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.

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

U.S. License No. 43

Abbott
NAME AND INTENDED USE

The ABBOTT PRISM HBsAg assay is an in vitro chemiluminescent immunosay (CHLIA) for the qualitative detection of hepatitis B surface antigen (HBsAg) in human serum and plasma specimens. The ABBOTT PRISM HBsAg (CHLIA) system 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 plasma 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 Hepadnavirus family. The HBV genome contains 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, world-wide, 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. 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 error-prone.2 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. HBsAg, hepatitis B surface antigen, is the first viral antigen to circulate in the infected individual.

HBsAg is primarily found in serum, whole blood, plasma, and bile. It is present in less than 1% of infected interstitial fluid. The serum ALT level correlates with the viral load of HBV. The amount of light emitted is proportional to the amount of HBsAg in the sample. The chemiluminescent signal is generated by addition of an alkaline hydrogen peroxide generator to the sample. The light generated is detected by a photomultiplier and converted to an electronic signal. The assay is performed in an automated system. The chemiluminescent signal is generated by addition of an alkaline hydrogen peroxide generator to the sample. The light generated is detected by a photomultiplier and converted to an electronic signal. The assay is performed in an automated system.

The ABBOTT PRISM HBsAg assay meets FDA potency requirements. The ABBOTT PRISM HBsAg Wash Kit, ABBOTT PRISM Positive Run Control Kit (10.4 mL wash), and ABBOTT PRISM Positive Run Control Kit (3.3 mL wash) 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 for detailed handling and use instructions.

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 (281 mL) Conjugate Wash. Borate buffered saline and recalcified human plasma. Preservative: 0.1% sodium azide. (Symbol: ●)
• CAL 3 Bottles (10.4 mL each) Calibrator Wash. Recalcified plasma. Preservative: 0.1% sodium azide. (Symbol: NC)

ABBOTT PRISM Run Control Kit (REF 1A75-02 or 3L27-02)

• ACTIVATOR CONCENTRATE
  1 Bottle (300 mL) Activator Concentrate. 0.4% hydrogen peroxide/0.06% diethylenetriaminepentaacetic acid.
• ACTIVATOR DILUENT
  1 Bottle (900 mL) Activator Diluent. 0.3 sodium hydroxide.

ABBOTT PRISM Run Control Kit (REF 3E60-10)

ABBOTT PRISM Positive Run Control Kit (REF 3E60-11)

NOTE: Each batch MUST be used in a release control (ABBOTT PRISM Positive Control). The ABBOTT PRISM Positive Control (including 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 Pathogens32. Biosafety Level 215 or other appropriate biosafety practices32,36 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.32,27,28
• Decontaminate and dispose of all specimens, reagents, and other potentially contaminated materials in accordance with local, state, and federal regulations.32,29,30
• 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 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.

- MICROPARTICLES
  • 1 Bottle (333 mL) Antibody to Hepatitis B Surface Antigen (Mouse Monoclonal lgM) Coated Microparticles in phosphate buffered saline with bovine serum albumin, 1 mg/mL, 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); Acridinium 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). Recalibrated plasma. Preservative: 0.1% sodium azide. (Symbol: NC)

- CAL 3 Bottles (10.4 mL each) Positive Calibrator (Human). Recalibrated, 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 (281 mL) Conjugate Wash. Borate buffered saline and recalcified, human plasma. Preservative: 0.1% sodium azide. (Symbol: ●)

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

- ACTIVATOR CONCENTRATE
  1 Bottle (300 mL) Activator Concentrate. 0.4% hydrogen peroxide/0.06% diethylenetriaminepentaacetic acid.
- ACTIVATOR DILUENT
  1 Bottle (900 mL) Activator Diluent. 0.3 sodium hydroxide.

ABBOTT PRISM Run Control Kit (REF 3E60-10)

ABBOTT PRISM Positive Run Control Kit (REF 3E60-11)

NOTE: Each batch MUST be used in a release control (ABBOTT PRISM Positive Control). The ABBOTT PRISM Positive Control (including 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.
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 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
Serum (including serum collected in serum 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 S/CO (Sample Net Counts/Cutoff Value). 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 to avoid hemolysis and stored frozen. 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.

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 phlebotomy, that have not been frozen, do not require centrifugation. All other specimens (including previously frozen phlebotomy specimens) must be centrifuged as follows:

- Gently invert each component several times prior to loading the original container on the ABBOTT PRISM System to ensure a homogenous solution.
- The activator solution must be used within 24 hours of preparation.
- 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 Activator Kits.
- Do not use kits beyond the expiration date.
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>Centrifugation 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: $RCF = \frac{1.12 \times f_{max}}{(rpm/1000)^2}$

Convert RCF to rpm as follows: $rpm = 1000 \left(\frac{RCF}{1.12 \times f_{max}}\right)$

<table>
<thead>
<tr>
<th>Centrifugation 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>

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>Centrifugation 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>

Any specimen (excluding non-frozen plasmapheresis) not tested within 24 hours of initial centrifugation must be recentrifuged from 30,000 to 75,000 g-minutes as defined for non-frozen specimens. For Cadaveric Specimens Only

Failure to follow the specified centrifugation procedure may give erroneous or inconsistent test results.

Centrifuged cadaveric SERUM specimens tested with ABBOTT PRISM HBsAg may be filtered using the instructions indicated below. If testing includes ABBOTT PRISM HIV O Plus, then the following instructions must be performed.

NOTE: Failure to adhere to the following instructions may result in erroneous or inconsistent test results for ABBOTT PRISM HIV O Plus.

Filtration of Centrifuged Cadaveric SERUM Specimens

Wear personal protective equipment, including eyewear.

After centrifugation, filter each cadaveric specimen through a Millipore GV Filter as follows:

1. Label an empty tube with the specimen identification number matching the original tube.
2. Remove the plunger from a sterile 10 cc syringe.

NOTE: Do not use a syringe smaller than 10 cc because excess pressure may build up, potentially causing damage to the filter unit or personal injury.
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 section 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).

For a description of the error codes that appear on ABBOTT PRISM System error lists, refer to the ABBOTT PRISM Operations Manual, Section 5: Operating Instructions, Reports. The ABBOTT PRISM System reports sample results in Net Counts and S/CO values.

S/CO = 2.00

Calculation of Cutoff and S/CO Values

The ABBOTT PRISM System calculates the ABBOTT PRISM HBsAg assay cutoff value using the following formula:

Cutoff Value = Mean Negative Calibrator (NC) Net Counts + (0.19 \times Mean Positive Calibrator [PC] Net Counts)

Example:

Mean NC Net Counts = 100
Mean PC Net Counts = 1,000

100 + (0.19 \times 1,000) = 290

Cutoff Value = 290

The ABBOTT PRISM System calculates the ABBOTT PRISM HBsAg assay S/CO for each sample and control using the following formula:

S/CO = Sample Net Counts / Cutoff Value

Example:

S/CO = Sample Net Counts / Cutoff Value

S/CO = 2.00

Interpretation of Results

• In the ABBOTT PRISM HBsAg assay, specimens with Net Counts less than the cutoff value are nonreactive and need not be tested further. Nonreactive specimens are considered negative for HBsAg by the criteria of ABBOTT PRISM HBsAg.

• Specimens with Net Counts greater than or equal to the cutoff value are considered initially reactive by the criteria of the ABBOTT PRISM HBsAg assay. All specimens (excluding non-frozen plasmapheresis specimens) that are reactive on initial testing must be centrifuged prior to retesting according to the table in the SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS section of this package insert. Initially reactive specimens must be retested in duplicate using the ABBOTT PRISM HBsAg assay.

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

• If the sample Net Counts for both retests are less than the cutoff value, the specimen is nonreactive. Nonreactive specimens are considered negative for HBsAg by the criteria of ABBOTT PRISM HBsAg.

• If the sample Net Counts for either duplicate retest are greater than or equal to the cutoff value, the specimen is considered repeatedly reactive.

Repeatedly reactive specimens must be tested by the ABBOTT PRISM HBsAg Confirmatory assay, a licensed neutralizing confirmatory test. Only the specimens which are confirmed by specific neutralization with anti-HBs are considered positive for HBsAg.

• Individuals who are repeatedly reactive may be referred for medical evaluation which may include additional testing.

Although the association of infectivity of donated blood or plasma and the presence of HBsAg is strong, it is recognized that presently available methods for HBsAg detection are not sensitive enough to detect all potentially infectious units of blood, plasma or possible cases of HBV infection. A nonreactive test result does not exclude infection.

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. The ABBOTT PRISM System reports sample results in Net Counts and S/CO. Net Counts are used by the ABBOTT PRISM System to interpret results. The S/CO value is provided in reports to show reactivity relative to the cutoff value. In the ABBOTT PRISM HBsAg assay, specimens with S/CO values of less than 1.00 are considered nonreactive. Specimens with an S/CO value of greater than or equal to 1.00 are considered reactive.

System Errors

For a description of the error codes that appear on ABBOTT PRISM System reports, refer to the ABBOTT PRISM Operations Manual, Section 10.
SIMPLIFIED PERFORMANCE CHARACTERISTICS

ASSAY REPRODUCIBILITY

ASSAY SPECIFICITY

SPECIFIC PERFORMANCE CHARACTERISTICS

ASSAY SPECIFICITY

A total of 25,238 fresh 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.

Specificity 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 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 by 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 (10 tested), and five pregnant females (555 tested). The estimated specificity in this population was 98.80% (820/830).

TABLE IV
Reactivity of the ABBOTT PRISM HBsAg Assay in Whole Blood and Plasmapheresis Donors, in Specimens from Individuals with Medical Conditions Unrelated to HBV Infection, and in Specimens Containing Potentially Interfering Substances

<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)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Volunteer Blood Donors</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Serum</td>
<td>8,246</td>
<td>5 (0.06)</td>
<td>3 (0.04)</td>
<td>2 (66.67)</td>
</tr>
<tr>
<td>Plasma</td>
<td>13,911</td>
<td>8 (0.06)</td>
<td>5 (0.04)</td>
<td>4 (80.00)</td>
</tr>
<tr>
<td>Plasmapheresis Donors</td>
<td>3,081</td>
<td>1 (0.03)</td>
<td>0 (0.00)</td>
<td>0 (0.00)</td>
</tr>
<tr>
<td>Total Donors</td>
<td>25,238</td>
<td>14 (0.06)</td>
<td>8 (0.03)</td>
<td>6 (24.00)</td>
</tr>
<tr>
<td>Medical Conditions</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unrelated to HBV Infection</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>and Potentially Interfering</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Substances</td>
<td>870</td>
<td>59 (6.78)</td>
<td>50 (5.75)</td>
<td>40 (46.00)</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.

Specimens from individuals with medical conditions unrelated to HBV infection and specimens containing potentially interfering substances included the following categories: anti-CMV positive (11), anti-EBV positive (12), anti-HIV-1 positive (12), anti-HIV-2 positive (5), anti-HTLV-I positive (12), anti-HTLV-II positive (12), non-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 (555).

The 50 repeatedly reactive specimens included the following: anti-EBV positive (1), anti-HIV-1 positive (1), anti-HIV-2 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).

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

TABLE III
ABBOTT PRISM HBsAg Assay Reproducibility

<table>
<thead>
<tr>
<th>Panel Number of Replicates</th>
<th>Mean S/CO*</th>
<th>SD %CV</th>
<th>SD %CV</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>6.98</td>
<td>0.283</td>
<td>4.1</td>
</tr>
<tr>
<td>2</td>
<td>4.4</td>
<td>0.160</td>
<td>3.9</td>
</tr>
<tr>
<td>3</td>
<td>1.39</td>
<td>0.068</td>
<td>4.9</td>
</tr>
<tr>
<td>4</td>
<td>3.99</td>
<td>0.866</td>
<td>5.3</td>
</tr>
<tr>
<td>5</td>
<td>4.62</td>
<td>0.162</td>
<td>3.5</td>
</tr>
<tr>
<td>6</td>
<td>3.7</td>
<td>0.078</td>
<td>5.7</td>
</tr>
<tr>
<td>7</td>
<td>0.34</td>
<td>0.036</td>
<td>10.6</td>
</tr>
</tbody>
</table>

Negative Control 439* 0.26 0.038 14.6 0.041 15.6

Positive Control 440 2.63 0.138 5.2 0.204 7.8

Cutoff Value = Mean Negative Calibrator Net Counts + (0.19 x Mean Positive Calibrator Net Counts)

<table>
<thead>
<tr>
<th>Category Number of Replicates</th>
<th>Mean Net Counts</th>
<th>SD %CV</th>
<th>SD %CV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Negative</td>
<td>660</td>
<td>89</td>
<td>10.8</td>
</tr>
<tr>
<td>Positive</td>
<td>660</td>
<td>1299</td>
<td>73.3</td>
</tr>
</tbody>
</table>

a Inter-assay variability contains intra-assay variability.
b One replicate was invalid due to instrument detection of sample dispense errors.
c Two replicates were invalid due to instrument detection of sample dispense errors.
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 (63.28%) 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 Reactively Positive (% of Total)</th>
<th>Number Confirmed Positive (% of Reactively Positive)</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>96</td>
<td>96 (100.00)</td>
<td>96 (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>47 (0.00)</td>
<td></td>
</tr>
</tbody>
</table>

In Increased Risk for HBV Infection:

<table>
<thead>
<tr>
<th>Risk for HBV Infection</th>
<th>Number Confirmed Positive (%)</th>
<th>Number Reactively Positive (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Increased Risk</td>
<td>452 (11.95)</td>
<td>41+ (75.93)</td>
</tr>
</tbody>
</table>

Total 1,212: 767 (63.28%) and 754 (98.31%)

a Specimens from the preselected HBsAg positive category were tested only once.

Assay 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 dispenser 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 (99.27%)</td>
<td>1 (0.73%)</td>
</tr>
<tr>
<td>Normal Donor</td>
<td>54</td>
<td>162</td>
<td>0.24</td>
<td>162 (100.00%)</td>
<td>(0.0%)</td>
</tr>
</tbody>
</table>

a 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.

b 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% (95% CI = [99.00%, 99.98%]) 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) ratio for the 142 postmortem replicates (51 specimens, with three reagent lots; see Table IX, footnote a) was 2.05, and the mean S/CO ratio 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>142 (100.00%)</td>
<td>(0.0%)</td>
</tr>
<tr>
<td>Normal Donor</td>
<td>54</td>
<td>162</td>
<td>2.07</td>
<td>162 (100.00%)</td>
<td>(0.0%)</td>
</tr>
</tbody>
</table>

a 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