VITROS Products Immunodiagnostic

Intended for Use in the United States

INSTRUCTIONS FOR USE

HBeAg

VITROS Immunodiagnostic Products HBeAg Reagent Pack REF 680 1819

VITROS Immunodiagnostic Products HBeAg Calibrator REF 680 1820

CAUTION: Federal law restricts this device to sale by or on the order of a physician.

Intended Use

VITROS Immunodiagnostic Products HBeAg Reagent Pack

For the *in vitro* qualitative detection of hepatitis B e antigen (HBeAg) in human adult and pediatric (2 to 21 years old) serum from individuals who have symptoms of hepatitis or who may be at risk for hepatitis B virus (HBV) infection using the VITROS ECi/ECiQ Immunodiagnostic Systems.

Test results, in conjunction with other serological and clinical information, may be used for the laboratory diagnosis of individuals with acute or chronic hepatitis B, or recovery from hepatitis B infection.

VITROS Immunodiagnostic Products HBeAg Calibrator

For use in the calibration of the VITROS ECi/ECiQ Immunodiagnostic Systems for the *in vitro* qualitative detection of hepatitis B e antigen (HBeAg) in human adult and pediatric (2 to 21 years old) serum from individuals who have symptoms of hepatitis or who may be at risk for hepatitis B virus (HBV) infection.

WARNING: *The VITROS HBeAg test should not be used to test cord blood samples.*

Test performance characteristics have not been established in patients under the age of 2, or in populations of immunocompromised or immunosuppressed patients.

This test has not been FDA licensed for the screening of blood, plasma and tissue donors.

Summary and Explanation of the Test

HBeAg is a small polypeptide which is found in a free form in the serum of HBV infected persons.[1] It appears shortly after the rise of HBsAg and is detectable in samples taken early in the course of acute HBV infection and in some chronic carriers. During the early phase of infection, anti-HBe is not detected. The presence of HBeAg in serum is a strong indication of high infectivity for both maternofetal and horizontal transmission of HBV.[2] Seroconversion to anti-HBe usually occurs before the appearance of antibodies to HBsAg, and infectivity during this phase is not clear. As the anti-HBe levels rise, so the levels of circulating HBeAg fall and eventually become undetectable.

Treatment of HBeAg positive patients with α-interferon can often induce seroconversion to anti-HBe.[3] Monitoring HBeAg/anti-HBe status allows the detection of seroconversion which is important in the management of HBV infection.[2]

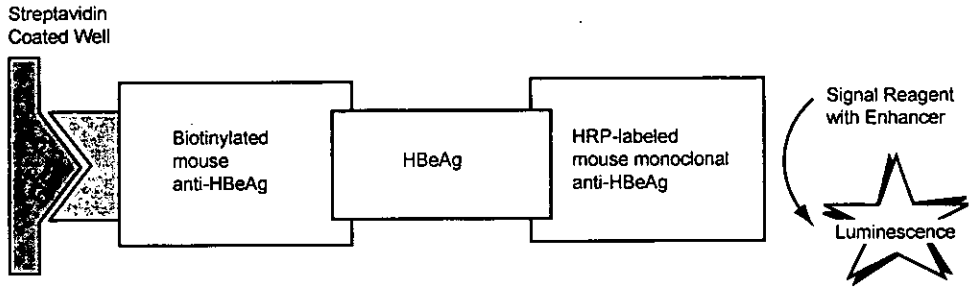
Principles of the Procedure

The VITROS HBeAg test is performed using the VITROS Immunodiagnostic Products HBeAg Reagent Pack and the VITROS Immunodiagnostic Products HBeAg Calibrator on the VITROS ECi/ECiQ Immunodiagnostic Systems using Intellicheck® Technology. An immunometric technique is used. This involves the simultaneous reaction of HBeAg in the sample with biotinylated mouse monoclonal HBeAg antibody and horseradish peroxidase (HRP)-labeled mouse monoclonal HBeAg antibody in the conjugate. The immune complex is captured by streptavidin on the wells, unbound materials are removed by washing.

The bound HRP conjugate is measured by a luminescent reaction.[4] A reagent containing luminogenic substrates (a luminol derivative and a peracid salt) and an electron transfer agent,is added to the wells. The HRP in the bound conjugate catalyzes the oxidation of the luminol derivative, producing light. The electron transfer agent (a substituted acetanilide) increases the level of light produced and prolongs its emission. The light signals are read by the system. The amount of HRP conjugate bound is indicative of the level of HBeAg present in the sample.

Test Type	System	Incubation Time	Time to first result	Test Temperature	Reaction Sample Volume
Immunometric immunoassay	ECi/ECiQ	29 minutes	37 minutes	37 °C	80 µL

Reaction Scheme



Warnings and Precautions

For *in vitro* diagnostic use only.

WARNING: Potentially Infectious Material

Treat as if capable of transmitting infection.

Use caution when handling material of human origin. Consider all samples potentially infectious. No test method can offer complete assurance that hepatitis B virus, hepatitis C virus (HCV), human immunodeficiency virus (HIV 1+2) or other infectious agents are absent. Handle, use, store and dispose of solid and liquid waste from samples and test components in accordance with procedures defined by appropriate national biohazard safety guideline or regulation (e.g. CLSI document M29).[5,6]

The VITROS HBeAg Calibrator contains:

Human HBeAg and Anti-HBe negative plasma obtained from donors who were tested individually and who were found to be negative for hepatitis B surface antigen, and for antibodies to hepatitis C virus (HCV) and human immunodeficiency virus (HIV 1+2), using FDA approved methods (enzyme immunoassays, EIA).

Human HBeAg positive plasma obtained from donors who were tested individually and who were found to be negative for antibodies to HCV and HIV 1+2, using FDA approved methods (EIA). The HBeAg positive plasma has been treated in order to reduce the titer of potentially infectious virus. However, as no testing method can rule out the risk of potential infection, handle as if capable of transmitting infection.

WARNING: Contains Kathon (CAS 55965-84-9) and 2-Choroacetamide (CAS 79-07-2)

The VITROS HBeAg Reagent Pack and VITROS HBeAg Calibrator contain Kathon. The VITROS HBeAg Reagent Pack contains 2-Choroacetamide. R43: May cause sensitization by skin contact. S24: Avoid contact with skin. S37: Wear suitable gloves.

Reagents

Reagent Pack Contents

1 reagent pack containing:

- 100 coated wells (streptavidin source, bacterial; binds ≥3 ng biotin/well)
- 6.6 mL assay reagent (buffer with mouse serum, bovine serum albumin and antimicrobial agent)
- 6.6 mL conjugate reagent (HRP-mouse monoclonal anti-HBe 0.3 µg/mL and biotin-mouse monoclonal anti-HBe 5.0 µg/mL) in buffer with sheep and mouse serum, bovine serum albumin, bovine gamma globulin and antimicrobial agent

Reagent Pack Handling

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

Reagent Pack Storage and Preparation

Reagent	Storage Condition		Stability
Unopened	Refrigerated	2–8 °C (36–46 °F)	expiration date
Opened	On system	System turned on	≤12 weeks
Opened	Refrigerated	2–8 °C (36–46 °F)	≤12 weeks

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

Calibrator Contents

- 3 vials of VITROS HBeAg Calibrator (freeze-dried; HBeAg positive human plasma in HBeAg and anti-HBe defibrinated and delipidized negative human plasma; 0.7±0.3 Units*/mL with antimicrobial agent), reconstitution volume 1.0 mL
- Lot calibration card
- Protocol card
- 8 calibrator bar code labels

* Paul-Ehrlich-Institute Reference Serum

Calibrator Handling

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

Calibrator Storage and Preparation

Calibrator	Storage Condition		Stability
Unopened	Refrigerated	2–8 °C (36–46 °F)	expiration date
Opened-reconstituted	Refrigerated	2–8 °C (36–46 °F)	≤5 weeks
Opened-reconstituted	Frozen	-20 °C (-4 °F)	≤13 weeks

- The VITROS HBeAg Calibrator is supplied freeze-dried.
- The VITROS HBeAg Calibrator is suitable for use until the expiration date on the carton when stored and handled as specified. Do not use beyond the expiration date.
- Reconstitute with 1.0 mL distilled water.
- Opened, reconstituted calibrators may be stored frozen. Do not freeze-thaw more than once.
- The VITROS HBeAg test uses 80 µL of calibrator for each determination. Transfer an aliquot of each calibrator into a sample container (taking account of the minimum fill volume of the container), which may be bar coded with the labels provided. For details on minimum fill volume of sample cups or containers, refer to the operator's guide for your system.
- The VITROS HBeAg Calibrator is automatically processed in duplicate.

Specimen Collection, Preparation and Storage

Patient Preparation

No special patient preparation is necessary.

Specimens Recommended

Serum

Specimens Not Recommended

- Do not use turbid specimens. Turbidity in specimens may affect test results.
- Do not use plasma samples.
- Do not use hemolysed samples as hemolysis may affect test results.
- Do not use cord blood samples.

Special Precautions

Important: *Certain collection devices have been reported to affect other analytes and tests.[7] Owing to the variety of specimen collection devices available, Ortho-Clinical Diagnostics is unable to provide a definitive statement on the performance of its products with these devices. Confirm that your collection devices are compatible with this test.*

Specimen Collection and Preparation

- Collect specimens using standard procedures.[8,9]
- Samples should be thoroughly separated from all cellular material on the day of collection. Failure to do so may lead to an erroneous result.
- Thoroughly mix samples by inversion and bring to 15–30 °C (59–86 °F) before use.
- The VITROS HBeAg test uses 80 µL of sample for each determination. This does not take account of the minimum fill volume of the chosen sample container. For details on minimum fill volume of sample cups or containers, refer to the operator's guide for your system.

Handling and Storage Conditions

- Handle samples in stoppered containers to avoid contamination and evaporation. Use a separate disposable tip if samples are manually pipetted. Avoid splashing, forming an aerosol, or cross-contaminating sample tube stoppers.
- The amount of time samples are on the system prior to analysis should be limited to avoid evaporation. This time should not exceed two hours. Refer to the operator's guide for your system.
- Return to 2–8 °C (36–46 °F) as soon as possible after use, or load sufficient volume for a single determination.
- Samples are not to be repeatedly frozen and thawed because this can cause analyte deterioration. Samples are to be thawed only once.
- The Clinical and Laboratory Standards Institute (CLSI) provides the following recommendations for storing serum specimens:[10]
  - Store samples at room temperature for no longer than 8 hours.
  - If the test will not be completed within 8 hours, refrigerate the serum at 2–8 °C (36–46 °F).
  - If the test will not be completed within 48 hours, or for shipment, freeze the serum at or below -20 °C (-4 °F).

Specimen Stability

Serum samples may be stored up to 4 weeks at -20°C with no loss of HBeAg reactivity in the VITROS HBeAg test.

Testing Procedure

Materials Provided

- VITROS Immunodiagnostic Products HBeAg Reagent Pack
- VITROS Immunodiagnostic Products HBeAg Calibrator

Materials Required but not Provided

- VITROS Immunodiagnostic Products Signal Reagent
- VITROS Immunodiagnostic Products Universal Wash Reagent
- Quality control materials such as VITROS Immunodiagnostic Products HBeAg Controls
- VITROS Immunodiagnostic Products Reagent Pack Storage Box (optional) with desiccant
- Calibrated pipette, distilled water and sample containers for reconstitution of VITROS HBeAg Calibrator

Operating Instructions

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

For detailed information on running the test on the VITROS ECi/ECiQ Immunodiagnostic Systems, refer to the operator's guide for your system.

NOTE: Do not use visibly damaged product.

Default Test Name

The default test name which will appear on patient reports is HBeAg. The default short name that will appear on the test selection menus and laboratory reports is HBeAg. These defaults may be reconfigured, if required. For detailed information refer to the operator's guide for your system.

Calibration

Calibration Procedure

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

  Cutoff value = (a x Signal of Cal 1)
- Ensure that the Master Calibration for each new reagent lot is available on your system.
- Process the calibrator in the same manner as samples. Load sufficient volume for the automatic duplicate determination. Calibration need not be programmed if bar code labels are used; Calibration will be initiated automatically.
- When the calibrator is processed the validity of the calibration is assessed against quality parameters that compare the actual signal of the calibrator with the expected signal. If the calibration is acceptable the cutoff value is calculated and stored for use with any reagent pack of that lot.
- The quality of calibration cannot be completely described by a single parameter. The calibration report should be used in conjunction with acceptable control values to determine the validity of the calibration.
- Recalibration is required after a pre-determined calibration interval, or when a different reagent lot is loaded.
- Calibration results are assessed against two quality parameters. Failure to meet either of the defined quality parameter ranges will be coded in the calibration report. For actions to be taken following a failed calibration, refer to the operator's guide for your system.

Refer to the operator's guide for your system for detailed instructions on the calibration process.

When to Calibrate

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

For additional information on when to calibrate, refer to the operator's guide for your system.

Traceability of Calibration

The calibration of the VITROS HBeAg test is traceable to an in-house reference calibrator which has been value-assigned to optimize the clinical sensitivity and specificity performance.

Calibration Model

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

Quality Control

Quality Control Material Selection

VITROS HBeAg Controls are recommended for use with the VITROS ECi/ECiQ Immunodiagnostic Systems. There are 2 VITROS HBeAg Controls (C1 – negative and C2 - HBeAg positive). The performance of other commercial control fluids should be evaluated for compatibility with this test before they are used for quality control.

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

Choose control material that has a composition similar to or identical with the patient sample matrix being analyzed.[11]

Appropriate quality control value ranges must be established for all quality control materials used with the VITROS HBeAg test.

Quality Control Procedure Recommendations

- Good laboratory practice requires that controls be processed to verify the performance of the test.
- Choose control levels that check the clinically relevant concentrations. The recommendation is to run a negative control and a positive control close to the HBeAg decision point [signal/cutoff (S/C) 1.00].
- To verify system performance, analyze control materials:
  - After calibration
  - According to local regulations or at least once each day that the test is being performed
  - After specified service procedures or maintenance to critical parts or subsystems that might influence performance of the test

  If quality control procedures within your laboratory require more frequent use of controls, follow those procedures.
- Analyze quality control materials in the same manner as patient specimens.
- If control results fall outside your acceptable range, investigate the cause before deciding whether to report patient results. Investigate and determine the cause for the unacceptable control results. When the condition is corrected, retest the controls and confirm that results are within acceptable limits. Repeat all patient specimens before reporting results for this run.
- Refer to the published guidelines for general quality control recommendations.[11]
- Additional controls may be tested according to guidelines or requirements of local, state, and/or federal regulations or accrediting organizations.

  For more detailed information, refer to the operator's guide for your system.

Quality Control Material Preparation and Storage

Refer to the manufacturer's product literature for preparation, storage, and stability information.

Results

Results are automatically calculated by the VITROS ECi/ECiQ Immunodiagnostic Systems.

Result Calculation

Results are calculated as a normalized signal, relative to the cutoff value (signal/cutoff, S/C). During the calibration process, a lot-specific parameter, is used to determine a valid stored cutoff value for the VITROS ECi/ECiQ Immunodiagnostic Systems.

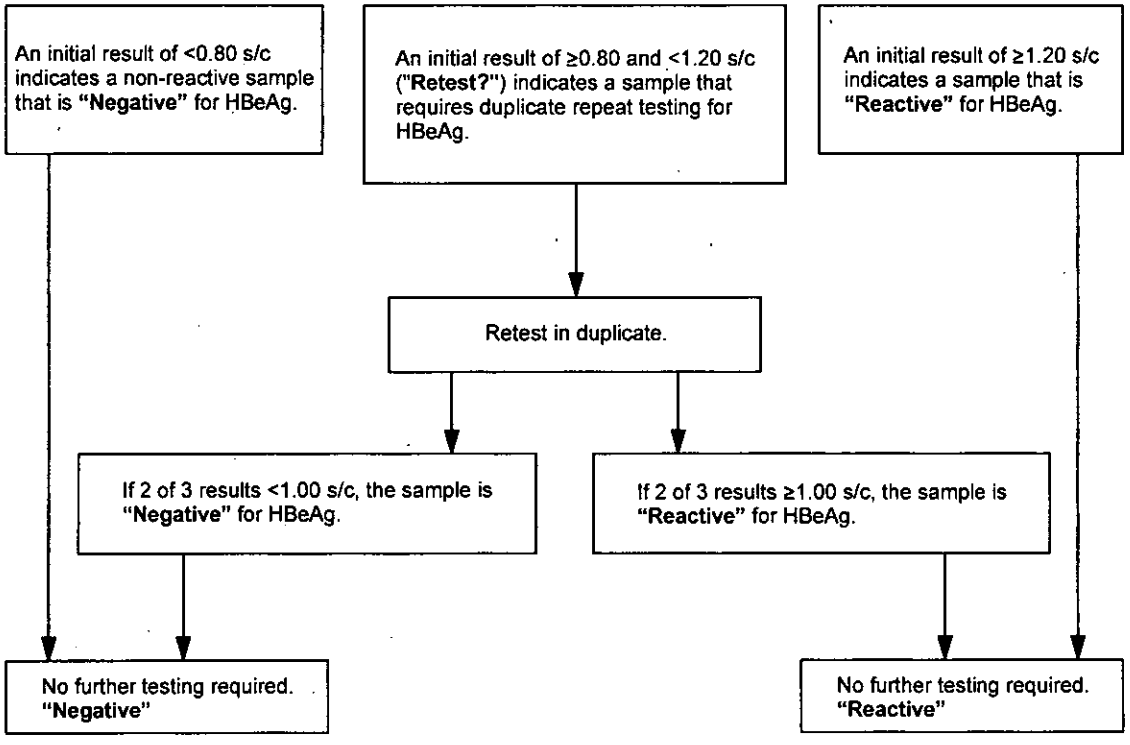
$$\text{Result} = \frac{\text{Signal for test sample}}{\text{Signal at the Cutoff (cutoff value)}}$$

Patient sample results will be displayed with a "Negative", "Retest?" or "Reactive" label. An initial result labeled with "Retest?" indicates a sample that requires duplicate repeat testing for HBeAg.

VITROS HBeAg Test Result (S/C)	<0.80	≥0.80 and <1.20	≥1.20
Result Text	Negative	Retest?	Reactive

Final results should be manually interpreted using the following algorithm.

Testing Algorithm



Interpretation of Results

The following table summarizes the interpretation of results obtained with the VITROS HBeAg test upon completion of all testing steps required in the testing algorithm.

VITROS HBeAg Test Result (S/C)	Conclusion from Testing Algorithm	Interpretation
<0.80	Negative	Sample is non-reactive and presumed negative for HBeAg. This result should not be used alone, but in conjunction with other hepatitis B serological markers to determine disease state.
≥0.80 and <1.20	Retest in duplicate	Sample is non-reactive and presumed negative for HBeAg if 2 of 3 results are <1.00. Sample is reactive and presumed positive for HBeAg if 2 of 3 results are ≥1.00.
≥1.20	Reactive	Sample is reactive and presumed positive for HBeAg. This result should not be used alone, but in conjunction with other hepatitis B serological markers to determine disease state.

Limitations of the Procedure

Known Interferences

The VITROS HBeAg test was evaluated for interference consistent with CLSI document EP7.[12] Commonly encountered substances were tested on 2 lots of reagents. Of the compounds tested, hemoglobin may interfere with the VITROS HBeAg test. Hemoglobin, when tested, caused the bias shown at the concentration indicated. Refer to "Substances that do not Interfere" for a list of other compounds tested that did not show interference.

Interferent	Interferent Concentration		Units = s/c	
			Mean Result*	Bias**
Hemoglobin	0.038 mmol/L	62 mg/dL	0.42***	+50.0%

* Mean result of replicate determinations using 2 different lots of reagent.

** Estimate of the average difference observed.

*** Negative sample showed a positive bias.

NOTE: These results are representative. The degree of interference at concentrations other than those listed might not be predictable from these results. Other interfering substances may be encountered in the patient population.

Other Limitations

- The effect of elevated serum protein levels on the VITROS HBeAg test was not evaluated. Each clinical laboratory should verify the performance of this assay with samples with high protein content.[12]
- Materials such as commercially available controls or proficiency panel members containing azide at concentrations greater than or equal to 0.25% have been shown to interfere with this test.
- The results from this or any other diagnostic kit should be used and interpreted only in the context of the overall clinical picture.
- A negative test result does not guarantee that HBeAg is not present. HBV mutants lacking the ability to produce HBeAg have been reported.[13] These may occur as 'escape' mutants in the presence of anti-HBe and such patients may be infectious.
- Heterophilic antibodies in serum or plasma samples may cause interference in immunoassays.[14] These antibodies may be present in blood samples from individuals regularly exposed to animals or who have been treated with animal serum products. Results which are inconsistent with clinical observations indicate the need for additional testing.
- Biotin levels in serum remain elevated for up to 24 hours after oral or intravenous biotin administration.[15] Cautions should be taken when testing samples collected from patients receiving biotin therapy.

Expected Results

Approximately 65.6% (1296/1976) of the prospective subjects in Population 1 were at risk for viral hepatitis due to lifestyle, behavior, occupation or known exposure event, but reported no recent or current signs or symptoms of hepatitis. Of these 1296 asymptomatic individuals, 45.8% were enrolled in Miami, FL, 19.1% were enrolled in Dallas, TX, 7.1% were enrolled in Newark, NJ, and 28.0% were enrolled in Chicago, IL. The group was Caucasian (20.0%), African American (49.5%), Hispanic (23.6%), and Asian (3.2%) with the remaining 3.7% represented by other ethnic groups. The group was 52.9% male and 47.1% female and ranged in age from 5 to 89 years. The VITROS HBeAg test was reactive in 0.8% (11/1296) of the individuals in this group. The percent VITROS HBeAg reactive results observed in the asymptomatic population at each site was 1.0% at Miami, FL, 0.0% at Dallas, TX, 1.1% at Newark, NJ and 1.1% at Chicago, IL. The expected results for the VITROS HBeAg test are presented in the following table.

Expected Results for the VITROS HBeAg Test in Study Subjects without Signs or Symptoms of Hepatitis in Population 1 (N=1296)

Age Range	Gender	Reactive		Negative		Total§
		N*	Percent**	N***	Percent†	
≤15	Female	0	0.0	1	100	1
	Male	0	0.0	4	100	4
16–20	Female	0	0.0	24	100	24
	Male	0	0.0	11	100	11
21–30	Female	1	1.0	95	99.0	96
	Male	0	0.0	106	100	106
31–40	Female	1	0.7	134	99.3	135
	Male	4	2.4	162	97.6	166
41–50	Female	0	0.0	157	100	157
	Male	3	1.4	212	98.6	215
51–60	Female	1	0.8	121	99.2	122
	Male	1	0.8	126	99.2	127

Expected Results for the VITROS HBeAg Test in Study Subjects without Signs or Symptoms of Hepatitis in Population 1 (N=1296)

Age Range	Gender	Reactive		Negative		Total§
		N*	Percent**	N***	Percent†	
61–70	Female	0	0.0	49	100	49
	Male	0	0.0	41	100	41
>70	Female	0	0.0	26	100	26
	Male	0	0.0	14	100	14
Unknown	Female	0	0.0	1	100	1
	Male	0	0.0	1	100	1
Total		11	0.8	1285	99.2	1296

* The total number (N) of subjects in each age range/gender category with reactive VITROS HBeAg results.

** The total number (N) of subjects in each age range/gender category that are reactive; expressed as a percentage (%) of all subjects in that category.

*** The total number (N) of subjects in each age range/gender category with negative VITROS HBeAg results.

† The total number (N) of subjects in each age range/gender category that are negative; expressed as a percentage (%) of all subjects in that category.

§ The total number (N) of subjects in each age range/gender category.

Approximately 34.4% (680/1976) of the prospective subjects in Population 1 were at risk for viral hepatitis due to lifestyle, behavior, occupation, or known exposure event, and reported recent or current signs or symptoms of hepatitis. Of these 680 symptomatic individuals, 68.4% were enrolled in Miami, FL, 5.0% were enrolled in Dallas, TX, 4.4% were enrolled in Newark, NJ, and 22.2% were enrolled in Chicago, IL. The group was Caucasian (16.8%), African American (53.8%), Hispanic (24.3%), and Asian (1.2%) with the remaining 3.9% represented by other ethnic groups. The group was 55.7% male and 44.3% female and ranged in age from 12 to 81 years. The VITROS HBeAg test was reactive in 3.5% (24/680) of the individuals in this group. The percent VITROS HBeAg reactive results observed in the symptomatic population at each site was 4.3% at Miami, FL, 0.0% at Dallas, TX, 0.0% at Newark, NJ and 2.6% at Chicago, IL. The expected results for the VITROS HBeAg test in subjects from Population 1 with signs or symptoms of hepatitis are presented in the following table.

Expected Results for the VITROS HBeAg Test in Study Subjects with Signs or Symptoms of Hepatitis in Population 1 (N= 680)

Age Range	Gender	Reactive		Negative		Total§
		N*	Percent**	N***	Percent†	
≤15	Female	0	0.0	1	100	1
16–20	Female	0	0.0	5	100	5
	Male	0	0.0	8	100	8
21–30	Female	1	2.7	36	97.3	37
	Male	1	3.0	32	97.0	33
31–40	Female	1	2.0	50	98.0	51
	Male	1	1.6	62	98.4	63
41–50	Female	3	3.2	90	96.8	93
	Male	8	6.0	126	94.0	134
51–60	Female	1	1.3	74	98.7	75
	Male	6	5.7	99	94.3	105
61–70	Female	0	0.0	27	100	27
	Male	2	6.5	29	93.5	31
>70	Female	0	0.0	12	100	12
	Male	0	0.0	5	100	5
Total		24	3.5	656	96.5	680

* The total number (N) of subjects in each age range/gender category with reactive VITROS HBeAg results.

** The total number (N) of subjects in each age range/gender category that are reactive; Expressed as a percentage (%) of all subjects in that category.

*** The total number (N) of subjects in each age range/gender category with negative VITROS HBeAg results.

† The total number (N) of subjects in each age range/gender category that are negative; Expressed as a percentage (%) of all subjects in that category.

§ The total number (N) of subjects in each age range/gender category.

All subjects enrolled in Population 2 (N=311) were from an area in India with a high prevalence of HBV infection and all presented with signs or symptoms of viral hepatitis. The mean age of the population was 38.5 years and the median age was 40 years. Approximately 87% of the study subjects were ≤50 years of age. The minimum age was 18 years and the maximum age was 90 years. The population was 27% female and 73% male. The VITROS HBeAg test was reactive in 23.8% (74/311) of the individuals in this group. The expected results for the subjects in Population 2 are shown in the following table.

Expected Results for the VITROS HBeAg Test in Study Subjects with Signs or Symptoms of Hepatitis in Population 2 (N=311)

Age Range	Gender	Reactive		Negative		Total§
		N*	Percent**	N***	Percent†	
18–20	Female	2	28.6	5	71.4	7
	Male	3	15.0	17	85.0	20
21–30	Female	9	26.5	25	73.5	34
	Male	8	16.3	41	83.7	49
31–40	Female	12	38.7	19	61.3	31
	Male	14	22.2	49	77.8	63
41–50	Female	3	37.5	5	62.5	8
	Male	11	19.0	47	81.0	58
51–60	Female	1	33.3	2	66.7	3
	Male	7	24.1	22	75.9	29
61–70	Female	0	0.0	1	100	1
	Male	4	57.1	3	42.9	7
>70	Male	0	0.0	1	100	1
Total		74	23.8	237	76.2	311

* The total number (N) of subjects in each age range/gender category with reactive VITROS HBeAg results.

** The total number (N) of subjects in each age range/gender category that are reactive; Expressed as a percentage (%) of all subjects in that category.

*** The total number (N) of subjects in each age range/gender category with negative VITROS HBeAg results.

† The total number (N) of subjects in each age range/gender category that are negative; Expressed as a percentage (%) of all subjects in that category.

§ The total number (N) of subjects in each age range/gender category.

Expected results for the VITROS HBeAg test were also determined using prospective samples from a population of pediatric subjects in Florida at high risk for viral hepatitis (N=165). The group was 47.9% male and 52.1% female, and the subjects' ages ranged from 2 through 21 years. The expected results for the VITROS HBeAg test in the 103 (62.4%) pediatric subjects without signs or symptoms of hepatitis are presented in the following table.

Expected Results for the VITROS HBeAg Test in Pediatric Subjects without Signs or Symptoms of Hepatitis (N=103)

Age Range	Gender	Reactive		Negative		
		N*	Percent**	N***	Percent†	Total§
2–4	Female	0	0.0	9	100	9
	Male	0	0.0	7	100	7
5–8	Female	0	0.0	12	100	12
	Male	0	0.0	4	100	4
9–12	Female	0	0.0	10	100	10
	Male	0	0.0	13	100	13
13–16	Female	0	0.0	13	100	13
	Male	0	0.0	8	100	8
17–21	Female	0	0.0	14	100	14
	Male	0	0.0	13	100	13
Total		0	0.0	103	100	103

* The total number (N) of subjects in each age range/gender category with reactive VITROS HBeAg results.

** The total number (N) of subjects in each age range/gender category that are reactive; Expressed as a percentage (%) of all subjects in that category.

*** The total number (N) of subjects in each age range/gender category with negative VITROS HBeAg results.

† The total number (N) of subjects in each age range/gender category that are negative; Expressed as a percentage (%) of all subjects in that category.

§ The total number (N) of subjects in each age range/gender category.

All 103 samples from subjects without signs or symptoms of hepatitis were negative with the VITROS HBeAg test.