

**E****ABBOTT PRISM HIV Ag/Ab Combo****REF** 7G46-48

77-4254/R6

**B7G460****Read Highlighted Changes**  
Revised February, 2008

# ABBOTT PRISM HIV Ag/Ab Combo

Human Immunodeficiency Virus Type 1 and 2 (*E. coli*, *B. megaterium*, Recombinant)  
Antigen, HIV p24 Antibody (mouse, monoclonal) and Synthetic Peptide

## Customer Service


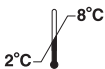
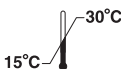



For additional product information, please contact your local customer service organization.

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

**NOTE:** If you receive reagents, calibrators, controls, or bulk solutions that are in a condition contrary to the package insert or label recommendation, or that are damaged, contact your local customer service organization.

For use with software version 3.11 or higher

## Key to symbols used

<b>REF</b>	List Number	<b>MASTER LOT</b>	Master Lot
<b>IVD</b>	<i>In Vitro</i> Diagnostic Medical Device	<b>SAMPLE CUPS</b>	Sample Cups
<b>LOT</b>	Lot Number	<b>CALIBRATORS</b>	Calibrators
	Expiration Date	<b>PIPETTE TIPS</b>	Pipette Tips
	Store at 2-8°C	<b>ASSAY KIT CARD</b>	Assay Kit Card
	Store at 15-30°C	<b>REACTION TRAYS</b>	Reaction Trays
	CAUTION: Consult accompanying documents.	<b>REAGENT COMPONENTS</b>	Reagent Components
	Manufacturer	<b>RUN CONTROL ADAPTERS</b>	Run Control Adapters
			Consult instructions for use

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

## NAME AND INTENDED USE

The ABBOTT PRISM HIV Ag/Ab Combo assay is an *in vitro* chemiluminescent immunoassay (ChLIA) for the simultaneous qualitative detection of HIV p24 antigen and/or antibodies to human immunodeficiency virus type 1 and/or type 2 (HIV-1/HIV-2) in human serum or plasma. The ABBOTT PRISM HIV Ag/Ab Combo assay is intended to be used as a screening test for donated blood and plasma to prevent transmission of HIV-1/HIV-2 to recipients of blood and blood components, and as an aid in the diagnosis of HIV-1/HIV-2 infection. The ABBOTT PRISM HIV Ag/Ab Combo assay does not discriminate between HIV p24 antigen, HIV-1 antibody, or HIV-2 antibody reactivity.

## SUMMARY AND EXPLANATION OF TEST

Acquired immunodeficiency syndrome (AIDS) is caused by two types of human immunodeficiency viruses, collectively designated as HIV.<sup>1-7</sup> HIV is the etiologic agent of AIDS.<sup>1,3,6,7</sup> HIV is transmitted by sexual contact, exposure to blood or blood products, and prenatal or perinatal infection of a fetus or newborn, respectively.<sup>8</sup> Antibodies against HIV are nearly always detected in AIDS patients and HIV infected asymptomatic individuals.<sup>8,9</sup> and HIV infection is always detected in AIDS patients and seropositive individuals by culture or amplification of viral RNA and/or proviral DNA.<sup>8,10</sup>

Phylogenetic analysis classifies HIV-1 into groups M (major), N (non-M, non-O), and O (outlier).<sup>4,5</sup> Group M viruses have spread throughout the world to cause the global AIDS pandemic. In contrast, groups N and O are relatively rare and endemic to west central Africa.<sup>11-17</sup> However, group O infections have been identified in Europe and the USA.<sup>18-22</sup> HIV-1 group M is composed of genetic subtypes (A, B, C, D, F, G, H, J, and K) and circulating recombinant forms (CRFs).<sup>5,23</sup> The geographic distribution and regional predominance of HIV-1 subtypes and CRFs vary. All subtypes and many recombinant strains exist in Africa with CRF02\_AG the predominant strain in west and west central Africa, subtypes A, C, and D predominant in east central Africa, and subtype C predominant in southern Africa.<sup>23-28</sup> HIV-1 subtype B is the predominant subtype in the USA, Europe, Japan, and Australia. However, a significant percentage of new HIV-1 infections in Europe are caused by non-B subtypes.<sup>29,30</sup> In Asia, subtype C is found in India, and CRF01\_AE (formerly called subtype E) and subtype B are found in Thailand and southeast Asia.<sup>31</sup> South America predominantly has subtypes B and F.<sup>32,33</sup>

Human immunodeficiency virus type 2 (HIV-2) is similar to HIV-1 in its structural morphology, genomic organization, cell tropism, *in vitro* cytopathogenicity, transmission routes and ability to cause AIDS.<sup>6-8</sup> However, HIV-2 is less pathogenic than HIV-1, and HIV-2 infections have a longer latency period with slower progression to disease, lower viral titers, and lower rates of vertical and horizontal transmission.<sup>34-37</sup> HIV-2 is endemic to west Africa but HIV-2 infections, at a low frequency compared to HIV-1, have been identified in the USA, Europe, Asia, and other regions of Africa.<sup>31,37</sup> HIV-2 is classified into genetic subtypes A-G with most infections caused by subtypes A and B.<sup>38,39</sup>

The key immunogenic protein and antigenic target for serodetection of HIV infection is the viral (HIV) transmembrane protein (TMP). Antibodies against the TMP (anti-TMP) consistently are among the first to appear at seroconversion of HIV infected individuals.<sup>9,40-44</sup> The anti-TMP response remains relatively strong throughout the course of the disease, as evidenced by the near universal presence of antibodies against the TMP in asymptomatic and symptomatic stages of HIV infection.<sup>9,40-44</sup> TMPs from HIV-1 groups M and O and HIV-2 are represented in ABBOTT PRISM HIV Ag/Ab Combo reagents by three pairs of recombinant antigens and a synthetic peptide derived from native TMP sequences. The rationale for including three pairs of TMPs is derived from the genetic diversity within HIV-1 and between HIV-1 and HIV-2.<sup>4,5,45,46</sup> Serologic studies indicate that although HIV-1 and HIV-2 share multiple common epitopes in their core antigens, the envelope glycoproteins are much less cross-reactive.<sup>7,47-51</sup> Antibodies elicited against the TMP (or portions of the TMP) of a viral strain within one group or type may or may not react with the TMP (or portions of the TMP) from a viral strain of a different group or type.<sup>15,52-57</sup> An exception may be antibodies elicited against HIV-1 group N.<sup>11,12</sup>

Early after infection with HIV, prior to seroconversion, HIV antigen(s) may be detected in serum or plasma specimens.<sup>57-65</sup> The HIV structural protein most often used as the marker of antigenemia is the core protein, p24. The ABBOTT PRISM HIV Ag/Ab Combo assay uses anti-HIV p24 in the reagents to detect HIV p24 antigen prior to seroconversion, thereby decreasing the seroconversion window and improving early detection of HIV infection.

## BIOLOGICAL PRINCIPLES OF THE PROCEDURE

ABBOTT PRISM HIV Ag/Ab Combo is a two-step sandwich ChLIA to determine the presence of HIV p24 antigen and antibodies to HIV-1 and HIV-2 in human serum and plasma. The reactions occur in the following sequence:

- A reaction mixture is formed by combining specimen diluent, sample, and microparticles in the incubation well of the reaction tray. HIV p24 antigens and HIV-1/HIV-2 antibodies present in the sample bind to the HIV-1/HIV-2 antigen (recombinant) and HIV p24 antibody (mouse, monoclonal) coated microparticles.
- After the first incubation is complete, the reaction mixture is transferred to the glass fiber matrix of a reaction tray using the transfer wash. The microparticles are captured on the matrix with the unbound materials being washed through the matrix.
- After washing, the HIV p24 antigen and HIV-1/HIV-2 antibodies bind to acridinium labeled conjugates (HIV-1/HIV-2 antigens [recombinant], HIV-1 synthetic peptide, and HIV p24 antibodies [mouse, monoclonal]) added in the second step.
- A chemiluminescent signal is generated by addition of an alkaline hydrogen peroxide solution and the resulting photons are counted.

The amount of light emitted is proportional to the amount of HIV p24 antigen or HIV-1/HIV-2 antibodies in the sample. For further information on ChLIA technology, refer to the ABBOTT PRISM Operations Manual, Section 3. The presence or absence of HIV p24 antigen or HIV-1/HIV-2 antibodies in a sample is determined by comparing the number of photons collected from the sample to a cutoff value determined from a calibration performed in the same batch.

If the number of photons collected from a test sample is less than the cutoff value, the sample is considered nonreactive for HIV p24 antigen or HIV-1/HIV-2 antibodies by the criteria of the ABBOTT PRISM HIV Ag/Ab Combo assay. These specimens need not to be further tested.

If the number of photons collected from a test sample is greater than or equal to the cutoff value, the sample is considered reactive for HIV p24 antigen or HIV-1/HIV-2 antibodies by the criteria of the ABBOTT PRISM HIV Ag/Ab Combo assay. Specimens which are initially reactive should be handled according to the table in the **SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS** section and retested in duplicate. Reactivity in either or both of these tests (i.e., repeatedly reactive) is highly predictive of the presence of HIV p24 antigen, anti-HIV-1, and/or anti-HIV-2 in individuals at risk for HIV infection. However, as with all immunoassays, the ABBOTT PRISM HIV Ag/Ab Combo assay may yield nonspecific reactions due to other causes, particularly when testing low prevalence populations (e.g., blood donors). A specimen which is found repeatedly reactive should be investigated further by sensitive supplemental HIV-specific tests, such as one or more of the following: immunoblots, antigen tests, HIV nucleic acid tests.

Supplemental testing of repeat reactive specimens obtained from individuals at risk for HIV infection usually confirms the presence of HIV antigen or HIV antibodies, and HIV nucleic acid. A full differential diagnostic workup for the diagnosis of AIDS and AIDS-related conditions necessarily includes an examination of the patient's immune status and clinical history.

## REAGENTS

**NOTE: Each specific component description in this section is accompanied by a unique symbol. These symbols appear on both the component labels and on the corresponding instrument tubing identifier labels. They are meant to facilitate identification and installation of reagent bottles within the ambient reagent bay and refrigerator.**

ABBOTT PRISM HIV Ag/Ab Combo Assay Kit (7G46-48)

- **[MICROPARTICLES]** 1 Bottle (324 mL) HIV-1/HIV-2 antigen (*E. coli* and *B. megaterium*, recombinant) and HIV p24 antibody (mouse, monoclonal) coated microparticles in TRIS buffered saline. Minimum concentration: 0.08% solids. Preservative: sodium azide. (Symbol: ●)
- **[CONJUGATE 20X CONC]** 1 Bottle (16 mL) Conjugate 20x Concentrate. Conjugates are acridinium-labeled HIV-1/HIV-2 antigens (*E. coli*, recombinant), acridinium-labeled HIV-1 synthetic peptide, and acridinium-labeled HIV p24 antibodies (mouse, monoclonal) in phosphate buffered saline with Triton X-100. Minimum concentration: 0.8 µg/mL. Preservative: ProClin 300. (No Symbol).

and

**CONJUGATE DILUENT** 1 Bottle (306 mL) Conjugate Diluent. Phosphate buffered saline with Triton X-405 and protein (bovine) stabilizers. Preservative: ProClin 300. (Symbol: ▲)

**NOTE: Conjugate 20x Concentrate MUST be mixed with Conjugate Diluent prior to use. Refer to the PREPARATION OF DILUTED CONJUGATE section for details.**

- **SPECIMEN DILUENT** 1 Bottle (324 mL) Specimen Diluent. Citrate/phosphate buffer with Triton X-100, Tween 20, protein (bovine) stabilizer, and 10% calf serum. Preservative: ProClin 300. (Symbol: X).
- **CAL -** 3 Bottles (10.4 mL each) Negative Calibrator (Human). Recalcified plasma, nonreactive for HBsAg, HIV-1 RNA or HIV-1 Ag, anti-HCV, and anti-HIV-1/HIV-2. Preservative: sodium azide. (Symbol: NC)
- **CAL +** 3 Bottles (10.4 mL each) Positive Calibrator (Human). Chemically and physically inactivated purified HIV viral lysate in TRIS buffered saline with protein (bovine) stabilizer. Preservative: sodium azide. (Symbol: PC)

#### Standardization:

To maintain a consistent HIV-1 p24 antigen sensitivity of < 50 pg/mL, the ABBOTT PRISM HIV Ag/Ab Combo Positive Calibrator has a HIV-1 p24 antigen concentration matched against the international standard Agence française de sécurité sanitaire des produits de santé (AFSSAPS) panel.

- **CONTROL +** 3 Bottles (10.4 mL each) HIV-1 Positive Assay Control (Human). Recalcified, heat-inactivated plasma reactive for anti-HIV-1, nonreactive for HBsAg, HIV-1 RNA or HIV-1 Ag, and anti-HCV. Preservative: sodium azide. (Symbol: PC1)

#### ABBOTT PRISM HIV Ag/Ab Combo Wash Kit (7G46-38)

- **TRANSFER WASH** 1 Bottle (3360 mL) Transfer Wash. Borate buffered saline with Tween 20. Preservative: sodium azide. (Symbol: ~)
- **CONJUGATE WASH** 1 Bottle (3615 mL) Conjugate Wash. TAPS/TRIS buffer with Triton X-100. Preservative: sodium azide. (Symbol: ★) Important: Handle Conjugate Wash Bottle carefully to avoid spillage.

#### OTHER REAGENTS REQUIRED

##### ABBOTT PRISM Activator Concentrate (1A75-02 or 3L27-02)

- **ACTIVATOR CONCENTRATE** 4 Bottles (900 mL each) Activator Concentrate. 0.4% hydrogen peroxide/0.06% diethylenetriaminepentaacetic acid.

##### ABBOTT PRISM Activator Diluent (1A75-01 or 3L27-01)

- **ACTIVATOR DILUENT** 4 Bottles (900 mL each) Activator Diluent. 0.3 N sodium hydroxide.

#### OTHER REAGENTS AVAILABLE

##### ABBOTT PRISM Run Control Kit (2K24-10)


##### ABBOTT PRISM Positive Run Control Kit (2K24-11)

**NOTE:** Each batch **MUST** end in a release control. An HIV Ag/Ab Combo release control is any control reactive for anti-HIV-1, anti-HIV-2, or both, which has been configured to validate system function and release sample results. The configuration criteria are defined in the Run Controls file of the PRISM Resource Management software. Any customer specified control reactive for anti-HIV-1, anti-HIV-2, or both may be used.

#### WARNINGS AND PRECAUTIONS

- **IVD** For *In Vitro* Diagnostic Use.
- Failure to adhere to instructions in the ABBOTT PRISM Operations Manual or package insert may result in erroneous results.

#### SAFETY PRECAUTIONS

-  **CAUTION:** This product contains human sourced infectious and/or potentially infectious components. Refer to the REAGENTS section of this package insert. No known test method can offer complete assurance that products derived from human sources or inactivated microorganisms will not transmit infection. Therefore, it is recommended that all human sourced materials be considered potentially infectious and handled with appropriate biosafety practices.

- The ABBOTT PRISM Line Cleaner contains 2% tetraethylammonium hydroxide (TEAH), which may cause mild eye irritation. If this solution comes in contact with eyes, flush immediately with water (for additional information, refer to the ABBOTT PRISM Operations Manual, Section 8).
- Conjugate 20x Concentrate, Conjugate Diluent, and Specimen Diluent contain methylisothiazolines, which are components of ProClin and 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.

- The ABBOTT PRISM Activator Diluent contains sodium hydroxide and is classified per applicable European Community (EC) Directives as: Irritant (Xi). The following are the appropriate Risk (R) and Safety (S) phrases.



- R36 Irritating to eyes.
- S26 In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.
- S35 This material and its container must be disposed of in a safe way.
- S46 If swallowed, seek medical advice immediately and show this container or label.

- This product contains sodium azide; for a specific listing, refer to the **REAGENTS** section. Contact with acids liberates very toxic gas. This material and its container must be disposed of in a safe way.
- For product not classified as dangerous per European Directive 1999/45/EC as amended - Safety data sheet available for professional user on request.

#### HANDLING PRECAUTIONS

- Do not use kits beyond the expiration date.
- **Gently** invert each component several times prior to loading on the ABBOTT PRISM System to ensure a homogenous solution. Additional gentle inversion may be required to thoroughly resuspend microparticles. Avoid foaming. Each component of the ABBOTT PRISM HIV Ag/Ab Combo Wash Kit should be at room temperature (15 to 30°C) before mixing.
- Do not mix reagents from different bottles. Do not mix reagents from different assay kit lots.
- Any lot of ABBOTT PRISM HIV Ag/Ab Combo Wash Kit can be used with any lot of ABBOTT PRISM HIV Ag/Ab Combo Assay Kit.
- Avoid microbial and chemical contamination of samples, reagents and equipment. The use of disposable pipette tips is recommended for any preliminary sample transfer.
- Do not freeze reagents.
- Use caution when handling samples, reagent bottles and reagent caps to prevent cross contamination.

Additional safety and handling precautions and limitations for the assay kit, calibrators, specimens, controls, and other reagents are described in the ABBOTT PRISM Operations Manual, Section 7.

#### PREPARATION OF DILUTED CONJUGATE

1. Carefully empty the entire contents of the Conjugate 20x Concentrate dropper bottle (small bottle) into the larger Conjugate Diluent bottle by slowly squeezing the dropper bottle while maintaining the dropper tip within the opening of the Conjugate Diluent bottle (large bottle). Squeeze the dropper bottle repeatedly until empty. Avoid foaming.
2. Write the date of dilution and lot number of the Conjugate 20x Concentrate on the label of Conjugate Diluent, in the spaces provided.

**NOTE:** The diluted conjugate must be used within 56 days of preparation.



3. Reseal the large bottle and mix thoroughly by slowly inverting several times. Do not vortex.
4. Place in the onboard refrigerator. Refer to the ABBOTT PRISM Operations Manual, Section 5, under *Prepare and Load Reagents*, for additional information.

### PREPARATION OF ACTIVATOR SOLUTION

Activator Solution must be prepared daily by mixing equal parts of Activator Concentrate and Activator Diluent. The volume of Activator Solution required for multiple tests is calculated by the ABBOTT PRISM software. Refer to the ABBOTT PRISM Operations Manual, Section 5, under *Plan Workload*, for additional information. Use clean pipettes and/or metal-free containers (such as plasticware or acid-washed and purified water-rinsed glassware) to measure. Prepare in the bottle provided in the ABBOTT PRISM Accessory Kit. Cover the bottle opening securely with the cap provided and invert gently five to ten times to mix. Load the Activator Solution on the ABBOTT PRISM System. Refer to the ABBOTT PRISM Operations Manual, Section 5, under *Prepare and Load Activator Solution*, for additional information.

**NOTE: The Activator Solution must be used within 24 hours of preparation.**

### STORAGE INSTRUCTIONS

-  2°C - 8°C Store the ABBOTT PRISM HIV Ag/Ab Combo Assay Kit, ABBOTT PRISM Run Control Kit, ABBOTT PRISM Positive Run Control Kit, and ABBOTT PRISM Activator Concentrate at 2 to 8°C.
-  15°C - 30°C Store the ABBOTT PRISM HIV Ag/Ab Combo Wash Kit and ABBOTT PRISM Activator Diluent at room temperature (15 to 30°C).
- Store ABBOTT PRISM Pipette Tips and ABBOTT PRISM Reaction Trays in their original package until use.

### INDICATIONS OF INSTABILITY OR DETERIORATION OF REAGENTS

The ABBOTT PRISM System will not continue to process samples when calibrator or positive control values do not meet specifications. This may indicate either deterioration or contamination of reagents, or instrument failure. Refer to the ABBOTT PRISM Operations Manual, Section 10, for additional information.

### INSTRUMENT PROCEDURE

- Refer to the ABBOTT PRISM Operations Manual for a detailed description of Instrument Procedures.
- Solutions required for instrument cleaning and maintenance are described in detail in the ABBOTT PRISM Operations Manual, Sections 5 and 9.
- For optimal performance, it is important to follow the routine maintenance procedures defined in the ABBOTT PRISM Operations Manual, Section 9.

### SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS

- Either human serum (including serum collected in serum separator tubes) or plasma collected in potassium EDTA, kaolin, potassium oxalate, sodium citrate, ACD, CPD, or CPDA-1 anticoagulants may be used with the ABBOTT PRISM HIV Ag/Ab Combo assay. Follow the manufacturer's processing instructions for serum and plasma collection tubes.

**NOTE: Do not use specimens collected in heparin as it may cause a reduction in the number of photons in the test sample and in the S/CO (Sample/Cutoff) for the ABBOTT PRISM HCV assay; therefore, heparin is not recommended for any PRISM assay.**

- The PRISM does not provide the capability to verify specimen type. It is the responsibility of the operator to verify the correct specimen types are used in the PRISM HIV Ag/Ab Combo assay.
- Use caution when handling patient specimens to prevent cross-contamination. Use of disposable pipettes or pipette tips is recommended.
- Performance has not been established using cadaver specimens or body fluids other than human serum or plasma.

- The use of specimens with obvious microbial contamination should be avoided.
- The use of specimens with gross lipemia or gross hemolysis should be avoided.
- Ensure that complete clot formation in serum specimens has taken place prior to centrifugation. Some specimens, especially those from patients receiving anticoagulant or thrombolytic therapy may exhibit increased clotting time. If the specimen is centrifuged before a complete clot forms, the presence of fibrin may cause erroneous results or aspiration errors.
- This assay was designed for use with human serum or plasma from individual donor specimens. This assay has not been validated for use with pooled specimens.
- Do not use heat-inactivated specimens.
- Specimens collected by plasmapheresis, which have not been frozen, do not require centrifugation. All other specimens (including previously frozen plasmapheresis specimens) must be centrifuged as follows:

**Nonfrozen specimens** (excluding plasmapheresis specimens) must be centrifuged such that g-minutes (the product of relative centrifugal force [RCF] and centrifugation time [minutes]) is between 30000 and 75000. The following chart lists examples of acceptable time and force ranges that meet this criterion.

Time (minutes)	RCF (x g)	RCF x Time (g-minutes)
10	3000	30000
15	2000 - 3000	30000 - 45000
20	1500 - 3000	30000 - 60000
25	1300 - 3000	32500 - 75000

Convert rpm to RCF as follows:  $RCF = 1.12 \times r_{max} (rpm/1000)^2$

Convert RCF to rpm as follows:  $rpm = 1000 \times \sqrt{\frac{RCF}{1.12 \times r_{max}}}$

- RCF - The relative centrifugal force generated during centrifugation.
- rpm - The rotation per minute of the rotor on which the specimens are being spun (usually the digital readout on the centrifuge will indicate rpm).
- Time - Centrifugation time should be measured from the time the rotor reaches the required RCF or rpm to the time it begins decelerating.
- $r_{max}$  - Radius of the rotor in millimeters. The radius measured is dependant on whether the rotor is a fixed angle rotor or a swinging bucket rotor. This value is typically provided with the rotor, by the manufacturer.
- g-minutes - The unit of measure for the product of RCF (x g) and time (minutes).

**NOTE:** For fixed angle,  $r_{max}$  is a measure of the distance from the rotor axis (center) to the bottom of the tube cavity. For the swinging bucket,  $r_{max}$  is a measure of the distance from the rotor axis (center) to the bottom of the tube bucket while it is extended during rotation.

**Previously frozen specimens** must be centrifuged such that g-minutes (the product of relative centrifugal force [RCF] and centrifugation time [minutes]) is between 180000 and 300000. The following chart lists examples of acceptable time and force ranges that meet this criterion.

Time (minutes)	RCF (x g)	RCF x Time (g-minutes)
15	12000	180000
20	9000 - 12000	180000 - 240000
25	7200 - 12000	180000 - 300000

**ANY specimen** (excluding nonfrozen plasmapheresis) not tested within 24 hours of initial centrifugation, must be recentrifuged from 30000 to 75000 (g-minutes) as defined for nonfrozen specimens.

**NOTE:** Specimens retested within 24 hours of initial centrifugation do not require recentrifugation.



## FAILURE TO FOLLOW THE SPECIFIED CENTRIFUGATION PROCEDURE MAY GIVE ERRONEOUS OR INCONSISTENT RESULTS.

- Specimens may be stored for up to 7 days at 2 to 8°C. If storage periods greater than 7 days are anticipated, the serum or plasma should be removed from the clot or red blood cells to avoid hemolysis. Serum or plasma may be stored for up to 28 days frozen (-10°C or colder).
- Specimens must be mixed after thawing by inverting specimens 10 times. Visually inspect the specimens for the absence of stratification. If layering or stratification is observed, repeat until specimens are visibly homogeneous. Centrifuge in accordance to the table in this section prior to use.
- Clear, nonhemolyzed specimens should be used when possible. Specimens containing particulate matter may give erroneous or inconsistent test results.
- 22 nonreactive and 23 reactive specimens showed no qualitative performance differences when subjected to 3 freeze/thaw cycles. However, specimens that have undergone multiple freeze/thaw cycles or have been stored frozen for prolonged periods may give erroneous results.
- No qualitative performance differences were observed when nonreactive and reactive specimens were spiked with elevated levels of bilirubin ( $\leq 20$  mg/dL), hemoglobin ( $\leq 500$  mg/dL), red blood cells ( $\leq 0.4\%$  v/v), triglycerides ( $\leq 3000$  mg/dL), or protein ( $\leq 12$  g/dL).
- When shipped, specimens must be packaged and labeled in compliance with applicable regulations covering the transport of clinical specimens and infectious substances. Specimens may be shipped at 30°C or colder (ambient, refrigerated on wet ice, or frozen on dry ice). Prior to freezing, the specimen should be removed from the clot or red cells.

### SPECIMEN VOLUME

The specimen volume required to perform a single assay on the ABBOTT PRISM System varies according to the number and type of assays, and the different specimen containers. The ABBOTT PRISM HIV Ag/Ab Combo assay requires a 100  $\mu$ L sample dispense. For ABBOTT PRISM Sample Cups, the minimum specimen volume required for one ABBOTT PRISM HIV Ag/Ab Combo assay is 400  $\mu$ L. For primary or aliquot tubes, or additional assay volume requirements, refer to the ABBOTT PRISM Operations Manual, Section 5.

### ABBOTT PRISM HIV AG/AB COMBO PROCEDURE

#### MATERIALS PROVIDED

- 7G46-48 ABBOTT PRISM HIV Ag/Ab Combo Assay Kit
- 7G46-38 ABBOTT PRISM HIV Ag/Ab Combo Wash Kit

#### MATERIALS REQUIRED BUT NOT PROVIDED

- 1A75-02 or 3L27-02 ABBOTT PRISM **ACTIVATOR CONCENTRATE**
- 1A75-01 or 3L27-01 ABBOTT PRISM **ACTIVATOR DILUENT**
- 5A07-01 ABBOTT PRISM **REACTION TRAYS**
- 5A07-10 ABBOTT PRISM **PIPETTE TIPS**
- 6A36-31 ABBOTT PRISM **RUN CONTROL ADAPTERS**
- 6A36-60 ABBOTT PRISM Accessory Kit

#### ADDITIONAL MATERIALS AVAILABLE

- 1A75-10 or 3L27-10 ABBOTT PRISM **ACTIVATOR LINE TREATMENT**
- 2K24-10 ABBOTT PRISM Run Control Kit
- 2K24-11 ABBOTT PRISM Positive Run Control Kit
- 7A03-01 or 3L00-01 ABBOTT PRISM **PRIME/PURGE ACCESSORIES**
- 7A03-30 or 3L00-30 ABBOTT PRISM **PURGE CONCENTRATE**
- 7A03-31 ABBOTT PRISM **LINE CLEANER**
- 7B36-01 ABBOTT PRISM **SAMPLE CUPS**

### ABBOTT PRISM ASSAY PROCEDURE

For detailed information concerning batch time and maximum batch size, refer to the ABBOTT PRISM Operations Manual, Section 2.

#### STEP 1:

- Enter a Plan Workload (refer to the ABBOTT PRISM Operations Manual, Section 5).
- Replace reagents as needed.  
**NOTE:** Gently invert each component several times prior to loading on the ABBOTT PRISM System to ensure a homogeneous solution. Additional gentle inversion may be required to thoroughly resuspend microparticles. Avoid foaming. Each component of the ABBOTT PRISM HIV Ag/Ab Combo Wash Kit should be at room temperature (15 to 30°C) before mixing.
- Verify that all tubing label symbols match the symbols on each reagent label. (Refer to the symbol key in the **REAGENTS** section.)
- Verify that all tubing is securely fastened to the corresponding wash and reagent bottles.**

#### STEP 2:

- Inspect the waste containers. Empty and clean as defined in the ABBOTT PRISM Operations Manual, Section 9, if necessary.

#### STEP 3:

- Prepare Activator Solution (refer to the **PREPARATION OF ACTIVATOR SOLUTION** section of this package insert) and load into the ABBOTT PRISM System.

#### STEP 4:

- Verify an adequate number of Reaction Trays are in the Tray Loader.

#### STEP 5:

- Verify an adequate number of Pipette Tips are in the Pipette Tip Racks.

#### STEP 6:

- Perform the prime procedure (refer to the ABBOTT PRISM Operations Manual, Section 5).

#### STEP 7:

- Initiate sample processing. Open the bottles in the Calibrator Pack and place in the Calibrator Rack. Load the Calibrator Rack and Sample Racks, including the Controls (refer to the Control Handling Procedure under CONTROLS).**

#### STEP 8:

- After the calibrators have been pipetted, remove the Calibrator Rack. Close the calibrator bottles and return to 2 to 8°C storage.

#### STEP 9:

- Each sample is initially tested once. Sample Racks may be removed after the samples have been pipetted.

#### STEP 10:

- Any specimen (excluding nonfrozen plasmapheresis) that is initially reactive must be centrifuged according to the table in the SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS section and retested in duplicate (refer to the ABBOTT PRISM Operations Manual, Section 5).**

**NOTE:** Specimens retested within 24 hours of initial centrifugation do not require recentrifugation.

#### STEP 11:

- After the run is complete, perform the purge procedure (refer to the ABBOTT PRISM Operations Manual, Section 5).

Refer to the ABBOTT PRISM Operations Manual, Section 3, for a detailed description of ChLIA procedures.

### QUALITY CONTROL PROCEDURES

#### CALIBRATION

The ABBOTT PRISM HIV Ag/Ab Combo Calibrators (both Negative and Positive) and HIV-1 Positive Assay Control are automatically tested in triplicate at the beginning of each batch. The ABBOTT PRISM System will not generate results when calibrator or positive assay control values do not meet specifications. This may indicate deterioration or contamination of reagents or instrument failure.

## CONTROLS

1. A release control **MUST** be included as the last sample in each batch. A control such as the ABBOTT PRISM Positive Control or any customer-specified control reactive for anti-HIV-1 and/or anti-HIV-2 may be used. This control must test reactive in order to validate system function and to release results. If this control does not test reactive, refer to the ABBOTT PRISM Operations Manual, Section 10. Sites using a release control other than the ABBOTT PRISM Positive Control must validate its performance on the ABBOTT PRISM System.
2. Additional controls may be run at the operator's discretion. Validity specifications may be assigned such that if these controls fail, no results are reported for that assay batch.
3. Control Handling Procedure  
(ABBOTT PRISM Run Control Kit, ABBOTT PRISM Positive Run Control Kit, and Customer Specified Controls)
  - a. Determine the volume of controls required if using Sample Cups.  
The control volume required to perform a single assay on the ABBOTT PRISM System varies according to the different specimen containers. For ABBOTT PRISM Sample Cups, the minimum control volume required for one assay is 600 µL (400 µL + 200 µL sample cup dead volume). For every additional assay performed from the same control container, an additional 200 µL is required.  
For volume requirements in primary or aliquot tubes, refer to the ABBOTT PRISM Operations Manual, Section 5.  
Place one control barcode on each Negative and Positive Control Sample Cup.
  - b. Preparing Run Controls  
Place each Run Control bottle into an adapter such that when the flip-top cap is opened, it can be snapped into an open position within the adapter. Place each Run Control with adapter onto the Sample Rack. **The control can be placed in any rack position except 1, 2, 27, or 28.**
  - c. Place an ABBOTT PRISM Positive Control, or any other release control reactive for anti-HIV-1 or anti-HIV-2 **AFTER** the last sample in the Sample Rack. **The controls can be placed in any rack position except 1, 2, 27, or 28.**

## ASSAY PARAMETER SPECIFICATIONS

The PRISM assay parameter specifications have been factory set. These parameters cannot be printed, displayed, or edited.

## RESULTS

### CALCULATION OF CUTOFF AND S/CO VALUES

The ABBOTT PRISM System calculates the ABBOTT PRISM HIV Ag/Ab Combo assay cutoff value using the following formula:

$$\text{Cutoff} = \text{Mean Negative Calibrator Net Counts} + (0.360 \times \text{Mean Positive Calibrator Net Counts})^*$$

Example: If the Mean NCC = 200,  
and the Mean PCC = 4300,  
 $200 + (0.360 \times 4300) = 1748$   
Cutoff = 1748

- \* The Mean Net Counts of the Positive Calibrator, Negative Calibrator, and the HIV-1 Positive Assay Control are calculated using the two lowest values of the three replicates. An Instrument Code will be displayed in place of the count value for the third replicate. Each of the two remaining replicates of the Positive Calibrator and Negative Calibrator used to calculate the cutoff must meet all specifications.

The ABBOTT PRISM System calculates the ABBOTT PRISM HIV Ag/Ab Combo assay S/CO using the following formula:

$$\text{S/CO} = \text{Sample Net Counts} \div \text{Cutoff}$$

Example: If the Sample Net Counts = 7980,  
and the Cutoff = 1748,  
 $7980 \div 1748 = 4.56$   
S/CO = 4.56

## INTERPRETATION OF RESULTS

### ABBOTT PRISM HIV Ag/Ab Combo Retest Instructions

Initial Results (S/CO)	Interpretation	Retest Procedure
$\geq 1.00$	Reactive (R)	Retest in duplicate*
$< 1.00$	Nonreactive (NR)	No retest required

- \* All specimens (excluding nonfrozen plasmapheresis) that are reactive on initial testing must be centrifuged according to the table in the **SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS** section of this package insert and retested in duplicate.

- If repeat testing shows the S/CO to be  $\geq 1.00$  for either retest value, the specimen is considered repeatedly reactive.
- Repeatedly reactive specimens should be investigated further by sensitive supplemental HIV-specific tests for either antigen or HIV nucleic acids.
- For details on configuring the ABBOTT PRISM System to use Grayzone Interpretations, refer to the ABBOTT PRISM Operations Manual, Section 2.

## READING RESULTS

Some S/CO values may be flagged with "<" or ">" symbols. For more information on sample reports, refer to the ABBOTT PRISM Operations Manual, Section 5: *Operating Instructions, Reports*.

## SYSTEM ERRORS

For a description of the error codes that appear in the ABBOTT PRISM Report, refer to the ABBOTT PRISM Operations Manual, Section 10.

## LIMITATIONS OF THE PROCEDURE

- This assay was designed for use with human serum or plasma from individual patient and donor specimens. This assay has not been validated for use with pooled specimens.
- The ABBOTT PRISM HIV Ag/Ab Combo assay does not discriminate between HIV p24 antigen and HIV-1/HIV-2 antibody reactivity.
- The presence of HIV p24 antigen or HIV-1/HIV-2 antibodies is not a diagnosis of AIDS. It is recommended that repeatedly reactive specimens be investigated by supplemental testing. Individuals who are repeatedly reactive should be referred for medical evaluation which may include additional testing.
- Although the association of infectivity and the presence of HIV p24 antigen or HIV-1/HIV-2 antibodies is strong, it is recognized that presently available methods for HIV p24 antigen or HIV-1/HIV-2 antibody detection are not sensitive enough to detect all potentially infectious units of blood, plasma, or possible cases of HIV infection. A nonreactive test result does not exclude an infection.
- Because of possible nonspecific reactions due to causes other than HIV infection, particularly when testing low prevalence populations (e.g., blood donors), it is appropriate to further investigate specimens found to be repeatedly reactive by the ABBOTT PRISM HIV Ag/Ab Combo assay to demonstrate whether or not HIV antigen or antibody is indeed present. Supplemental testing of repeatedly reactive specimens obtained from individuals at risk for HIV infection usually confirms the presence of HIV antigen or HIV antibodies, and HIV nucleic acid. A full differential diagnostic workup for the diagnosis of AIDS and AIDS-related conditions necessarily includes an examination of the patient's immune status and clinical history.
- Performance has not been established using cadaver specimens or body fluids other than human serum or plasma.
- The use of specimens with obvious microbial contamination should be avoided.
- Specimens from patients who have received preparations of mouse monoclonal antibodies for diagnosis or therapy may contain human anti-mouse antibodies (HAMA). Such specimens may show either falsely elevated or depressed values when tested with assay kits that employ mouse monoclonal antibodies.<sup>66,67</sup> ABBOTT PRISM HIV Ag/Ab Combo reagents contain a component that reduces the effect of HAMA reactive specimens. Additional clinical or diagnostic information may be required to determine patient status.
- The use of specimens with gross lipemia or gross hemolysis should be avoided.
- Previously frozen specimens must be centrifuged per the **SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS** section of this package insert prior to running the assay.

## SPECIFIC PERFORMANCE CHARACTERISTICS

### PRECISION

The imprecision of the ABBOTT PRISM HIV Ag/Ab Combo assay is  $\leq 14\%$  for reactive samples in a study where a panel, consisting of five assay-specific reactive samples, three Run Control lots, and three calibrator lots (including the HIV-1 Positive Assay Control), was tested. The study was performed at three external sites using one instrument and one internal site using two instruments. All panel members were tested across two reagent lots at the external site and three reagent lots at the internal site. Each combination of instruments, panel members, and reagent lots was tested in four runs. Each panel member was tested in replicates of six using three replicates for each subchannel. The intra-run and inter-run standard deviation (SD) and percent coefficient of variation (%CV) were analyzed with a variance components analysis<sup>68</sup> using a mixed analysis of variance model.<sup>69</sup> The data from the study are summarized in Table I.\*

**Table I**  
**ABBOTT PRISM HIV Ag/Ab Combo Precision**

Panel Member	n	Mean S/CO	Intra-run SD	%CV	Inter-run SD	%CV
1 (High Anti-HIV-1)	288	7.69	0.344	4.47	0.456	5.93
2 (Low Anti-HIV-1)	288	2.30	0.110	4.78	0.150	6.51
3 (HIV-1 gO)	288	2.17	0.086	3.97	0.119	5.48
4 (Anti-HIV-2)	288	2.83	0.176	6.22	0.208	7.34
5 (HIV-1 p24 Ag)	288	2.48	0.142	5.72	0.152	6.11
Calibrators and Controls	n	Mean S/CO	Intra-run SD	%CV	Inter-run SD	%CV
Negative Run Control	864	0.13	0.042	32.53	0.046	35.70
HIV-1 Positive Run Control						
Control	864	2.61	0.155	5.97	0.200	7.66
SUP Run Control	864	2.50	0.140	5.57	0.162	6.47
Negative Calibrator	288	0.13	0.025	19.08	0.029	22.65
Positive Calibrator	286	2.47	0.110	4.44	0.134	5.41
Positive Assay Control	286	3.64	0.199	5.47	0.265	7.28

\* Representative performance data are shown. Results obtained at individual laboratories may vary.

### SPECIFICITY

The specificity of the ABBOTT PRISM HIV Ag/Ab Combo assay is  $\geq 99.5\%$ . The specificity of the assay was estimated assuming a zero prevalence of HIV infection in volunteer blood donors and hospitalized patients.

A total of 10940 serum, plasma, and plasmapheresis specimens were collected from five blood donor centers.

Three repeatedly reactive specimens were confirmed as having HIV infection by confirmatory testing and were excluded from the study.

The data from the remaining 10937 specimens are summarized in Table II.\*

The initial reactive rate obtained at individual laboratories varied.

**Table II**  
**Specificity Evaluation with Specimens from Blood Donors**

Specimen Type	n	Repeatedly Reactive	Specificity (%)	Specificity 95% CI <sup>a</sup>
Serum	5202	4	99.92	99.80 - 99.98
Plasma	5136	2	99.96	99.86 - 100.00
Plasmapheresis	599	0	100.00	99.39 - 100.00
Total	10937	6	99.95	99.88 - 99.98

\* Representative performance data are shown. Results obtained at individual laboratories and with different populations may vary. The repeat reactive rate varied from 0.03% (95% CI = 0.00 - 0.18%) to 0.08% (95% CI = 0.02 - 0.21%) between the individual clinical sites.

<sup>a</sup> CI = Confidence Interval

Specimens from individuals with medical conditions unrelated to HIV infection or containing potentially interfering substances and specimens from random hospital patients were tested with the ABBOTT PRISM HIV Ag/Ab Combo assay. Of the 1138 hospitalized patients specimens (HP) and the 322 specimens containing potentially interfering substances (IS), 6 HP and 13 IS specimens were confirmed as having HIV infection by supplemental testing. These specimens were excluded from the study. The data from the remaining 1132 HP specimens and the 309 IS specimens are summarized in Table III and Table IV.\*

The 309 specimens containing potentially interfering substances belonged to the following categories: (viral infection) CMV, EBV, HSV, HAV, HBV, HCV, HTLV-I, HTLV-II, Rubella; (bacterial/fungal infection/yeast/protozoa) *C. albicans*, *T. gondii*, *C. trachomatis*, *T. pallidum*, *E. coli*, *N. gonorrhoea*; (autoimmune) rheumatoid factor (RF), antinuclear antibodies (ANA); (other conditions) pregnant females all trimesters, multiparous females, elevated IgG, elevated IgM, monoclonal gammopathy, flu vaccine recipients, hemodialysis, hemophiliacs, multiple transfusion recipients, and human antimouse antibodies (HAMA).

**Table III**  
**Specificity Evaluation with Hospitalized Patient Specimens**

n	Initially Reactive	Repeatedly Reactive	Specificity (%)
1132	3	1	99.91

**Table IV**  
**Evaluation with Potentially Interfering Conditions**

n	Initially Reactive	Repeatedly Reactive	Interference (%)
309	4	5 <sup>a</sup>	1.62

<sup>a</sup> During the clinical evaluation, a grayzone was established for the ABBOTT PRISM HIV Ag/Ab Combo for this study. One specimen in this study fell within the grayzone and was retested and was found to be repeatedly reactive. Five samples which were repeatedly reactive and could not be confirmed by supplemental testing belonged to the following categories: HAMA (3) and dialysis (2).

\* Representative performance data are shown. Results obtained at individual laboratories and with different populations may vary.

One hundred specimens from patients at increased risk for HIV infection were tested by the ABBOTT PRISM HIV Ag/Ab Combo assay. Of these 100, 70 were repeatedly reactive\* by ABBOTT PRISM HIV Ag/Ab Combo and confirmation testing. The specimens from the high risk population belonged to the following categories: intravenous drug users, homosexual males and sexually transmitted diseases.

\* Representative performance data are shown. Results obtained at individual laboratories and with different populations may vary.

### SENSITIVITY

The ABBOTT PRISM HIV Ag/Ab Combo assay demonstrated the sensitivity shown in Table V in a study where specimens were obtained from individuals clinically diagnosed with HIV infection and classified disease status. Specimens were precharacterized as to antibody type and the data from this study are summarized in Table V.\*

**Table V**  
**Sensitivity**

Antibody Type	Number of Specimens tested	Number of Reactive Specimens	(%)
Anti-HIV-1	522	522	100.0
Anti-HIV-2	111	111	100.0
Anti-HIV gO	37	37	100.0

\* Representative performance data are shown. Results obtained at individual laboratories and with different populations may vary.

Sensitivity of the ABBOTT PRISM HIV Ag/Ab Combo assay to detect anti-HIV-1 and anti-HIV-2 was evaluated by testing sequential specimens from 20 seroconverting donors. These specimens are commercially available and precharacterized as to HIV antibodies. Sensitivity in seroconverting donors was better than the method of comparison. Table VI shows a subset of four representative seroconversion panels.

The analytical sensitivity of the ABBOTT PRISM HIV Ag/Ab Combo assay is  $< 50$  pg/mL. Analytical sensitivity of the ABBOTT PRISM HIV Ag/Ab Combo to detect HIV-1 p24 antigen was evaluated by testing the HIV-Ag 2003 AFSSAPS panel (n = 53) across three reagent lots. In this study, the assay demonstrated an analytical sensitivity on average of 33.44 pg/mL HIV-1 p24 antigen.\*

**Table VI**  
**Results for Seroconverting Donors (S/CO)**

Donor	Days since first bleed	ABBOTT PRISM HIV Ag/Ab Combo	Western Blot <sup>a</sup>	PCR <sup>a</sup>
BBI PRB958	0	0.13	NEG (no band)	70
	2	0.11	NEG (no band)	1,000
	7	<b>1.13</b>	NEG (no band)	100,000
	9	<b>3.22</b>	NEG (no band)	300,000
	15	<b>7.32</b>	NEG (no band)	500,000
	17	<b>8.65</b>	NEG (no band)	>500,000
BBI PRB951	0	0.10	NEG (no band)	BLD <sup>b</sup>
	2	0.09	NEG (no band)	BLD <sup>b</sup>
	8	<b>1.13</b>	NEG (no band)	200,000
	11	<b>12.25</b>	NEG (no band)	1,000,000
	15	<b>75.45</b>	NEG (no band)	5,000,000
	19	<b>41.36</b>	NEG (no band)	2,000,000
BBI PRB954	0	0.09	NEG (no band)	BLD <sup>b</sup>
	2	0.10	NEG (no band)	BLD <sup>b</sup>
	7	0.12	NEG (no band)	BLD <sup>b</sup>
	10	0.10	NEG (no band)	1,000
	14	0.31	NEG (no band)	60,000
	17	<b>2.31</b>	NEG (no band)	600,000
BCP 9016	21	<b>38.68</b>	NEG (no band)	>800,000
	0	0.13	no band	<50
	2	0.11	no band	<50
	7	0.11	no band	<50
	9	0.11	no band	<50
	15	0.11	no band	<50
	18	0.09	no band	<50
	23	0.10	no band	<50
	27	0.24	no band	7,473
	30	<b>1.24</b>	no band	69,010
	34	<b>6.82</b>	no band	286,400

\* Representative performance data are shown. Results obtained at individual laboratories may vary.

a Data from the vendor.

b Below limit of detection.

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