VITROS Immunodiagnostic Products

Intended for Use in the United States

# INSTRUCTIONS FOR USE aHIV

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| VITROS Immunodiagnostic Products Anti-HIV 1+2 Reagent Pack | REF | 680 1861 |
| VITROS Immunodiagnostic Products Anti-HIV 1+2 Calibrator | REF | 680 1862 |

Rx ONLY

## Intended Use

### VITROS Immunodiagnostic Products Anti-HIV 1+2 Reagent Pack

For the *in vitro* qualitative detection of antibodies to Human Immunodeficiency Virus types 1 and/or 2 (anti-HIV-1 and anti-HIV-2) in human serum and plasma (heparin, EDTA or citrate) using the VITROS ECi/ECiQ/3600 Immunodiagnostic Systems and the VITROS 5600/XT 7600 Integrated Systems.

The results of the VITROS Anti-HIV 1+2 assay, in conjunction with other serological evidence and clinical information, may be used as an aid in the diagnosis of infection with HIV-1 and/or HIV-2 in persons with signs or symptoms of, or at risk for, HIV infection.

**WARNING:** This assay has not been FDA cleared, licensed or approved for the screening of blood or plasma donors.

### VITROS Immunodiagnostic Products Anti-HIV 1+2 Calibrator

For *in vitro* use in the calibration of the VITROS ECi/ECiQ/3600 Immunodiagnostic Systems and the VITROS 5600/XT 7600 Integrated Systems for the qualitative detection of antibodies to Human Immunodeficiency Virus types 1 and/or 2 (anti-HIV-1 and anti-HIV-2) in human serum and plasma (heparin, EDTA or citrate).

## Summary and Explanation of the Test

Acquired Immunodeficiency Syndrome (AIDS) is caused by at least two types of Human Immunodeficiency Viruses designated HIV-1 and HIV-2. In addition, some HIV-1 strains have been isolated from AIDS patients in West Africa and designated as HIV-1 subtype O. Serological studies have shown that antibodies may develop to epitopes present in the peptides of the viral core and glycoprotein envelope. While antibodies to HIV-1 and HIV-2 core peptides demonstrate considerable cross reactivity, the antibodies generated by the glycoprotein envelope show less cross-reactivity.

The VITROS Anti-HIV 1+2 test uses 4 recombinant antigens (HIV-1 Env 13, HIV-1 Env 10, HIV-1 p24, and HIV-2 Env AL) derived from HIV-1 core, HIV-1 envelope and HIV-2 envelope. HIV-1 Env 13, envelope SOD fusion protein, contains regions from both gp 120 and gp 41 regions. HIV-1 Env 10, envelope SOD fusion protein, contains a gp 41 region which extends beyond the C-terminus of Env 13. HIV-1 p24 is derived from full length core protein of HIV-1. HIV-2 Env AL, envelope SOD fusion protein, contains a region from gp 36 of HIV-2.

These antigens detect antibodies to HIV-1 and antibodies to HIV-2 in the same test. The use of these recombinant antigens improves test specificity by avoiding non-specific reactions due to cross-reaction with human cell proteins, which are present in cell lysates.

The host organism for all four HIV recombinant antigens is S. cerevisiae (yeast).

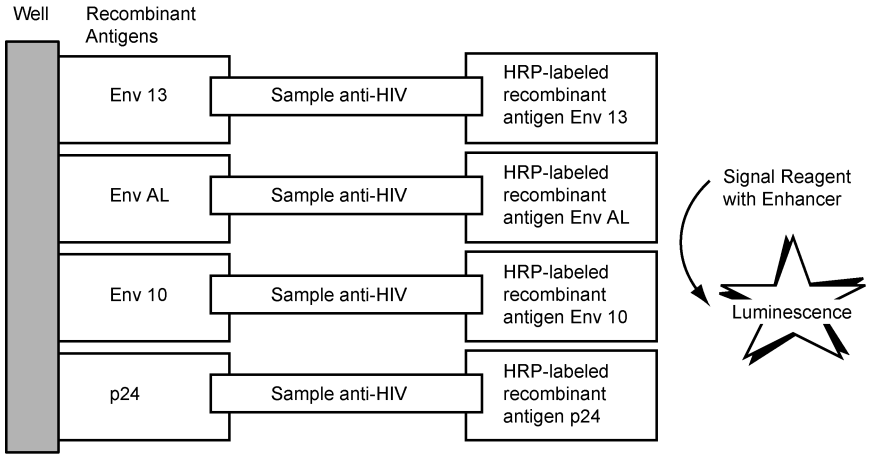
## Principles of the Procedure

An immunometric bridging technique is used; this involves a two-stage reaction. In the first stage HIV antibody present in the sample binds with HIV recombinant antigen coated on the wells. Unbound sample is removed by washing. In the second stage horseradish peroxidase (HRP)-labeled recombinant HIV antigens are added in the conjugate reagent. The conjugate binds specifically to any human anti-HIV-1 or anti-HIV-2 (IgG and IgM) captured on the well in the first stage. Unbound conjugate is removed by washing.

The bound HRP conjugate is measured by a luminescent reaction. [1] A reagent containing luminogenic substrates (a luminol derivative and a peracid salt) and an electron transfer agent is added to the wells. The HRP in the bound conjugate catalyzes the oxidation of the luminol derivative, producing light. The electron transfer agent (a substituted acetanilide) increases the level of light produced and prolongs its emission. The light signals are read by the system. The amount of HRP conjugate bound is indicative of the level of anti-HIV-1 and anti-HIV-2 present.

| Test Type | System | Incubation Time | Time to first result | Test Temperature | Reaction Sample Volume |
|---|---|---|---|---|---|
| Immunometric | ECi/ECiQ, 3600, 5600, XT 7600 | 37 minutes | 48 minutes | 37 °C | 80 µL |

***Reaction Scheme***




## Warnings and Precautions

For *in vitro* diagnostic use only.

**WARNING:** Potentially Infectious Material

Treat as if capable of transmitting infection.

Handling of samples and test components, their use, storage, and solid and liquid waste disposal should be in accordance with the procedures defined by the appropriate national biohazard safety guideline or regulation (e.g. CLSI document M29). [2, 3]

The VITROS Anti-HIV 1+2 Calibrator contains:

HIV antibody positive plasma obtained from donors who were tested individually and found to be negative for hepatitis B surface antigen (HBsAg), and for antibodies to hepatitis C virus (HCV), using FDA approved methods (enzyme immunoassays, EIA). The HIV antibody positive plasma has been heat-treated to inactivate viruses. However, as no testing method can rule out the risk of potential infection, handle as if capable of transmitting infection.

HIV antibody negative plasma obtained from donors who were tested individually and found to be negative for hepatitis B surface antigen (HBsAg), and for antibodies to hepatitis C virus (HCV) and HIV, using FDA approved methods (enzyme immunoassays, EIA).

Care should be taken when handling material of human origin. All samples should be considered potentially infectious. No test method can offer complete assurance that hepatitis B virus, HCV, HIV 1+2 or other infectious agents are absent.

**WARNING:** Contains Kathon or ProClin 200 (CAS 55965-84-9) [4]

The VITROS Anti-HIV 1+2 Reagent Pack and VITROS Anti-HIV 1+2 Calibrators contain 1.0% and 2% Kathon or ProClin 200. H317: May cause an allergic skin reaction. P280: Wear protective gloves/protective clothing/eye protection/face protection. P302 + P352: IF ON SKIN: Wash with plenty of soap and water. P333 + P313: If skin irritation or rash occurs: Get medical advice/attention. P363: Wash contaminated clothing before reuse.

Refer to www.Orthoclinicaldiagnostics.com for the Safety Data Sheets and for Ortho contact information.

WARNING

# Reagents

## Reagent Pack Contents

1 reagent pack containing:

- 100 coated wells (Human Immunodeficiency Virus Recombinant Antigens Env 13, Env AL, Env 10, p24 derived from yeast [S. cerevisiae]; coated at 0.36 µg/well)
- 6.2 mL Assay Reagent (buffer with anti-microbial agent)
- 13.3 mL Conjugate Reagent (HRP labeled HIV –1 [Env 13, Env 10, p24] and HIV- 2 [Env AL] recombinant antigens [1 to 3 µg/well] in buffered fetal calf serum with anti-microbial agent)

## Reagent Pack Handling

- The reagent pack is supplied ready for use.
- The reagent pack contains homogeneous liquid reagents that do not require shaking or mixing prior to loading onto the system.
- Handle the reagent pack with care. Avoid the following:
  - allowing condensation to form on the pack
  - causing reagents to foam
  - agitation of the pack

## Reagent Pack Storage and Preparation

| Reagent | Storage Condition | | Stability |
|---|---|---|---|
| Unopened | Refrigerated | 2–8 °C (36–46 °F) | expiration date |
| Opened | On system | System turned on | ≤8 weeks |
| Opened | Refrigerated | 2–8 °C (36–46 °F) | ≤8 weeks |

- The VITROS Anti-HIV 1+2 Reagent Pack is suitable for use until the expiration date on the carton when stored and handled as specified. Do not use beyond the expiration date.
- Do not freeze unopened reagent packs.
- Load reagent packs directly from refrigerated storage to minimize condensation.
- Store opened refrigerated reagent packs in a sealed reagent pack storage box that contains dry desiccant.

## Calibrator Contents

- 1 VITROS Anti-HIV 1+2 calibrator (2 mL, heat-inactivated anti-HIV 1+2 positive human plasma in anti-HIV 1+2 negative human plasma with antimicrobial agent)
- Lot calibration card
- Protocol card - Major Protocol Version 2
- 8 calibrator bar code labels

## Calibrator Handling

- Use only with reagent packs of the same lot number. Mix thoroughly by inversion and bring to 15–30 °C (59–86 °F) before use. Each pack contains sufficient volume for a minimum of 6 calibration events of each calibrator.
- Handle calibrators in stoppered containers to avoid contamination and evaporation. To avoid evaporation, limit the amount of time calibrators are on the system. Refer to the operating instructions for your system. Return to 2–8 °C (36–46 °F) as soon as possible after use, or load only sufficient volume for a single determination.

## Calibrator Storage and Preparation

| Calibrator | Storage Condition | | Stability |
|---|---|---|---|
| Unopened | Refrigerated | 2–8 °C (36–46 °F) | expiration date |
| Opened | Refrigerated | 2–8 °C (36–46 °F) | ≤13 weeks |
| Opened | Frozen | ≤-20 °C (≤-4 °F) | ≤13 weeks |

- VITROS Anti-HIV 1+2 Calibrator is supplied ready for use.

- The VITROS Anti-HIV 1+2 Calibrator is suitable for use until the expiration date on the carton when they are stored and handled as specified. Do not use beyond the expiration date.
- Opened calibrators may be stored frozen (with no more than 1 freeze-thaw cycle).
- The VITROS Anti-HIV 1+2 test uses 80 µL of calibrator for each determination. The VITROS Anti-HIV 1+2 Calibrator may be used directly on the VITROS Immunodiagnostic and VITROS Integrated Systems. Alternatively, transfer an aliquot of each calibrator into a sample container (taking account of the minimum fill volume of the container), which may be bar coded with the labels provided. For details on minimum fill volume of sample cups or containers, refer to the operating instructions for your system.
- The VITROS Anti-HIV 1+2 Calibrator is automatically processed in duplicate.

# Specimen Collection, Preparation and Storage

## Patient Preparation

No special patient preparation is necessary.

## Specimens Recommended

- Serum
- Heparin plasma
- EDTA plasma
- Citrate plasma

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| **Note:** | Results from citrated plasma will be proportionately lower due to dilution by the anticoagulant. |

## Specimens Not Recommended

Do not use turbid specimens. Turbidity in specimens may affect test results.

## Special Precautions

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| **IMPORTANT:** | Certain collection devices have been reported to affect other analytes and tests. [5] Owing to the variety of specimen collection devices available, Ortho Clinical Diagnostics is unable to provide a definitive statement on the performance of its products with these devices. Confirm that your collection devices are compatible with this test. |

## Specimen Collection and Preparation

- Collect specimens using standard procedures. [6, 7]
- Samples should be thoroughly separated from all cellular material. Failure to do so may lead to an erroneous result.
- Thoroughly mix samples by inversion and bring to 15–30 °C (59–86 °F) before use.
- The VITROS Anti-HIV 1+2 test uses 80 µL of sample for each determination. This does not take account of the minimum fill volume of the chosen sample container. For details on minimum fill volume of sample cups or containers, refer to the operating instructions for your system.

## Handling and Storage Conditions

- Handle samples in stoppered containers to avoid contamination and evaporation.
- The amount of time samples are on the system prior to analysis should be limited to avoid evaporation. This time should not exceed 3 hours. Refer to the operating instructions for your system.
- Return to 2–8 °C (36–46 °F) as soon as possible after use, or load sufficient volume for a single determination.
- The Clinical and Laboratory Standards Institute (CLSI) provides the following recommendations for storing specimens: [8]
  - Store samples at 22 °C (72 °F) for no longer than 8 hours.
  - If the test will not be completed within 8 hours, refrigerate samples at 2–8 °C (36–46 °F).
  - If the test will not be completed within 48 hours, or for shipment, freeze samples at or below -20 °C (-4 °F).
- Samples are not to be repeatedly frozen and thawed because this can cause analyte deterioration. Samples are to be thawed only once.

# Testing Procedure

## Materials Provided

- VITROS Immunodiagnostic Products Anti-HIV 1+2 Reagent Pack
- VITROS Immunodiagnostic Products Anti-HIV 1+2 Calibrators

### Materials Required but Not Provided

- VITROS Immunodiagnostic Products Signal Reagent
- VITROS Immunodiagnostic Products Universal Wash Reagent
- Quality control materials such as VITROS Immunodiagnostic Products Anti-HIV 1+2 Controls
- VITROS Immunodiagnostic Products Reagent Pack Storage Box (optional) with desiccant

### Operating Instructions

Check the inventory regularly to aid the management of reagents and ensure that sufficient VITROS Signal Reagent, VITROS Universal Wash Reagent and calibrated reagent lots are available for the work planned. When performing panels of tests on a single sample, ensure that the sample volume is sufficient for the tests ordered.

For detailed instructions refer to the operating instructions for your system.

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| **Note:** | Do not use visibly damaged product. |

### Sample Dilution

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| **WARNING:** | Automatic dilution is not available for this test on the VITROS Immunodiagnostic and VITROS Integrated Systems.<br><br>Do not use manually diluted samples. |

### Default Test Name

The default test name which will appear on patient reports is Anti-HIV 1+2. The default short name that will appear on the test selection menus and laboratory reports is aHIV. These defaults may be reconfigured, if required. For detailed information refer to the operating instructions for your system.

## Calibration

### Calibration Procedure

- Calibration is lot specific; Reagent Packs and Calibrators are linked by lot number. Reagent Packs from the same lot may use the same calibration.
- A Master Calibration is established for each new reagent lot by performing multiple tests. This is the process by which a lot-specific parameter [a] which links the signal at the cutoff (cutoff value) to the calibrator signal is determined.

Cutoff value = (a x Signal of Cal 1)

- Ensure that the Master Calibration for each new reagent lot is available on your system.
- Process the Calibrator in the same manner as samples. Load sufficient for the automatic duplicate determination. Calibration need not be programmed if bar code labels are used; Calibration will be initiated automatically.
- When the Calibrator is processed the validity of the calibration is assessed against quality parameters which compares the actual signal of the calibrator with the expected signal. If the calibration is acceptable the cutoff value is calculated and stored for use with any reagent pack of that lot.
- The quality of calibration cannot be completely described by a single parameter. The calibration report should be used in conjunction with acceptable control values to determine the validity of the calibration.
- Recalibration is required after a pre-determined calibration interval, or when a different reagent lot is loaded.
- Calibration results are assessed against a range of quality parameters. Failure to meet any of the defined quality parameter ranges will be coded in the calibration report. For actions to be taken following a failed calibration, refer to the operating instructions for your system.

Refer to the operating instructions for your system for detailed instructions on the calibration process.

### When to Calibrate

- Calibrate when the Reagent Pack and Calibrator lot changes.
- Calibrate every 28 days.
- After specified service procedures have been performed.
- If quality control results are consistently outside of your acceptable range.

For additional information on when to calibrate, refer to the operating instructions for your system.

### Calibration Model

Results are calculated as a normalized signal, relative to a cutoff value. During the calibration process a lot-specific parameter is used to determine a valid stored cutoff value for the VITROS Immunodiagnostic and VITROS Integrated Systems.

# Quality Control

## Quality Control Material Selection

VITROS Anti-HIV 1+2 Controls or similar materials are recommended for use with the VITROS Immunodiagnostic and VITROS Integrated Systems. There are 3 VITROS Anti-HIV 1+2 Controls (negative, anti-HIV 1 positive and anti-HIV 2 positive). The performance of other commercial or non-commercial control fluids should be evaluated for compatibility with this test before they are used for quality control.

Control materials may show a difference when compared with other anti-HIV 1+2 methods if they contain high concentrations of preservatives, stabilizers, or other nonphysiological additives, or otherwise depart from a true human sample matrix.

Appropriate quality control value ranges must be established for all quality control materials used with the VITROS Anti-HIV 1+2 test.

## Quality Control Procedure Recommendations

- Good laboratory practice requires that controls be processed to verify the performance of the test.
- Choose control levels that check the clinically relevant concentrations.
- The recommendation is to run a positive control close to the anti-HIV 1+2 decision point (the cutoff) and a negative control.
- To verify system performance, analyze control materials:
  - After calibration
  - At least once every 24 hours
  - After specified service procedures are performed
  - After testing Research Use Only (RUO) reagents, Investigational Use Only (IUO) reagents or User Defined Assays (UDAs) and prior to testing HIV specimens and reporting HIV results. **NOTE**: Additional Quality Control testing is not required after testing Manufacturers Validated Applications (MVAs) which utilize the UDA channel. MVAs are included on Ortho Clinical Diagnostic menu listings and have been validated by the original reagent manufacturer, who has tested and documented performance parameters.

If quality control procedures within your laboratory require more frequent use of controls, follow those procedures.

- Analyze quality control materials in the same manner as patient specimens.
- If control results fall outside your acceptable range, investigate the cause before deciding whether to report patient results. When the condition is corrected, retest the controls and specimens.
- Refer to Internal Quality Control Testing: Principles and Definitions or other published guidelines for general quality control recommendations. [9]
- Additional controls may be tested according to guidelines or requirements of local, state, and/or federal regulations or accrediting organizations.

For more detailed information, refer to the operating instructions for your system.

## Quality Control Material Preparation and Storage

Refer to the manufacturer's product literature for preparation, storage, and stability information or established preparation, storage and stability specifications for non-commercial material.

# Results

Results are automatically calculated by the VITROS Immunodiagnostic and VITROS Integrated Systems.

## Result Calculation

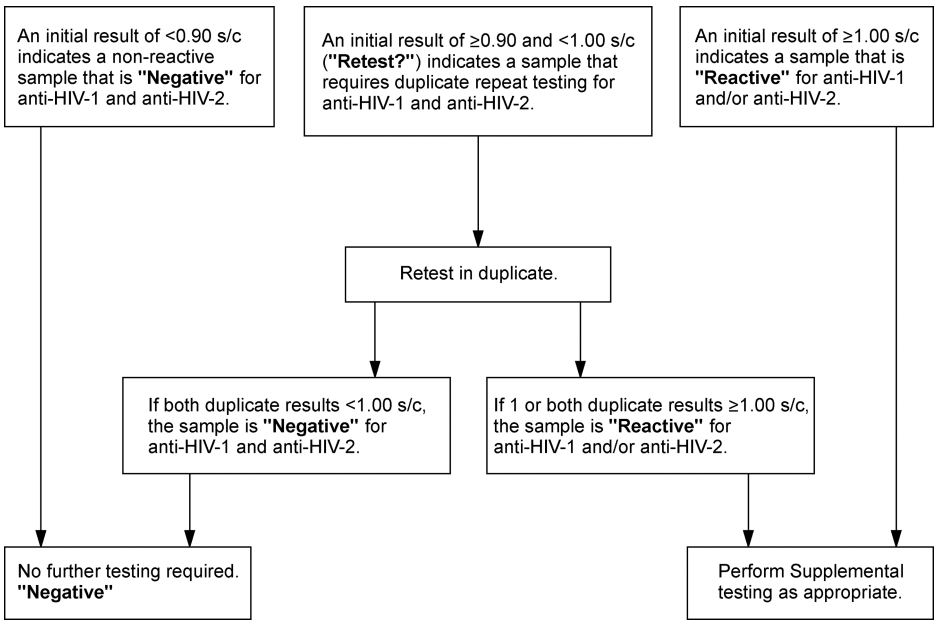
Results are calculated as a normalized signal, relative to a cutoff value. During the calibration process, a lot-specific parameter is used to determine a valid stored cutoff value for the VITROS Immunodiagnostic and VITROS Integrated Systems.

$$\text{Result} = \frac{\text{Signal for test sample}}{\text{Cutoff value}}$$

| | |
|---|---|
| **Caution:** | This test will not report aHIV results associated with VITROS ECi/ECiQ System codes CE (Calibration Expired), ED (Edited Results), EM (Expired Maintenance), IT (Incubator Temperature is outside Specifications), LT (Luminometer Temperature is outside specifications), M1 and/or M2 (Calibration data used for generating a calibration curve or patient results have changed from the default |

values), RC (Luminometer or Incubator reference readings are outside specifications), RE (Reagent Expired), and WT (Well wash temperature is outside specifications).

## Testing Algorithm




## Interpretation of Results

The following table summarizes the interpretation of results obtained with the VITROS Anti-HIV 1+2 test on the VITROS Immunodiagnostic and VITROS Integrated Systems.

| VITROS Anti-HIV 1+2 Test Result (s/c) | Conclusion from Testing Algorithm | Interpretation |
|---|---|---|
| <0.90 | Negative | Specimen is negative for anti-HIV-1 and anti-HIV-2. |
| ≥0.90 and <1.00 | Retest in duplicate | Specimen is negative for anti-HIV-1 and anti-HIV-2 if both duplicate results are <1.00 s/c. Specimen is reactive for anti-HIV-1 and/or anti-HIV-2 if 1 or both duplicate results are ≥1.00 s/c. |
| ≥1.00 | Reactive | Specimen is reactive for anti-HIV-1 and/or anti-HIV-2. |

Patient sample results will be displayed with a **"Negative"**, **"Retest?"** or "**Reactive**" label. An initial result labeled with **"Retest?"** indicates a sample that requires duplicate repeat testing. A duplicate result labeled with **"Retest?"** does not require further testing using the VITROS Anti-HIV 1+2 test. Any result labeled with **"Reactive"** requires supplemental testing.

| Result (s/c) | <0.90 | ≥0.90 and <1.00 | ≥1.00 |
|---|---|---|---|
| Result Text | Negative | Retest? | Reactive |

- If a specimen is reactive (result ≥1.00 s/c) the probability that HIV antibodies are present is high, especially in subjects at high risk for HIV infection. In most settings, it is appropriate to investigate reactive results by additional, more specific tests. Specimens found reactive by the VITROS Anti-HIV 1+2 test and positive by additional, more specific tests are considered positive for antibodies to HIV-1 and/or HIV-2. Clinical correlation is indicated with appropriate counseling, medical intervention and possibly additional testing to decide whether a diagnosis of HIV infection is accurate.
- Interpretation of results from specimens found to be reactive by the VITROS Anti-HIV 1+2 test and negative by additional, more specific tests is unclear. Further clarification may be obtained by testing another specimen obtained three to six months later.
- The magnitude of a VITROS Anti-HIV 1+2 test result cannot be correlated to an endpoint titer.

## Limitations of the Procedure

- The results from this or any other diagnostic test should be used and interpreted only in the context of the overall clinical picture. A negative test result does not exclude the possibility of exposure to or infection with HIV. Levels of HIV antibodies may be undetectable in the early stages of infection.
- This test has not been validated for use with specimens from individuals less than 2 years of age.
- Samples with Total Protein >9 g/dL may give falsely reactive results.
- Heterophilic antibodies in serum or plasma samples may cause interference in immunoassays. [10] These antibodies may be present in blood samples from individuals regularly exposed to animals or who have been treated with animal serum products. Results, which are inconsistent with clinical observations, indicate the need for additional testing.

### Other Limitations

- A person who has antibodies to HIV-1 or HIV-2 is presumed to be infected with the virus, except that a person who has participated in an HIV vaccine study may develop antibodies to the vaccine and may or may not be infected with HIV. Clinical correlation is indicated with appropriate counseling, medical evaluation and possibly additional testing to decide whether a diagnosis of HIV infection is accurate.
- If your laboratory processes patient samples using a finite set of sample IDs (i.e. re-use of sample IDs over time), refer to the Deleting Programs section of the operating instructions for your system regarding the steps that need to be followed to avoid the potential of a sample programming mismatch.
- When processing this test on the VITROS 3600 Immunodiagnostic or VITROS Integrated Systems, the MicroSensor must be enabled.

## Performance Characteristics

### Clinical Performance

A multi center study was conducted to establish the performance characteristics of the VITROS Anti-HIV 1+2 test using samples obtained in the U.S. and internationally from individuals at low or high risk for HIV infection, or known to be HIV antibody positive. Statistical testing was performed to ensure that the distribution of VITROS Anti-HIV 1+2 s/c values was homogeneous across the three testing sites participating in the study.

The specificity of the VITROS Anti-HIV 1+2 test was evaluated among individuals at low risk for HIV infection. The sensitivity of the VITROS Anti-HIV 1+2 test was evaluated among individuals known to be HIV antibody positive, and by testing serially collected samples from individuals with HIV infection (seroconversion panels). Test performance was further evaluated among individuals with signs or symptoms of HIV infection and among individuals belonging to groups recognized to be at risk for HIV infection due to lifestyle, behavior, occupation or known exposure event.

#### *Results by Specimen Classification*

Samples from subjects at high or low risk for HIV infection were tested with an FDA-licensed anti-HIV 1/2 test, and with the VITROS Anti-HIV 1+2 test at the three testing sites. The HIV antibody status (HIV Antibody Positive, HIV Antibody Negative or HIV antibody status Not Determined) of the individual subject was defined according to the following licensed test and supplemental testing algorithm. In those instances where the licensed test was negative but the VITROS Anti-HIV 1+2 test was reactive, supplemental testing was performed to determine the HIV antibody status of the sample.

| Licensed Anti-HIV 1/2 Test Result | Supplemental Testing Result(s) | HIV Antibody Status |
|---|---|---|
| Negative | Not Applicable | HIV Antibody Negative |
| Reactive | Western blot (WB) Negative | HIV Antibody Negative* |
| Reactive | Western blot Positive | HIV Antibody Positive |
| Reactive | Western blot Indeterminate<br>Indirect Immunofluorescence assay (IFA) HIV-1 Negative | HIV Antibody Negative* |
| Reactive | Western blot Indeterminate<br>IFA HIV-1 Positive | HIV Antibody Positive |
| Reactive | Western blot Indeterminate<br>IFA HIV-1 Indeterminate | HIV Antibody Status<br>Not Determined ** |

* Samples from the high risk population whose anti-HIV 1/2 test results were discordant were tested with an HIV-2 EIA/HIV-2 IFA. The HIV antibody status remained “Negative” if the HIV-2 EIA was negative. If the HIV-2 EIA was repeatedly reactive and the HIV-2 IFA was negative or indeterminate, the HIV status was “Not Determined”. The HIV antibody status was “Positive” if the HIV-2 IFA was positive.

** These samples were tested with an HIV-2 EIA/IFA. HIV antibody status remained “Not Determined” if the HIV-2 EIA was negative, or if the HIV-2 EIA was repeatedly reactive but the HIV-2 IFA was negative or indeterminate. The HIV antibody status was “Positive” if the HIV-2 IFA was positive.

Samples from an additional 4600 subjects at low risk for HIV infection were tested with an FDA-licensed anti-HIV 1/2 test, and with the VITROS Anti-HIV 1+2 test at the three testing sites. The HIV antibody status (HIV Antibody Positive, HIV Antibody Negative or HIV antibody status Not Determined) of the individual subject was defined according to the following licensed test and supplemental testing algorithm. In those instances where the licensed test was negative but the VITROS Anti-HIV 1+2 test was reactive, supplemental testing was performed to determine the HIV antibody status of the sample.

| Licensed Anti-HIV 1/2 Test Result | HIV-1 Supplemental Testing Result(s) | HIV-2 Supplemental Testing Result(s) | HIV Antibody Status |
|---|---|---|---|
| Negative | Not Required | Not Required | HIV Antibody Negative |
| Negative | HIV-1 Western Blot Negative | HIV-2 EIA Non-Reactive<br>or<br>Rapid EIA Non-Reactive | HIV Antibody Negative |
| | | HIV-2 EIA RR<br>and<br>Rapid EIA Positive | HIV Antibody Positive |
| Negative | HIV-1 Western Blot Positive | Not Required | HIV Antibody Positive |
| Negative | HIV-1 Western Blot Indeterminate<br>HIV-1 IFA Negative | HIV-2 EIA Non-Reactive<br>or<br>Rapid EIA Non-Reactive | HIV Antibody Negative |
| | | HIV-2 EIA RR<br>and<br>Rapid EIA Positive | HIV Antibody Positive |
| Negative | HIV-1 Western Blot Indeterminate<br>HIV-1 IFA Positive | Not Required | HIV Antibody Positive |
| Negative | HIV-1 Western Blot Indeterminate<br>HIV-1 IFA Indeterminate | HIV-2 EIA Non-Reactive<br>or<br>Rapid EIA Non-Reactive | HIV Antibody Status<br>Not Determined |
| | | HIV-2 EIA RR<br>and<br>Rapid EIA Positive | HIV Antibody Positive |
| Repeatedly reactive | HIV-1 Western Blot Negative | HIV-2 EIA Non-Reactive<br>or<br>Rapid EIA Non-Reactive | HIV Antibody Negative |
| | | HIV-2 EIA RR<br>and<br>Rapid EIA Positive | HIV Antibody Positive |
| Repeatedly reactive | HIV-1 Western Blot Positive | Not Required | HIV Antibody Positive |
| Repeatedly reactive | HIV-1 Western Blot Indeterminate<br>HIV-1 IFA Negative | HIV-2 EIA Non-Reactive<br>or<br>Rapid EIA Non-Reactive | HIV Antibody Negative |
| | | HIV-2 EIA RR<br>and<br>Rapid EIA Positive | HIV Antibody Positive |
| Repeatedly reactive | HIV-1 Western Blot Indeterminate<br>HIV-1 IFA Positive | Not Required | HIV Antibody Positive |
| Repeatedly reactive | HIV-1 Western Blot Indeterminate<br>HIV-1 IFA Indeterminate | HIV-2 EIA Non-Reactive<br>or<br>Rapid EIA Non-Reactive | HIV Antibody Status<br>Not Determined |
| | | HIV-2 EIA RR<br>and<br>Rapid EIA Positive | HIV Antibody Positive |

The specificity/negative percent agreement of the VITROS Anti-HIV 1+2 test was calculated as the percentage of the combined HIV Antibody Negative and status Not Determined subjects that tested negative with the VITROS test. The sensitivity/positive percent agreement of the VITROS Anti-HIV 1+2 test was calculated as the percentage of HIV Antibody Positive subjects that tested reactive with the test.

### *Specificity in Individuals at Low Risk for HIV Infection*

Samples from 1444 subjects at low risk for HIV infection were tested with the VITROS Anti-HIV 1+2 and licensed anti-HIV 1/2 tests (with supplemental testing by licensed HIV-1 Western blot as required). These samples were obtained from

pregnant women in the U.S. (N=297), pregnant women in the U.S. in the period around labor and delivery (N=49), from insurance applicants in the U.S. for whom HIV testing was required (N=999), and from pediatric subjects ages 2-17 years (N=99).

Samples from 4600 additional subjects at low risk for HIV infection were tested with the VITROS Anti-HIV 1+2 and licensed anti-HIV 1/2 tests (with supplemental testing by licensed HIV-1 Western blot as required). These samples were obtained from first time donors of blood or blood derivatives (N=2300), from persons submitting samples for annual physicals (N=700), and from geriatric subjects aged ≥65 years (N=1600). The combined low risk population consisted of 6044 subjects.

There were 99 unlinked samples from low risk pediatric subjects tested with the VITROS Anti-HIV 1+2 test. The group was 49.5% male and 50.5% female and ranged in age from 2-17 years. None of the 99 samples was reactive with the VITROS Anti-HIV 1+2 test. The distribution of VITROS Anit-HIV 1+2 test negative results among the low risk pediatric subjects by age and gender is presented in the following table.

**Demographics of Seroreactivity in Low Risk Pediatric Population (N=99)**

| | | VITROS Anti-HIV 1+2 Test Results | | | | |
|---|---|---|---|---|---|---|
| | | Reactive | | Negative | | |
| Age Range | Gender | N | Percent | N | Percent | Total |
| 2–4 | Female | 0 | 0.0 | 13 | 100.0 | 13 |
| | Male | 0 | 0.0 | 12 | 100.0 | 12 |
| 5–9 | Female | 0 | 0.0 | 11 | 100.0 | 11 |
| | Male | 0 | 0.0 | 14 | 100.0 | 14 |
| 10–14 | Female | 0 | 0.0 | 10 | 100.0 | 10 |
| | Male | 0 | 0.0 | 14 | 100.0 | 14 |
| 15–17 | Female | 0 | 0.0 | 16 | 100.0 | 16 |
| | Male | 0 | 0.0 | 9 | 100.0 | 9 |
| Total | | 0 | 0.0 | 99 | 100.0 | 99 |

The results obtained from the 6044 low risk subjects are summarized in the following table.

**VITROS Anti-HIV 1+2 and Licensed Anti-HIV 1/2 Test Results in Low Risk Populations (N=6044)**

| Population Description | Number Tested | Licensed Anti-HIV 1/2 Test | | | VITROS Anti-HIV 1+2 Test | | Total WB Positive |
|---|---|---|---|---|---|---|---|
| | | NR | IR | RR (WB+) | NR | Reactive (WB+) | |
| Pregnancy (Low Risk–U.S.) | 297 | 295 | 2 | 2 (0) | 294 | 3 (1) | 1 |
| Labor & Delivery (Low Risk–U.S.) | 49 | 49 | 0 | 0 (0) | 48 | 1 (0) | 0 |
| Insurance Applicants (Low Risk–U.S.) | 999 | 993 | 6 | 6 (2) | 991 | 8 (5) | 5 |
| Pediatric (Low Risk–U.S.) | 99 | 99 | 0 | 0 (0) | 99 | 0 (0) | 0 |
| First Time Donors (Low Risk-US) | 2300 | 2296 | 13 | 4 (2) | 2292 | 8 (2) | 2 |
| Annual Physical (Low Risk-US) | 700 | 699 | 17 | 1 (0) | 697 | 3 (0) | 0 |
| Geriatric (Low Risk-US) | 1600 | 1596 | 38 | 4 (0) | 1594 | 6 (0) | 0 |
| Total | 6044 | 6027 | 76 | 17 (4) | 6015 | 29 (8) | 8 |

NR = non reactive (negative); IR = initially reactive; RR = repeatedly reactive; WB = licensed HIV-1 Western blot

Eight of the 6044 low risk samples were reactive with the VITROS Anti-HIV 1+2 test and positive on Western blot. Only four of those Western blot positive samples were repeatedly reactive with the licensed test.

The performance of the VITROS Anti-HIV 1+2 test compared with HIV antibody status in low risk populations is summarized in the following table:

### Agreement of the VITROS Anti-HIV 1+2 Test with HIV Antibody Status in Low Risk Populations (N=6044)

| VITROS Anti-HIV 1+2 Test Results | HIV Antibody Status | | | Total |
|---|---|---|---|---|
| | Positive | Negative | Not Determined* | |
| Reactive | 8 | 20** | 1 | 29 |
| Negative | 0 | 6015 | 0 | 6015 |
| Total | 8 | 6035 | 1 | 6044 |

* This sample remained HIV status "Not Determined" following VITROS, licensed and supplemental testing for anti-HIV-1 and anti-HIV-2 (EIA, WB, IFA). This sample was considered anti-HIV negative when calculating specificity.

** Three samples were initially reactive with the VITROS test during the clinical study and were retested in duplicate: both duplicate results for the three samples were negative. All three were initially non reactive with the reference test. HIV-1 Western blot results are not available for the three samples The VITROS test results for these three specimens were considered reactive per the VITROS testing algorithm shown in the Interpretation of Results and Expected results section when calculating specificity.

The specificity of the VITROS Anti-HIV 1+2 test was calculated as the percentage of the combined HIV antibody Negative and status "Not Determined" subjects that tested negative with the VITROS test.

The specificity of the VITROS Anti-HIV 1+2 test in the low risk populations was 99.65% (6015/6036 in this study with a 95% exact confidence interval (CI) of 99.47% to 99.78%) compared with 99.78% (6023/6036) for the licensed anti-HIV 1/2 test.

### *Specificity in Individuals at Low Risk for HIV Infection*

Samples from 1444 subjects at low risk for HIV infection were tested with the VITROS Anti-HIV 1+2 and licensed anti-HIV 1/2 tests (with supplemental testing by licensed HIV-1 Western blot as required). These samples were obtained from pregnant women in the U.S. (N=297), pregnant women in the U.S. in the period around labor and delivery (N=49), from insurance applicants in the U.S. for whom HIV testing was required (N=999), and from pediatric subjects ages 2–17 years (N=99).

There were 99 unlinked samples from low risk pediatric subjects tested with the VITROS Anti-HIV 1+2 test. The group was 49.5% male and 50.5% female and ranged in age from 2–17 years. None of the 99 samples was reactive with the VITROS Anti-HIV 1+2 test. The distribution of VITROS Anti-HIV 1+2 test negative results among the low risk pediatric subjects by age and gender is presented in the following table.

### Demographics of Seroreactivity in Low Risk Pediatric Population (N=99)

| Age Range | Gender | VITROS Anti-HIV 1+2 Test Results | | | | Total |
|---|---|---|---|---|---|---|
| | | Reactive | | Negative | | |
| | | N | Percent | N | Percent | |
| 2–4 | Female | 0 | 0.0 | 13 | 100.0 | 13 |
| | Male | 0 | 0.0 | 12 | 100.0 | 12 |
| 5–9 | Female | 0 | 0.0 | 11 | 100.0 | 11 |
| | Male | 0 | 0.0 | 14 | 100.0 | 14 |
| 10–14 | Female | 0 | 0.0 | 10 | 100.0 | 10 |
| | Male | 0 | 0.0 | 14 | 100.0 | 14 |
| 15–17 | Female | 0 | 0.0 | 16 | 100.0 | 16 |
| | Male | 0 | 0.0 | 9 | 100.0 | 9 |
| Total | | 0 | 0.0 | 99 | 100.0 | 99 |

The results obtained from the 1444 low risk subjects are summarized in the following table.

**VITROS Anti-HIV 1+2 and Licensed Anti-HIV 1/2 Test Results in Low Risk Populations (N=1444)**

| Population Description | Number Tested | Licensed Anti-HIV 1/2 Test | | | VITROS Anti-HIV 1+2 Test | | Total WB Positive |
|---|---|---|---|---|---|---|---|
| | | NR | IR | RR (WB+) | NR | Reactive (WB+) | |
| Pregnancy (Low Risk–U.S.) | 297 | 295 | 2 | 2 (0) | 294 | 3 (1) | 1 |
| Labor & Delivery (Low Risk–U.S.) | 49 | 49 | 0 | 0 (0) | 48 | 1 (0) | 0 |
| Insurance Applicants (Low Risk–U.S.) | 999 | 993 | 6 | 6 (2) | 991 | 8 (5) | 5 |
| Pediatric (Low Risk–U.S.) | 99 | 99 | 0 | 0 (0) | 99 | 0 (0) | 0 |
| Total | 1444 | 1436 | 8 | 8 (2) | 1432 | 12 (6) | 6 |

NR = non reactive (negative); IR = initially reactive; RR = repeatedly reactive; WB = licensed HIV-1 Western blot

Six of the 1444 low risk samples were reactive with the VITROS Anti-HIV 1+2 test and positive on Western blot. Only two of those Western blot positive samples were repeatedly reactive with the licensed test.

The performance of the VITROS Anti-HIV 1+2 test compared with HIV antibody status in low risk populations is summarized in the following table:

**Agreement of the VITROS Anti-HIV 1+2 Test with HIV Antibody Status in Low Risk Populations (N=1444)**

| VITROS Anti-HIV 1+2 Test Results | HIV Antibody Status | | | Total |
|---|---|---|---|---|
| | Positive | Negative | Not Determined* | |
| Reactive | 6 | 5** | 1 | 12 |
| Negative | 0 | 1432 | 0 | 1432 |
| Total | 6 | 1437 | 1 | 1444 |

* This sample remained HIV status “Not Determined” following VITROS, licensed and supplemental testing for anti-HIV-1 and anti-HIV-2 (EIA, WB, IFA). This sample was considered anti-HIV negative when calculating specificity.

** Three samples were initially reactive with the VITROS test during the clinical study and were retested in duplicate: both duplicate results for the three samples were negative. All three were initially non reactive with the reference test. HIV-1 Western blot results are not available for the three samples

The specificity of the VITROS Anti-HIV 1+2 test was calculated as the percentage of the combined HIV antibody Negative and status “Not Determined” subjects that tested negative with the VITROS Anti-HIV 1+2 test.

The specificity of the VITROS Anti-HIV 1+2 test in the low risk populations was 99.58% (1432/1438 in this study with a 95% exact confidence interval (CI) of 99.09% to 99.85%) compared with 99.58% (1432/1438) for the licensed anti-HIV 1/2 test. The six low risk samples that were HIV-1 Western blot positive (HIV antibody positive) were excluded from the specificity calculation.

### *Sensitivity in Individuals Positive for Antibodies to HIV-1 and HIV-2*

### *Sensitivity in Individuals Positive for Antibodies to HIV-1*

Samples from 1121 HIV-1 infected adults were tested with the VITROS Anti-HIV 1+2 test. These subjects were enrolled in Florida (72.8%; N=816), California (16.3%; N=183), New Jersey (8.9%; N=100) and Texas (2.0%; N=22). Clinical and laboratory documentation of HIV-1 infection and HIV antibody positive status were obtained from medical records for each of the 1121 individuals. In addition, 40 unlinked residual samples from HIV-1 antibody positive pediatric subjects, ages 1–16 years of age, were also tested with the VITROS Anti-HIV 1+2 test.

CD4+ counts were available for 1094 of the 1121 HIV-1 infected adults. As shown in the following table, the VITROS Anti-HIV 1+2 test was reactive with all 1121 samples regardless of the subjects’ CD4+ counts.

**VITROS Anti-HIV 1+2 Test Results Among U.S. HIV Infected Adults with Documented CD4+ Counts (N=1121)**

| CD4+ Count | N | VITROS Anti-HIV 1+2 Test |
|---|---|---|
| | | Reactive |
| <200 | 149 | 149 |
| 200–499 | 429 | 429 |
| >499 | 516 | 516 |
| Unknown | 27 | 27 |

The VITROS Anti-HIV 1+2 test results in the HIV-1 antibody positive populations are summarized in the following table:

**VITROS Anti-HIV 1+2 Test Results in Known HIV Antibody Positive Subjects (N=1161)**

| Population Description | VITROS Anti-HIV 1+2 Test | | |
|---|---|---|---|
| | N | NR | Reactive |
| Adult HIV-1 Positive (U.S.) | 1121 | 0 | 1121 |
| Pediatric HIV-1 Positive (U.S.) | 40 | 0 | 40 |
| Total | 1161 | 0 | 1161 |

All 1161 samples were reactive with the VITROS Anti-HIV 1+2 test. The sensitivity of the VITROS Anti-HIV 1+2 test for U.S. subjects known to be positive for HIV-1 antibody was 100% (1161/1161; 95% CI = 99.68% to 100%) in this study.

### *Sensitivity in Individuals Positive for Antibodies to HIV-2*

Sensitivity of the VITROS Anti-HIV 1+2 test was also determined among 208 mono-infected HIV-2 antibody positive individuals from the Ivory Coast. Testing results are summarized in the following table:

**VITROS Anti-HIV 1+2 Test Results in Mono-Infected HIV-2 Antibody Positive Subjects (N=208)**

| Population Description | VITROS Anti-HIV 1+2 Test | | |
|---|---|---|---|
| | N | NR | Reactive |
| HIV-2 Positive (Ivory Coast) | 208 | 0 | 208 |

All 208 anti-HIV-2 positive samples were reactive with the VITROS Anti-HIV 1+2 test. The sensitivity of the VITROS Anti-HIV 1+2 test in mono-infected HIV-2 antibody positive individuals was 100% (208/208; 95% CI = 98.24% to 100.0%) in this study.

### *Sensitivity in International Populations Known to be HIV Antibody Positive*

Sensitivity of the VITROS Anti-HIV 1+2 test was determined among 194 HIV antibody positive individuals from four geographic locations outside the U.S. Samples were obtained from archives in Africa (9.8%; N=19), Asia (53.1%; N=103), Europe (34.0%; N=66) and Latin America (3.1%; N=6). The VITROS Anti-HIV 1+2 test results are presented in the following table:

**VITROS Anti-HIV 1+2 Test Results in International Populations Known to be HIV Antibody Positive (N=194)**

| Population Description | VITROS Anti-HIV 1+2 Test | | |
|---|---|---|---|
| | N | NR | Reactive |
| HIV Positive (Africa) | 19 | 1* | 18 |
| HIV Positive (Asia) | 103 | 0 | 103 |
| HIV Positive (Europe) | 66 | 0 | 66 |
| HIV Positive (Latin America) | 6 | 0 | 6 |
| Total | 194 | 1* | 193 |

* This archived sample was obtained from a rural clinic in Keffi, Nigeria. The VITROS Anti-HIV 1+2 test gave an initial s/c result of 0.53, and repeat results of 0.55 and 0.56. Singleton testing with the licensed test gave an s/c result of 1.93. An HIV-1 Western blot showed bands of 1+ intensity at the p17, p24 and gp160 positions. The sample is unlinked to the donor's identity. No clinical information or follow-up sample is available.

One sample was non-reactive and 193 samples were reactive with the VITROS Anti-HIV 1+2 test.

### *Reactivity in Populations at High Risk for HIV-1 Infection and HIV-2 Infection*

### *Demographics of Seroreactivity Study in High Risk Populations*

Among the 2175 high risk subjects from the U.S. participating in the VITROS Anti-HIV 1+2 test clinical study, 1975 (90.8%) reported no current signs or symptoms of HIV infection. Of these 1975 asymptomatic individuals, 14.7% (291) were enrolled in Florida, 11.2% (222) were enrolled in New Jersey, 1.1% (21) were enrolled in Texas and 73.0% (1441) were

enrolled in California. The group was Caucasian (28.2%), African American (18.1%) Hispanic (49.6%), and Asian (1.1%) with the remaining 3.0% represented by other ethnic groups. The group was 50.3% male and 49.7% female and ranged in age from 14 to 82 years. All were at risk for HIV infection due to lifestyle, behavior, occupation or known exposure event, or belonged to groups at risk for HIV infection. The VITROS Anti-HIV 1+2 test was reactive in 2.2% (43/1975) of the individuals in this group. The percent VITROS Anti-HIV reactive results observed in the asymptomatic population in each collection area was 3.8% (11/291) in Florida, 0.0% (0/21) in Texas, 3.6% (8/222) in New Jersey, and 1.7% (24/1441) in California. The distribution of VITROS Anti-HIV 1+2 test reactive and negative results among the high risk subjects without signs or symptoms of HIV infection by age and gender is presented in the following table.

**Seroreactivity for the VITROS Anti-HIV 1+2 Test in High Risk Subjects Without Signs or Symptoms of HIV Infection (N=1975)**

| | | VITROS Anti-HIV 1+2 Test Results | | | | |
|---|---|---|---|---|---|---|
| | | Reactive | | Negative | | |
| Age Range | Gender | N | Percent | N | Percent | Total |
| 14–19 | Female | 0 | 0.0 | 28 | 100.0 | 28 |
| | Male | 0 | 0.0 | 25 | 100.0 | 25 |
| 20–29 | Female | 1 | 0.4 | 233 | 99.6 | 234 |
| | Male | 5 | 3.0 | 162 | 97.0 | 167 |
| 30–39 | Female | 1 | 0.4 | 284 | 99.6 | 285 |
| | Male | 5 | 2.0 | 249 | 98.0 | 254 |
| 40–49 | Female | 7 | 2.4 | 289 | 97.6 | 296 |
| | Male | 11 | 3.5 | 307 | 96.5 | 318 |
| 50–59 | Female | 2 | 1.7 | 116 | 98.3 | 118 |
| | Male | 11 | 5.6 | 184 | 94.4 | 195 |
| 60–69 | Female | 0 | 0.0 | 14 | 100.0 | 14 |
| | Male | 0 | 0.0 | 36 | 100.0 | 36 |
| 70–79 | Female | 0 | 0.0 | 1 | 100.0 | 1 |
| | Male | 0 | 0.0 | 3 | 100.0 | 3 |
| 80–82 | Female | 0 | 0.0 | 1 | 100.0 | 1 |
| | Male | 0 | 0.0 | 0 | 0.0 | 0 |
| Total | | 43 | 2.2 | 1932 | 97.8 | 1975 |

### *Reactivity in Populations at High Risk for HIV-1 Infection*

The performance of the VITROS Anti-HIV 1+2 test was evaluated among 2175 individuals at high risk for HIV-1 infection prospectively enrolled in California (69.8%; N=1517), Florida (14.6; N=317), New Jersey (14.6%; N=317) and Texas (1.1%; N=24), and in 249 pregnant women at high risk for HIV infection enrolled in California (49.0%; N=122) and Florida (51.0%; N=127). Testing results are presented in the following table:

**VITROS and Licensed Anti-HIV 1/2 Test Results in U.S. High Risk Populations (N=2424)**

| Population Description | Number Tested | Licensed Anti-HIV 1/2 Test | | | VITROS Anti-HIV 1+2 Test | | Total WB Positive |
|---|---|---|---|---|---|---|---|
| | | NR | IR | RR (WB+) | NR | Reactive (WB+) | |
| High Risk (U.S.) | 2175 | 2106 | 69 | 68 (53) | 2116 | 59 (54) | 54 |
| Pregnancy (High Risk - U.S.) | 249 | 244 | 5 | 4 (4) | 243 | 6 (5) | 5 |
| Total | 2424 | 2350 | 74 | 72 (57) | 2359 | 65 (59) | 59 |

The VITROS Anti-HIV 1+2 test was reactive in 65 samples; 59 of those were Western blot positive. The licensed test was repeatedly reactive in 57 (57/59; 99.61% detection). All of the 57 Western blot positives that were RR in the licensed test were reactive in the VITROS Anti-HIV 1+2 test.

The performance of the VITROS Anti-HIV 1+2 test compared with HIV antibody status in the U.S. high risk populations is summarized in the following table:

### Agreement of the VITROS Anti-HIV 1+2 Test with HIV Antibody Status in U.S. High Risk Populations (N=2424)

| VITROS Anti-HIV 1+2 Test Results | HIV Antibody Status | | | Total |
|---|---|---|---|---|
| | Positive | Negative | Not Determined* | |
| Reactive | 59 | 5** | 1 | 65 |
| Negative | 0 | 2359 | 0 | 2359 |
| Total | 59 | 2364 | 1 | 2424 |

* This sample remained HIV status "Not Determined" following VITROS, licensed and supplemental test testing for anti-HIV-1 and anti-HIV-2 (EIA, WB, IFA). This sample was considered anti-HIV negative when calculating percent agreement.

** One sample was initially reactive with the VITROS test during the clinical study and was retested in duplicate: both duplicate results were negative. The sample was initially non reactive with the reference test. HIV-1 Western blot results are not available for the sample.

The positive percent agreement of the VITROS Anti-HIV 1+2 test with HIV antibody status in U.S. high risk populations was 100% (59/59; 95% CI = 93.94% to 100%) compared with 96.61% (57/59) for the licensed test. The negative percent agreement was 99.75% (2359/2365; 95% CI = 99.45% to 99.91%) in this study compared with 99.37% (2350/2365) for the licensed test.

#### *Reactivity in Populations at High Risk for HIV-2 Infection*

The performance of the VITROS Anti-HIV 1+2 test was evaluated among individuals at high risk for HIV-2 infection due to residence in an HIV-2 endemic area. The 488 subjects in this group were prospectively enrolled in the Ivory Coast. Testing results are presented in the following table:

### VITROS and Licensed Anti-HIV 1/2 Test Results in Individuals at High Risk for HIV-2 Infection (N=488)

| Population Description | Number Tested | Licensed Anti-HIV 1/2 Test | | | VITROS Anti-HIV 1+2 Test | | Total WB Positive |
|---|---|---|---|---|---|---|---|
| | | NR | IR | RR (WB+) | NR | Reactive (WB+) | |
| High Risk (Ivory Coast) | 488 | 453 | 35 | 33 (26) | 457 | 31 (26) | 26 |

The VITROS Anti-HIV 1+2 test was reactive in 31 samples; 26 of these were HIV-1 Western blot positive. The licensed anti-HIV 1/2 test was repeatedly reactive in 33 samples; 26 of these were HIV-1 Western blot positive. All of the 26 HIV-1 positives that were RR on the licensed test were reactive in the VITROS Anti-HIV 1+2 test. The HIV-1 Western blot positive samples did not undergo further supplemental testing. Samples with discordant anti-HIV 1/2 EIA results that were negative or indeterminate by supplemental testing for antibodies to HIV-1 were tested for antibodies to HIV-2 (EIA and IFA). None were positive for anti-HIV-2.

The performance of the VITROS Anti-HIV 1+2 test compared with HIV antibody status in the HIV-2 high risk population is summarized in the following table:

### Agreement of the VITROS Anti-HIV 1+2 Test with HIV Antibody Status in the HIV-2 High Risk Population (N=488)

| VITROS Anti-HIV 1+2 Test Results | HIV Antibody Status | | | Total |
|---|---|---|---|---|
| | Positive | Negative | Not Determined* | |
| Reactive | 26 | 1 | 4 | 31 |
| Negative | 0 | 452 | 5 | 457 |
| Total | 26 | 453 | 9 | 488 |

* These samples remained HIV status "Not Determined" following VITROS, licensed and supplemental test testing for anti-HIV-1 and anti-HIV-2 (EIA, WB, IFA). These samples were considered anti-HIV negative when calculating percent agreement.

The positive percent agreement of the VITROS Anti-HIV 1+2 test with HIV antibody status in the HIV-2 high risk population was 100% (26/26; 95% CI = 86.77% to 100%) compared with 100% (26/26) for the licensed test. The negative percent agreement was 98.92% (457/462; 95% CI = 97.49% to 99.65%) compared with 98.48% (455/462) for the licensed test.

#### *Positive and Negative Percent Agreement of the VITROS Anti-HIV 1+2 Test with HIV Antibody Status by Study Population*

The positive and/or negative percent agreement of the VITROS Anti-HIV 1+2 test with HIV antibody status in the high risk, low risk and HIV antibody positive populations tested in this clinical study are summarized in the following table:

**Positive and Negative Percent Agreement of the VITROS Anti-HIV 1+2 Test with HIV Antibody Status by Study Population (N=5919)**

| Population | Positive Percent Agreement | 95% Exact Confidence Intervals | Negative Percent Agreement | 95% Exact Confidence Intervals |
|---|---|---|---|---|
| High Risk (U.S.)* | 100% (59/59) | 93.94%–100% | 99.75% (2359/2365) | 99.45%–99.91% |
| High Risk (Ivory Coast) | 100% (26/26) | 86.77%–100% | 98.92% (457/462) | 97.49%–99.65% |
| HIV Positive (U.S.)** | 100% (1161/1161) | 99.68%–100% | ***** | ***** |
| HIV Positive (International) | 99.48% (193/194) | 97.16%–99.99% | ***** | ***** |
| HIV-2 Positive (Ivory Coast) | 100% (208/208) | 98.24%–100% | ***** | ***** |
| Low Risk (U.S.)*** | 100% (6/6) | N/A**** | 99.58% (1432/1438) | 99.09%–99.85% |

* Adult (N=2175); pregnant (N=249).

** Adult (N=1121); pediatric (N=40).

*** Pregnant (N=297); labor and delivery (N=49); insurance applicants (N=999); pediatric (N=99).

**** N/A=Not applicable. Confidence intervals calculated on small numbers are not meaningful.

***** No information available.

### *Seroconversion Panels*

Twenty commercially available seroconversion panels were tested. Results for the twenty panels are summarized in the following table. The table presents the days elapsed from the date of the initial bleed to the last negative sample and first reactive sample. Data are presented for both tests for each of the seroconversion panels.

**Days to Evidence of HIV Infection**

| | Licensed Anti-HIV 1/2 Test | | VITROS Anti-HIV 1+2 Test | | Difference in Days to Anti-HIV Reactive Result |
|---|---|---|---|---|---|
| Panel ID | –* | +** | –*** | +**** | Licensed Test minus VITROS Anti-HIV 1+2 Test |
| PRB904 | 49 | 92 | 49 | 92 | 0 |
| PRB910 | 14 | 26 | 14 | 26 | 0 |
| PRB916 | 15 | 30 | 15 | 30 | 0 |
| PRB923 | 37 | 47 | 37 | 47 | 0 |
| PRB924 | 26 | 33 | 26 | 33 | 0 |
| PRB925 | 22 | 44 | 22 | 44 | 0 |
| PRB926 | 9 | 27 | 9 | 27 | 0 |
| PRB927 | 0 | 28 | 28 | 33 | -5 |
| PRB929 | 21 | 25 | 18 | 21 | 4 |
| PRB931 | 15 | 28 | 15 | 28 | 0 |
| PRB933 | 0 | 21 | 0 | 21 | 0 |
| PRB934 | 0 | 7 | 0 | 0 | 7 |
| PRB935 | 28 | 43 | 28 | 43 | 0 |
| PRB940 | 7 | 11 | 0 | 7 | 4 |
| PRB941 | 9 | 18 | 9 | 18 | 0 |
| PRB944 | 9 | 14 | 9 | 14 | 0 |
| PRB945 | 7 | 13 | 7 | 13 | 0 |
| PRB947 | 0 | 9 | 0 | 9 | 0 |
| PRB952 | 14 | 17 | 10 | 14 | 3 |
| PRB959 | 7 | 9 | 0 | 7 | 2 |

* Post bleed day of last non-reactive result, usually denotes previous bleed from first repeatedly reactive result.

** Post bleed day of first repeatedly reactive result.

*** Post bleed day of last non-reactive result, usually denotes previous bleed from first reactive result.

**** Post bleed day of first reactive result.

The VITROS Anti-HIV 1+2 and licensed anti-HIV 1/2 tests were in agreement for 14 of the 20 panels. The VITROS Anti-HIV 1+2 test became reactive one bleed (from two to seven days) earlier for five of the twenty panels. The licensed test became repeatedly reactive one bleed (five days) earlier for the final panel.

### *Genotype Detection*

Genotype detection was assessed using the Boston Biomedica, Inc. Worldwide HIV Performance Panel. This panel consists of 25 naturally occurring plasma specimens originating from diverse geographic locations. Twenty three of these specimens have been characterized to be anti-HIV reactive, while two are anti-HIV nonreactive. The reactive specimens represent HIV Group M (subtypes A, B, C, D, E, F, and G) Group O, and HIV-2 genotypes. All 23 of the anti-HIV reactive panel members were also reactive in the VITROS anti-HIV 1+2 test, while the two anti-HIV nonreactive panel members were negative in the VITROS Anti-HIV 1+2 test. Two lots of VITROS Anti-HIV 1+2 Reagent Packs and Calibrators were included in this study.

**BBI Worldwide HIV Performance Panel Test Results with VITROS Anti-HIV 1+2 Test**

| | VITROS Anti-HIV 1+2 Test | | |
|---|---|---|---|
| Panel ID Number | Genotype | Result (s/c) | Classification |
| WWRB302(M)-01 | O | 9.45 | Reactive |
| WWRB302(M)-02 | A | 55.3 | Reactive |
| WWRB302(M)-03 | G | 85.3 | Reactive |
| WWRB302(M)-04 | G | 56.4 | Reactive |
| WWRB302(M)-05 | A | 54.6 | Reactive |
| WWRB302(M)-06 | G | 65.0 | Reactive |
| WWRB302(M)-08 | G | 60.0 | Reactive |
| WWRB302(M)-09 | A | 82.6 | Reactive |
| WWRB302(M)-10 | NEG | 0.14 | Negative |
| WWRB302(M)-11 | HIV-2* | 26.3 | Reactive |
| WWRB302(M)-12 | C | 89.4 | Reactive |
| WWRB302(M)-14 | D | 61.2 | Reactive |
| WWRB302(M)-15 | D | 59.2 | Reactive |
| WWRB302(M)-16 | D | 64.1 | Reactive |
| WWRB302(M)-17 | D | 73.0 | Reactive |
| WWRB302(M)-19 | C | 55.2 | Reactive |
| WWRB302(M)-21 | B' | 81.4 | Reactive |
| WWRB302(M)-22 | E | 72.7 | Reactive |
| WWRB302(M)-24 | E | 78.4 | Reactive |
| WWRB302(M)-25 | HIV-2* | 40.6 | Reactive |
| WWRB302(M)-26 | B | 74.1 | Reactive |
| WWRB302(M)-27 | B/D | 54.1 | Reactive |
| WWRB302(M)-28 | F | 79.6 | Reactive |
| WWRB302(M)-29 | B | 86.7 | Reactive |
| WWRB302(M)-30 | NEG | 0.10 | Negative |

* The HIV-2 status of these specimens was determined by serological testing.

### *Detection of HIV-1 Group O Specimens*

Thirteen confirmed HIV 1 Group O antibody positive samples were tested with the VITROS Anti-HIV 1+2 test. All 13 samples gave a reactive result. All samples were confirmed as HIV O reactive using Western Blots developed at the Institut Alfred Fournier. The results are shown in the following table.

| HIV-1 Group O Sample ID | VITROS Anti-HIV 1+2 Test Result (s/c) |
|---|---|
| R407/937 | 25.4 |
| R407/939 | 23.3 |
| R407/941 | 5.85 |
| R407/942 | 27.1 |
| R407/943 | 10.1 |
| R407/946 | 2.73 |
| R407/947 | 4.08 |
| R407/948 | 11.1 |
| R407/949 | 26.1 |
| R407/950 | 5.18 |
| R407/951 | 8.69 |
| R407/952 | 19.0 |
| R407/953 | 36.6 |

One additional sample from the Worldwide HIV Performance Panel obtained from Boston Biomedica, Inc. (sample WWRB302(M)-01) was also tested with the VITROS Anti-HIV 1+2 test, and gave a reactive s/c result of 9.45. Thus, all 14 HIV-1 Group O samples gave reactive results with the VITROS Anti-HIV 1+2 test.

### *Potentially Cross-Reacting Sub-groups*

A total of 236 patient samples from the following 14 potentially cross-reacting sub-groups were tested in the VITROS anti-HIV 1+2 test: HCV infection, HBV infection, HTLV I/II antibody positive, EBV infection, Influenza vaccine recipients, multiply transfused patients, multiparous females, dialysis patients, hemophilia patients, autoimmune disease patients, high rheumatoid factor, yeast (Candida) reactive, SOD reactive samples, and cord blood samples from neonates. An additional 15 samples from patients who had not received the Influenza vaccine were tested as a control for the Influenza vaccine recipients group. In all, 251 patient samples were tested for this study.

In these 251 samples tested, 3 were reactive in the VITROS Anti-HIV 1+2 test. In the 14 clinical categories, 1 sample from the hemophilia group was found to give a reactive result in the VITROS Anti-HIV 1+2 test yielding results just above the cutoff. This sample yielded negative results with an FDA licensed anti-HIV 1/2 test. The second reactive sample was from the pre-Influenza vaccine group, this sample was also reactive with an FDA licensed anti-HIV 1/2 test. A third sample in the SOD reactive subgroup tested reactive on initial determination and negative upon repeat testing.

**Summary of VITROS Anti-HIV 1+2 Test Results with Potentially Cross-Reacting Specimens**

| Sample Category | No. Samples Tested | No. Negative | No. Reactive | No. Confirmed Positive |
|---|---|---|---|---|
| HCV Infection | 16 | 16 | 0 | 0 |
| HTLV I/II Positive | 16 | 16 | 0 | 0 |
| EBV Infection | 15 | 15 | 0 | 0 |
| Multiparous Females | 16 | 16 | 0 | 0 |
| Pre-Influenza Vaccine | 15 | 14 | 1 | 0* |
| Post-Influenza Vaccine | 15 | 15 | 0 | 0 |
| Rheumatoid Factor | 16 | 16 | 0 | 0 |
| Autoimmune Disease | 16 | 16 | 0 | 0 |
| Multiply Transfused Patients | 16 | 16 | 0 | 0 |
| HBV Infection | 16 | 16 | 0 | 0 |
| Hemophilia | 16 | 15 | 1 | 0 |
| Dialysis | 16 | 16 | 0 | 0 |
| Yeast Reactive | 20 | 20 | 0 | 0 |
| SOD Reactive | 22 | 21 | 1 | 0 |
| Cord Blood (Neonates) | 20 | 20 | 0 | 0 |

* Pre-Influenza specimen was tested and found reactive with an FDA licensed anti-HIV 1/2 test. There was insufficient sample to perform a Western blot.

### *Potentially Cross-Reacting Sub-Groups - Microbiological Studies*

The potential for bacterial contamination to affect the performance of the VITROS Anti-HIV 1+2 test was evaluated further by testing samples spiked with *Staphylococcus aureus, Escherichia coli* and *Pseudomonas aeruginosa*. The samples were tested with and without a spike of anti-HIV 1+2.

Of the samples that were tested none of the anti-HIV 1+2 unspiked (negative) samples were found to be false reactive and none of the anti-HIV 1+2 spiked samples were observed to be false negative in the VITROS Anti-HIV 1+2 test.

## Potentially Interfering Substances

The potentially interfering effects of hemoglobin, bilirubin and triolein were evaluated using samples from 30 patients. The results demonstrate that hemoglobin (up to 500 mg/dL), bilirubin (up to 20 mg/dL) and triolein (up to 3000 mg/dL) cause no misclassification of results. Samples spiked with anti-HIV-1 and anti-HIV-2 reactive plasma were tested near the cutoff (cutoff s/c=1.00) and were observed to remain reactive at all levels tested with each potential interferent. Similarly, no interference was observed in samples not spiked with anti-HIV-1 and anti-HIV-2 reactive plasma, with results remaining below 1.00 s/c.

| HIV-1 | | | Mean Result at 0 Interferent Level | | Mean Result at Maximum Interferent Level | |
|---|---|---|---|---|---|---|
| Test Substance | Sample | Maximum Level Tested | s/c | Classification | s/c | Classification |
| Hemoglobin | HIV-1 Spiked sample | 500 mg/dL | 1.11 | Reactive | 1.16 | Reactive |
| | Negative sample | 500 mg/dL | 0.06 | Negative | 0.09 | Negative |
| | | | | | | |
| Bilirubin | HIV-1 Spiked sample | 20 mg/dL | 1.53 | Reactive | 1.51 | Reactive |
| | Negative sample | 20 mg/dL | 0.08 | Negative | 0.08 | Negative |
| | | | | | | |
| Triolein | HIV-1 Spiked sample | 3000 mg/dL | 1.34 | Reactive | 1.36 | Reactive |
| | Negative sample | 3000 mg/dL | 0.06 | Negative | 0.06 | Negative |

| HIV-2 | | | Mean Result at 0 Interferent Level | | Mean Result at Maximum Interferent Level | |
|---|---|---|---|---|---|---|
| Test Substance | Sample | Maximum Level Tested | s/c | Result | s/c | Result |
| Hemoglobin | HIV-2 Spiked sample | 500 mg/dL | 1.21 | Reactive | 1.29 | Reactive |
| | Negative sample | 500 mg/dL | 0.06 | Negative | 0.09 | Negative |
| | | | | | | |
| Bilirubin | HIV-2 Spiked sample | 20 mg/dL | 1.39 | Reactive | 1.40 | Reactive |
| | Negative sample | 20 mg/dL | 0.17 | Negative | 0.18 | Negative |
| | | | | | | |
| Triolein | HIV-2 Spiked sample | 3000 mg/dL | 1.55 | Reactive | 1.55 | Reactive |
| | Negative sample | 3000 mg/dL | 0.06 | Negative | 0.06 | Negative |

A total of 60 samples was tested from the following subsets: patients with Cholesterol <200 mg/dL, patients with Cholesterol >300 mg/dL, patients with Total Protein between 6 and 8 g/dL, patients with Total Protein >9 g/dL, normal patient samples (assumed normal IgG concentration) and patient samples spiked with Human IgG to achieve a concentration of >2620 mg/dL. Samples for Cholesterol and Total Protein testing were naturally occurring. Samples for IgG testing were normal patient samples that were spiked with a purified IgG preparation to achieve a level above the normal range.

All samples in each subset were spiked with anti-HIV-1 or anti-HIV-2 reactive plasma to evaluate performance with positive samples. All samples in each subset were also tested after being spiked with negative plasma to evaluate performance with negative samples.

All samples spiked with anti-HIV-1 or anti-HIV-2 reactive plasma yielded reactive results. Among samples spiked with negative plasma 47 of 60 yielded negative results. A total of 13 samples, all in the high total protein group, yielded reactive results. Twelve of 13 reactive samples also tested reactive with an FDA licensed method.

Patient samples spiked with cholesterol up to 415 mg/dL or IgG up to 2620 mg/dL do not interfere with the clinical interpretation of results.

| Sample Category | No. Samples Tested | No. Negative | VITROS Anti-HIV 1+2 Test Reactive | No. Reactive in FDA Licensed Test |
|---|---|---|---|---|
| Cholesterol <200 mg/dL | 10 | 10 | 0 | 0 |
| Cholesterol >300 mg/dL * | 10 | 10 | 0 | 0 |
| Total Protein between 6 and 8 g/dL | 10 | 10 | 0 | 0 |
| Total Protein >9 g/dL | 20 | 7 | 13 | 12 |
| Serum spiked with Human IgG** | 10 | 10 | 0 | 0 |

* Maximum cholesterol level in the tested sample was 415 mg/dL.

** Maximum IgG concentration was 2620 mg/dL.

A total of 22 additional samples were tested from patients with Total Protein >9 g/dL. Samples were tested to determine if the presence of high total protein would cause negative samples to yield reactive test results.

A total of 21 of the 22 samples yielded negative results with the VITROS Anti-HIV 1+2 test. A single sample yielded reactive results with the VITROS Anti-HIV 1+2 test. This sample was determined to be reactive in an FDA licensed test and confirmed HIV antibody positive by Western blot.

Patient samples containing protein >9 g/dL do not consistently interfere with the clinical interpretation of results.

**Summary of VITROS anti-HIV 1+2 Data from Potentially Interfering Sample Conditions**

| Sample Category | No. Samples Tested | No. Negative | VITROS Anti-HIV 1+2 Test Reactive | No. Reactive in FDA Licensed Test | No. Confirmed HIV Antibody Positive in Western Blot |
|---|---|---|---|---|---|
| Total Protein >9 g/dL | 22 | 21 | 1 | 1 | 1 |

## Precision

### *VITROS ECi/ECiQ Immunodiagnostic System*

Precision was evaluated on a different VITROS ECi/ECiQ Immunodiagnostic System at three external sites, using one reagent pack and calibrator kit lot. At least 2 replicates each of a four member panel were tested on a single occasion per day on 20 different days. The data shown in the table were rounded following all calculations.

| Clinical Site | Mean VITROS Anti-HIV 1+2 Test Results (s/c) | | Repeatability* | | Between Day** | | Total*** | | No. of Obs. | No. of Days |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | S.D. | C.V.(%) | S.D. | C.V.(%) | S.D. | C.V.(%) | | |
| Site 1 | 0.07 | Negative | 0.007 | 9.9 | 0.005 | 6.9 | 0.008 | 12.1 | 40 | 20 |
| | 6.06 | HIV-1 | 0.071 | 1.2 | 0.172 | 2.8 | 0.186 | 3.1 | 40 | 20 |
| | 4.12 | HIV-2 | 0.046 | 1.1 | 0.163 | 3.9 | 0.169 | 4.1 | 40 | 20 |
| | 1.32 | HIV-1 | 0.023 | 1.8 | 0.046 | 3.5 | 0.052 | 4.0 | 40 | 20 |
| Site 2 | 0.08 | Negative | 0.006 | 7.6 | 0.003 | 4.3 | 0.007 | 8.7 | 40 | 20 |
| | 6.66 | HIV-1 | 0.169 | 2.5 | 0.163 | 2.5 | 0.235 | 3.5 | 40 | 20 |
| | 4.39 | HIV-2 | 0.060 | 1.4 | 0.100 | 2.3 | 0.117 | 2.7 | 40 | 20 |
| | 1.39 | HIV-1 | 0.045 | 3.2 | 0.035 | 2.5 | 0.057 | 4.1 | 40 | 20 |
| Site 3 | 0.07 | Negative | 0.003 | 4.4 | 0.004 | 5.6 | 0.005 | 7.1 | 40 | 20 |
| | 6.22 | HIV-1 | 0.085 | 1.4 | 0.131 | 2.1 | 0.156 | 2.5 | 40 | 20 |
| | 4.42 | HIV-2 | 0.033 | 0.7 | 0.158 | 3.6 | 0.162 | 3.7 | 40 | 20 |
| | 1.34 | HIV-1 | 0.028 | 2.1 | 0.034 | 2.5 | 0.044 | 3.3 | 40 | 20 |

* Repeatability: Variability of the test performance from replicate to replicate.

** Between Day: Variability of the test performance from day to day.

*** Total: Variability of the test combining the effects of repeatability and between day.

Precision was further evaluated incorporating between site and between lot variation. The study was performed at three external sites using three reagent lots. At least three replicates each of a four member panel were tested on a single

occasion per day on six different days. The between site, between lot, and total precision estimates CV (%) were derived from a variance component analysis. The data shown in the table were rounded following all calculations.

| Mean VITROS Anti-HIV 1+2 Test Results (s/c) | | Between Site* | | Between Lot** | | Total*** | | |
|---|---|---|---|---|---|---|---|---|
| | | S.D. | C.V. (%) | S.D. | C.V. (%) | S.D. | C.V. (%) | No. Observ. |
| 0.10 | Negative | 0.004 | 3.9 | 0.031 | 30.0 | 0.034 | 32.0 | 162 |
| 1.09 | HIV-2 | 0.000 | 0.0 | 0.152 | 14.0 | 0.164 | 15.0 | 162 |
| 1.34 | HIV-1 | 0.000 | 0.0 | 0.017 | 1.3 | 0.070 | 5.2 | 162 |
| 3.76 | HIV-1 | 0.049 | 1.3 | 0.210 | 5.6 | 0.275 | 7.3 | 162 |

* Between Site: Variability of the test performance from site to site.

** Between Lot: Variability of the test performance from lot to lot calculated using data across all sites.

*** Total: Variability of the test incorporating factors of site, lot and day.

### *VITROS 3600 Immunodiagnostic System and VITROS 5600 Integrated System*

Precision was evaluated consistent with NCCLS document EP5. [11] Two replicates each of 7 patient sample pools were tested on 2 separate occasions per day on at least 20 different days. The experiment was performed using 1 reagent lot on each system. The data presented are a representation of the product performance.

| System | Mean VITROS Anti-HIV 1+2 Test Results (s/c) | | Repeatability* | | Between Day** | | Total*** | | No. Observ. | No. Days |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | S.D. | C.V.(%) | S.D. | C.V.(%) | S.D. | C.V.(%) | | |
| ECi/ECiQ | 0.11 | Neg | 0.003 | 2.7 | 0.006 | 5.5 | 0.007 | 6.4 | 92 | 23 |
| | 0.57 | HIV-1 | 0.024 | 4.2 | 0.017 | 3.0 | 0.037 | 6.5 | 92 | 23 |
| | 1.00 | HIV-1 | 0.044 | 4.4 | 0.035 | 3.5 | 0.072 | 7.2 | 92 | 23 |
| | 1.98 | HIV-1 | 0.071 | 3.6 | 0.000 | 0.0 | 0.126 | 6.4 | 92 | 23 |
| | 0.80 | HIV-2 | 0.022 | 2.8 | 0.045 | 5.6 | 0.056 | 7.0 | 92 | 23 |
| | 1.24 | HIV-2 | 0.033 | 2.6 | 0.064 | 5.2 | 0.076 | 6.1 | 92 | 23 |
| | 2.55 | HIV-2 | 0.090 | 3.5 | 0.129 | 5.1 | 0.165 | 6.5 | 92 | 23 |
| 3600 | 0.14 | Neg | 0.007 | 5.0 | 0.004 | 2.9 | 0.010 | 7.1 | 92 | 23 |
| | 0.60 | HIV-1 | 0.017 | 2.8 | 0.026 | 4.3 | 0.042 | 7.0 | 92 | 23 |
| | 1.04 | HIV-1 | 0.036 | 3.5 | 0.053 | 5.1 | 0.074 | 7.1 | 92 | 23 |
| | 2.01 | HIV-1 | 0.062 | 3.1 | 0.092 | 4.6 | 0.136 | 6.8 | 92 | 23 |
| | 0.81 | HIV-2 | 0.019 | 2.4 | 0.042 | 5.2 | 0.052 | 6.4 | 92 | 23 |
| | 1.30 | HIV-2 | 0.032 | 2.5 | 0.057 | 4.4 | 0.071 | 5.5 | 92 | 23 |
| | 2.67 | HIV-2 | 0.060 | 2.3 | 0.125 | 4.7 | 0.160 | 6.0 | 92 | 23 |
| 5600**** | 0.13 | Neg | 0.007 | 5.4 | 0.007 | 5.4 | 0.011 | 8.5 | 92 | 23 |
| | 0.60 | HIV-1 | 0.017 | 2.8 | 0.013 | 2.2 | 0.037 | 6.2 | 92 | 23 |
| | 1.03 | HIV-1 | 0.038 | 3.7 | 0.045 | 4.4 | 0.064 | 6.2 | 92 | 23 |
| | 1.97 | HIV-1 | 0.070 | 3.6 | 0.041 | 2.1 | 0.110 | 5.6 | 92 | 23 |
| | 0.84 | HIV-2 | 0.035 | 4.1 | 0.066 | 7.9 | 0.079 | 9.4 | 92 | 23 |
| | 1.35 | HIV-2 | 0.029 | 2.2 | 0.084 | 6.2 | 0.100 | 7.4 | 92 | 23 |
| | 2.72 | HIV-2 | 0.063 | 2.3 | 0.153 | 5.6 | 0.186 | 6.8 | 92 | 23 |

* Repeatability: Variability of the test performance from replicate to replicate.

** Between Day: Variability of the test performance from day to day.

*** Total: Variability of the test combining the effects of repeatability and between day.

**** Performance characteristics for the VITROS 5600 System are applicable to the VITROS XT 7600 System.

### *Percent Agreement Between VITROS ECi/ECiQ Immunodiagnostic Systems, VITROS 3600 Immunodiagnostic System and VITROS 5600 Integrated System*

Percent agreement was evaluated in a method comparison study using 720 serum and plasma patient samples (491 negative, 169 HIV-1 reactive, 60 HIV-2 reactive). Results from samples analyzed on the VITROS ECi/ECiQ Immunodiagnostic System were compared with those analyzed on the VITROS 3600 Immunodiagnostic System and the VITROS 5600 Integrated System at 3 different sites with a single reagent lot. Positive and negative percent confidence intervals were determined using an exact confidence interval analysis.

**Percent Agreement of Samples to VITROS ECi/ECiQ Immunodiagnostic**

| System | Positive Agreement (≥1.00 s/c) | 95% Confidence Intervals | Negative Agreement (<1.00 s/c) | 95% Confidence Intervals |
|---|---|---|---|---|
| 5600* | 94.76 % | 92.82–96.30% | 98.98% | 98.33–99.43% |
| 3600 | 94.32% | 92.32–95.93% | 98.91% | 98.24–99.38% |

* Performance characteristics for the VITROS 5600 System are applicable to the VITROS XT 7600 System.

## References

1. Summers M et al. Luminogenic Reagent Using 3-Chloro 4-Hydroxy Acetanilide to Enhance Peroxidase/Luminol Chemiluminescence. *Clin Chem*. 41:S73; 1995.
2. CDC-NIH. Biosafety in Microbiological and Biomedical Laboratories – 3rd Edition. HHS Publication No. (CDC) 93-8395. U.S. Government Printing Office, Washington, D.C., 1993.
3. CLSI. *Protection of laboratory Workers from Occupationally Acquired Infections; Approved Guideline- Fourth Edition*. CLSI Document M29-A4. Wayne, PA: Clinical and Laboratory Standards Institute; 2014.
4. Regulation (EC) No 1272/2008 of the European Parliament and of the Council of 16 December 2008 on classification, labelling and packaging of substances and mixtures, amending and repealing Directives 67/548/EEC and 1999/45/EC, and amending Regulation (EC) No 1907/2006.
5. Calam RR. Specimen Processing Separator Gels: An Update. *J Clin Immunoassay*. 11:86–90; 1988.
6. CLSI. *Collection of Diagnostic Venous Blood Specimens. 7th ed.*CLSI standard GP41.Wayne, PA: Clinical and Laboratory Standards Institute; 2017.
7. NCCLS. *Procedures and Devices for the Collection of Diagnostic Capillary Blood Specimens; Approved Standard – Fifth Edition*. NCCLS document H4-A5 [ISBN 1-56238-538-0]. CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898 USA, 2004.
8. CLSI. Procedures for the Handling and Processing of Blood Specimens; Approved Guideline – Third Edition. CLSI document H18-A3 (ISBN 1-56238-555-0). CLSI, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087, 1999.
9. CLSI. *Statistical Quality Control for Quantitative Measurements: Principles and Definitions; Approved Guideline - Third Edition*. CLSI document C24-A3 [ISBN 1-56238-613-1]. CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898 USA, 2006.
10. Levinson SS. The Nature of Heterophilic Antibodies and Their Role in Immunoassay Interference , J Clin Immunoassay 15: 108-115 (1992).
11. NCCLS. *Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline - Second Edition*. NCCLS document EP5-A2 [ISBN 1-56238-542-9]. CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898 USA, 2004.

## Glossary of Symbols

The following symbols may have been used in the labeling of this product.

| | | |
|---|---|---|
| Do Not Reuse | Upper Limit of Temperature | Range |
| Use by or Expiration Date (Year-Month-Day) | Lower Limit of Temperature | Range of Means |
| LOT Batch Code or Lot Number | Temperature Limitation | Midpoint |
| SN Serial Number | Consult Instructions for Use | Revised |
| REF Catalog Number or Product Code | Attention: The Instructions for Use (IFU) has been updated | Supersedes |
| Caution | S1 For use in Slide Supply 1 | Contains Sufficient for "n" Tests |
| Manufacturer | S2 For use in Slide Supply 2 | IVD *in vitro* Diagnostic Medical Device |
| Date of Manufacture | SI SI Units | Der Grüne Punkt (the Green Dot). Manufacturer follows certain packaging material waste disposal management regulations |
| EC REP Authorized Representative in the European Community | CONV Conventional Units | Estimated within-lab SD |
| | Value | |
| Corrosive | Flammable | Serious Health Hazards |
| Health Hazards | Acute Toxicity | Environmental or Aquatic Toxicity |

## Revision History

| Date of Revision | Version | Description of Technical Changes* |
|---|---|---|
| 2017-10-03 | 16.0-Draft | • Added information for the VITROS XT 7600 Integrated System<br>• Minor formatting and wording updates<br>• References: updated<br>• Glossary of Symbols: updated |

* The change bars indicate the position of a technical amendment to the text with respect to the previous version of the document.

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