ARCHITECT

SYSTEM

en

HBsAg Qualitative II Confirmatory

REF 2G23

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Read Highlighted Changes
Revised April 2019

# HBsAg Qualitative II Confirmatory

**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 used

| | | | |
|---|---|---|---|
| REF | List Number | CONTROL NO. | Control Number |
| IVD | *In Vitro* Diagnostic Medical Device | REACTION VESSELS | Reaction Vessels |
| LOT | Lot Number | REAGENT LOT | Reagent Lot |
| | Expiration Date | REPLACEMENT CAPS | Replacement Caps |
| SN | Serial Number | SAMPLE CUPS | Sample Cups |
| 2°C 8°C | Store at 2-8°C | SEPTUM | Septum |
| | | WARNING: SENSITIZER | Warning: May cause an allergic reaction |
| | Caution | CONTAINS: AZIDE | Contains Sodium Azide. Contact with acids liberates very toxic gas. |
| | Consult instructions for use | GTIN | Global Trade Item Number |
| | Manufacturer | PRODUCT OF IRELAND | Product of Ireland |

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

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## NAME

ARCHITECT HBsAg Qualitative II Confirmatory

## INTENDED USE

The ARCHITECT HBsAg Qualitative II Confirmatory assay is a chemiluminescent microparticle immunoassay (CMIA) for the confirmation of the presence of hepatitis B surface antigen (HBsAg) in human serum and plasma including specimens collected post-mortem (non-heart-beating) by means of specific antibody neutralization.

It is intended to be used for the confirmation of samples found to be repeatedly reactive by ARCHITECT HBsAg Qualitative II.

## SUMMARY AND EXPLANATION OF TEST

The causative agent of serum hepatitis is hepatitis B virus (HBV) which is an enveloped DNA virus. During infection, HBV produces an excess of hepatitis B surface antigen (HBsAg), also known as Australia antigen, which can be detected in the blood of infected individuals. It is responsible for binding the virus to the liver cell and is the target structure of neutralizing antibodies.[1,2] HBsAg is the first serological marker after infection with HBV appearing one to ten weeks after exposure and two to eight weeks before the onset of clinical symptoms.[3,4] HBsAg persists during this acute phase and clears late in the convalescence period. Failure to clear HBsAg within six months indicates a chronic HBsAg carrier state.

HBsAg assays are used to identify persons infected with HBV and to prevent transmission of the virus by blood and blood products as well as to monitor the status of infected individuals in combination with other hepatitis B serological markers.[5] In most countries, testing for HBsAg is part of the antenatal screening program to identify HBV infected mothers and to prevent perinatal HBV infection by subsequent immunization.[6]

The hepatitis B virus, 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.[7] Some of these mutations may cause changes in the antigenic structure of HBsAg, resulting in epitopes that are no longer recognized by anti-HBs. HBsAg mutants have been reported in a wide range of patient populations, including blood donors, vaccine recipients, renal dialysis patients, orthotopic liver transplant recipients, infants born to HBsAg-positive mothers, and patients undergoing nucleoside analog treatment for HBV.[7-14] HBsAg mutations may result in a less favorable outcome in some patients[7,8,10] and false negative results in some HBsAg assays.[7-9]

It is recommended that confirmatory testing be performed prior to disclosure of HBV status. ARCHITECT HBsAg Qualitative II Confirmatory uses the principle of specific antibody neutralization to confirm the presence of HBsAg in samples found to be repeatedly reactive. Antibody to hepatitis B surface antigen (anti-HBs, human) is incubated with a sample. If HBsAg is present in the sample, it will be neutralized by the antibody. The neutralized HBsAg is subsequently blocked from binding to the anti-HBs coated microparticles. A reduction of signal occurs when compared to the signal of a paired sample that has not been treated with the antibody reagent. A sample is considered confirmed if the signal for the non-neutralized sample (incubated with Pre-Treatment 2) result is greater than or equal to the cutoff of 0.70 S/CO and the RLU of the neutralized sample is reduced by at least 50% compared to the non-neutralized sample.

## BIOLOGICAL PRINCIPLES OF THE PROCEDURE

The ARCHITECT HBsAg Qualitative II Confirmatory assay consists of two single tests that are both one-step pre-treatment immunoassays. The ARCHITECT HBsAg Qualitative II Confirmatory assay is for the confirmation of the presence of hepatitis B surface antigen (HBsAg) in human serum and plasma, using CMIA technology with flexible assay protocols, referred to as Chemiflex. (Note: As Ancillary Wash Buffer is added in a second incubation step, the assay files indicate a two-step assay.)

In the ARCHITECT HBsAg Qualitative II Confirmatory assay, sample and Pre-Treatment 1 are combined in a reaction vessel (RV) and incubated. When HBsAg is present in the sample, it is neutralized by the antibody (anti-HBs) in Pre-Treatment 1. An aliquot of the pretreated sample, anti-HBs coated paramagnetic microparticles, and anti-HBs acridinium-labeled conjugate are combined to create a reaction mixture. Any non-neutralized HBsAg present in the sample binds to the anti-HBs coated microparticles and to the anti-HBs acridinium-labeled conjugate. The neutralized HBsAg is blocked from forming a sandwich with acridinium-labeled anti-HBs conjugate and anti-HBs coated microparticles. After washing, ancillary wash buffer is added to the RV and incubated. Following another wash cycle, pre-trigger and trigger solutions are added to the reaction mixture. The resulting chemiluminescent reaction is measured as relative light units (RLUs). A direct relationship exists between the amount of HBsAg in the sample and the RLUs detected by the ARCHITECT *i* System optics.

This sequence is repeated for the sample and Pre-Treatment 2, except Pre-Treatment 2 does not neutralize HBsAg in the sample. If the signal for the non-neutralized sample (incubated with Pre-Treatment 2) result is greater than or equal to the cutoff of 0.70 S/CO and the RLU of the neutralized sample (incubated with Pre-Treatment 1) is reduced by at least 50% compared to the non-neutralized sample, the sample is considered confirmed positive for HBsAg.

For additional information on system and assay technology, refer to the ARCHITECT System Operations Manual, Section 3.

## REAGENTS

**NOTE:** Some kit sizes are not available in all countries or for use on all ARCHITECT *i* Systems. Please contact your local distributor.

### Reagent Kit, 100 Tests (50 Determinations)

ARCHITECT HBsAg Qualitative II Confirmatory Reagent Kit (2G23)

- **MICROPARTICLES** 1 bottle (6.6 mL) anti-HBs (mouse, monoclonal, IgM, IgG) coated microparticles in MES buffer with protein (bovine serum albumin) stabilizer. Minimum concentration: 0.08% solids. Preservatives: ProClin 300 and ProClin 950.
- **CONJUGATE** 1 bottle (5.9 mL) anti-HBs (mouse, monoclonal, IgG) and anti-HBs (goat, IgG) acridinium-labeled conjugate in phosphate buffer with human plasma and protein (bovine serum albumin, fetal bovine serum, goat IgG, mouse IgG) stabilizers. Minimum concentration: 0.35 µg/mL. Preservatives: ProClin 300 and ProClin 950.
- **ANCILLARY WASH BUFFER** 1 bottle (5.9 mL) ancillary wash buffer containing MES buffer. Preservatives: ProClin 300 and ProClin 950.
- **PRE-TREATMENT 1** 1 bottle (2.4 mL) Pre-Treatment 1 containing recalcified human plasma reactive for anti-HBs. Preservatives: ProClin 300 and ProClin 950.
- **PRE-TREATMENT 2** 1 bottle (2.4 mL) Pre-Treatment 2 containing recalcified human plasma. Preservatives: ProClin 950 and sodium azide.

### Assay Diluent

ARCHITECT HBsAg Qualitative II Confirmatory Manual Diluent (2G23-40)

- **MANUAL DILUENT** 1 bottle (100 mL) ARCHITECT HBsAg Qualitative II Confirmatory Manual Diluent containing recalcified human plasma. Preservatives: ProClin 950 and sodium azide.

### Other Reagents

ARCHITECT *i* Pre-Trigger Solution

- **PRE-TRIGGER SOLUTION** Pre-trigger solution containing 1.32% (w/v) hydrogen peroxide.

ARCHITECT *i* Trigger Solution

- **TRIGGER SOLUTION** Trigger solution containing 0.35 N sodium hydroxide.

ARCHITECT *i* Wash Buffer

- **WASH BUFFER** Wash buffer containing phosphate buffered saline solution. Preservatives: antimicrobial agents.

## WARNINGS AND PRECAUTIONS

- **IVD**
- For *In Vitro* Diagnostic 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.

### Safety 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 or inactivated microorganisms will not transmit infection. Therefore, all human sourced materials should be considered potentially infectious. It is recommended that these reagents and human specimens be handled in accordance with the OSHA Standard on Bloodborne Pathogens[15]. Biosafety Level 2[16] or other appropriate biosafety practices[17,18] should be used for materials that contain or are suspected of containing infectious agents.

- The Conjugate contains human plasma that is nonreactive for HBsAg, HIV-1 Ag or HIV-1 RNA, anti-HIV-1/HIV-2, and anti-HCV.
- Pre-Treatment 1 contains human plasma that is reactive for anti-HBs, and nonreactive for HBsAg, HIV-1 Ag or HIV-1 RNA, anti-HIV-1/HIV-2, and anti-HCV.
- Pre-Treatment 2 contains human plasma that is nonreactive for HBsAg, HIV-1 Ag or HIV-1 RNA, anti-HIV-1/HIV-2, anti-HCV, and anti-HBs.
- The following warnings and precautions apply to these components:
  - Microparticles
  - Conjugate
  - Ancillary Wash Buffer
  - Pre-Treatment 1
  - Pre-Treatment 2

| | |
|---|---|
| **WARNING:** | Contains methylisothiazolones |
| H317 | 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. |
| P363 | Wash contaminated clothing before reuse. |

This material and its container must be disposed of in a safe way.

- Pre-Treatment 2 contains sodium azide. Contact with acids liberates very toxic gas. This material and its container must be disposed of in a safe way.
- Safety Data Sheets are available at www.abbottdiagnostics.com or contact your local representative.
- For information on the safe disposal of sodium azide and a detailed discussion of safety precautions during system operation, refer to the ARCHITECT System Operations Manual, Section 8.

**Handling Precautions**

- Do not use reagent kits beyond the expiration date.
- **Do not pool reagents within a kit or between reagent kits.**
- Before loading the ARCHITECT HBsAg Qualitative II Confirmatory Reagent Kit on the system for the first time, the microparticle bottle requires mixing to resuspend microparticles that may have settled during shipment. For microparticle mixing instructions, refer to the **PROCEDURE, Assay Procedure** section of this package insert.
- **Septums MUST be used to prevent reagent evaporation and contamination and to ensure reagent integrity. Reliability of assay results cannot be guaranteed if septums are not used according to the instructions in this package insert.**
- To avoid contamination, wear clean gloves when placing a septum on an uncapped reagent bottle.
  - When handling conjugate vials, change gloves that have contacted human serum or plasma, since introduction of human IgG or IgM will result in a neutralized conjugate.
  - Once a septum has been placed on the reagent bottle, **do not invert the bottle** as this will result in reagent leakage and may compromise assay results.
  - Over time, residual liquids may dry on the septum surface. These are typically dried salts, and have no effect on assay efficacy.
- For a detailed discussion of handling precautions during system operation, refer to the ARCHITECT System Operations Manual, Section 7.

**Storage Instructions**


- The ARCHITECT HBsAg Qualitative II Confirmatory Reagent Kit must be stored at 2-8˚C in an upright position and may be used immediately after removal from 2-8˚C storage.
- When stored and handled as directed, the reagents are stable until the expiration date.
- The ARCHITECT HBsAg Qualitative II Confirmatory Reagent Kit may be stored on board the ARCHITECT *i* System for a maximum of 30 days. After 30 days, the reagent kit must be discarded. For information on tracking onboard time, refer to the ARCHITECT System Operations Manual, Section 5.
- Reagents may be stored on or off the ARCHITECT *i* System. If reagents are removed from the system, store them at 2-8˚C (with septums and replacement caps) in an upright position. For reagents stored off the system, it is recommended that they be stored in their original trays and boxes to ensure they remain upright. **If the microparticle bottle does not remain upright (with a septum installed) while in refrigerated storage off the system, the reagent kit must be discarded.** For information on unloading reagents, refer to the ARCHITECT System Operations Manual, Section 5.

**Indications of Reagent Deterioration**

When a control value is out of the specified range, it may indicate deterioration of the reagents or errors in technique. Associated test results are invalid and samples must be retested. Assay recalibration may be necessary. For troubleshooting information, refer to the ARCHITECT System Operations Manual, Section 10.

## INSTRUMENT PROCEDURE

- The ARCHITECT HBsAg Qualitative II Confirmatory assay files must be installed on the ARCHITECT *i* System before performing the assay.
  - Install the software and assay files in the following order:
    1. ARCHITECT System Software Version 7.00 or higher
    2. HBsAgQ2 C2 (assay number 629)
    3. HBsAgQ2 C1 (assay number 630)
  - When assay files 629 and 630 are installed, assay HBsAgQ2 %N and patient panel HBsAgQ2 Pa are installed automatically.
    - HBsAgQ2 %N allows the instrument to automatically calculate a percent neutralization result from the HBsAgQ2 C2 and HBsAgQ2 C1 results.
    - HBsAgQ2 Pa provides a convenient method to order HBsAg confirmatory tests so the ARCHITECT *i* System will report the S/CO and % neutralization results required for the interpretation.
  - Recommended system configuration steps:
    - Configure result units and decimal places.
      - It is recommended to configure the result unit to % and the decimal places to 0 decimal places for HBsAgQ2 %N.
      - For information on configuring a result unit and decimal places, refer to the ARCHITECT System Operations Manual, Section 2.
    - Configure the Positive Control as a multiconstituent control (MCC)
      - It is recommended to configure the ARCHITECT HBsAg Qualitative II Positive Control as an MCC with the HBsAgQ2 %N and HBsAgQ2 C2 assays.
      - For information on configuring a multiconstituent control, refer to the ARCHITECT System Operations Manual, Section 2.
  - Refer to the **PROCEDURE, Assay Procedure** section of this package insert for information on ordering tests.
- For detailed information on assay file installation and viewing and editing assay parameters, refer to the ARCHITECT System Operations Manual, Section 2.
- For information on printing assay parameters, refer to the ARCHITECT System Operations Manual, Section 5.
- For a detailed description of system procedures, refer to the ARCHITECT System Operations Manual.

## SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS

### Specimen Types

- The specimen collection tubes listed below were verified for use with the ARCHITECT HBsAg Qualitative II Confirmatory assay. Other specimen collection tubes have not been tested with this assay.
  - Human serum (including serum collected in serum separator tubes)
  - Human plasma collected in:
    - Lithium heparin
    - Dipotassium EDTA
    - Tripotassium EDTA
    - Sodium citrate
    - Plasma separator tubes (lithium heparin)
    - Sodium heparin
    - CPD
    - CPDA-1
    - ACD
    - Potassium oxalate / sodium fluoride plasma
- Performance has not been established for the use of body fluids other than human serum or plasma.
- Performance has been established for the use of cadaveric blood specimens (specimens collected post-mortem, non-heart-beating), for details refer to section **TESTING OF CADAVERIC BLOOD SPECIMENS.**
- Liquid anticoagulants may have a dilution effect resulting in lower concentrations for individual patient specimens.
- The ARCHITECT *i* System does not provide the capability to verify specimen type. It is the responsibility of the operator to verify that the correct specimen types are used in the ARCHITECT HBsAg Qualitative II Confirmatory assay.

### Specimen Conditions

- Do not use specimens with the following conditions:
  - heat-inactivated
  - pooled
  - grossly hemolyzed
  - obvious microbial contamination
- For accurate results, serum and plasma specimens should be free of fibrin, red blood cells, and other particulate matter. Serum specimens from patients receiving anticoagulant or thrombolytic therapy may contain fibrin due to incomplete clot formation.
- Ensure that complete clot formation in serum specimens has taken place prior to centrifugation. If the specimen is centrifuged before a complete clot forms, the presence of fibrin may cause erroneous results.
- As specimens from heparinized patients may be partially coagulated and erroneous results could occur due to the presence of fibrin, draw the specimen prior to heparin therapy.
- Use caution when handling patient specimens to prevent cross contamination. Use of disposable pipettes or pipette tips is recommended.
- For optimal results, inspect all specimens for bubbles. Remove bubbles with an applicator stick before analysis. Use a new applicator stick for each specimen to prevent cross contamination.
- No qualitative performance differences were observed between experimental controls and nonreactive or spiked reactive specimens tested with elevated levels of conjugated and unconjugated bilirubin (20 mg/dL), triglycerides (3000 mg/dL), protein (12 g/dL), or hemoglobin (500 mg/dL).

### Preparation for Analysis

- Follow the tube manufacturer's processing instructions for serum and plasma collection tubes. Gravity separation is not sufficient for specimen preparation.
- Prepare frozen specimens as follows:
  - Frozen specimens must be completely thawed before mixing.
  - Mix thawed specimens thoroughly by inverting 10 times or by low speed vortexing. Visually inspect the specimens. If layering or stratification is observed, continue mixing until specimens are visibly homogeneous. If samples are not mixed thoroughly, inconsistent results may be obtained.
  - Centrifuge mixed specimens as described below.
- To ensure consistency in results, specimens must be transferred to a centrifuge tube and centrifuged at ≥ 10,000 RCF (Relative Centrifugal Force) for 10 minutes before testing if
  - they contain fibrin, red blood cells, or other particulate matter or
  - they were frozen and thawed.
- Centrifuged specimens with a lipid layer on the top must be transferred to a sample cup or secondary tube. Care must be taken to transfer only the clarified specimen without the lipemic material.
- Transfer clarified specimen to a sample cup or secondary tube for testing.

### Storage

- Specimens may be stored on or off the clot, red blood cells, or separator gel for
  - up to 24 hours at room temperature or
  - up to 6 days at 2-8°C.
- If testing will be delayed more than 6 days, remove serum or plasma from the clot, red blood cells, or separator gel and store at -20°C or colder.
- Avoid more than 3 freeze/thaw cycles.

### Shipping

- Before shipping specimens, it is recommended that specimens be removed from the clot, red blood cells, or separator gel.
- When shipping specimens, package and label specimens in compliance with applicable state, federal, and international regulations covering the transport of clinical specimens and infectious substances.
- Specimens may be shipped ambient, at 2-8°C (wet ice), or frozen (dry ice). Do not exceed the storage time limitations listed above.

## TESTING OF CADAVERIC BLOOD SPECIMENS

- Performance has been established for the use of cadaveric blood specimens (specimens collected post-mortem, non-heart-beating) that have been collected up to 18.5 hours after death. Performance was established using 50 spiked and 50 non-spiked cadaveric blood specimens[19].
- Testing of cadaveric blood specimens from patients with plasma dilution due to transfusions of > 2000 mL of blood or colloids within 48 hours, or > 2000 mL of crystalloids within 1 hour (or any combination thereof) prior to collection of the specimens have not been validated.
- Follow general standards and/or regulations for collection, storage and handling.
- Follow the tube manufacturer's processing instructions for serum or plasma collection tubes. After initial centrifugation, transfer the supernatant to a centrifuge tube and centrifuge at ≥ 10,000 RCF (Relative Centrifugal Force) for 10 minutes. If specimens are not processed directly after initial centrifugation, it is recommended to remove the supernatant from the clot or red blood cells until further processing.
- Cadaveric blood specimens can be stored for up to 6 days at 2-8°C or up to 24 hours at 15-30°C following collection.
- No qualitative differences were observed for cadaveric blood specimens (spiked reactive) when subjected to up to 3 freeze/thaw cycles. However, multiple freeze/thaw cycles should be avoided.

## PROCEDURE

### Materials Provided

- 2G23 ARCHITECT HBsAg Qualitative II Confirmatory Reagent Kit

### Materials Required but not Provided

- ARCHITECT *i* System
- ARCHITECT HBsAg Qualitative II Confirmatory Assay file, may be obtained from:
  - ARCHITECT *i* System e-Assay CD-ROM found on www.abbottdiagnostics.com
  - ARCHITECT *i* System Assay CD-ROM
- 2G22-01 ARCHITECT HBsAg Qualitative II Calibrators
- 2G22-10 ARCHITECT HBsAg Qualitative II Controls or other control material
- 2G23-40 ARCHITECT HBsAg Qualitative II Confirmatory Manual Diluent
- ARCHITECT *i* **PRE-TRIGGER SOLUTION**
- ARCHITECT *i* **TRIGGER SOLUTION**
- ARCHITECT *i* **WASH BUFFER**
- ARCHITECT *i* **REACTION VESSELS**
- ARCHITECT *i* **SAMPLE CUPS**
- ARCHITECT *i* **SEPTUM**
- ARCHITECT *i* **REPLACEMENT CAPS**
- Pipettes or pipette tips (optional) to deliver the specified volumes.

For information on materials required for maintenance procedures, refer to the ARCHITECT System Operations Manual, Section 9.

**Assay Procedure**

- Before loading the ARCHITECT HBsAg Qualitative II Confirmatory Reagent Kit on the system for the first time, the microparticle bottle requires mixing to resuspend microparticles that may have settled during shipment. After the first time the microparticles have been loaded, no further mixing is required.
  - **Invert the microparticle bottle 30 times.**
  - Visually inspect the bottle to ensure microparticles are resuspended. If microparticles are still adhered to the bottle, continue to invert the bottle until the microparticles have been completely resuspended.
  - **If the microparticles do not resuspend, DO NOT USE. Contact your Abbott representative.**
  - Once the microparticles have been resuspended, place a septum on the bottle. For instructions about placing septums on bottles, refer to the **Handling Precautions** section of this package insert.
- Load the ARCHITECT HBsAg Qualitative II Confirmatory Reagent Kit on the ARCHITECT *i* System.
  - Verify that all necessary reagents are present.
  - Ensure that septums are present on all reagent bottles.
- Order calibration, if necessary.
  - For information on ordering calibrations, refer to the **PROCEDURE, Calibration** section of this package insert.
- Order tests.
  - For each HBsAg confirmatory result, two tests must performed by the ARCHITECT *i* System: HBsAgQ2 C1 and HBsAgQ2 C2. The percent neutralization result is calculated from these two test results.
  - It is recommended that patient specimens be ordered using the patient panel HBsAgQ2 Pa. HBsAgQ2 Pa automatically selects the necessary assay files to test and reports the results required for assay interpretation (HBsAgQ2 C2 S/CO and percent neutralization).
  - If a rerun is required to ensure the calculated % neutralization result is based on constituent assay results, run on the same day,
    - perform the rerun on the same day the exception is generated or
    - perform the rerun on a different day using the calculated assay (HBsAgQ2 %N) and both constituent assays (HBsAgQ2 C1 and HBsAgQ2 C2).
  - For information on ordering patient specimens and controls and for general operating procedures, refer to the ARCHITECT System Operations Manual, Section 5.
- The minimum sample cup volume is calculated by the system and is printed on the Orderlist report. No more than 10 tests may be sampled from the sample cup. To minimize the effects of evaporation, verify adequate sample cup volume is present before running the test.
  - Priority: 242 µL (sample volume for HBsAgQ2 C1 and HBsAgQ2 C2).
  - ≤ 3 hours on-board: 242 µL (sample volume for HBsAgQ2 C1 and HBsAgQ2 C2).
  - > 3 hours on-board: replace with a fresh sample (patient specimens, controls, and calibrators).
  - If using primary or aliquot tubes, use the sample gauge to ensure sufficient patient specimen is present.
- Prepare calibrators and controls.
  - Refer to the **INSTRUMENT PROCEDURE** section of this package insert for information on configuring the Positive Control.
  - Mix the ARCHITECT HBsAg Qualitative II Calibrators and Controls by gentle inversion before use.
  - To obtain the recommended volume requirements for the ARCHITECT HBsAg Qualitative II Calibrators, hold the bottles **vertically** and dispense 14 drops of each calibrator (for 3 replicates) into each respective sample cup.
  - To obtain the recommended volume requirement for the ARCHITECT HBsAg Qualitative II Positive Control, hold the bottle **vertically** and dispense 10 drops of Positive Control (for two replicates, one for HBsAgQ2 C1 and one for HBsAgQ2 C2) into each respective sample cup.
  - If commercially available control material is used, follow the manufacturer's instructions for preparation.
- Load samples
  - For information on loading samples, refer to the ARCHITECT System Operations Manual, Section 5.
- Press RUN.
- For additional information on principles of operation, refer to the ARCHITECT System Operations Manual, Section 3.
- For optimal performance, it is important to perform routine maintenance as described in the ARCHITECT System Operations Manual, Section 9. Perform maintenance more frequently when required by laboratory procedures.

**Specimen Dilution Procedure**

- A Manual Dilution Procedure may be performed on a neat sample if the ARCHITECT HBsAg Qualitative II Confirmatory assay has a HBsAgQ2 C2 S/CO of ≥ 10.00 and a % Neutralization of < 50%. Refer to the **RESULTS** section of this package insert for further information.
- The suggested dilution for the ARCHITECT HBsAg Qualitative II Confirmatory assay is 1:500.
  - Add 25 µL of the patient specimen to 475 µL of ARCHITECT HBsAg Qualitative II Confirmatory Manual Diluent for a 1:20 dilution.
  - Add 20 µL of the 1:20 dilution to 480 µL of ARCHITECT HBsAg Qualitative II Confirmatory Manual Diluent for a 1:500 dilution.
- Additional specimen dilutions may be performed if the 1:500 dilution result is still reactive but not neutralized.
  - For a 1:20000 dilution, add 25 µL of the 1:500 dilution to 975 µL of ARCHITECT HBsAg Qualitative II Confirmatory Manual Diluent.
- **NOTE: Manual dilution factors cannot be entered into the Patient or Control order screen. However, for maintenance of detailed information (records) - Select Patient Order then select the appropriate Assay. Select Sample Details F2. Enter the Dilution Factor in the Comments Box.**
- Refer to **Interpretation of Results** section of this package insert for further information.

**Calibration**

- To perform an ARCHITECT HBsAg Qualitative II Confirmatory calibration, test ARCHITECT HBsAg Qualitative II Calibrators 1 and 2 in replicates of 3 with the HBsAgQ2 C2 assay. The HBsAgQ2 C1 assay uses the calibration generated from HBsAgQ2 C2.
  - The calibrators should be priority loaded.
  - The minimum sample cup volume required for ordering 3 replicates of each of the calibrators is 338 µL for each calibrator.
- A single sample of the ARCHITECT HBsAg Qualitative II Positive Control must be tested to evaluate the assay calibration.
  - Order the positive control as described in the **Assay Procedure** section of this package insert.
  - Ensure that the positive control S/CO and % Neutralization results are within the ranges specified in the control package insert.
- Once an ARCHITECT HBsAg Qualitative II Confirmatory calibration is accepted and stored, all subsequent samples may be tested without further calibration unless:
  - A reagent kit with a new lot number is used.
  - Controls are out of range.
- For detailed information on how to perform an assay calibration, refer to the ARCHITECT System Operations Manual, Section 6.

## QUALITY CONTROL PROCEDURES

The recommended control requirement for the ARCHITECT HBsAg Qualitative II Confirmatory assay is that a single sample of the positive control be tested once every 24 hours each day of use. If your laboratory quality control procedures require more frequent use of controls to verify test results, follow those procedures. Additional controls may be tested in conformance with local, state, and/or federal regulations or accreditation requirements and your laboratory's quality control policy.

Control values must be within the ranges specified in the control package insert. If a control result is out of its specified range, any test results generated since the last acceptable control results must be evaluated to determine if test results may have been adversely affected. Adversely affected test results are invalid, and these samples must be retested. For troubleshooting information, refer to the ARCHITECT System Operations Manual, Section 10.

## RESULTS

The ARCHITECT HBsAg Qualitative II Confirmatory result is based on the sample to cutoff ratio (S/CO) and % neutralization of the sample.

Note: If the sample's HBsAgQ2 C2 S/CO is < 0.70, % neutralization is not applicable. Obtain the final interpretation of results directly from the table in the **Interpretation of Results** section in this package insert.

### Calculation

- The ARCHITECT *i* System calculates the S/CO result for the ARCHITECT HBsAg Qualitative II Confirmatory assay using the ratio of the sample RLU to the cutoff RLU (S/CO) for each specimen and control.
  - Cutoff RLU = (0.0575 x Calibrator 1 Mean RLU) + (0.8 x Calibrator 2 Mean RLU)
  - S/CO = Sample RLU/Cutoff RLU
- The ARCHITECT *i* System calculates the % Neutralization result for the ARCHITECT HBsAg Qualitative II Confirmatory assay using the HBsAgQ2 C1 and HBsAgQ2 C2 results for each specimen and control using the following equation:

$$\% \text{ Neutralization} = \frac{\text{(Sample HBsAgQ2 C2 RLU)-(Sample HBsAgQ2 C1 RLU)}}{\text{(Sample HBsAgQ2 C2 RLU)-(Calibrator 2 HBsAgQ2 C2 Mean RLU)}} \times 100$$

### Interpretation of Results

- The table below summarizes the various final interpretations from the neat and dilution results:

| Dilution | HBsAgQ2 C2 S/CO | % Neutralization* | Final Interpretation |
|---|---|---|---|
| Neat (Undiluted) | < 0.70 | Not applicable | Not confirmed |
| | < 10.00 | < 50% | Not confirmed |
| | ≥ 0.70 | ≥ 50% | Confirmed positive |
| | ≥ 10.00 | < 50% | Repeat test using a 1:500 dilution |
| 1:500 | < 0.70 | Not applicable | Not confirmed |
| | ≥ 0.70 | ≥ 50% | Confirmed positive |
| | ≥ 0.70 | < 50% | Repeat test using a 1:20000 dilution |
| 1:20000 | < 0.70 | Not applicable | Not confirmed |
| | ≥ 0.70 | ≥ 50% | Confirmed positive |
| | ≥ 0.70 | < 50% | Not confirmed |

* If the % neutralization is < -15%, then the results should be considered invalid and the specimen should be retested. Perform the retest using the calculated assay (HBsAgQ2 %N) and both constituent assays (HBsAgQ2 C1 and HBsAgQ2 C2).

- NOTES:
  - Follow the dilution and final interpretation routine as outlined in the table above, even if % neutralization results of -15% to 0%, or > 100% are obtained.
  - For specimen dilution instructions, refer to the **Specimen Dilution Procedure** section of this package insert.
  - The interpretation of not confirmed for HBsAg indicates the presence of HBsAg cannot be confirmed via neutralization. The repeatedly reactive result obtained with the ARCHITECT HBsAg Qualitative II assay may be the result of a nonspecific reaction (false reactive). As the presence of nonspecific binding may obscure low levels of HBsAg in the specimen due to early infection or early recovery, it is recommended that the patient be evaluated for other serologic markers of HBV infection (*i.e.*, total anti-HBc or IgM anti-HBc)[20] and that the patient be retested for HBsAg in 4 to 6 weeks.[21]
  - Although there is an association between the presence of HBsAg, infectivity and a reactive result, it is recognized that presently available methods for HBsAg confirmation may not confirm all possible cases of HBV infection.

### Flags

Some results may contain information in the Flags field. For a description of the flags that may appear in this field, refer to the ARCHITECT System Operations Manual, Section 5.

## LIMITATIONS OF THE PROCEDURE

- If the ARCHITECT HBsAg Qualitative II Confirmatory results are inconsistent with clinical evidence, additional testing is suggested to confirm the result.
- For diagnostic purposes, results should be used in conjunction with patient history and other hepatitis markers for diagnosis of acute and chronic infection.
- Specimens from patients who have received preparations of mouse monoclonal antibodies for diagnosis or therapy may contain human anti-mouse antibodies (HAMA).[22,23] Specimens containing HAMA may produce anomalous values when tested with assay kits such as ARCHITECT HBsAg Qualitative II Confirmatory that employ mouse monoclonal antibodies.[22]
- Heterophilic antibodies in human serum can react with reagent immunoglobulins, interfering with *in vitro* immunoassays.[24] Patients routinely exposed to animals or to animal serum products can be prone to this interference and anomalous results may be observed. Additional information may be required for diagnosis.
- Refer to the **SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS** section of this package insert for specimen limitations.

## SPECIFIC PERFORMANCE CHARACTERISTICS

Assay results obtained in individual laboratories may vary from data presented.

### Confirmation of HBsAg Reactive Specimens

A total of 7505 specimens from the categories presented in the table below were evaluated with the ARCHITECT HBsAg Qualitative II assay. There were a total of 338 repeatedly reactive specimens, which were tested with the ARCHITECT HBsAg Qualitative II Confirmatory assay to confirm the presence of HBsAg. In 332 of the 338 repeatedly reactive specimens (98.22%) the presence of HBsAg was confirmed. The remaining 6 samples were tested with a commercially available HBsAg assay. Four specimens were nonreactive and two specimens were repeat reactive and not confirmed. The data are summarized in the following table.

| Category | N Total Number Tested | ARCHITECT HBsAg Qualitative II Number of Reactive Specimens (% of Total) | | ARCHITECT HBsAg Qualitative II Confirmatory Number of Confirmed Specimens[a] (% of Reactive Specimens) | |
|---|---|---|---|---|---|
| Overall Blood Donors | 5401 | 6 | (0.11%) | 1 | (16.67%) |
| Diagnostics Patients | 1499 | 17 | (1.13%) | 16[b] | (94.12%) |
| Preselected HBsAg Positive | 311 | 311 | (100.00%) | 311 | (100.00%) |
| Potentially Interfering Substances | 294 | 4[c] | (1.36%) | 4[c] | (100.00%) |
| Total | 7505 | 338 | (4.50%) | 332 | (98.22%) |

[a] A specimen is considered as confirmed if the signal for the non-neutralized specimen (incubated with Pre-Treatment 2) result is greater than or equal to the cutoff (S/CO ≥ 0.70) and the RLU of the neutralized specimen is reduced by at least 50% compared to the non-neutralized specimen.

[b] One aberrant result was observed. Without this aberrant result, the number of confirmed specimens was 15 (88.24%).

[c] Samples were from the Human Immunodeficiency Virus (HIV-2) and Pregnant 2nd Trimester categories.

### Analytical Sensitivity

Analytical sensitivity was evaluated using serial dilutions of the WHO 2nd International HBsAg Standard (subtype *adw2*, genotype A, NIBSC Code 00/588). The dilutions ranged from 0.010 to 0.5 IU/mL. The dilutions that were repeat reactive by the ARCHITECT HBsAg Qualitative II assay (2G22) were tested across 3 ARCHITECT HBsAg Qualitative II Confirmatory reagent lots on 2 instrument types (1 $i2000_{SR}$ and 1 $i1000_{SR}$). In this study, the ARCHITECT HBsAg Qualitative II Confirmatory assay confirmed as positive all dilutions detected as repeat reactive by the ARCHITECT HBsAg Qualitative II assay.

## BIBLIOGRAPHY


1. Neurath AR, Kent SB, Strick N, *et al*. Identification and chemical synthesis of a host cell receptor binding site on hepatitis B virus. *Cell* 1986;46:429-36.
2. Szmuness W, Stevens CE, Harley EJ, *et al*. Hepatitis B vaccine-demonstration of efficacy in a controlled clinical trial in a high-risk population in the United States. *N Engl J Med* 1980;303:833-41.
3. Krugman S, Giles JP. Viral hepatitis, type B (MS-2-Strain)- further observations on natural history and prevention. *N Engl J Med* 1973;288:755-60.
4. Krugman S, Overby LR, Mushahwar IK, *et al*. Viral hepatitis, type B-studies on natural history and prevention re-examined. *N Engl J Med* 1979;300:101-6.
5. Perrillo RP, Aach RD. The clinical course and chronic sequelae of hepatitis B virus infection. *Seminars in Liver Disease* 1981;1:15-25.
6. CDC. A comprehensive immunization strategy to eliminate transmission of Hepatitis B virus infection in the United States: recommendations of the Advisory Committee on Immunization Practices (ACIP) Part 1: Immunization of Infants, Children, and Adolescents. *MMWR* 2005;54(RR-16):1-23.
7. Hunt CM, McGill JM, Allen MI, *et al*. Clinical relevance of hepatitis B viral mutations. *Hepatology* 2000;31(5):1037-44.
8. Locarnini SA. Hepatitis B virus surface antigen and polymerase gene variants: potential virological and clinical significance. *Hepatology* 1998;27(1):294–7.
9. Zuckerman AJ. Effect of hepatitis B virus mutants on efficacy of vaccination. *Lancet* 2000;355:1382-4.
10. Carman WF, Trautwein C, Van Deursen FJ, *et al*. Hepatitis B virus envelope variation after transplantation with and without hepatitis B immune globulin prophylaxis. *Hepatology* 1996;24(3):489-93.
11. Grethe S, Monazahian M, Böhme I, *et al*. Characterization of unusual escape variants of hepatitis B virus isolated from a hepatitis B surface antigen-negative subject. *J Virology* 1998;72(9):7692-6.
12. Nainan OV, Stevens CE, Taylor PE, *et al.* Hepatitis B virus (HBV) antibody resistant mutants among mothers and infants with chronic HBV infection. In: Rizzetto M, Purcell RH, Gerin JL, *et al.,* eds. *Viral Hepatitis and Liver Disease.* Minerva Medica: Torino; 1997:132-134.
13. Jongerius JM, Wester M, Cuypers HTM, *et al*. New hepatitis B virus mutant form in a blood donor that is undetectable in several hepatitis B surface antigen screening assays. *Transfusion* 1998;38:56-9.
14. Bock CT, Tillmann HL, Torresi J, *et al*. Selection of hepatitis B virus polymerase mutants with enhanced replication by lamivudine treatment after liver transplantation. *Gastroenterology* 2002;122:264-73.
15. US Department of Labor, Occupational Safety and Health Administration, 29 CFR Part 1910.1030, Bloodborne pathogens.
16. US Department of Health and Human Services. *Biosafety in Microbiological and Biomedical Laboratories*. 5th ed. Washington, DC: US Government Printing Office; December 2009.
17. World Health Organization. *Laboratory Biosafety Manual 3*rd *ed.* Geneva: World Health Organization; 2004.
18. Clinical and Laboratory Standards Institute. *Protection of Laboratory Workers from Occupationally Acquired Infections: Approved Guideline—Third Edition*. CLSI Document M29-A3. Wayne, PA: Clinical and Laboratory Standards Institute; 2005.
19. U.S. Department of Health and Human Services, Food and Drug Administration, Center for Biologics Evaluation and Research. Guidance for Industry: Recommendations for Obtaining a Labeling Claim for Communicable Disease Donor Screening Tests Using Cadaveric Blood Specimens from Donors of Human Cells, Tissues, and Cellular and Tissue-Based Products (HCT/Ps), November 2004. http://www.fda.gov/BiologicsBloodVaccines/GuidanceComplianceRegulatoryInformation/Guidances/Tissue/ucm073972.htm Accessed September 01, 2013.
20. Centers for Disease Control and Prevention. *Guidelines for Viral Hepatitis Surveillance and Case Management*. Atlanta, GA: 2005.
21. Koziel MJ, Siddiqui A. Hepatitis B virus and hepatitis delta virus. In: Mandell GL, Bennett JE, Dolin R, eds. *Mandell, Douglas and Bennett's Principles and Practice of Infectious Diseases*. 6th ed. New York, NY: Elsevier/Churchill Livingstone; 2005:1864-90.
22. Primus FJ, Kelley EA, Hansen HJ, *et al*. "Sandwich"-type immunoassay of carcinoembryonic antigen in patients receiving murine monoclonal antibodies for diagnosis and therapy. *Clin Chem* 1988;34(2):261-4.
23. Schroff RW, Foon KA, Beatty SM, *et al*. Human anti-murine immunoglobulin responses in patients receiving monoclonal antibody therapy. *Cancer Res* 1985;45:879-85.
24. Boscato LM, Stuart MC. Heterophilic antibodies: a problem for all immunoassays. *Clin Chem* 1988;34(1):27-33.


The following U.S. Patents are relevant to the ARCHITECT System or its components. There are other such patents and patent applications in the United States and worldwide.

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| 5 468 646 | 5 543 524 | 5 545 739 |
| 5 565 570 | 5 669 819 | 5 783 699 |




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