Antibody to Hepatitis B Surface Antigen (Mouse Monoclonal IgM)

Customer Service: Contact your local representative or find country specific contact information on www.abbottdiagnostics.com

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.

Key to Symbols

Lot Number
List Number
In Vitro Diagnostic Medical Device
Consult instructions for use
Caution
Store at 2-8°C
Store at 15-30°C
Expiry Date
Authorized Representative in the European Community
Manufacturer
Activator Line Treatment
Assay Kit Card
Calibrators
Contains Sodium Azide. Contact with acids liberates very toxic gas.

Danger: Reproductive Hazard
Distributed by
Line Cleaner
Master Lot
Pipette Tips
Prime/Purge Accessories
Produced for Abbott by
Product of USA
Purge Concentrate
Reaction Trays
Reagent Components
Run Control Adapters
Sample Cups
Warning: May cause an allergic reaction
Warning: Severe Irritant

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

U.S. License No. 43
NAME AND INTENDED USE
The ABBOTT PRISM HBsAg assay is an in vitro chemiluminescent immunoassay (CLIA) for the qualitative detection of hepatitis B surface antigen (HBsAg) in human serum and plasma specimens. The ABBOTT PRISM HBsAg (CLIA) is intended to screen individual human donors, including volunteer donors of whole blood and blood components, and other living donors for the presence of HBsAg. It is also intended for use in testing blood and blood specimens to screen organ donors when specimens are obtained while the donor’s heart is still beating, and in testing blood specimens to screen cadaveric (non-heart-beating) donors. It is not intended for use on cord blood specimens.

SUMMARY AND EXPLANATION OF THE TEST
Hepatitis B virus (HBV) is a small, partially double-stranded, DNA virus and a member of the Hepadnaviridae family. The HBV genome consists of four overlapping reading frames representing the core, polymerase, surface, and X genes. This virus is responsible for infecting approximately one third of the global population. Approximately 350 million individuals, worldwide, are chronic carriers of HBV.1 HBV is primarily transmitted through sexual, parenteral, and perinatal routes. Premature mortality from chronic liver disease occurs in 15-25% of the chronically infected HBV patients.2 HBsAg, hepatitis B surface antigen, is the first viral antigen to circulate in the infected individual. HBV, unlike other DNA viruses, replicates through reverse transcription. The reverse transcription process lacks proofreading capability; therefore, HBV is subject to a mutation rate 10 times higher than the mutation rate of other DNA viruses.3 Some of these mutations may cause changes in the antigenic structure of HBsAg, resulting in epitopes that are no longer recognized by anti-HBs.4,5 Sensitive immunoassays for the detection of HBsAg were first described in the early 1970s10-16 and were subsequently used to screen blood and blood products for the presence of HBsAg to prevent transmission of HBV infection to recipients of blood or blood products.17 In addition, assays for HBsAg are routinely used to diagnose suspected HBV infection and to monitor the status of infected individuals, i.e., whether the patient has resolved infection or has become a chronic carrier of the virus.18,19 The Centers for Disease Control and Prevention have recommended the prenatal screening of all pregnant women so that newborns from HBV carrier mothers may obtain prophylactic therapy. Prenatal transmission of HBV infection from mother to neonate is a major mode of transmission in an HBV endemic population.20

BIOLGICAL PRINCIPLES OF THE PROCEDURE
The ABBOTT PRISM HBsAg assay is a two-step sandwich CLIA. The reactions occur within the ABBOTT PRISM System in the following sequence:
- Microparticles coated with mouse monoclonal anti-HBs are incubated with sample (either plasma, serum, calibrator, or control) in the incubation well of the reaction tray. During incubation, HBsAg present in the sample binds to the antibody on the Microparticles.
- After this first incubation is complete, the reaction mixture is transferred to the glass fiber matrix (matrix) of the reaction tray using the Transfer Wash. The Microparticles are captured by the matrix while the remaining matrix flows through to the absorbent pad.
- The Acidinium-Labeled Goat Polyclonal Anti-HBs Conjugate is added to the Microparticles on the matrix and incubated. After this second incubation, the unbound Conjugate is washed into the blotter with the Conjugate Wash.
- The chemiluminescent signal is generated by addition of an alkaline hydrogen peroxide solution. The resultant photons are counted.

The amount of light emitted is proportional to the amount of HBsAg in the sample. The presence or absence of HBsAg in the 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 HBsAg by the criteria of the ABBOTT PRISM HBsAg assay. These specimens need not 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 HBsAg by the criteria of the ABBOTT PRISM HBsAg assay. Specimens that are initially reactive must be handled according to the table in the SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS section of this package insert and retested in duplicate. A specimen that is repeatedly reactive must be confirmed by the ABBOTT PRISM HBsAg Confirmatory assay, a licensed neutralizing confirmatory test. Only the specimens that are confirmed by specific neutralization with anti-HBs are considered positive for HBsAg. For further information regarding CLIA technology, refer to the ABBOTT PRISM Operations Manual, Section 3.

REAGENTS
NOTE: Each specific component description that follows is accompanied by a unique symbol. These symbols appear on both the component labels and on corresponding instrument tubing identifier labels. They are meant to facilitate identification and installation of reagent bottles within the ABBOTT PRISM System ambient reagent bay and refrigerator.

ABBOTT PRISM HBsAg Assay Kit (REF: 6D19-68)
NOTE: Do not mix reagents from different bottles. Do not mix or interchange reagents from different ABBOTT PRISM HBsAg Assay Kits.
- MicroParticulates: 1 Bottle (333 mL) Antibody to Hepatitis B Surface Antigen (Mouse Monoclonal IgM). Coated Microparticles in phosphate buffered saline with bovine serum albumin, 0.001% polysorbate 20, and protein stabilizers. Minimum concentration: 0.03% solids. Preservative: 0.1% sodium azide. (Symbol: ☢)
- Conjugate: 1 Bottle (328 mL) Antibody to Hepatitis B Surface Antigen (Goat Polyclonal); Acidinium Conjugate in phosphate buffered saline with calf serum and recalcified, human plasma. Minimum concentration: 0.025 µg/mL. Preservative: 0.1% sodium azide. (Symbol: ☢)
- CAL: 3 Bottles (10.4 mL each) Negative Calibrator (Human). Recalculated plasma. Preservative: 0.1% sodium azide. (Symbol: NC)
- CAL: 3 Bottles (10.4 mL each) Positive Calibrator (Human). Recalculated, inactivated plasma reactive for HBsAg. HBsAg concentration: 0.25-0.65 ng/mL. Preservative: 0.1% sodium azide. (Symbol: ☢)

Other Reagents Required
ABBOTT PRISM HBsAg Wash Kit (REF: 6D19-58)
- TRANSFER WASH: 1 Bottle (3393 mL) Transfer Wash. Phosphate buffered saline. Preservative: 0.1% sodium azide. (Symbol: ☢)
- CONJUGATE WASH: 1 Bottle (2811 mL) Conjugate Wash. Borate buffered saline. Preservative: 0.1% sodium azide. (Symbol: ☢)

ABBOTT PRISM Activator Concentrate (REF: 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 (REF: 1A75-01 or 3L27-01)
- ACTIVATOR DILUENT: 4 Bottles (900 mL each) Activator Diluent. 0.3% sodium hydioxide.

ABBOTT PRISM Run Control Kit (REF: 3E60-10)
- Or
ABBOTT PRISM Positive Run Control Kit (REF: 3E60-11)
NOTE: Each batch MUST end in a release control (ABBOTT PRISM Positive Control). The ABBOTT PRISM Positive Control (included in Kit [REF: 3E60-10 or 3E60-11]) must be used as the release control which has been configured to validate the system functionality and release sample results. Refer to the ABBOTT PRISM Run Control Kit package insert or the ABBOTT PRISM Positive Run Control Kit package insert for detailed handling and use instructions.

WARNINGS AND PRECAUTIONS
CAUTION: This product contains human sourced 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 will not transmit infection. Therefore, all human sourced materials must be considered potentially infectious. It is recommended that these reagents and human specimens be handled in accordance with the OSHA Standard on Bloodborne Pathogens21,22. Biosafety Level 223 or other appropriate biosafety practices should be used for materials that contain or are suspected of containing infectious agents. These precautions include, but are not limited to the following:
- Wear gloves when handling specimens or reagents.
- Do not pipette by mouth.
- Do not eat, drink, smoke, apply cosmetics, or handle contact lenses in work areas where specimens or reagents are handled.
- Clean and disinfect all spills of specimens or reagents using an appropriate disinfectant, such as 0.1% sodium hypochlorite, or other suitable disinfectants.24,25
- Decontaminate and dispose of all specimens, reagents, and other potentially contaminated materials in accordance with local, state, and federal regulations.26,27
- The human plasma used in the Conjugate is nonreactive for HBsAg, HIV-1 RNA or HIV-1 Ag. anti-HIV-1/HIV-2, and anti-HCV.
- The human plasma used in the Negative Calibrator is nonreactive for HBsAg, HIV-1 RNA or HIV-1 Ag. anti-HIV-1/HIV-2, and anti-HCV.
- The human plasma used in the Positive Calibrator is reactive for HBsAg and nonreactive for HIV-1 RNA or HIV-1 Ag. anti-HIV-1/HIV-2 and anti-HCV.

ABBOTT P
• 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.

• The following warnings and precautions apply to the Purge Concentrate.

  **WARNING:** Contains methylisothiazolones. May cause an allergic skin reaction.
  Prevention
  P261 Avoid breathing mist / vapours / spray.
  P272 Contaminated work clothing should not be allowed out of the workplace.
  P280 Wear protective gloves / protective clothing / eye protection.
  Response
  P302+P352 IF ON SKIN: Wash with plenty of water.
  P333+P313 If skin irritation or rash occurs: Get medical advice / attention.
  P362+P364 Take off contaminated clothing and wash it before reuse.
  Disposal
  P501 Dispose of containers/container in accordance with local regulations.

• Safety Data Sheets are available at www.abbottdiagnostics.com or contact your local representative.

**Handling Precautions**

• Do not use kits beyond the expiration date.

• Gently invert each component several times prior to loading the original container on the ABBOTT PRISM System to ensure a homogenous solution.

• Additional gentle inversion may be required to thoroughly resuspend microparticles. Avoid foaming.

• Gently invert calibrators in the calibrator pack several times prior to each use.

• Each component of the ABBOTT PRISM HBsAg Wash Kit should be at room temperature (15 - 30°C) and then mixed before loading onto the ABBOTT PRISM System.

• Do not mix reagents or calibrators from different bottles. Do not mix or interchange reagents from different ABBOTT PRISM HBsAg Assay Kits.

• Any lot of ABBOTT PRISM HBsAg Wash Kit can be used with any lot of ABBOTT PRISM HBsAg Assay Kit.

• Any lot of ABBOTT PRISM Activator Concentrate, ABBOTT PRISM Activator Diluent, and Control from ABBOTT PRISM Run Control Kit or ABBOTT PRISM Positive Run Control Kit may be used with any lot of ABBOTT PRISM Assay Kit.

• Treat Negative and Positive Calibrators and Controls as specimens.

• Avoid microbial and chemical contamination of samples, reagents and equipment. The use of disposable pipette tips is recommended for any preliminary sample transfer.

• Use accurately calibrated equipment.

• Do not freeze reagents.

• Failure to adhere to instructions in the ABBOTT PRISM Operations Manual or package insert may result in erroneous test results.

• 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, Sections 7 and 8.

**Preparation of Activator Solution**

Activator solution must be prepared by mixing equal parts of ABBOTT PRISM Activator Concentrate and ABBOTT PRISM Activator Diluent. The activator solution expires 24 hours from preparation. The ABBOTT PRISM Activator Concentrate may be used immediately after removing from the refrigerator. The volume of activator solution required for multiple tests is calculated by the ABBOTT PRISM System software. Refer to the ABBOTT PRISM Operations Manual, Section 5, PLAN WORK LOAD for additional information. Use clean pipettes and/or metal-free containers (such as plasticware or acid-washed and purified or equivalent water-rinsed glassware) to measure. Refer to the ABBOTT PRISM Operations Manual Glossary for the definition of purified water. Prepare the activator solution in the bottle provided in the ABBOTT PRISM Accessory Kit (Part #13636-60). Cover the bottle opening securely with the cap provided and 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, PREPARE AND LOAD ACTIVATOR SOLUTION, for additional information.

**NOTE:** The activator solution must be used within 24 hours of preparation.

**Storage Instructions**

• Store the ABBOTT PRISM HBsAg Assay Kit, ABBOTT PRISM Run Control Kit, ABBOTT PRISM Positive Run Control Kit, and ABBOTT PRISM Activator Concentrate at 2 - 8°C.

• Store the ABBOTT PRISM HBsAg Wash Kit and ABBOTT PRISM Activator Diluent at room temperature (15 - 30°C).

• Store ABBOTT PRISM Pipette Tips and ABBOTT PRISM Reaction Trays in their original packaging until use.

• The activator solution must be stored at 15 - 30°C and used within 24 hours of preparation.

**Indications of Instability or Deterioration of Reagents**

The ABBOTT PRISM System will not continue to process samples when calibrator 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**

• For the software versions that may be used to perform the assay, refer to the ABBOTT PRISM Assay / Software Version Matrix located in the Supplemental Information tab of the ABBOTT PRISM Operations Manual.

• Refer to the ABBOTT PRISM Operations Manual for a detailed description of Instrument Procedures.

• Refer to the ABBOTT PRISM Operations Manual, Section 7, for limitations associated with test management.

• Solutions required for instrument cleaning and maintenance are described in detail in the ABBOTT PRISM Operations Manual, Sections 9 and 10.

• 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**

- Serum (including serum collected in separator tubes), plasma collected in EDTA, potassium oxalate, sodium citrate, ACD-A, ACD-B, CP2D, CPD, or CPDA-1 anticoagulants, or plasma collected from segmented tubing may be used with the ABBOTT PRISM HBsAg assay. Follow the manufacturer’s processing instructions for serum and plasma collection tubes.

**CAUTION:** Do not use specimens collected in heparin. Use of heparin as an anticoagulant may cause a reduction in Sample Net Counts and in Sample Net Counts/Cutoff Value (S/CO) for ABBOTT PRISM HCV; therefore, heparin is not recommended for any ABBOTT PRISM assay.

- This assay was designed and validated for use with individual human serum and plasma specimens. This assay has not been validated for use with pooled specimens.

- Do not use cadaveric plasma specimens.

- Do not use heat-inactivated specimens.

- Do not use specimens with obvious microbial contamination.

- 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 for a period not to exceed 7 days. Prior to freezing, the serum or plasma should be removed from the clot or red blood cells.

- Failure to follow the specified centrifugation procedure on specimens tested with the ABBOTT PRISM HBsAg assay may cause a reduction in Sample Net Counts and in Sample Net Counts/Cutoff Value (S/CO) for ABBOTT PRISM HCV.

- Specimens may be stored for up to 14 days at 2 - 8°C. If storage periods greater than 14 days are anticipated, the serum or plasma should be removed from the clot or red blood cells to avoid hemolysis. Store the serum or plasma frozen (-20°C or colder).

- For cadaveric specimens, follow general standards and/or regulations for collection, storage and handling. Cadaveric specimens may be stored frozen (-20°C or colder) or stored for up to 2 days at 2 - 8°C. If storage periods greater than 2 days at 2 - 8°C are anticipated, the serum should be removed from the clot to avoid hemolysis and stored frozen.

- Previously frozen specimens must be mixed gently and thoroughly after thawing and centrifuged according to Table II in this section.

- Twenty nonreactive and 20 low-level reactive specimens showed no qualitative performance differences when subjected to 6 freeze-thaw cycles. However, some specimens that have undergone multiple freeze-thaw cycles or have been stored frozen for prolonged periods may give erroneous or inconsistent test results.

- Clear, non-hemolyzed specimens should be used when possible. Specimens containing visible particulate matter may give erroneous or inconsistent test results.

**PREPARE AND LOAD ACTIVATOR SOLUTION**
- No qualitative performance differences were observed when 20 nonreactive and 18 low-level reactive specimens were spiked with elevated levels of bilirubin (≤ 20 mg/dL), hemoglobin (≤ 500 mg/dL), red blood cells (≤ 0.4% v/v), triglycerides (≤ 3000 mg/dL), or protein (≤ 12 g/dL). However, specimens that contain greater concentrations of these potentially interfering substances have not been tested. The impact of greater concentrations of these potentially interfering substances on the ABBOTT PRISM HBsAg assay is unknown.

- Performance has not been established using umbilical cord blood, or body fluids such as urine, saliva, semen, amniotic fluid, cerebrospinal fluid, or pleural fluid. These specimens should not be tested using the ABBOTT PRISM HBsAg assay.

- Specimens collected by plasmapheresis, that have not been frozen, do not require centrifugation. All other specimens (including previously frozen plasmapheresis specimens) must be centrifuged as follows:
  - Non-frozen specimens (excluding non-frozen plasmapheresis specimens) must be centrifuged such that g-minutes is between 30,000 and 75,000. A refrigerated or non-refrigerated centrifuge is acceptable for use. The acceptable time and force ranges that meet this criterion are listed in Table I.

<table>
<thead>
<tr>
<th>Time (minutes)</th>
<th>RCF (x g)</th>
<th>g-minutes</th>
</tr>
</thead>
<tbody>
<tr>
<td>10</td>
<td>3,000</td>
<td>30,000</td>
</tr>
<tr>
<td>15</td>
<td>2,000 - 3,000</td>
<td>30,000 - 45,000</td>
</tr>
<tr>
<td>20</td>
<td>1,500 - 3,000</td>
<td>30,000 - 60,000</td>
</tr>
<tr>
<td>25</td>
<td>1,300 - 3,000</td>
<td>32,500 - 75,000</td>
</tr>
</tbody>
</table>

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

Convert RPM to g-minutes as follows: g-minutes = 1,000 x RCF

- Centrifugation:
  - Time: The time should be measured from the time the rotor reaches the required RPM or rpm to the time it begins decelerating.
  - RCF: The relative centrifugal force generated during centrifugation.
  - RPM: The revolutions per minute of the rotor on which the specimens are being spun (usually the digital readout on the centrifuge will indicate the rpm).

Centrifugation:
- Time: The time should be measured from the time the rotor reaches the required RPM or rpm to the time it begins decelerating.
- RCF: The relative centrifugal force generated during centrifugation.
- RPM: The revolutions per minute of the rotor on which the specimens are being spun (usually the digital readout on the centrifuge will indicate the rpm).

Previously frozen specimens must be centrifuged such that g-minutes is between 180,000 and 300,000. A refrigerated or non-refrigerated centrifuge is acceptable for use. The acceptable time and force ranges that meet this criterion are listed in Table II.

<table>
<thead>
<tr>
<th>Time (minutes)</th>
<th>RCF (x g)</th>
<th>g-minutes</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>12,000</td>
<td>180,000</td>
</tr>
<tr>
<td>20</td>
<td>9,000 - 12,000</td>
<td>180,000 - 240,000</td>
</tr>
<tr>
<td>25</td>
<td>7,200 - 12,000</td>
<td>180,000 - 300,000</td>
</tr>
</tbody>
</table>

**PROCEDURE**

**Materials Provided**
- REF 6D19-68 ABBOTT PRISM HBsAg Assay Kit

**Materials Required but not Provided**
- REF 6D19-58 ABBOTT PRISM HBsAg Wash Kit
- REF 1A75-02 or 3L27-02 ABBOTT PRISM ACTIMON CONCENTRATE
- REF 1A75-01 or 3L27-01 ABBOTT PRISM ACTIMON DILUENT
- REF 5A07-01 ABBOTT PRISM REACTION TRAYS
- REF 6A36-60 ABBOTT PRISM Accessory Kit
- REF 3E60-10 ABBOTT PRISM Run Control Kit
- REF 3E60-11 ABBOTT PRISM Positive Run Control Kit
- REF 6A36-31 ABBOTT PRISM RUN CONTROL ADAPTERS
- Protective Disposable Gloves
- Disinfectant
- Purified Water-rinsed or Clean Disposable Measuring Equipment

**Additional Materials Available**
- REF 7B36-01 ABBOTT PRISM SAMPLE CUPS
- REF 1A75-10 or 3L27-10 ABBOTT PRISM ACTIMON LINE TREATMENT
- REF 7A03-01 or 3L00-01 ABBOTT PRISM PRIME/PURGE ACCESSORIES
- REF 7A03-30 or 3L00-30 ABBOTT PRISM PURGE CONCENTRATE
- REF 4A03-31 ABBOTT PRISM LINE CLEANER

For Cadaveric Specimens Only
- REF 2P41-01 Millipore GV Filters
- 10 cc Sterile Syringes
ABBOTT PRISM HBsAg ASSAY PROCEDURE

Key procedures that require operator interaction for testing samples are listed below. For detailed information concerning batch time, maximum batch size, reagent handling and loading, and associated procedural steps, refer to the ABBOTT PRISM Operations Manual, Sections 2, 5, and 7.

- Enter a Plan Work Load (refer to the ABBOTT PRISM Operations Manual, Section 5).
- Replace reagents as needed (refer to the ABBOTT PRISM Operations Manual, Sections 5 and 7).

NOTE: 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. Gently invert calibrators in the calibrator pack several times prior to each use. Each component of the ABBOTT PRISM HBsAg Wash Kit should be at room temperature (15 - 30°C) and then mixed before loading onto the ABBOTT PRISM System.
- Verify that all tubing label symbols match the symbols on each reagent label. (Refer to the symbol key in the REAGENTS section of this package insert, and the ambient reagent bay and refrigerator diagrams provided with the ABBOTT PRISM System).
- Verify that all tubing is securely fastened to the corresponding wash and reagent bottles.
- Inspect the waste containers. Empty and clean as defined in the ABBOTT PRISM Operations Manual, Section 9, if necessary.
- Prepare activator solution (Refer to the Preparation of Activator Solution description of this package insert) and load onto the ABBOTT PRISM System.
- Verify that an adequate number of ABBOTT PRISM Reaction Trays are in the Tray Loader.
- Verify that an adequate number of ABBOTT PRISM Pipette Tips are in the Pipette Tip Racks.
- Perform the prime procedure (Refer to the ABBOTT PRISM Operations Manual, Section 5).
- Initiate sample processing. Gently invert calibrators in the calibrator pack several times. Open the bottles in the calibrator pack and place in the calibrator rack. Load the calibrator rack and sample racks, including the run controls. (Refer to the QUALITY CONTROL PROCEDURES, Controls, Control Handling Procedure, in this package insert.)
- After the calibrators have been automatically pipetted, remove the calibrator rack. Close the calibrator bottles and return them to 2 - 8°C storage.
- Each specimen is initially tested once, unless the operator overrides this automatic function of the ABBOTT PRISM System.
- Sample racks may be removed after the samples have been pipetted.

NOTE: No operator interaction is required for the following steps, which are automatically carried out by the ABBOTT PRISM System: reaction tray transport, calibrator/sample/release control pipetting, incubation, reagent dispense, sample reading, data reduction, run validity and result determination.
- After specimen processing 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. The ABBOTT PRISM HBsAg assay is a two-step ChLIA procedure.

QUALITY CONTROL PROCEDURES

Calibration

The ABBOTT PRISM HBsAg Negative and Positive Calibrators are automatically tested in triplicate at the beginning of each batch. The ABBOTT PRISM System will not generate test results when calibrator values do not meet specifications. This may indicate either deterioration or contamination of reagents, or instrument failure.

Controls

1. The ABBOTT PRISM Positive Control MUST be included as the last sample in each batch as a release control. The operator is prompted to include this control as the last sample in every batch, and the ABBOTT PRISM Positive Control is then automatically tested as a single replicate. This control must meet specifications defined in the ABBOTT PRISM Run Control Kit package insert or the ABBOTT PRISM Positive Run Control Kit package insert in order to validate the system functionality and release sample results. If this control does not meet specifications defined in the ABBOTT PRISM Run Control Kit package insert or the ABBOTT PRISM Positive Run Control Kit package insert, refer to the ABBOTT PRISM Operations Manual, Section 10, for additional information.

2. Additional controls may be run at the operator’s discretion (refer to the ABBOTT PRISM Operations Manual, Section 3).

Invalidate controls: Additional controls may be run anywhere within a batch as an invalidate control. Specifications may be assigned to invalidating controls. If an invalidate control fails to meet assigned specifications, sample processing is shutdown and no sample results are calculated or provided by the instrument. When an invalidate control meets assigned specifications, sample processing continues and a valid release control (ABBOTT PRISM Positive Control) result is required to release data.
ASSAY SPECIFICITY

A total of 25,238 serum and plasma specimens from volunteer whole blood donors and plasmapheresis donors were collected and tested at six geographically distinct blood centers (Table IV). Two sites tested a total of 8,246 serum specimens with initial and repeat reactive rates of 0.06% (5/8,246) and 0.04% (3/8,246), respectively. Three sites tested a total of 13,911 plasma specimens with initial and repeat reactive rates of 0.06% (8/13,911) and 0.04% (5/13,911), respectively. One site tested a total of 3,081 plasmapheresis donor specimens with initial and repeat reactive rates of 0.03% (1/3,081) and 0.00% (0/3,081), respectively. A total of eight specimens were repeatedly reactive. In six of the eight specimens (75.00%), the presence of HBsAg was confirmed by specific neutralization with anti-HBs. Two of the eight specimens were not confirmed as positive.

Specifically based on assumed zero prevalence of HBsAg in whole blood and plasmapheresis donors was estimated in these studies to be 99.99% (25,230/25,232) with a 95% confidence interval (CI) of 99.97% to 100.00%. The six repeatedly reactive specimens that confirmed positive for HBsAg were excluded from these calculations.

Three sites evaluated 870 serum and plasma specimens either collected from individuals with medical conditions unrelated to HBV infection or containing potentially interfering substances (Table IV). Fifty-nine of the 870 specimens (6.78%) were initially reactive, and 50 of the 870 specimens (5.75%) were repeatedly reactive. Forty of the 50 specimens (80.00%) confirmed positive for HBsAg, and ten specimens did not confirm specific antibody neutralization. The ten specimens included one anti-EBV positive (12 tested), one anti-HSV positive (12 tested), one rubella antibody positive (12 tested), one anti-nuclear antibody positive (12 tested), one elevated triglycerides (12 tested), and five pregnant females (553 tested). The estimated specificity in this population was 98.80% (850/863).

### TABLE IV

<table>
<thead>
<tr>
<th>Category</th>
<th>Number Tested</th>
<th>IR (% of Total) (95% CI)</th>
<th>RR (% of Total) (95% CI)</th>
<th>Number Confirmed Positive (% of RR) CI</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Volunteer Blood Donors</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Serum</td>
<td>8,246</td>
<td>6.78% (5/25,232)</td>
<td>0.03% (1/3,081)</td>
<td>100.00% (48/48)</td>
</tr>
<tr>
<td>Plasma</td>
<td>13,911</td>
<td>5.75% (8/13,911)</td>
<td>0.04% (5/13,911)</td>
<td>100.00% (50/50)</td>
</tr>
<tr>
<td>Plasmapheresis</td>
<td>3,081</td>
<td>5.75% (1/3,081)</td>
<td>0.00% (0/3,081)</td>
<td>100.00% (0/0)</td>
</tr>
<tr>
<td><strong>Total Donors</strong></td>
<td>25,238</td>
<td>5.75% (14/25,238)</td>
<td>0.00% (0/25,238)</td>
<td>100.00% (48/48)</td>
</tr>
<tr>
<td><strong>Medical Conditions</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unrelated to HBV infection</td>
<td>870</td>
<td>5.75% (50/870)</td>
<td>0.00% (0/870)</td>
<td>100.00% (50/50)</td>
</tr>
<tr>
<td>Potentially Interfering Substances</td>
<td>870</td>
<td>5.75% (50/870)</td>
<td>0.00% (0/870)</td>
<td>100.00% (50/50)</td>
</tr>
</tbody>
</table>

* IR = Initial Reactive; RR = Repeat Reactive; CI = Confidence Interval

**A specimen was confirmed positive for HBsAg if the non-neutralized specimen (with ABBOTT PRISM HBsAg Confirmatory assay Reagent B added) exhibited a net count greater than or equal to the ABBOTT PRISM HBsAg Confirmatory assay cutoff value and if the neutralization with anti-HBs (Reagent A) was 50% or greater.

**B specimens from individuals with medical conditions unrelated to HBV infection and specimens containing potentially interfering substances included the following categories: anti-CMV positive (12), anti-EBV positive (12), anti-HIV positive (12), anti-HIV-1 positive (12), anti-HIV-2 positive (5), anti-HTLV-I positive (12), anti-HTLV-II positive (12), anti-HIV-1 positive (12), anti-HIV-2 positive (6), anti-HIV-1 positive (12), anti-HIV-2 positive (5), anti-HTLV-I positive (12), anti-HTLV-II positive (12), anti-viral liver diseases (42), rubella antibody positive (12), toxoplasma antibody positive (11), E.coli infections (5), syphilis serology positive (12), anti-nuclear antibody positive (12), rheumatoid factor positive (12), influenza vaccine recipients (52), elevated IgG (12), elevated IgM (12), elevated triglycerides (10), elevated bilirubin (12), elevated hemoglobin (11), and pregnant females (553).
The 50 repeatedly reactive specimens included the following: anti-EBV positive (1), anti-HSV positive (1), anti-HCV positive (1), anti-HIV-1 positive (5), anti-HIV-2 positive (1), non-viral liver diseases (5), rubella antibody positive (1), anti-nuclear antibody positive (1), influenza vaccine recipients (1), elevated triglycerides (1), and pregnant females (32).

d The following 40 specimens confirmed positive for HBsAg: anti-HCV positive (1), anti-HIV-1 positive (6), anti-HIV-2 positive (1), non-viral liver diseases (5), influenza vaccine recipients (1), and pregnant females (27).

ASSAY SENSITIVITY

A total of 1,212 serum and plasma specimens from 514 individuals known to be positive for HBsAg, 98 individuals with acute HBV infection, 101 individuals with chronic HBV infection, 47 individuals who have recovered from HBV infection, and 452 individuals at increased risk for HBV infection were tested with the ABBOTT PRISM HBsAg assay. A total of 767 specimens (63.2%) were repeatedly reactive, of which 754 (98.31%) were confirmed positive by specific antibody neutralization (Table V). The overall sensitivity was estimated in these studies to be 100.00% (754/754) with a 95% CI of 99.51% to 100.00%.

TABLE V
Reactivity of the ABBOTT PRISM HBsAg Assay in Selected Populations with HBV Infection and at Increased Risk for HBV Infection

<table>
<thead>
<tr>
<th>Category</th>
<th>Number Tested</th>
<th>Number Repeatedly Reactive (% of Total)</th>
<th>Number Confirmed Positive (% of Repeatedly Reactive)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Preselected HBsAg Positive</td>
<td>514</td>
<td>514 (100.00)</td>
<td>514 (100.00)</td>
</tr>
<tr>
<td>Acute HBV Infection</td>
<td>98</td>
<td>98 (100.00)</td>
<td>98 (100.00)</td>
</tr>
<tr>
<td>Chronic HBV Infection</td>
<td>101</td>
<td>101 (100.00)</td>
<td>101 (100.00)</td>
</tr>
<tr>
<td>Recovered HBV Infection</td>
<td>47</td>
<td>0 (0.00)</td>
<td></td>
</tr>
<tr>
<td>Increased Risk for HBV Infection</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>1,212</td>
<td>767 (63.28)</td>
<td>754 (98.31)</td>
</tr>
</tbody>
</table>

- Specimens from the preselected HBsAg positive category were tested only once.
- Preselected HBsAg positive specimens were previously confirmed positive by specific antibody neutralization.
- Individuals at increased risk for HBV infection included the following categories: intravenous drug users (254), hemodialysis patients (50), hemophila patients (50), and STD clinic patients (148).
- The 54 repeatedly reactive specimens included the following: intravenous drug users (25), hemodialysis patients (6), hemophila patients (4), and STD clinic patients (19).
- The 41 specimens that confirmed positive for HBsAg included the following: intravenous drug users (15), hemodialysis patients (5), hemophila patients (3), and STD clinic patients (18). Of these 41 specimens, 32 were confirmed positive by a licensed reference HBsAg test. The PRISM assay confirmed an additional 9 specimens. In addition, there were no specimens in this category (452 specimens) that were confirmed positive by the licensed reference HBsAg test that were not confirmed positive by the PRISM assay. The sensitivity of the ABBOTT PRISM HBsAg assay was estimated using a seven-member panel comprised of specimens from an Abbott Laboratories HBsAg Sensitivity Panel. Panel members were prepared in recalcified human plasma. Three panel members were reactive for HBsAg subtype, three members were reactive for HBsAg ay subtype, and one member was nonreactive for HBsAg. The panel was tested as described in the ASSAY REPRODUCIBILITY section of this package insert. The detection of HBsAg ad and ay subtypes is presented in Tables VI and VII, respectively.

TABLE VI
Detection of Purified HBsAg ad and ay Subtypes by the ABBOTT PRISM HBsAg Assay

<table>
<thead>
<tr>
<th>HBsAg Concentration (ng/mL)</th>
<th>Mean S/CO Value</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.917</td>
<td>6.98</td>
<td>+</td>
</tr>
<tr>
<td>0.525</td>
<td>4.06</td>
<td>+</td>
</tr>
<tr>
<td>1.134</td>
<td>1.39</td>
<td>-</td>
</tr>
<tr>
<td>0.000</td>
<td>0.34</td>
<td>-</td>
</tr>
</tbody>
</table>

Reproducibility

Inter-assay reproducibility of PRISM HBsAg was assessed using 10 postmortem donor sera. These sera specimens were spiked with human plasma positive for HBsAg to create low-level reactive specimens. Each of the specimens was tested in triplicate on three different days on each of three lots of PRISM HBsAg at one site for a total of 270 replicates. Three replicates generated dispense errors and 16 replicates generated drain time errors and were excluded from the analysis. For intra-assay reproducibility, the %CV ranged from 2.9 to 5.5 for the low-level reactive specimens. For inter-assay reproducibility over all lots, the percent coefficient of variation (%CV) ranged from 4.4 to 8.7 for the low-level reactive specimens. The total reproducibility ranged from 5.3 to 9.7 for the low level reactive specimens. Note: Inter-assay reproducibility includes intra-assay and inter-assay variation. Total reproducibility includes intra-assay, inter-assay and inter-lot variations.

Specificity

Specificity was evaluated using 51 postmortem donor specimens and 54 normal donor specimens. Each of the specimens was tested once on each of three lots of PRISM HBsAg. The mean sample to cutoff (S/CO) ratio for the 136 nonreactive postmortem replicates (51 specimens with three reagent lots; see Table VIII, footnotes a and b) was 0.37, and the mean S/CO for 162 normal donor replicates (54 specimens with three reagent lots) was 0.24. Results are presented in Table VIII.

TABLE VII
Detection of Purified HBsAg ay by the ABBOTT PRISM HBsAg Assay

<table>
<thead>
<tr>
<th>HBsAg Concentration (ng/mL)</th>
<th>Mean S/CO Value</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.002</td>
<td>8.86</td>
<td>+</td>
</tr>
<tr>
<td>0.485</td>
<td>4.62</td>
<td>+</td>
</tr>
<tr>
<td>0.131</td>
<td>1.37</td>
<td>+</td>
</tr>
<tr>
<td>0.000</td>
<td>0.34</td>
<td>-</td>
</tr>
</tbody>
</table>

The ability of the ABBOTT PRISM HBsAg assay to detect HBsAg was evaluated by testing 12 HBV seroconversion panels from blood and plasmapheresis donors who seroconverted over the course of their donation history. All specimens were also tested by a FDA licensed assay. The ABBOTT PRISM HBsAg assay detected HBsAg three to 13 days (one to three bleeds) earlier in ten of the 12 panels and five to 48 days (one to three bleeds) longer in four of the 12 panels when compared to the licensed assay. Both assays detected HBsAg in the first available bleed for two of the 12 panels.

PERFORMANCE CHARACTERISTICS OF CADAVERIC SERUM TESTING

Reproducibility

Inter-assay reproducibility of PRISM HBsAg was assessed using 10 postmortem donor sera. These sera specimens were spiked with human plasma positive for HBsAg to create low-level reactive specimens. Each of the specimens was tested in triplicate on three different days on each of three lots of PRISM HBsAg at one site for a total of 270 replicates. Three replicates generated dispense errors and 16 replicates generated drain time errors and were excluded from the analysis. For intra-assay reproducibility, the %CV ranged from 2.9 to 5.5 for the low-level reactive specimens. For inter-assay reproducibility over all lots, the percent coefficient of variation (%CV) ranged from 4.4 to 8.7 for the low-level reactive specimens. The total reproducibility ranged from 5.3 to 9.7 for the low level reactive specimens. Note: Inter-assay reproducibility includes intra-assay and inter-assay variation. Total reproducibility includes intra-assay, inter-assay and inter-lot variations.

Specificity

Specificity was evaluated using 51 postmortem donor specimens and 54 normal donor specimens. Each of the specimens was tested once on each of three lots of PRISM HBsAg. The mean sample to cutoff (S/CO) ratio for the 136 nonreactive postmortem replicates (51 specimens with three reagent lots; see Table VIII, footnotes a and b) was 0.37, and the mean S/CO for 162 normal donor replicates (54 specimens with three reagent lots) was 0.24. Results are presented in Table VIII.

TABLE VIII
Reactivity with PRISM HBsAg

<table>
<thead>
<tr>
<th>Population</th>
<th>No. of Specimens</th>
<th>No. of Replicates</th>
<th>Mean S/CO</th>
<th>Nonreactive</th>
<th>Initial Reactive</th>
</tr>
</thead>
<tbody>
<tr>
<td>Postmortem</td>
<td>51</td>
<td>137</td>
<td>0.37</td>
<td>136</td>
<td>1(1)</td>
</tr>
<tr>
<td>Normal Donor</td>
<td>54</td>
<td>162</td>
<td>0.24</td>
<td>162</td>
<td>0(100)</td>
</tr>
</tbody>
</table>

- No results were obtained for 15 specimens on one lot due to drain time errors and one specimen on one lot due to an invalid result.
- Specimen was not retested due to insufficient specimen volume.

Assuming the specimen with the initial reactive result would have a reactive result upon retest, the PRISM HBsAg assay has an estimated specificity of 99.27% (136/137) and sensitivity of 99.27% (1/1) in these studies of postmortem serum specimens collected up to 16.1 hours after death.

Sensitivity

Sensitivity was evaluated using 51 postmortem specimens and 54 normal donor specimens that were pre-screened for anti-HBs and HBsAg and found to be negative. The 105 specimens were spiked with human plasma positive for HBsAg to create low-level reactive specimens. Each of the specimens was tested once on each of three lots of PRISM HBsAg. The mean sample to cutoff (S/CO) for the 142 postmortem replicates (51 specimens, with three reagent lots; see Table IX, footnote a) was 2.05, and the mean S/CO for the 162 normal donor replicates (54 specimens, with three reagent lots) was 2.07. Results are presented in Table IX.
Table IX
Reactivity with PRISM HBsAg

<table>
<thead>
<tr>
<th>Population</th>
<th>No. of Specimens</th>
<th>No. of Replicates</th>
<th>Mean S/CO</th>
<th>Nonreactive</th>
<th>Initial Reactive</th>
</tr>
</thead>
<tbody>
<tr>
<td>Postmortem</td>
<td>51</td>
<td>142</td>
<td>2.05</td>
<td>0</td>
<td>142</td>
</tr>
<tr>
<td>Normal Donor</td>
<td>54</td>
<td>162</td>
<td>2.07</td>
<td>0</td>
<td>162</td>
</tr>
</tbody>
</table>

- No results were obtained for 7 unique specimens, and 2 specimens using 2 reagent lots due to drain time errors.
- The PRISM HBsAg assay has an estimated sensitivity of 100.00% (142/142) (binomial confidence interval = [97.44%, 100.00%]) in these studies of postmortem serum specimens collected up to 16.1 hours after death.

BIBLIOGRAPHY


ABBOTT PRISM is a trademark of Abbott Laboratories in various jurisdictions. All trademarks are property of their respective owners.