTICS

#### Size

Total Length 34 cm (13.4 in)

Total Height 49 cm (19.3 in)

Total Depth 45 cm (17.8 in)

**Weight** 34.1 kg (75.2 lbs)

**Power Source** 100-240 V



## Abbott RealTime *mPlus*

# The performance you need now. *Plus.*

Have you put the Plus in your Efficiency?

The *m2000* system's flexible, lab-ready automation is now better than ever with new *mPlus*.

***mPlus* enables you to:**

- Extend the use of the amplification reagent
- Customize your workflow to your sample arrival pattern
- Reduce waste, costs and turnaround time
- Perform new automatic inventory capabilities
- Provide faster results

*mPlus*. One more reason the *m2000* gives you a total solution for automation, flexibility, platform consolidation and added efficiencies.



## Abbott RealTime support. Ready now.

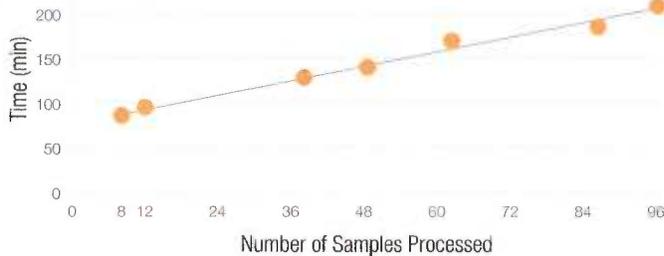
You're in the real-time business of helping physicians care for patients, so we make it our business to serve you. Whether you need installation, implementation, training or preventive maintenance, our certified, knowledgeable field representatives and technical specialists are committed to your success.

# RealTime *mPlus* Putting the Plus in your Efficiency.

*mPlus* is an enabling solution that helps you do more with less.



***m2000sp* Sample Flexibility Matrix\***

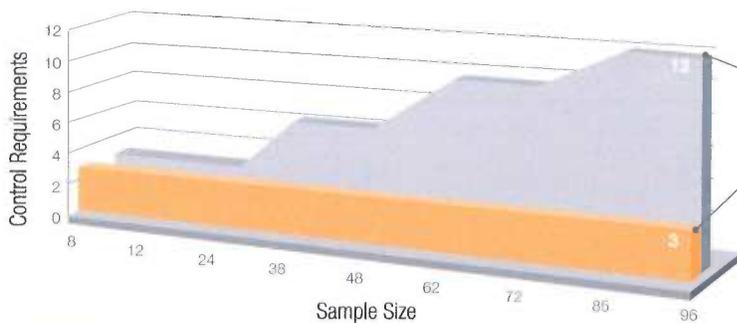


## Time Efficiency

Allows run of 1–96 for customized workflow; processing time reduced by up to 38% by running <24 samples for RealTime CT/NG

Example: 48 vs. 8 samples  
141 min. – 87 min. = 54 min. reduction

**Minimum Control Requirements/Sample Size\***

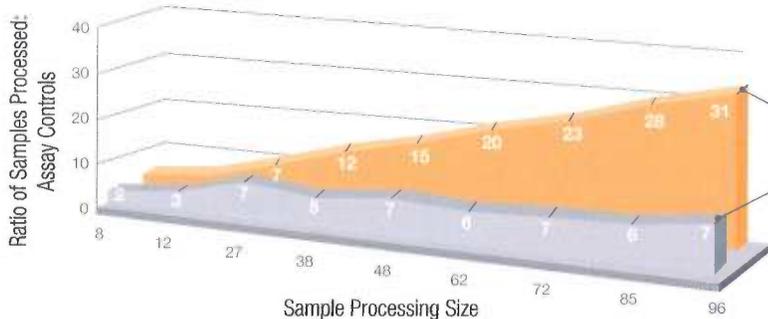


## Cost Efficiency

Minimum control requirements per sample size (Abbott *m2000* = 3; Comparator = 3–12)

3 vs. 12 controls at 96 samples

**Assay Efficiency Ratio of Patient Sample to Controls\***



## Processing Efficiency

More patient results per assay control (Abbott *m2000* ≤31:1; Comparator = 7:1)

31 results per control (Abbott *m2000*)  
7 results per control (Comparator)

Enabling solutions. Leading science. Trusted partner.  
Only with the Abbott *m2000*, now with NEW *mPlus*.

\*Abbott data on file

**Abbott RealTime**  
The **performance** you need. Now.



Contact your Abbott Molecular representative today.  
[www.AbbottMolecular.com](http://www.AbbottMolecular.com)

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# Abbott RealTime HIV-1

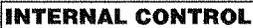
REF 6L18

51-602146/R7

REF 6L18

51-602146/R7

Note: Changes Highlighted

Key to Symbols Used	
	Manufacturer
	Reference Number
	Lot Number
	In Vitro Diagnostic Medical Device
	Internal Control
	Amplification Reagent Pack
	Calibrator A
	Calibrator B
	Negative Control
	Low Positive Control
	High Positive Control
	Store at -10°C or colder
	Use by
	Consult instructions for use
	CAUTION: Handle human sourced materials as potentially infectious. Consult instructions for use. (Infection Risk)

See REAGENTS section for a full explanation of symbols used in reagent component naming.

**CUSTOMER SERVICE: 1-800-553-7042**

**INTERNATIONAL: CALL YOUR ABBOTT REPRESENTATIVE**

This package insert must be read carefully prior to use. Package insert instructions must be followed accordingly. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this package insert

## NAME

Abbott RealTime HIV-1

## INTENDED USE

The Abbott RealTime HIV-1 assay is an in vitro reverse transcription-polymerase chain reaction (RT-PCR) assay for the quantitation of Human Immunodeficiency Virus type 1 (HIV-1) on the automated *m2000* System in human plasma from HIV-1 infected individuals over the range of 40 to 10,000,000 copies/mL. The Abbott RealTime HIV-1 assay is intended for use in conjunction with clinical presentation and other laboratory markers for disease prognosis and for use as an aid in assessing viral response to antiretroviral treatment as measured by changes in plasma HIV-1 RNA levels. This assay is not intended to be used as a donor screening test for HIV-1 or as a diagnostic test to confirm the presence of HIV-1 infection.

## SUMMARY AND EXPLANATION OF THE TEST

Human Immunodeficiency Virus (HIV) is the etiologic agent of Acquired Immunodeficiency Syndrome (AIDS).<sup>1-3</sup> It can be transmitted through sexual contact, exposure to infected blood or blood products, or from an infected mother to the fetus.<sup>4</sup> Acute HIV syndrome, characterized by flu-like symptoms, develops 3 to 5 weeks after initial infection and is associated with high levels of viremia.<sup>5,6</sup> Within 4 to 6 weeks of the onset of symptoms, HIV specific immune response is detectable.<sup>7,8</sup> After seroconversion, viral load in peripheral blood declines and most patients enter an asymptomatic phase that can last for years.<sup>9</sup>

Quantitative measurement of HIV levels in peripheral blood has greatly contributed to the understanding of the pathogenesis of HIV infection<sup>10,11</sup> and has been shown to be an essential parameter in prognosis and management of HIV infected individuals.<sup>12-17</sup> Decisions regarding initiation or changes in antiretroviral therapy are guided by monitoring plasma HIV RNA levels (viral load), CD4+ T cell count, and the patient's clinical condition.<sup>17,18</sup> The goal of antiretroviral therapy is to reduce the HIV virus in plasma to below detectable levels of available viral load tests.<sup>17,19</sup>

HIV RNA levels in plasma can be quantitated by nucleic acid amplification or signal amplification technologies.<sup>20-22</sup> The Abbott RealTime HIV-1 assay uses Polymerase Chain Reaction (PCR) technology with homogenous real-time fluorescent detection. Partially double-stranded fluorescent probe design allows detection of diverse group M subtypes and group O isolates. The assay is standardized against a viral standard from the Virology Quality Assurance (VQA) Laboratory of the AIDS Clinical Trial Group,<sup>23</sup> and against World Health Organization (WHO) 1<sup>st</sup> International Standard for HIV-1 RNA (97/656).<sup>24,25</sup> The assay results can be reported in copies/mL or International Units/mL (IU/mL).

## BIOLOGICAL PRINCIPLES OF THE PROCEDURE

The Abbott RealTime HIV-1 assay uses RT-PCR<sup>26</sup> to generate amplified product from the RNA genome of HIV-1 in clinical specimens. An RNA sequence that is unrelated to the HIV-1 target sequence is introduced into each specimen at the beginning of sample preparation. This unrelated RNA sequence is simultaneously amplified by RT-PCR, and serves as an internal control (IC) to demonstrate that the process has proceeded correctly for each sample. The amount of HIV-1 target sequence that is present at each amplification cycle is measured through the use of fluorescent-labeled oligonucleotide probes on the Abbott *m2000rt* instrument. The probes do not generate signal unless they are specifically bound to the amplified product. The amplification cycle at which fluorescent signal is detected by the Abbott *m2000rt* is proportional to the log of the HIV-1 RNA concentration present in the original sample.

## Sample Preparation

The purpose of sample preparation is to extract and concentrate the target RNA molecules to make the target accessible for amplification, and to remove potential inhibitors of amplification from the extract.

The Abbott *m2000sp* instrument prepares samples for the Abbott RealTime HIV-1 assay using the Abbott *mSample* Preparation System (4 × 24 Preps) reagents. The *m2000sp* uses magnetic particle technology to capture nucleic acids and washes the particles to remove unbound sample components. The bound nucleic acids are eluted and transferred to a 96 deep-well plate. The nucleic acids are then ready for amplification. The IC is taken through the entire sample preparation procedure along with the calibrators, controls, and specimens.

## Reagent Preparation and Reaction Plate Assembly

The Abbott *m2000sp* combines the Abbott RealTime HIV-1 amplification reagent components (HIV-1 Oligonucleotide Reagent, Thermostable rTth Polymerase Enzyme, and Activation Reagent). The Abbott *m2000sp* dispenses the resulting master mix to the Abbott 96-Well Optical

Reaction Plate along with aliquots of the nucleic acid samples prepared by the Abbott *m2000sp*. The plate is ready, after manual application of the optical seal, for transfer to the Abbott *m2000rt*.

## Amplification

During the amplification reaction on the Abbott *m2000rt*, the target RNA is converted to cDNA by the reverse transcriptase activity of the thermostable rTth DNA polymerase. First, the HIV-1 and IC reverse primers anneal to their respective targets and are extended during a prolonged incubation period. After a denaturation step, in which the temperature of the reaction is raised above the melting temperature of the double-stranded cDNA:RNA product, a second primer anneals to the cDNA strand and is extended by the DNA polymerase activity of the rTth enzyme to create a double-stranded DNA product.

During each round of thermal cycling, amplification products dissociate to single strands at high temperature allowing primer annealing and extension as the temperature is lowered. Exponential amplification of the product is achieved through repeated cycling between high and low temperatures, resulting in a billion-fold or greater amplification of target sequences. Amplification of both targets (HIV-1 and IC) takes place simultaneously in the same reaction.

The target sequence for the Abbott RealTime HIV-1 assay is in the *pol* integrase region of the HIV-1 genome. This region is highly conserved.<sup>27</sup>

The IC target sequence is derived from the hydroxypyruvate reductase gene from the pumpkin plant, *Cucurbita pepo*, and is delivered in an Armored RNA<sup>®</sup> particle that has been diluted in negative human plasma.

## Detection

During the read cycles of amplification on the Abbott *m2000rt*, the temperature is lowered further to allow fluorescent detection of amplification products as the HIV-1 and IC probes anneal to their targets (real-time fluorescence detection). The HIV-1 probe has a fluorescent moiety that is covalently linked to the 5' end. A short oligonucleotide (quencher oligonucleotide) is complementary to the 5' end of the HIV-1 probe and has a quencher molecule 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 target sequences, probe fluorescence is quenched. In the presence of IC target sequences, probe hybridization to complementary sequences separates the fluorophore and the quencher and allows fluorescent emission and detection.

The HIV-1 and IC specific probes are each labeled with a different fluorophore, thus allowing for simultaneous detection of both amplified products at each cycle. The amplification cycle at which fluorescent signal is detected by the Abbott *m2000rt* is proportional to the log of the HIV-1 RNA concentration present in the original sample.

## Quantitation

A calibration curve is required to quantitate the HIV-1 RNA concentration of specimens and controls. Two assay calibrators are run in replicates of 3 to generate a calibration curve. The calibration curve slope and intercept are calculated from the assigned HIV-1 RNA concentration and the median observed threshold cycle for each calibrator and are stored on the instrument. The concentration of HIV-1 RNA in specimens and controls is calculated from the stored calibration curve, and the results are automatically reported on the *m2000rt* workstation. The Abbott RealTime HIV-1 Negative Control, Low Positive Control, and High Positive Control must be included in each run to verify run validity. The *m2000* System verifies that the controls are within the assigned ranges.

## PREVENTION OF NUCLEIC ACID CONTAMINATION

The possibility of nucleic acid contamination is minimized because:

- Reverse transcription, PCR amplification, and oligonucleotide hybridization occur in a sealed 96-Well Optical Reaction Plate.
- Detection is carried out automatically without the need to open the 96-Well Optical Reaction Plate.
- Aerosol barrier pipette tips are used for all pipetting. The pipette tips are discarded after use.
- Separate dedicated areas are used to perform the Abbott RealTime HIV-1 assay. Refer to the **SPECIAL PRECAUTIONS** section of this package insert.

## REAGENTS

The Abbott RealTime Reagents are intended for single-use only and unused reagents should be discarded.

## Abbott RealTime HIV-1 Amplification Reagent Kit (List No. 6L18-90)

1. **INTERNAL CONTROL** Abbott RealTime HIV-1 Internal Control (List No. 2G31Y)  
(4 vials, 1.2 mL per vial)  
Noninfectious Armored RNA with internal control sequences in negative human plasma. Negative human plasma tested and found to be nonreactive for HBsAg, HIV RNA, HCV RNA, HBV DNA, anti-HIV-1/HIV-2, and anti-HCV. Preservatives: 0.1% ProClin<sup>®</sup> 300 and 0.15% ProClin 950.
2. **AMPLIFICATION REAGENT PACK** Abbott RealTime HIV-1 Amplification Reagent Pack (List No. 6L18)  
Four packs of single-use reagents, 24 tests/pack. **Unused reagents should be discarded.**  
Each pack contains:
  - 1 bottle (0.141 mL) Thermostable rTth Polymerase Enzyme (2.9 to 3.5 Units/ $\mu$ L) in buffered solution.
  - 1 bottle (1.10 mL) HIV-1 Oligonucleotide Reagent. Synthetic oligonucleotides (4 primers, 2 probes, and 1 quencher oligonucleotide), and dNTPs in a buffered solution with a reference dye. Preservatives: 0.1% ProClin 300 and 0.15% ProClin 950.
  - 1 bottle (0.40 mL) Activation Reagent. 30 mM manganese chloride solution. Preservatives: 0.1% ProClin 300 and 0.15% ProClin 950.

## Abbott RealTime HIV-1 Control Kit (List No. 6L18-80) and Lot-specific Kit Card

1. **CONTROL<sub>-</sub>** Abbott RealTime HIV-1 Negative Control (List No. 2G31Z)  
(8 vials, 1.8 mL per vial)  
Negative human plasma tested and found to be nonreactive for HBsAg, HIV RNA, HCV RNA, HBV DNA, anti-HIV-1/HIV-2, and anti-HCV. Preservatives: 0.1% ProClin 300 and 0.15% ProClin 950.
2. **CONTROL<sub>L</sub>** Abbott RealTime HIV-1 Low Positive Control (List No. 2G31W)  
(8 vials, 1.8 mL per vial)  
Noninfectious Armored RNA with HIV-1 sequences in negative human plasma. Negative human plasma tested and found to be nonreactive for HBsAg, HIV RNA, HCV RNA, HBV DNA, anti-HIV-1/HIV-2, and anti-HCV. Preservatives: 0.1% ProClin 300 and 0.15% ProClin 950.
3. **CONTROL<sub>H</sub>** Abbott RealTime HIV-1 High Positive Control (List No. 2G31X)  
(8 vials, 1.8 mL per vial)  
Noninfectious Armored RNA with HIV-1 sequences in negative human plasma. Negative human plasma tested and found to be nonreactive for HBsAg, HIV RNA, HCV RNA, HBV DNA, anti-HIV-1/HIV-2, and anti-HCV. Preservatives: 0.1% ProClin 300 and 0.15% ProClin 950.

## Abbott RealTime HIV-1 Calibrator Kit (List No. 6L18-70) and Lot-specific Kit Card

1. **CAL<sub>A</sub>** Abbott RealTime HIV-1 Calibrator A (List No. 2G31A)  
(12 vials, 1.8 mL per vial)  
Noninfectious Armored RNA with HIV-1 sequences in negative human plasma. Negative human plasma tested and found to be nonreactive for HBsAg, HIV RNA, HCV RNA, HBV DNA, anti-HIV-1/HIV-2, and anti-HCV. Preservatives: 0.1% ProClin 300 and 0.15% ProClin 950.
2. **CAL<sub>B</sub>** Abbott RealTime HIV-1 Calibrator B (List No. 2G31B)  
(12 vials, 1.8 mL per vial)  
Noninfectious Armored RNA with HIV-1 sequences in negative human plasma. Negative human plasma tested and found to be nonreactive for HBsAg, HIV RNA, HCV RNA, HBV DNA, anti-HIV-1/HIV-2, and anti-HCV. Preservatives: 0.1% ProClin 300 and 0.15% ProClin 950.

**NOTE: Control kit lots, calibrator kit lots, and amplification reagent kit lots can be used interchangeably. If a new amplification reagent kit lot is used, then the assay needs to be recalibrated. Do not interchange kit components from different kit lots. For example, do not use the negative control from control kit lot X with the positive controls from control kit lot Y.**

## WARNINGS AND PRECAUTIONS

**IVD** In Vitro Diagnostic Medical Device

For In Vitro Diagnostic Use

- This assay is not intended to be used as a screening test for HIV-1 or as a diagnostic test to confirm the presence of HIV-1 infection.

- The Abbott RealTime HIV-1 reagents are intended to be used only on the Abbott m2000 System consisting of the m2000sp for sample processing and the m2000rt for amplification and detection.
- Do not use expired reagents.
- The Abbott m2000sp Master Mix Addition protocol must be initiated within 1 hour after completion of Sample Preparation. If the Abbott m2000sp master mix addition protocol is aborted, seal the Abbott 96-Well Optical Reaction Plate in a sealable plastic bag and dispose according to the m2000sp Operations Manual, Hazards section, along with the gloves used to handle the plate. Do not import the test order onto the m2000rt.
- The appropriate PCR plate must be selected when samples are loaded into the m2000rt instrument.
- The m2000rt protocol must be started within 50 minutes of the initiation of the Master Mix Addition protocol. If the Abbott m2000rt instrument run is not initiated within 50 minutes, or is interrupted or aborted, seal the Abbott 96-Well Optical Reaction Plate in a sealable plastic bag and dispose according to the Abbott m2000rt Operations Manual along with the gloves used to handle the plate.

## Safety Precautions

Refer to the Abbott m2000sp and Abbott m2000rt Operations Manuals, Hazard Section, for instructions on safety precautions.



**CAUTION:** This product contains human sourced and/or potentially infectious components. For a specific listing, refer to the **REAGENTS** section of this package insert. Human sourced material has been tested and found to be nonreactive to HBsAg, HCV RNA, HIV RNA, HBV DNA, anti-HIV-1/HIV-2, and anti-HCV. No known test method can offer complete assurance that products derived from human sources or inactivated microorganisms will not transmit infection. Therefore, all human sourced materials should be considered potentially infectious. It is recommended that these reagents and human specimens be handled in accordance with the OSHA Standard on Bloodborne Pathogens,<sup>28</sup> Biosafety Level 2<sup>29</sup> or other appropriate biosafety practices<sup>30,31</sup> should be used for materials that contain or are suspected of containing infectious agents. These precautions include, but are not limited to, the following:

- Wear gloves when handling specimens or reagents.
- Do not pipette by mouth.
- Do not eat, drink, smoke, apply cosmetics, or handle contact lenses in areas where these materials are handled.
- Clean and disinfect spills of specimens by including the use of a tuberculocidal disinfectant such as 1.0% sodium hypochlorite or other suitable disinfectant.<sup>32,33</sup>
- Decontaminate and dispose of all potentially infectious materials in accordance with local, state, and federal regulations.<sup>34,35</sup>

The Abbott RealTime HIV-1 Calibrator Kit, Control Kit, Internal Control, HIV-1 Oligonucleotide Reagent, and Activation Reagent contain a mixture of 5-chloro-2-methyl-4-isothiazolin-3-one and 2-methyl-4-isothiazolin-3-one which are components of ProClin. The components are classified per applicable European Community (EC) Directives as: Irritant (Xi). The following are the appropriate Risk (R) and Safety (S) phrases:



- |     |   |
|-----|---|
| R43 | May cause sensitization by skin contact.  |
| S24 | Avoid contact with skin.  |
| S35 | This material and its container must be disposed of in a safe way.              |
| S37 | Wear suitable gloves.   |
| S46 | If swallowed, seek medical advice immediately and show this container or label. |

## SPECIAL PRECAUTIONS

### Handling Precautions

The Abbott RealTime HIV-1 assay is only for use with plasma specimens that have been handled and stored in capped tubes as described in the **SPECIMEN COLLECTION, STORAGE, AND TRANSPORT TO THE TEST SITE** section.

During preparation of samples, compliance with good laboratory practices is essential to minimize the risk of cross-contamination between samples, and the inadvertent introduction of ribonucleases (RNases) into samples during and after the extraction procedure. Proper aseptic technique should always be used when working with RNA. Amplification technologies such as PCR are sensitive to accidental introduction of product from previous amplification reactions. Incorrect results could occur if either the clinical specimen or the RealTime reagents used in the amplification step become contaminated by accidental introduction of even a few molecules of amplification product.

Measures to reduce the risk of contamination in the laboratory include physically separating the activities involved in performing PCR in compliance with good laboratory practices.

## Work Areas

Two dedicated areas, Sample Preparation Area and Amplification Area, are recommended.

- The Sample Preparation Area is dedicated to processing samples (specimens, Abbott RealTime HIV-1 Controls, and Calibrators), and to adding processed specimens, controls, and calibrators to the Abbott 96-Well Optical Reaction Plate. **All reagents used in the Sample Preparation Area should remain in this dedicated area at all times. Laboratory coats, pipettes, pipette tips, and vortexers used in the Sample Preparation Area must remain in this area and not be moved to the Amplification Area. Do not bring amplification product into the Sample Preparation Area.**
- The Amplification Area is dedicated to the amplification and detection of amplified product. Laboratory coats and equipment used in the Amplification Area must remain in this area and not be moved to the Sample Preparation Area.

Control kit lots, calibrator kit lots, and amplification reagent kit lots can be used interchangeably. If a new amplification reagent kit lot is used, then the assay needs to be recalibrated. Do not interchange kit components from different kit lots. For example, do not use the negative control from control kit lot X with the positive controls from control kit lot Y.

The Amplification Reagent Kit, Control Kit, and Calibrator Kit can be thawed and refrozen up to 3 times before use.

Work area and instrument platforms must be considered potential sources of contamination. Change gloves after contact with potential contaminants (specimens, eluates, and/or amplified product) before handling unopened reagents, negative control, positive controls, calibrators, or specimens. Refer to the Abbott m2000sp and m2000rt Operations Manuals for instrument cleaning procedures.

If the Abbott m2000sp instrument run is aborted, dispose of all commodities and reagents according to the Abbott m2000sp Operations Manual. If the Abbott m2000sp master mix addition protocol is aborted, seal the Abbott 96-Well Optical Reaction Plate in a sealable plastic bag and dispose according to the m2000sp Operations Manual, Hazards section, along with the gloves used to handle the plate.

If the Abbott m2000rt instrument run is interrupted or aborted, seal the Abbott 96-Well Optical Reaction Plate in a sealable plastic bag and dispose according to the Abbott m2000rt Operations Manual along with the gloves used to handle the plate.

Decontaminate and dispose of all potentially biohazardous materials in accordance with local, state, and federal regulations.<sup>34,35</sup> All materials should be handled in a manner that minimizes the chance of potential contamination of the work area.

**NOTE: Autoclaving the sealed Reaction Plate will not degrade the amplified product and may contribute to the release of the amplified product by opening the sealed plate. The laboratory area can become contaminated with amplified product if the waste materials are not carefully handled and contained.**

## Aerosol Containment

To reduce the risk of nucleic acid contamination due to aerosols formed during manual pipetting, aerosol barrier pipette tips must be used for all manual pipetting. The pipette tips must be used only one time. Clean and disinfect spills of specimens and reagents as stated in the Abbott m2000sp and Abbott m2000rt Operations Manuals.

## Contamination and Inhibition

The following precautions should be observed to minimize the risks of RNase contamination, cross-contamination between samples, and inhibition:

- Wear appropriate personal protective equipment at all times.
- Use powder-free gloves.
- Change gloves after having contact with potential contaminants (such as specimens, eluates, and/or amplified product).
- To reduce the risk of nucleic acid contamination due to aerosols formed during pipetting, pipettes with aerosol barrier tips must be used for all pipetting. The length of the tip should be sufficient to prevent contamination of the pipette barrel. While pipetting, care should be taken to avoid touching the pipette barrel to the inside of the sample tube or container. The use of extended aerosol barrier pipette tips is recommended.
- Change aerosol barrier pipette tips between ALL manual liquid transfers.

- The Abbott *mSample* Preparation System (4 × 24 Preps) reagents are single use only. Use new reagent vessels, reaction vessels, and newly opened reagents for every new Abbott RealTime HIV-1 assay run. At the end of each run, discard all remaining reagents from the worktable as stated in the Abbott *m2000sp* Operations Manual and the Abbott *m Sample* Preparation System (4 × 24 Preps) product information sheet.

#### STORAGE INSTRUCTIONS

##### Abbott RealTime HIV-1 Amplification Reagent Kit (List No. 6L18-90)

 <sup>-10°C</sup> The Abbott RealTime HIV-1 Amplification Reagent Pack and Internal Control vials must be stored at –10°C or colder when not in use. Care must be taken to separate the Abbott RealTime HIV-1 Amplification Reagent Pack that is in use from direct contact with samples, calibrators and controls.

##### Abbott RealTime HIV-1 Control Kit (List No. 6L18-80)

 <sup>-10°C</sup> The Abbott RealTime HIV-1 Negative and Positive Controls must be stored at –10°C or colder.

##### Abbott RealTime HIV-1 Calibrator Kit (List No. 6L18-70)

 <sup>-10°C</sup> The Abbott RealTime HIV-1 Calibrator A and Calibrator B must be stored at –10°C or colder.

#### SHIPPING CONDITIONS

- Abbott RealTime HIV-1 Amplification Reagent Kit: Ship on dry ice.
- Abbott RealTime HIV-1 Control Kit: Ship on dry ice.
- Abbott RealTime HIV-1 Calibrator Kit: Ship on dry ice.

#### INDICATION OF INSTABILITY OR DETERIORATION OF REAGENTS

When a positive or negative control value is out of the expected range, it may indicate deterioration of the reagents. Associated test results are invalid and samples must be retested. Assay recalibration may be necessary.

#### INSTRUMENT PROCEDURE

The Abbott RealTime HIV-1 application files must be installed on the Abbott *m2000sp* and Abbott *m2000rt* systems from the Abbott RealTime HIV-1 *m2000* System Combined Application CD-ROM prior to performing the assay. For detailed information on application file installation, refer to the Abbott *m2000sp* and Abbott *m2000rt* Operations Manuals, Operating Instructions section.

#### SPECIMEN COLLECTION, STORAGE, AND TRANSPORT TO THE TEST SITE

##### Specimen Collection and Storage

Human plasma (ACD-A and EDTA) specimens may be used with the Abbott RealTime HIV-1 assay. Follow the manufacturer's instructions for processing plasma collection tubes.

Freshly drawn specimens (whole blood) may be held at 15 to 30°C for up to 6 hours or at 2 to 8°C for up to 24 hours, prior to centrifugation.

After centrifugation, remove plasma from cells. Plasma specimens may be stored at 15 to 30°C for up to 24 hours or at 2 to 8°C for up to 5 days. Plasma specimens may be stored at –20°C for up to 60 days; if longer storage is required, plasma specimens must be stored at –70°C or lower.<sup>36,37</sup> Multiple freeze-thaw cycles should be avoided and should not exceed 3 freeze/thaw cycles. Thaw plasma specimens at 15 to 30°C or at 2 to 8°C. Once thawed, if plasma specimens are not being processed immediately, they can be stored at 2 to 8°C for up to 6 hours.

##### Specimen Transport

Ship specimens frozen on dry ice. For domestic shipments, specimens should be packaged and labeled in compliance with applicable local, state, and federal regulations covering the transport of clinical, diagnostic, or biological specimens.

#### ABBOTT REALTIME HIV-1 ASSAY PROCEDURE

The Abbott RealTime HIV-1 assay provides four sample volume options (0.2 mL, 0.5 mL, 0.6 mL, and 1.0 mL). See assay protocol step 9 and Interpretation of Results section

#### Materials Provided

- Abbott RealTime HIV-1 Amplification Reagent Kit (List No. 6L18-90)

#### Materials Required But Sold Separately

- Abbott RealTime HIV-1 Calibrator Kit (List No. 6L18-70)
- Abbott RealTime HIV-1 Control Kit (List No. 6L18-80)

#### Materials Required But Not Provided (Each available separately)

##### Sample Preparation Area

- Abbott *m2000sp* instrument (List No. 9K14) with Version 4.0 or higher Software
- Abbott *mSample* Preparation System (4 × 24 Preps) (List No. 04J70-24)
- 5 mL Reaction Vessels (List No. 4J71-20)
- 13 mm to 16 mm Sample Tubes
- 200 mL Reagent Vessels (List No. 4J71-60)
- 200 µL (List No. 4J71-15) Disposable Tips
- 1000 µL (List No. 4J71-10) Disposable Tips
- Abbott Optical Adhesive Covers (List No. 4J71-75)
- Abbott Adhesive Cover Applicators (List No. 9K32-01)
- Abbott Splash-Free Support Base (List No. 9K31-01)
- Abbott 96-Deep-Well Plate (List No. 4J71-30)
- Abbott RealTime HIV-1 *m2000* System Combined Application CD-ROM (List No. 6L83)
- Abbott 96-Well Optical Reaction Plate (List No. 4J71-70)
- Aerosol Barrier Pipette Tips for 20 to 1000 µL pipettes
- Calibrated Pipettes capable of delivering 20 to 1000 µL
- Centrifuge capable of 2000g
- Master Mix Tube (List No. 4J71-80)
- Vortex Mixer

##### Amplification Area

- Abbott *m2000rt* instrument (List No. 9K15) with Version 2.0 or higher Software
- Abbott RealTime HIV-1 *m2000* System Combined Application CD-ROM (List No. 6L83)
- Abbott *m2000rt* Optical Calibration Kit (List No. 4J71-93)

##### Other Materials

- Biological safety cabinet approved for working with infectious materials
- Sealable plastic bags
- RNase-free water (Eppendorf or equivalent)<sup>†</sup>
- 1.7 mL RNase-free Microcentrifuge Tubes (Dot Scientific, Inc. or equivalent)<sup>†</sup>
- Cotton Tip Applicators (Puritan or equivalent)<sup>†</sup>

<sup>†</sup>Note: These 3 items are used in the procedure for Monitoring the Laboratory for the Presence of Contamination. Refer to the QUALITY CONTROL PROCEDURES section of this package insert.

#### Procedural Precautions

- Read the instructions in this package insert carefully before processing samples.
- The Abbott RealTime HIV-1 Calibrators, Internal Control, Negative Control, Low Positive Control, and High Positive Control vials are intended for single-use only and should be discarded after use.
- The Abbott *m2000sp* Master Mix Addition protocol must be initiated within 1 hour after completion of Sample Preparation. If the Abbott *m2000sp* master mix addition protocol is not initiated, recap the Amplification Reagent vials and return the Amplification Reagent Pack to –10°C storage. Once thawed, the Abbott RealTime HIV-1 Amplification Reagent Pack can be frozen and thawed a maximum of 3 times. If the Abbott *m2000sp* master mix addition protocol is aborted, then discard the amplification reagents.
- The *m2000rt* protocol must be started within 50 minutes of the initiation of the Master Mix Addition protocol. If the Abbott *m2000rt* instrument run is not initiated within 50 minutes, or is interrupted or aborted, seal the Abbott 96-Well Optical Reaction Plate in a sealable plastic bag and dispose according to the Abbott *m2000rt* Operations Manual along with the gloves used to handle the plate.
- Use aerosol barrier pipette tips or disposable pipettes only 1 time when pipetting specimens or IC. To prevent contamination to the pipette barrel while pipetting, care should be taken to avoid touching the pipette barrel to the inside of the sample tube or container. The use of extended aerosol barrier pipette tips is recommended.
- Monitoring procedures for the presence of amplification product can be found in the QUALITY CONTROL PROCEDURES section in this package insert.
- To reduce the risk of nucleic acid contamination, clean and disinfect spills of specimens by including the use of a tuberculocidal disinfectant such as 1.0% sodium hypochlorite or other suitable disinfectant.

- The Abbott RealTime HIV-1 Calibrators and Controls must be prepared in conjunction with the specimens to be tested. The use of the Abbott RealTime HIV-1 Controls and Calibrators is integral to the performance of the Abbott RealTime HIV-1 assay. Refer to the **QUALITY CONTROL PROCEDURES** section of this package insert for details.

**ASSAY PROTOCOL**

For a detailed description on how to operate the Abbott *m2000sp* instrument or the Abbott *m2000rt* instrument, refer to the Abbott *m2000sp* and *m2000rt* Operations Manuals, Operating Instructions sections.

Laboratory personnel must be trained to operate the Abbott *m2000sp* and *m2000rt* instruments. The operator must have a thorough knowledge of the applications run on the instruments and must follow good laboratory practices.

- Thaw assay controls and IC at 15 to 30°C or at 2 to 8°C. Thaw calibrators at 15 to 30°C or at 2 to 8°C only if performing a calibration run; see **QUALITY CONTROL PROCEDURES** section of this package insert for description of assay calibration. Once thawed, assay controls, IC, and calibrators can be stored at 2 to 8°C for up to 24 hours before use.
- Vortex each assay calibrator and each control 3 times for 2 to 3 seconds. Ensure that the contents of each vial are at the bottom after vortexing by tapping the vials on the bench to bring liquid to the bottom of the vial. Ensure bubbles or foam are not generated; if present, remove with a sterile pipette tip, using a new tip for each vial.
- Thaw amplification reagents at 15 to 30°C or at 2 to 8°C and store at 2 to 8°C until required for the amplification master mix procedure. Once thawed, the amplification reagents can be stored at 2 to 8°C for up to 24 hours if not used immediately.

**NOTE: Use 1 bottle of mLysis Buffer, 1 vial of IC, and 1 RealTime HIV-1 Amplification Reagent Pack to support up to 24 reactions. Use a second set of reagents to support 25 to 48 reactions, a third set of reagents to support 49 to 72 reactions, and a fourth set of reagents to support 73 to 96 reactions WITH THE EXCEPTION OF mMICROPARTICLES. USE ONLY 2 BOTTLES OF mMICROPARTICLES WHEN PROCESSING 25 TO 96 SAMPLES.**

- Invert gently the Abbott *mSample* Preparation bottles to ensure a homogeneous solution without generating any bubbles. If crystals are observed in any of the reagent bottles upon opening, allow the reagent to equilibrate at room temperature until the crystals disappear. Do not use the reagents until the crystals have dissolved. Ensure bubbles or foam are not generated; if present, remove with a sterile pipette tip, using a new tip for each bottle.
- Vortex each IC vial 3 times for 2 to 3 seconds before use. Ensure bubbles or foam are not generated; if present, remove with a sterile pipette tip, using a new tip for each vial.
- Use a calibrated precision **PIPETTE DEDICATED FOR INTERNAL CONTROL USE ONLY** to add 500 µL of IC to each bottle of mLysis Buffer. Mix by gently inverting the container 5 to 10 times to minimize foaming.
- A maximum of 96 reactions can be performed per run.** A negative control, a low positive control, and a high positive control are included in each run, therefore allowing a maximum of 93 specimens to be processed per run.
- Thaw specimens at 15 to 30°C or at 2 to 8°C. Once thawed, specimens can be stored at 2 to 8°C for up to 6 hours if not processed immediately.
- Check sample volume. The Abbott RealTime HIV-1 assay minimum sample volume and associated rack requirements on the Abbott *m2000sp* are described below.

**CAUTION: Do not put a 13 mm tube in a 16 mm rack.**

Rack	Tube Diameter <sup>a</sup>	Abbott RealTime HIV-1 Minimum Sample Volume Assay Application			
		0.2 mL	0.5 mL	0.6 mL	1.0 mL <sup>b</sup>
13 mm	11.6 mm - 14.0 mm	0.7 mL	1.0 mL	1.1 mL	1.5 mL
16 mm	15.0 mm - 16.0 mm	1.0 mL	1.3 mL	1.4 mL	1.8 mL

<sup>a</sup> Refers to sample tube outer diameter.

<sup>b</sup> 1.0 mL sample volume option is only available for up to 48 samples.

**NOTE: Sample tubes containing insufficient sample volume will not be processed during the run and will be identified by an insufficient liquid volume error message code in the Process Log and Plate Result screen.**

**CAUTION: Steps 10 and 11 must be done in the order described. Vortex the specimens first, and follow with centrifugation. If these two steps are not performed in this order, then invalid results may occur.**

- Vortex each specimen 3 times for 2 to 3 seconds.
- Centrifuge specimens at 2000g for 5 minutes before loading on the Abbott *m2000sp* worktable.

**NOTE: The “g” refers to g force, not revolutions per minute (rpm).**

- Aliquot each specimen into clean tubes or vials if necessary. Refer to the Abbott *m2000sp* Operations Manual for tube sizes. Avoid touching the inside of the cap when opening tubes. Take care not to disturb contents of the tube while removing the tube from the centrifuge and that the bottom of the tube is not touched by the pipette tip. Ensure that the newly aliquotted sample retains the minimum volume indicated in the preceding table.
- Place the low and high positive controls, the negative control, the calibrators, if applicable, and the patient specimens into the Abbott *m2000sp* sample rack.
- Place the 5 mL Reaction Vessels into the *m2000sp* 1 mL subsystem carrier.
- Load the Abbott *mSample* Preparation System reagents and the Abbott 96 Deep-Well Plate on the Abbott *m2000sp* worktable as described in the following table and in the Abbott *m2000sp* Operations Manual, Operating Instructions section.
- Select the appropriate application file from the Run Sample Extraction screen that corresponds to the sample volume being tested. Initiate the sample extraction protocol as described in the Abbott *m2000sp* Operations Manual, Operating Instruction section.
- Enter calibrator (needed if a calibration curve has not been stored on the *m2000rt*) and control lot specific values in the **Sample Extraction: Worktable Setup, Calibrator and Control** fields. Lot specific values are specified in each Abbott RealTime HIV-1 Calibrator and Control Kit Card.

**NOTE: Verify the values entered match the values provided in the lot specific kit cards.**

**NOTE: The Abbott *m2000sp* Master Mix Addition protocol (step 21) must be initiated within 1 hour after completion of Sample Preparation.**

**NOTE: Change gloves before handling the amplification reagents.**

**NOTE: The plate-fill setup will automatically be enabled in the following run conditions:**

- A batch size of 49 to 95 samples is processed and any valid samples are detected in column locations 7 through 12.
- A batch size of 49 to 95 samples is processed and any samples are detected with a Well Status of “Error.”
- A full batch size of 96 samples is processed and any samples are detected with a Well Status of “Error.”

In these conditions, Reagent Carrier 2 should remain in place, minimally containing the reagent vessel for Elution Buffer (Reagent Carrier 2, location 6). If this reagent vessel has been unloaded, place a new reagent vessel containing the Elution Buffer label within Reagent Carrier 2, location 6. This reagent vessel may remain empty and system fluid will be added to the reagent vessel.

**NOTE: System instructions for use of the automated plate-filling feature are found in the *m2000sp* Operations Manual (List No. 9K20-04 or higher), Section 5, Operating Instructions, Sample Extraction – Closed Mode.**

- Load the amplification reagents and the master mix vial on the *m2000sp* worktable after sample preparation is completed. Each Amplification Reagent Pack supports up to 24 reactions.

**NOTE: A second Amplification Reagent Pack is required if performing 25 to 48 reactions.**

**A third Amplification Reagent Pack is required for 49 to 72 reactions.**

**A fourth Amplification Reagent Pack is required for 73 to 96 reactions.**

- Ensure that the contents are at the bottom of the vials prior to opening the amplification reagents by tapping the vials in an upright position on the bench.
- Remove and discard the amplification vial caps.
- Select the appropriate deep well plate from the Run Master Mix Addition screen that matches the corresponding sample preparation

extraction. Initiate the Abbott *m2000sp* Master Mix Addition protocol. Follow the instructions as described in the Abbott *m2000sp* Operations Manual, Operating Instructions section. **The *m2000rt* protocol must be started within 50 minutes of the initiation of the Master Mix Addition protocol.**

**NOTE: If the run is aborted for any reason subsequent to Step 21, a new 96-well PCR plate must be used if the Abbott *m2000sp* Master Mix Addition Protocol (Step 21) will be repeated.**

22. Switch on and initialize the Abbott *m2000rt* instrument in the Amplification Area.

**NOTE: The Abbott *m2000rt* requires 15 minutes to warm-up.**

**NOTE: Remove gloves before returning to the sample preparation area.**

23. Seal the Abbott 96-Well Optical Reaction Plate after the Abbott *m2000sp* instrument has completed addition of samples and master mix according to the Abbott *m2000sp* Operations Manual, Operating Instructions section. Export completed PCR plate results to a CD.

24. Place the sealed optical reaction plate into the Splash-Free Support Base for transfer to the Abbott *m2000rt* instrument.

25. Place the Abbott 96-Well Optical Reaction Plate in the Abbott *m2000rt* instrument. Import *m2000sp* test order via CD per the Import Order instructions in the Abbott *m2000rt* Operations Manual, Operating Instructions section.

**NOTE: If unable to transfer the *m2000sp* test order, enter sample IDs manually in the *m2000rt* in the correct PCR tray locations according to the “Wells for Selected Plate” grid, found on a detail screen under “PCR Plate Results” on the *m2000sp*. See the *m2000sp* Operations Manual, Operating Instructions section.**

## POST PROCESSING PROCEDURES

1. Remove the Abbott 96 Deep-Well Plate from the worktable and dispose of according to the Abbott *m2000sp* Operations Manual.
2. Place the Abbott 96-Well Optical Reaction Plate in a sealable plastic bag and dispose according to the Abbott *m2000rt* Operations Manual along with the gloves used to handle the plate.
3. Clean the Splash Free Support Base before next use, according to the Abbott *m2000rt* Operations Manual.

## QUALITY CONTROL PROCEDURES

### Abbott *m2000rt* Optical Calibration

Refer to the Calibration Procedures section in the Abbott *m2000rt* Operations Manual for a detailed description of how to perform an Abbott *m2000rt* Optical Calibration.

Optical calibration of the Abbott *m2000rt* instrument is required for the accurate measurement and discrimination of dye fluorescence during the Abbott RealTime HIV-1 assay.

The following Abbott *m2000rt* Optical Calibration Plates are used to calibrate the Abbott *m2000rt* instrument for the Abbott RealTime HIV-1 assay:

- FAM™ Plate (Carboxyfluorescein)
- ROX™ Plate (Carboxy-X-rhodamine)
- VIC® Plate (Proprietary dye)

### Assay Calibration

For a detailed description of how to perform an Assay Calibration refer to the Abbott *m2000sp* and *m2000rt* Operations Manuals, Operating Instructions sections.

A calibration curve is required to quantitate the HIV-1 RNA concentration of specimens and controls. Two assay calibrators are run in replicates of 3 to generate a calibration curve (HIV-1 concentration versus the threshold cycle [ $C_T$ ] at which a reactive level of fluorescent signal is detected). The calibration curve slope and intercept are calculated and stored on the instrument. The concentration of HIV-1 RNA in a sample is calculated from the stored calibration curve. Results are automatically reported on the *m2000rt* workstation.

Follow the procedure for sample extraction, master mix addition, amplification and detection protocols as stated in the Abbott *m2000sp* Operations Manual and the Abbott *m2000rt* Operations Manual.

Once an Abbott RealTime HIV-1 calibration is accepted and stored, it may be used for 6 months. During this time, all subsequent samples may be tested without further calibration unless:

- An Abbott RealTime HIV-1 Amplification Reagent Kit with a new lot number is used.

- An Abbott *mSample* Preparation System (4 × 24 Preps) with a new lot number is used.
- An Abbott RealTime HIV-1 application file for a different sample volume is used.
- A new Abbott RealTime HIV-1 application specification file is installed.
- Pure Dye optical recalibration of the Abbott RealTime HIV-1 assay-specific dyes (FAM, VIC, or ROX) is performed per the Calibration Procedures section of the *m2000rt* Operations Manual.

## Detection of Inhibition

An IC threshold cycle [ $C_T$ ] assay validity parameter is established during a calibration run.

A defined, consistent quantity of IC is introduced into each specimen, calibrator, and control at the beginning of sample preparation and detected on the Abbott *m2000rt* instrument to demonstrate proper specimen processing and assay validity. The IC is comprised of an RNA sequence unrelated to the HIV-1 target sequence.

The median amplification cycle at which the IC target sequence fluorescent signal is detected in calibration samples establishes an IC  $C_T$  validity range to be met by all subsequent processed specimens and controls.

An error is displayed when a specimen or control fails to meet this specification. Refer to the *m2000rt* Operations Manual for an explanation of the corrective actions for the error. Specimens whose IC  $C_T$  value exceeds the established range must be retested starting with sample preparation.

## Negative and Positive Controls

A negative control, a low positive control, and a high positive control are included in each test order to evaluate run validity.

The lot specific values for the low positive control and high positive control are specified on each Abbott RealTime HIV-1 Control Kit Card and must be entered into the assay test order when a run is performed.

An error is displayed when a control result is out of range. Refer to the Abbott *m2000rt* Operations Manual for an explanation of the corrective actions for the error. If negative or positive controls are out of range, all of the specimens and controls from that run must be reprocessed, beginning with sample preparation.

The presence of HIV-1 must not be detected in the negative control. HIV-1 detected in the negative control is indicative of contamination by other samples or by amplified product introduced during sample preparation or during preparation of the Abbott 96-Well Optical Reaction Plate. To avoid contamination, clean the Abbott *m2000sp* instrument and the Abbott *m2000rt* instrument and repeat sample processing for controls and specimens following the **Procedural Precautions**. If negative controls are persistently reactive, contact your Abbott representative.

## Monitoring the Laboratory for the Presence of Contamination

It is recommended that this test be done at least once a month to monitor laboratory surfaces and equipment for contamination by amplification product. It is very important to test all areas that may have been exposed to processed specimens, controls, and calibrators, and/or amplification product. This includes routinely handled objects such as pipettes, the Abbott *m2000sp* and Abbott *m2000rt* function keys, laboratory bench surfaces, microcentrifuges, and centrifuge adaptors.

1. Add 0.8 mL RNase-free water to a 1.7 mL RNase-free microcentrifuge tube.
2. Saturate the cotton tip of an applicator (Puritan or equivalent) in the RNase-free water from the microcentrifuge tube.
3. Using the saturated cotton tip of the applicator, wipe the area to be monitored using a sweeping motion. Place the applicator into the microcentrifuge tube.
4. Swirl the cotton tip in RNase-free water 10 times, and then press the applicator along the inside of the tube so that the liquid drains back into the solution at the bottom of the microcentrifuge tube. Discard the applicator.
5. Pipette 0.5 mL of *mWash* 1 buffer to a clean tube using the pipette dedicated for Internal Control use.
6. Add 20  $\mu$ L of the *mWash* 1 buffer to each microcentrifuge tube.
7. Cap the microcentrifuge tube.
8. Test the samples according to the assay procedure section of this package insert.

- Transfer liquid from the microcentrifuge tube to a 5 mL Reaction Vessel.
  - Bring the volume to a minimum of 1.5 mL with RNase-free water.
9. The presence of contamination is indicated by the detection of HIV-1 nucleic acid in the swab samples.
10. If HIV-1 nucleic acid is detected on equipment, follow the cleaning and decontaminating guidelines given in that equipment's operations manual. If HIV-1 nucleic acid is detected on surfaces, clean the contaminated areas with 1.0% (v/v) sodium hypochlorite solution, followed by 70% ethanol or water.

**NOTE: Chlorine solutions may pit equipment and metal. Use sufficient amounts or repeated applications of 70% ethanol or water until chlorine residue is no longer visible.**

11. Repeat testing of the contaminated area by following Steps 1 through 10.

## RESULTS

### Calculation

The concentration of viral HIV-1 RNA in a sample or control is calculated from the stored calibration curve. The Abbott *m2000rt* instrument automatically reports the results on the Abbott *m2000rt* workstation. Assay results can be reported in Copies/mL, Log [Copies/mL], International Units (IU)/mL, or Log [IU/mL]; (1 IU = 0.58 copies, 1 copy = 1.74 IU), with WHO 1st International Standard for HIV-1 RNA (97/656).

### INTERPRETATION OF RESULTS

Sample Volume	Result	Interpretation
1.0 mL	Not Detected	Target not detected
	< 1.60 Log [Copies/mL] <sup>a</sup>	Detected
	1.60 to 7.00 Log [Copies/mL]	
	> 7.00 Log [Copies/mL]	> ULQ <sup>d</sup>
0.6 mL	Not Detected	Target not detected
	< 1.60 Log [Copies/mL] <sup>a</sup>	Detected
	1.60 to 7.00 Log [Copies/mL]	
	> 7.00 Log [Copies/mL]	> ULQ
0.5 mL	Not Detected	Target not detected
	< 1.88 Log [Copies/mL] <sup>b</sup>	Detected
	1.88 to 7.00 Log [Copies/mL]	
	> 7.00 Log [Copies/mL]	> ULQ
0.2 mL	Not Detected	Target not detected
	< 2.18 Log [Copies/mL] <sup>c</sup>	Detected
	2.18 to 7.00 Log [Copies/mL]	
	> 7.00 Log [Copies/mL]	> ULQ

<sup>a</sup> 40 Copies/mL

<sup>b</sup> 75 Copies/mL

<sup>c</sup> 150 Copies/mL

<sup>d</sup> ULQ = upper limit of quantitation

- A result of "Not Detected" signifies that no target was detected.
- A result of "<1.60, <1.60, <1.88, <2.18 Log [copies/mL]" indicates that target was detected but is less than the lower limit of quantitation (LLQ) for the respective input volumes of 1.0, 0.6, 0.5, and 0.2 mL.
- For 1.0 mL and 0.6 mL input volumes, a result of "1.60 to 7.00 Log [copies/mL]" indicates that the target was detected and the concentration falls between 1.6 log copies per mL (LLQ) and 7.0 log copies per mL (ULQ). For a 0.5 mL input volume, a result of "1.88 to 7.00 Log [copies/mL]" indicates that the target was detected and the concentration falls between 1.88 log copies per mL (LLQ) and 7.0 log copies per mL (ULQ). For a 0.2 mL input volume, a result of "2.18 to 7.00 Log [copies/mL]" indicates that the target was detected and the concentration falls between 2.18 log copies per mL (LLQ) and 7.0 log copies per mL (ULQ). Note that no interpretation is reported on the *m2000rt* printout when results fall between LLQ and ULQ.
- A result of ">7.00 Log [copies/mL]" indicates that the target was detected and is greater than ULQ.

### LIMITATIONS OF THE PROCEDURE

#### FOR IN VITRO DIAGNOSTIC USE

- Optimal performance of this test requires appropriate specimen collection, handling, preparation, and storage (refer to the **SPECIMEN COLLECTION, STORAGE, AND TRANSPORT TO THE TEST SITE** section of this package insert).
- Human plasma specimens (collected in ACD-A or EDTA tubes) may be used with the Abbott RealTime HIV-1 assay.

- Use of the Abbott RealTime HIV-1 assay is limited to personnel who have been trained in the procedures of a molecular diagnostic assay and the Abbott *m2000sp* and the Abbott *m2000rt* instruments.
- The instruments and assay procedures reduce the risk of contamination by amplification product. However, nucleic acid contamination from the calibrators, positive controls, or specimens must be controlled by good laboratory practice and careful adherence to the procedures specified in this package insert.
- A specimen with a result of "Target not detected" cannot be presumed to be negative for HIV-1 RNA.
- As with any diagnostic test, results from the Abbott RealTime HIV-1 assay should be interpreted in conjunction with other clinical and laboratory findings.

### SPECIFIC PERFORMANCE CHARACTERISTICS

The performance characteristics were determined using the RealTime HIV-1 assay with the Abbott *m2000* system and 1.0 mL sample volume, unless otherwise specified.

#### Limit of Detection (LOD)

The limit of detection is defined as the HIV-1 RNA concentration detected with a probability of 95% or greater.

#### Limit of Detection, 1.0 mL Sample Volume

The LOD claim for the Abbott RealTime HIV-1 assay is 40 copies/mL with the 1.0 mL sample volume procedure.

The LOD was determined by testing dilutions of a viral standard from the Virology Quality Assurance (VQA) Laboratory of the AIDS Clinical Trial Group. Dilutions were made in HIV-1 negative human plasma. Testing was performed with 3 lots of amplification reagents on 3 *m2000* Systems. The results, representative of the analytical sensitivity performance of the RealTime HIV-1 assay, are summarized in Table 1.

**Table 1**

Conc. (copies/mL)	Number Tested	Number Detected	Percent Detected
100	57	57	100
75	57	57	100
60	57	57	100
50	57	57	100
40	57	57	100
30	57	55	96
20	57	50	88
10	56 <sup>a</sup>	38	68
5	57	30	53

<sup>a</sup> One replicate generated an invalid replicate error message and was deleted from the data analysis.

Probit analysis of the data determined that the concentration of HIV-1 RNA detected with 95% probability was 25 copies/mL (95% CI 20–33).

#### Limit of Detection, 0.6 mL Sample Volume

The LOD claim for the Abbott RealTime HIV-1 assay is 40 copies/mL with the 0.6 mL sample volume procedure.

The LOD for the 0.6 mL sample volume procedure was determined as described for the 1.0 mL sample volume procedure. The results, representative of the analytical sensitivity performance of the RealTime HIV-1 assay, are summarized in Table 2.

**Table 2**

Conc. (copies/mL)	Number Tested	Number Detected	Percent Detected
100	57	57	100
75	57	56	98
60	57	57	100
50	57	54	95
40	57	54	95
30	57	55	96
20	57	44	77
10	57	27	47
5	57	13	23

Probit analysis of the data determined that the concentration of HIV-1 RNA detected with 95% probability was 39 copies/mL (95% CI 33–49).

### Limit of Detection, 0.5 mL Sample Volume

The LOD claim for the Abbott RealTime HIV-1 assay is 75 copies/mL with the 0.5 mL sample volume procedure.

The LOD for the 0.5 mL sample volume procedure was determined as described for the 1.0 mL sample volume procedure. The results, representative of the analytical sensitivity performance of the RealTime HIV-1 assay, are summarized in Table 3.

**Table 3**

Conc. (copies/mL)	Number Tested	Number Detected	Percent Detected
100	57	57	100
75	57	57	100
60	57	54	95
50	56 <sup>a</sup>	52	93
40	57	47	82
30	57	46	81
20	57	42	74
10	57	26	46
5	57	21	37

<sup>a</sup> One replicate generated an invalid replicate error message and was deleted from the data analysis.

Probit analysis of the data determined that the concentration of HIV-1 RNA detected with 95% probability was 65 copies/mL (95% CI 51–88).

### Limit of Detection, 0.2 mL Sample Volume

The LOD claim for the Abbott RealTime HIV-1 assay is 150 copies/mL with the 0.2 mL sample volume procedure.

The LOD for the 0.2 mL sample volume procedure was determined as described for the 1.0 mL sample volume procedure. The results, representative of the analytical sensitivity performance of the RealTime HIV-1 assay, are summarized in Table 4.

**Table 4**

Conc. (copies/mL)	Number Tested	Number Detected	Percent Detected
250	57	57	100
200	57	56	98
150	57	56	98
100	57	54	95
75	57	47	82
60	57	38	67
50	57	39	68
40	54 <sup>a</sup>	30	56
30	52 <sup>a</sup>	19	37

<sup>a</sup> Eight replicates were invalid due to an instrument error and were deleted from the data analysis.

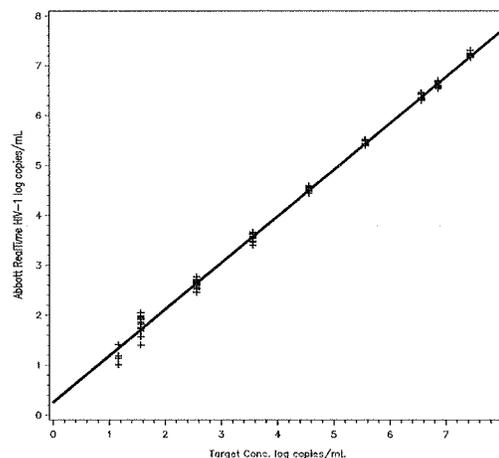
Probit analysis of the data determined that the concentration of HIV-1 RNA detected with 95% probability was 119 copies/mL (95% CI 102–150).

### Linear Range

The upper limit of quantitation (ULQ) for the Abbott RealTime HIV-1 assay is 10 million copies/mL, and the lower limit of quantitation is equivalent to the LOD (40 copies/mL for the 1.0 mL and 0.6 mL sample volume procedure, 75 copies/mL for the 0.5 mL sample volume procedure, and 150 copies/mL for the 0.2 mL sample volume procedure).

A 9-member panel prepared by diluting armored HIV-1 RNA from 7.44 log copies/mL to 1.16 log copies/mL in HIV-1 negative human plasma was tested. Linearity analysis was performed following the NCCLS EP6-A<sup>38</sup> guideline. The results, representative of the RealTime HIV-1 assay linearity, are shown in Figure 1.

**Figure 1**



The RealTime HIV-1 assay was shown to be linear across the range tested (n=99, r=0.999, slope=0.93, and intercept=0.26).

### Precision

The RealTime HIV-1 assay was designed to achieve an inter-assay standard deviation (SD) of less than or equal to 0.25 log copies/mL in samples that contain HIV-1 RNA concentrations between 5,000,000 to 500 copies/mL. Assay precision was demonstrated by testing a coded 45-member precision panel that consisted of 9 unique members repeated 5 times within the panel. The panel was prepared by diluting an HIV-1 viral stock in HIV-1 negative human plasma. The mean RNA concentrations of the panel members ranged from 6.51 to 1.46 log copies/mL. Testing was conducted using the CLSI EP10-A2 guideline.<sup>39</sup> A total of 3 reagent lots were used. Each of the 3 external sites tested 2 of the lots for 3 days for a total of 18 runs. A total of 90 replicates was tested for each panel member. The results of a variance component analysis are in Table 5.

**Table 5**  
Overall Precision Analysis

Panel	n <sup>c</sup>	Mean Conc. (log copies/mL)	Within-Run Component SD	Between-Run Component SD	Inter-Assay SD <sup>d</sup>	Between-Lot Component SD	Between-Site Component SD	Total SD <sup>b</sup>
1	89	6.51	0.06	0.05	0.08	0.00	0.14	0.16
2	86	5.83	0.06	0.03	0.06	0.02	0.11	0.13
3	87	5.21	0.05	0.03	0.06	0.04	0.11	0.13
4	87	4.58	0.06	0.03	0.06	0.06	0.07	0.11
5	88	3.96	0.06	0.00	0.06	0.07	0.03	0.09
6	87	3.38	0.06	0.01	0.06	0.04	0.05	0.09
7	88	2.77	0.07	0.02	0.08	0.06	0.06	0.11
8	89	2.13	0.13	0.04	0.13	0.10	0.07	0.19
9	86 <sup>d</sup>	1.46	0.24	0.00	0.24	0.18	0.00	0.30

<sup>a</sup> Includes within-run and between-run components

<sup>b</sup> Includes within-run, between-run, between-lot, and between-site components

<sup>c</sup> Valid test results

<sup>d</sup> One replicate reported as "Target not detected"

### Potentially Interfering Substances

The susceptibility of the Abbott RealTime HIV-1 assay to interference by elevated levels of endogenous substances and by drugs commonly prescribed to HIV-1 infected individuals was evaluated. HIV-1 negative samples and samples containing 10,000 copies/mL of HIV-1 RNA were tested.

No interference in the performance of the Abbott RealTime HIV-1 assay was observed in the presence of the following substances for all positive and negative samples tested:

- Hemoglobin 500 mg/dL
- Triglycerides 3000 mg/dL
- Bilirubin 20 mg/dL
- Protein 9 g/dL

Drugs at concentrations in excess of the peak plasma or serum levels were tested in 5 pools. No interference in the performance of the Abbott RealTime HIV-1 assay was observed in the presence of the following drug pools for all positive and negative samples tested:

Drug Pool	Drugs Tested
1	Zidovudine, Saquinavir, Ritonavir, Clarithromycin, Interferon 2a, Interferon 2b
2	Abacavir sulfate, Amprenavir, Peginterferon 2a, Peginterferon 2b, Ribavirin
3	Tenofovir disoproxil fumarate, Lamivudine, Indinavir sulfate, Ganciclovir, Valganciclovir hydrochloride, Acyclovir
4	Stavudine, Efavirenz, Lopinavir, Enfuvirtide, Ciprofloxacin
5	Zalcitabine, Nevirapine, Nelfinavir, Azithromycin, Valacyclovir

### Specificity

The specificity of the RealTime HIV-1 assay was evaluated at 3 external sites by testing 514 HIV-1 seronegative plasma specimens from volunteer blood donors. The specimens were tested on 3 m2000 instrument systems with 4 lots of amplification reagents.

In this representative study HIV-1 RNA was not detected for all 514 specimens and the RealTime HIV-1 assay specificity was estimated to be 100% (514/514), (95% CI 99.28 – 100%).

The specificity of the assay was further evaluated by testing 70 specimens that had been either obtained from individuals diagnosed or screened for an autoimmune disorder or serologically characterized as positive for the following markers: systemic lupus erythematosus (SLE), anti-nuclear antibodies (ANA), rheumatoid factor (RF), HBsAg, anti-HTLV-I/II, anti-HCV, and anti-HIV-2. HIV-1 RNA was not detected in any of the specimens tested. The results demonstrated that the presence of an autoimmune disorder or serologic markers for autoimmune disease or viral pathogens other than HIV-1 did not affect the Abbott RealTime HIV-1 assay.

### Cross-Reactivity

The following viruses and microorganisms were evaluated for potential cross-reactivity in the RealTime HIV-1 assay. Purified nucleic acid or viral lysate from each organism was added at a targeted concentration of 5.0 log copies/mL into HIV-1 RNA negative samples and samples that contained 10,000 copies/mL HIV-1 RNA.

Human Immunodeficiency virus 2	Vaccinia virus
Human T-lymphotropic virus 1	BK human polyomavirus
Hepatitis C virus	Human papilloma virus 16
Hepatitis B virus	Human papilloma virus 18
Epstein-Barr virus	Neisseria gonorrhoeae
Herpes simplex virus 1	Chlamydia trachomatis
Herpes simplex virus 2	Candida albicans
Cytomegalovirus	Staphylococcus aureus
Human herpesvirus 6B	Staphylococcus epidermidis
Human herpesvirus 8	Mycobacterium gordonae
Varicella-zoster virus	Mycobacterium smegmatis

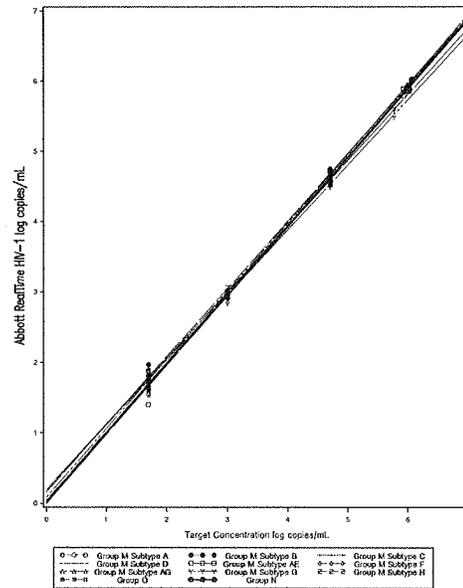
No interference in the performance of the Abbott RealTime HIV-1 assay was observed in the presence of the potential cross-reactants for all positive and negative samples tested.

### Detection of HIV-1 Subtypes and Groups

The performance of the RealTime HIV-1 assay with HIV-1 subtypes/groups was evaluated by analysis of purified RNA transcripts from Group M (subtypes A, B, C, D, CRF01-AE, F, CRF02-AG, G, and H), Group O, and Group N, and by testing 10 clinical specimens of each Group M subtype (A, B, C, D, CRF01-AE, F, CRF02-AG, G), and 10 specimens from Group O.

RNA transcripts of Group M (subtypes A, B, C, D, CRF01-AE, F, CRF02-AG, G, and H), Group O, and Group N with concentrations targeted to approximately 6.0 log copies/mL, 4.7 log copies/mL, 3.0 log copies/mL, and 1.7 log copies/mL were tested. Three replicates were tested at each concentration for each transcript. The results, representative of the dilution linearity for the 11 subtypes/groups tested, are shown in Figure 2.

Figure 2



The results showed that all subtypes and groups tested were detected, and dilution linearity was demonstrated for all groups and subtypes tested (correlation coefficients ranged from 0.997 to 1.000).

A total of 90 clinical specimens, 10 of each Group M subtype (A, B, C, D, CRF01-AE, F, CRF02-AG, G) and Group O, were tested with the RealTime HIV-1 assay and by 2 other approved HIV-1 quantitative assays referred to as Comparator 1 (FDA-approved version used) and Comparator 2 (CE-marked version used). The results are summarized in Table 6.

Table 6

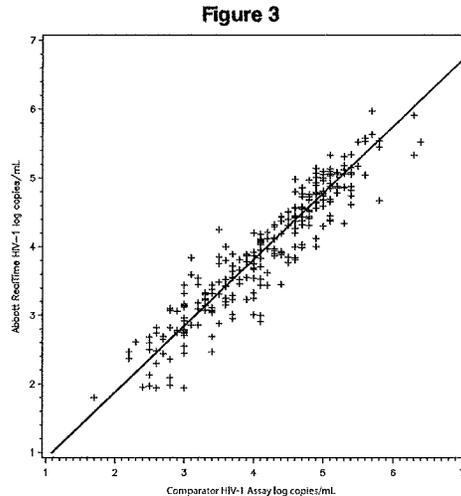
Group/Subtypes	n	RealTime Detected	Comparator 1 Detected <sup>a</sup>	Comparator 2 Detected <sup>a</sup>
M/Subtype A	10	10	10 (1)	10 (1)
M/Subtype B	10	10	10 (0)	10 (0)
M/Subtype C	10	10	10 (0)	10 (0)
M/Subtype D	10	10	10 (0)	10 (0)
M/Subtype AE	10	10	10 (0)	10 (0)
M/Subtype F	10	10	10 (0)	10 (0)
M/Subtype AG	10	10	10 (3)	10 (1)
M/Subtype G	10	10	10 (2)	10 (1)
Group O	10	10	0 (NA)	7 (7)

<sup>a</sup> The numbers in parentheses are the number of specimens that had lower quantitation values by more than 1.00 log copies/mL when compared to RealTime HIV-1 assay.

- The RealTime HIV-1 assay detected all subtypes and groups tested (quantitation range was 2.56 to 6.14 log copies/mL).
- Comparator 1 detected all Group M subtypes tested and did not detect the 10 Group O samples (quantitation range of those detected was 2.01 to 5.54 log copies/mL, and 3 samples were above the upper limit of quantitation [ULQ]).
- Comparator 2 detected all Group M subtypes tested and 7 out of 10 Group O samples (quantitation range of those detected was 1.75 to 5.41 log copies/mL).
- There were no samples that had RealTime assay quantitation values lower than Comparator 1 or Comparator 2 values by more than 1.00 log copies/mL.
- There were 6 Group M samples that had lower quantitation values with Comparator 1 by more than 1.00 log/copies/mL when compared to RealTime HIV-1 assay.
- There were 3 Group M samples and 7 Group O samples that had lower quantitation values with Comparator 2 by more than 1.00 log copies/mL when compared to RealTime HIV-1 assay.

## Correlation

HIV-1 RNA quantitation was compared between the Abbott RealTime HIV-1 assay and an FDA-approved comparator HIV-1 RNA quantitative assay. A total of 301 specimens collected from HIV-1 infected patients were tested with the RealTime HIV-1 assay at 3 external sites and with the comparator method at a central laboratory site. The results from a total of 259 specimens that fell within the common assay dynamic range were analyzed by the Passing-Bablok linear regression method (Figure 3).<sup>40</sup> The correlation coefficient was 0.936, the slope was 0.97 (95% CI 0.92–1.01), and the intercept was  $-0.05$  log copies/mL (95% CI  $-0.22$ – $0.14$ ).



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 Abbott RealTime HIV-1 Control Kit (List No. 6L18-80); and  
 Abbott RealTime HIV-1 Calibrator Kit (List No. 6L18-70).



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 March 2012  
 51-602146/R7



# Abbott *RealTime* HIV-1

At Abbott, we pride ourselves on improving global health. We are working on solutions for each of your laboratory needs. Now, with the Abbott RealTime HIV-1 assay you can have confidence that your results will be even more accurate across HIV-1 Subtypes. This means more complete results, engineered for confidence.

## Development Philosophy

Today's clinical molecular diagnostics laboratory must have confidence in the quality of the HIV-1 patient results. The Abbott RealTime HIV-1 is engineered to tolerate the genetic diversity of HIV-1 Subtypes.

### HIV-1 diversity can be attributed to:

- Error-prone reverse transcriptase enzyme
- Recombination of subtypes
- Cross-species transmission

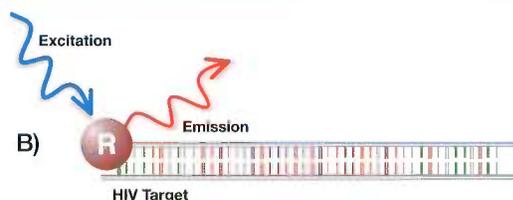
### Accurate quantitation is dependent upon a combination of:

- Primer design
- Probe design
- Cycling conditions

## Partially Double Stranded Probe Design



In the absence of target, the probe hybridizes to the quencher oligonucleotide, preventing fluorescent signal generation.



In the presence of target, the probe prefers to hybridize with the target sequence, disassociating from the quencher oligonucleotide and allowing fluorescent detection.

## Abbott RealTime HIV-1 Performance

<b>Sensitivity</b>	40 copies/mL for 1.0 mL input, 75 copies/mL for 0.5 mL input, 150 copies/mL for 0.2 mL input
<b>Linear Range</b>	40 copies/mL (1.6 log copies/mL) to 10 million copies/mL (7.0 log copies/mL)
<b>Precision</b>	Inter-assay standard deviation (SD) of $\leq 0.25$ log copies/mL
<b>Specificity</b>	100%† †The specificity of the RealTime HIV-1 assay was evaluated at three external sites by testing 514 HIV-1 seronegative plasma specimens from volunteer blood donors. HIV-1 RNA was not detected for all 514 specimens and the RealTime HIV-1 assay specificity was estimated to be 100% (514/514), (95% CI 99.28 to 100%).
<b>Specimen Type</b>	Plasma (ACD-A and EDTA)
<b>HIV-1 Subtype Detection</b>	Group M subtypes A–H, Group O and Group N
<b>Standardization</b>	Virology Quality Assurance (VQA) Laboratory of the AIDS Clinical Trial Group and against World Health Organization (WHO) 1st International Standard for HIV-1 RNA (97/656)
<b>Internal Control</b>	Added to lysis buffer during extraction and detected at all levels

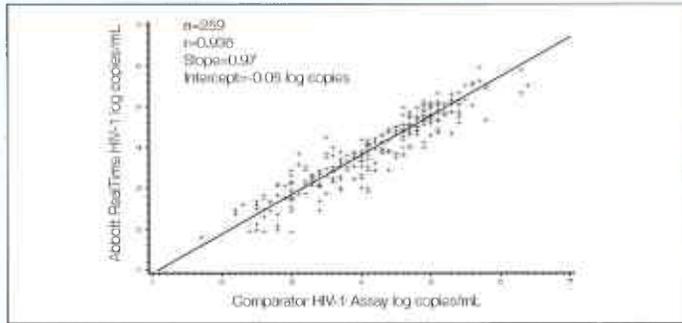
## INTENDED USE

The Abbott RealTime HIV-1 assay is an *in vitro* reverse transcription-polymerase chain reaction (RT-PCR) assay for the quantitation of Human Immunodeficiency Virus type 1 (HIV-1) on the automated *m2000* System in human plasma from HIV-1 infected individuals over the range of 40 to 10,000,000 copies/mL. The Abbott RealTime HIV-1 assay is intended for use in conjunction with clinical presentation and other laboratory markers for disease prognosis and for use as an aid in assessing viral response to antiretroviral treatment as measured by changes in plasma HIV-1 RNA levels. This assay is not intended to be used as a donor screening test for HIV-1 or as a diagnostic test to confirm the presence of HIV-1 infection.

**CAUTION:** United States Federal law restricts this device to sale and distribution by or on the order of a physician or to a clinical laboratory; and use is restricted to by or on the order of a physician.

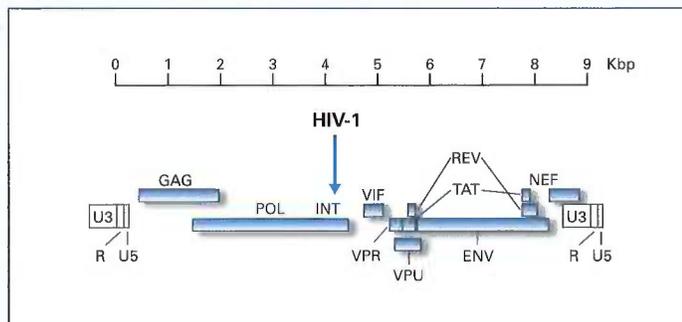
FOR WARNINGS AND LIMITATIONS SEE REVERSE.

## Correlation to Comparator Assay



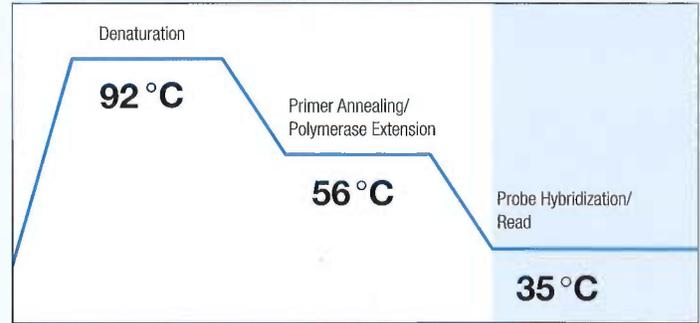
A total of 301 specimens collected from HIV-1 infected patients were tested with the RealTime HIV-1 assay at three external sites and with the comparator method at a central laboratory site. The results from a total of 259 specimens that fell within the common assay dynamic range were analyzed by the Passing-Bablok linear regression method.

## Primers and Probe are Targeted to the Integrase Region of the Polymerase Gene



The integrase region of the polymerase gene is a conserved region of the HIV-1 genome.

## Cycling Conditions: Low Temperature Read Cycles



The cycling conditions for Abbott RealTime HIV-1 encompass a low temperature read cycle. This read cycle allows the probe to tolerate mismatches more effectively than a probe required to bind during the extension phase.

## Detection of HIV-1 Subtypes and Groups

Group/Subtypes	RealTime Detected	Comparator 1 Detected	Comparator 2 Detected
M/Subtype A	10	10 (1)	10 (1)
M/Subtype B	10	10 (0)	10 (0)
M/Subtype C	10	10 (0)	10 (0)
M/Subtype D	10	10 (0)	10 (0)
M/Subtype AE	10	10 (0)	10 (0)
M/Subtype F	10	10 (0)	10 (0)
M/Subtype AG	10	10 (3)	10 (1)
M/Subtype G	10	10 (2)	10 (1)
Group O	10	0 (NA)	7 (7)

A total of 90 clinical specimens, ten of each Group M subtype (A, B, C, D, CRF01-AE, F, CRF02-AG, G) and of Group O, were tested with the RealTime HIV-1 assay and by two other approved HIV-1 quantitative assays referred to as Comparator 1 (FDA-approved version used) and Comparator 2 (CE-marked version used). The numbers in parentheses are the number of specimens that had lower quantitation values by more than 1.00 log copies/mL when compared to RealTime HIV-1 assay.

## Ordering Information

Product	List Number	Configuration
Abbott RealTime HIV-1 Amplification Reagent Kit	6L18-90	96 Assays (4 packs x 24 assays)
Abbott RealTime HIV-1 Control Kit	6L18-80	8 Low Positive, 8 High Positive, 8 Negative
Abbott RealTime HIV-1 Calibrator Kit	6L18-70	12 Cal A, 12 Cal B (4 Complete Calibration Sets)
Abbott RealTime HIV-1 Application CD-ROM	6L83	1 each

For additional information please visit [www.abbottmolecular.com](http://www.abbottmolecular.com) or contact your local Abbott Molecular Sales Representative.

## WARNINGS AND LIMITATIONS

- Human plasma specimens collected in ACD-A or EDTA tubes may be used.
- A specimen with a result of "Target not detected" cannot be presumed to be negative for HIV-1 RNA.
- As with any diagnostic test, results from the Abbott RealTime HIV-1 assay should be interpreted in conjunction with other clinical and laboratory findings.

Abbott RealTime  
A real difference

**Leading Science**

Proven precision provides confidence in results.

**Enabling Solutions**

Abbott mSystem portfolio enables menu consolidation, reduces training needs and decreases costs.

**Trusted Partner**

The commitment from Abbott to more than 25 years of global HIV leadership and innovation to meet the HIV challenges.



**Abbott RealTime HIV-1 ordering information**

Abbott RealTime HIV-1 Amplification Reagent Kit	06L18-090	1 kit (96 tests, 4 x 24 tests/pack)
Abbott RealTime HIV-1 Control Kit	06L18-080	1 kit (3 levels with 8 vials per level)
Abbott RealTime HIV-1 Calibrator Kit	06L18-070	1 kit (2 levels with 12 vials per level)
Abbott RealTime HIV-1 m2000 System ROW Combined Application CD-ROM	06L83	1 each

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Abbott RealTime HIV-1 Assay  
Meet HIV challenges



**Performance you expect.**  
**Precision you need.**  
**Deliver it now.**

**Abbott**  
A Promise for Life

## Abbott RealTime HIV-1: A difference by design Reliably detects all known groups and subtypes.<sup>1</sup>

The prevalence of non-B subtypes and recombinants in the U.S. is steadily increasing.<sup>2,3</sup>

"In a recent study of 24,484 samples from 46 states, non-B strains (4.12%) were present in 37 of them—including subtypes A1, A2, A3, A4, C, D, F1, F2, G, 23 CRFs, and 25 URFs."<sup>2</sup>

RealTime HIV-1 has the same level of detection for all groups and subtypes.



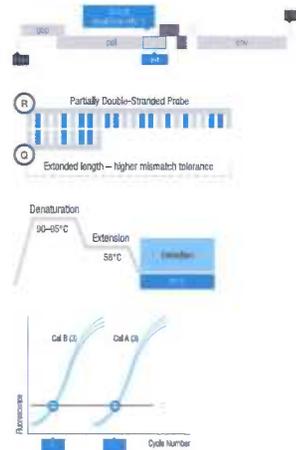
Unique assay design with a high degree of precision.

**Conserved Target Region (Integrase):** Demonstrates the capability to tolerate mutations, with no impact on assay performance.<sup>4</sup>

**Unique Probe Design:** Proprietary probe design allows for accurate quantification across genetically polymorphic targets.<sup>2</sup>

**Optimal Cycling Conditions:** Novel probe design allows for cycling conditions at a lower-read temperature, contributing to mismatch tolerance.

**Precise External Calibration:** Two-point external calibration allows for precise quantitation, eliminating competitive effects in PCR reaction that can occur in internal calibration methods.



Experience the difference with Abbott RealTime HIV-1. Designed for reliable detection of all known HIV-1 groups and subtypes.

1. Group M subtypes A-H, Group N, Group O. (Abbott RealTime HIV-1 Package Insert 51, Abbott RealTime HIV-1 802 146/R6). 2. Hackett. *J. Clin. Lab. Microbiol.* 2012;58:199-202. 3. Plantier et al. *Nature Medicine*, 2009;15:671-72. 4. Young. *J. Clin. Microbiol.* April 2011; 49:4:1631-1634; published ahead of print 2 February 2011 doi:10.1128/JCM.02253-10

**INTENDED USE:** The Abbott RealTime HIV-1 assay is an in vitro reverse transcription-polymerase chain reaction (RT-PCR) assay for the quantitation of human immunodeficiency virus type 1 (HIV-1) on the automated m2000 System in human plasma from HIV-1 infected individuals over the range of 40 to 10,000,000 copies/mL. The Abbott RealTime HIV-1 assay is intended for use in conjunction with clinical presentation and other laboratory markers for disease prognosis and for use as an aid in assessing viral response to antiretroviral treatment as measured by changes in plasma HIV-1 RNA levels. This assay is not intended to be used as a donor screening test for HIV-1 or as a diagnostic test to confirm the presence of HIV-1 infection.

## Abbott RealTime HIV-1: A difference by experience Physiological blip? Or random assay variation?

A detectable low-level viremia following a previous undetectable viral load may be:

- A **true increase** in the viral load level, such as the first step toward the development of resistance
- A **physiological blip** caused by, for example, release of virus from a reservoir
- An assay **artifact measurement**, such as random variations (imprecision) around clinical decision points

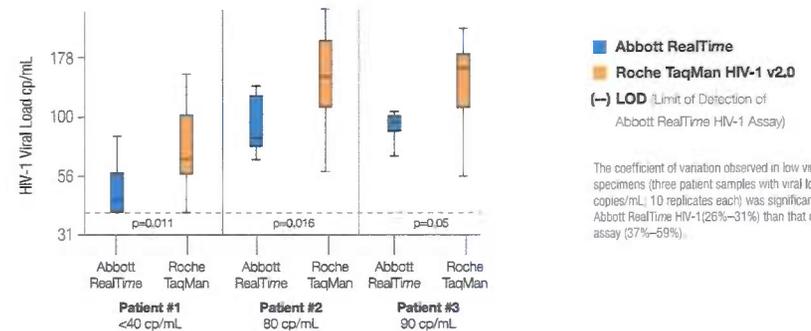
Choose the assay designed to minimize the risk of reporting low-level viremia due to random assay variation.

**According to DHHS guidelines** "...low-level positive viral load results (typically <200 copies/mL) appear to be more common with some viral load assays than others...The AIDS Clinical Trials Group (ACTG) currently defines virologic failure as a confirmed viral load >200 copies/mL, which eliminates most cases of apparent viremia caused by blips or assay variability."<sup>5</sup>

## Abbott RealTime HIV-1: Delivers a difference in precision.

Abbott RealTime HIV-1 provides precise performance when measuring low-level viremia.

Comparison of HIV-1 quantification in low-level viremia



Adapted from Naeth et al., 2012 *Med Microbiol Immunol*, 2012 Jun 15.

Experience the difference with Abbott RealTime HIV-1. Delivering precise performance when you and your customers count on it most.

5. Panel on Antiretroviral Guidelines for Adults and Adolescents. Guidelines for the use of antiretroviral agents in HIV-1-infected adults and adolescents. Department of Health and Human Services. Available at <http://aidsinfo.nih.gov/guidelines>. Accessed December 2012.

**LIMITATIONS:** 1. Human plasma specimens collected in ACD-A or EDTA tubes may be used. 2. A specimen with a result of "target not detected" cannot be presumed to be negative for HIV-1 RNA. 3. As with any diagnostic test, results from the Abbott RealTime HIV-1 assay should be interpreted in conjunction with other clinical and laboratory findings. **CAUTION:** United States federal law restricts this device to sale and distribution to or on the order of a physician or a clinical laboratory, and use is restricted to, by, or on the order of a physician.

**For In Vitro Diagnostic Use**



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- Reduction in troubleshooting time for your staff
- Faster time to resolution of instrument issues
- Identification of training opportunities for decreased user error
- Accurate information exchange through electronic system data downloads

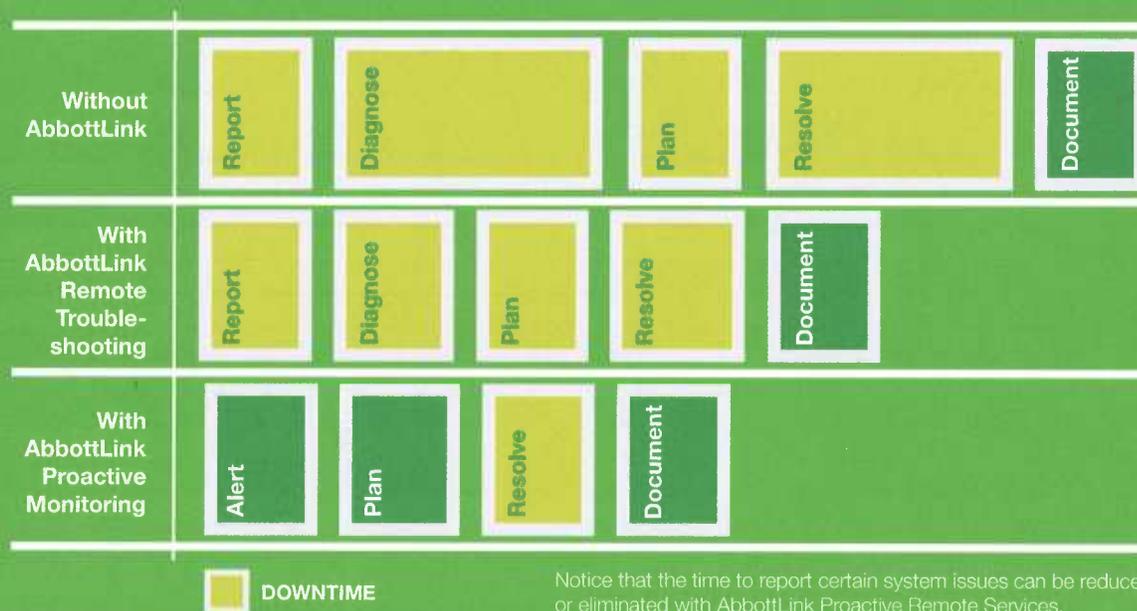
All Abbott Molecular *m2000* warranty and service plans include AbbottLink connectivity and reactive remote troubleshooting service.

Alert Abbott Support experts to critical changes to reduce downtime.

## AbbottLink **Proactive** Remote Troubleshooting Services

AbbottLink can be configured to monitor specific events and alarms that if left unchecked may potentially cause instrument downtime. Critical operational information is auto transmitted over a secure internet connection from your Abbott *m2000* instruments to our customer service team, once a predetermined threshold is reached.

The proactive support delivered through AbbottLink can reduce downtime and customer hands-on time.



AbbottLink Proactive Remote Services provides:

- All of the benefits of Reactive Remote Services, plus the following:
  - Abbott Customer Support can be auto-alerted to the instrument issue, once occurrences reach a certain threshold.
  - Less interference with your laboratory, as the technician spends less valuable time on the phone with Abbott Support
  - The proactive on-site visit or phone assistance can help prevent “down” status and the need for urgent care.

Remote Proactive Services is an available option built for labs with heightened sensitivity to downtime.



**Network security for you. Privacy for your patients.**

AbbottLink is a remote service solution that makes security the highest priority.

AbbottLink only transmits instrument operational data and usage count information, which is stored in a secure data center to be used for troubleshooting. AbbottLink does not calculate, adjust, or report patient results. AbbottLink allows the remote diagnosis of instrument alerts in a highly secured environment while providing necessary instrument information to Abbott experts.

**AbbottLink data security highlights:**

- Firewall-friendly communication
- No change to customer IT infrastructure
- Follow customer's current security policy to obtain internet access on port 443
- Data encrypted with 128 bit SSL encryption (standard technology used in internet banking)
- No PHI data leaves the customer site

AbbottLink is installed in several continents around the world. The wide spread acceptance achieved is due to the security principles that have been designed and implemented into the AbbottLink solution. With AbbottLink, labs are able to obtain the benefits of remote service in a secure manner.

For more information on AbbottLink please go to [www.abbottmolecular.com](http://www.abbottmolecular.com) or contact your local sales representative.

1-800-553-7042, option #2

