Abbott
AXSYM
SYSTEM

en

HAVAB 2.0

IVD REF 6C70-20

48-0384/R8

B6C700

Read Highlighted Changes
Revised September, 2008

HAVAB 2.0

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

This package insert must be read carefully prior to 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.

Key to symbols used

REF	List Number	CONTROL –	Negative Control
IVD	*In Vitro* Diagnostic Medical Device	CONTROL +	Positive Control
LOT	Lot Number	INDEX CAL	Index Calibrator
	Expiration Date	REAGENT PACK	Reagent Pack
2°C 8°C	Store at 2-8°C	REACTION VESSELS	Reaction Vessels
	CAUTION: Consult accompanying documents	MATRIX CELLS	Matrix Cells
	Consult instructions for use	SAMPLE CUPS	Sample Cups
	Manufacturer		

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

ABBOTT
Diagnostics Division

NAME

AxSYM HAVAB 2.0

INTENDED USE

AxSYM HAVAB 2.0 is a microparticle enzyme immunoassay (MEIA) for the detection of total antibody to hepatitis A virus (anti-HAV) in human serum or plasma. A test for anti-HAV is indicated as an aid in the diagnosis of previous or ongoing hepatitis A viral infection, or for the detection of anti-HAV after vaccination.

SUMMARY AND EXPLANATION OF THE TEST

The presence of anti-HAV in human serum or plasma is indicative of past or present infection with hepatitis A virus (HAV) or vaccination against HAV. anti-HAV is detectable during the acute stage of illness (IgM anti-HAV) and may persist for years after recovery (IgG anti-HAV). Virus specific IgM anti-HAV is the most reliable marker for determining the acute stage of disease. The test for total anti-HAV is primarily used to determine previous exposure to HAV. Increasing titers of total anti-HAV in sequential patient samples may indicate an ongoing hepatitis A viral infection.[1] The results from anti-HAV testing have been used to assess immune status[2,3] or for epidemiological studies.[4-9]

BIOLOGICAL PRINCIPLES OF THE PROCEDURE

AxSYM HAVAB 2.0 is based on MEIA technology and utilizes the principle of competitive binding between anti-HAV in the sample and anti-HAV (human): alkaline phosphatase conjugate for the HAV antigen coated on the microparticles. The AxSYM HAVAB 2.0 reagents and sample are pipetted in the following sequence:

SAMPLING CENTER

- Sample and hepatitis A virus (human) coated microparticles are combined in one reaction vessel (RV) well.
- When anti-HAV is present in the sample, it binds to the hepatitis A virus (human) coated microparticles, forming an antigen-antibody complex in the reaction mixture.

The RV is immediately transferred into the Processing Center. Further pipetting is done in the Processing Center by the Processing Probe.

PROCESSING CENTER

- A portion of the reaction mixture is transferred to the matrix cell. The microparticles bind irreversibly to the glass fiber matrix.
- Antibody to hepatitis A virus antigen (human): alkaline phosphatase conjugate is dispensed onto the matrix cell and binds with the HAV antigenic sites on the microparticles which are not bound with anti-HAV from the sample.
- The matrix cell is washed with Matrix Cell Wash to remove materials not bound to the microparticles.
- The substrate, 4-Methylumbelliferyl phosphate (MUP), is added. The alkaline phosphatase-labeled conjugate catalyzes the removal of a phosphate group from the substrate, yielding the fluorescent product, 4-Methylumbelliferone. This fluorescent product is measured by the MEIA optical assembly.

The presence or absence of anti-HAV in the sample is determined by comparing the rate of formation of fluorescent product to a cutoff rate which is calculated from a previous AxSYM HAVAB 2.0 Index Calibration. If the rate of formation of fluorescent product in the sample is less than or equal to the cutoff rate, the sample is considered reactive for anti-HAV.

For further information regarding MEIA technology, refer to the AxSYM System Operations Manual, Section 3.

REAGENTS

REAGENT PACK, 100 TESTS

AxSYM HAVAB 2.0 Reagent Pack (6C70-20)

- 1 Bottle (3.45 mL) hepatitis A virus (human) coated microparticles in TRIS buffer with protein stabilizers. Minimum concentration: 0.05% solids. Preservative: sodium azide. (Reagent Bottle 2)
- 1 Bottle (11.71 mL) antibody to hepatitis A virus antigen (human): alkaline phosphatase conjugate in TRIS buffer with protein stabilizers. Minimum concentration: 1.5 µg/mL. Preservatives: sodium azide and antimicrobial agents. (Reagent Bottle 1)
- **INDEX CAL** 1 Bottle (2.8 mL) AxSYM HAVAB 2.0 Index Calibrator is prepared in fetal bovine serum. Preservative: sodium azide. Dye: green (Acid Yellow No. 23 and Acid Blue No. 9).

CONTROLS

AxSYM HAVAB 2.0 Controls (6C70-10)

2 Bottles (9 mL each) of AxSYM HAVAB 2.0 Controls. The Negative Control is fetal bovine serum. The Positive Control contains purified anti-HAV (human, nonreactive for HBsAg, HIV-1 RNA or HIV-1 Ag, anti-HCV, and anti-HIV-1/HIV-2) prepared in fetal bovine serum. Preservative: sodium azide.

The AxSYM HAVAB 2.0 Controls have the following ranges:

Control	Color	Anti-HAV Concentration* (mIU/mL)	Control Range S/CO	Control Range % Inhibition
CONTROL −	Natural	NA**	1.400 to 2.600	-30.00 to 30.00
CONTROL +	Blue***	20.00 to 34.00	≤ 0.900	≥ 55.00

* Concentration standardized against the World Health Organization Reference Standard.

** Not Applicable

***Dye: Acid Blue No. 9

OTHER REAGENTS

AxSYM Probe Cleaning Solution (9A35-05)

- **PROBE CLEANING SOLUTION** 2 Bottles (220 mL each) AxSYM Probe Cleaning Solution containing 2% Tetraethylammonium hydroxide (TEAH).

Solution 1 (MUP) (8A47-04)

- **SOLUTION 1 MUP** 4 Bottles (230 mL each) Solution 1 (MUP) containing 4-Methylumbelliferyl phosphate, 1.2 mM, in AMP buffer. Preservative: sodium azide.

Solution 3 (Matrix Cell Wash) (8A81-04)

- **SOLUTION 3 MATRIX CELL WASH** 4 Bottles (1000 mL each) Solution 3 (Matrix Cell Wash) containing 0.3 M sodium chloride in TRIS buffer. Preservatives: sodium azide and antimicrobial agents.

Solution 4 (Line Diluent) (8A46)

- **SOLUTION 4 LINE DILUENT** 1 Bottle (10 L) Solution 4 (Line Diluent) containing 0.1 M phosphate buffer. Preservatives: sodium azide and antimicrobial agent.

WARNINGS AND PRECAUTIONS

For *In Vitro* Diagnostic Use.

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

SAFETY PRECAUTIONS

For product not classified as dangerous per European Directive 1999/45/EC as amended - Safety data sheet available for professional user on request.

For a detailed discussion of safety precautions during system operation, refer to the AxSYM System Operations Manual, Section 8.

HANDLING PRECAUTIONS

- **Do not use Solution 1 (MUP) beyond the expiration date or a maximum of 14 days on-board the AxSYM System. When loading new Solution 1 (MUP), it is important to immediately tighten the instrument cap for MUP to minimize exposure to air. Prolonged exposure of MUP to air may compromise performance.**
- Do not use kits beyond the expiration date or a maximum of 112 cumulative hours on-board the AxSYM System.
- Do not mix reagents from different reagent packs. Do not mix reagents and index calibrators from different lots.
- Avoid microbial contamination of specimens and reagents. Use of disposable pipettes or pipette tips is recommended.
- Avoid chemical contamination of reagents and equipment.

- Ensure that sufficient sample volume is present. If sample volume is insufficient, the AxSYM System will give an error code and no result will be reported. For a description of the system error codes, refer to the AxSYM System Operations Manual, Section 10.
- Inadequate adherence to package insert instructions may result in inconsistent results.
- Use accurately calibrated equipment.
- Use caution in handling patient specimens to prevent cross contamination.

Refer to the AxSYM System Operations Manual, Sections 7 and 8, for a more detailed discussion of the safety and handling precautions during system operation.

STORAGE INSTRUCTIONS

- 

Upon receipt, the AxSYM HAVAB 2.0 Reagent Pack, Index Calibrator, and Controls must be stored at 2-8°C. The AxSYM HAVAB 2.0 Reagent Pack, Index Calibrator, and Controls may be used immediately after removal from the refrigerator.

  When stored and handled as directed, reagents are stable until expiration date.

  The AxSYM HAVAB 2.0 Reagent Pack may be on-board the AxSYM System for a maximum of 112 cumulative hours. After 112 hours, the reagent pack and associated index calibrator must be discarded. Refer to the AxSYM System Operations Manual, Sections 2 and 5, and Appendix C, for further information on tracking on-board time.

  Solution 1 (MUP) must be stored at 2-8°C. It may be on-board the AxSYM System for a maximum of 14 days. After 14 days, it must be discarded.

- 

The AxSYM Probe Cleaning Solution, Solution 3 (Matrix Cell Wash), and Solution 4 (Line Diluent) must be stored at 15-30°C.

INDICATIONS OF INSTABILITY OR DETERIORATION OF REAGENTS

When an AxSYM HAVAB 2.0 Negative or Positive Control value is out of the expected range, it may indicate deterioration of the reagents or errors in technique. Associated test results are invalid and the specimens must be retested. Assay recalibration may be necessary.

INSTRUMENT PROCEDURE

Assay File Installation

The AxSYM HAVAB 2.0 Assay File Version 1.00.1 or higher must be installed on the AxSYM System from the software disks, 9C52-01 (HAVAB 2.0), 1C50-01 (HAVAB 2.0 %INH), or higher, prior to performing the AxSYM HAVAB 2.0 assay. Refer to the AxSYM System Operations Manual, Section 2, for proper installation procedures.

AxSYM HAVAB 2.0 Protocol Selection

The AxSYM HAVAB 2.0 assay has two protocol options which differ only in the method used to calculate and report results. One protocol (HAVAB2, Assay Number 147) reports the result as a “Sample to Cutoff” ratio (S/CO). The second protocol (HAV2INH, Assay Number 188) reports the result as “Percent Inhibition” (%INH).

AxSYM HAVAB 2.0 Assay Parameters

The default values for the assay parameters used for the AxSYM HAVAB 2.0 assay are listed below. Assay parameters that can be edited contain a (>) symbol. These parameters can be displayed and edited according to the procedure in the AxSYM System Operations Manual, Section 2. Press PRINT to print the assay parameters.

	Assay Parameters	S/CO	%INH
1	Long Assay Name (English):	HAVAB2	HAV2INH
6	Abbrev Assay Name (English):	HAVAB2	HAV2INH
11	Assay Number:	147	188
12	Assay Version:	1	1
13	Calibration Version:	00	00
14	Assay File Revision:	100	100
15	Assay Enabled >	ON	ON
17	Assay Type:	MEIA	MEIA
18	Standard Cal Reps >	2	2
19	Master Cal Reps:	0	0
20	Cal Adjust Reps:	0	0
21	Cal A Concentration:	1.000	50.00
22	Cal B Concentration:	0.000	0.00
23	Cal C Concentration:	0.000	0.00
24	Cal D Concentration:	0.000	0.00
25	Cal E Concentration:	0.000	0.00
26	Cal F Concentration:	0.000	0.00
27	Master Calibrator 1 Concentration:	0.000	0.00
28	Master Calibrator 2 Concentration:	0.000	0.00
29	Cal Adjust Concentration:	0.000	0.00
43	Default Dilution Protocol >	UNDILUTED	UNDILUTED
44	Default Calibration Method >	Index Cal	Index Cal
45	Selected Result Concentration Units >	S/CO	%INH
46*	Selected Result Decimal Places >	3	2
62	Blank I-Max background intensity:	0.0000	0.0000
63	Min Tracer-Min net intensity:	0.0000	0.0000
64	Max Intercept-Max MUP intercept:	20000.0000	20000.0000
65	Min Intercept-Min MUP intercept:	1833.0000	1833.0000
66	Upper limit for NRMSE for low rates:	9999.9900	9999.9900
67	Upper limit for NRMSE for high rates:	0.3000	0.3000
68	Max Rate-Max rate used to check Min MUP Intercept:	1833.0000	1833.0000
69	Min Rate-Rate cutoff for NRMSE and Corr. Coef:	60.0000	60.0000
70	Min correlation coefficient for low rates:	0.7000	0.7000
71	Min correlation coefficient for high rates:	0.9700	0.9700
72	MUP T Delay-Time delay following MUP:	2.6000	2.6000
73	Low Limit - Normal/Therapeutic Range lower limit >	0.000	0.00
74	High Limit - Normal/Therapeutic Range upper limit >	0.000	0.00
75	Low Extreme Value >	0.000	0.00
76	High Extreme Value >	0.000	0.00
77	Lo Norm - % Uptake Normal Range Low >	0.0000	0.0000
78	Hi Norm - % Uptake Normal Range High >	0.0000	0.0000
80	Interpretation Option to use >	1	1
84	Hold results with POS interpretation >	ON	ON
85	Hold results with NEG interpretation >	OFF	OFF
86	Hold results with GRY interpretation >	OFF	OFF
91	Low Range Undiluted:	0.000	-50.01
92	High Range Undiluted:	3.001	100.00
96	Low Range Dil1:	0.000	0.00
97	High Range Dil1:	0.000	0.00
101	Low Range Dil2:	0.000	0.00
102	High Range Dil2:	0.000	0.00
106	Low Range Dil3:	0.000	0.00
107	High Range Dil3:	0.000	0.00
112	Max End-Point Deviation:	0.0000	0.0000
113	Max Baseline Intensity:	0.0000	0.0000
114	Min Baseline Intensity:	0.0000	0.0000
115	Max Percent Transmission:	0.0000	0.0000

* **NOTE:** Parameter #46 must not be edited below 3 decimal places for S/CO and 2 decimal places for %INH.

Refer to the AxSYM System Operations Manual for a detailed description of Instrument Procedures.

SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS

- Human serum (including serum collected in separator tubes) or plasma (collected in sodium heparin, lithium heparin, sodium citrate, ACD-A, ACD-B, CPDA-1, or potassium EDTA) may be used in the AxSYM HAVAB 2.0 assay. Follow the manufacturer's processing instructions for serum and plasma collection tubes.
- The AxSYM System does not provide the capability to verify sample type. It is the responsibility of the operator to verify that the correct sample type(s) is (are) used in the AxSYM HAVAB 2.0 assay.
- This assay was designed and validated for use with human serum or plasma from individual specimens. Pooled specimens must not be used.
- Specimens containing clots, red blood cells, or particulate matter may give inconsistent results and must be clarified by centrifugation prior to testing. Gravity separation is not sufficient.
- **All patient specimens to be tested in Primary Tubes must be centrifuged to remove red blood cells or particulate matter. Follow the manufacturer's instructions for centrifugation.**
- **Each specimen that requires repeat testing or that has been frozen and thawed must be transferred to a centrifuge tube and centrifuged at a Relative Centrifugal Force (RCF) of 10,000 x g for 10 minutes. Transfer clarified specimen to a sample cup or secondary tube for testing. NOTE: AxSYM System Software version 3.00 and higher offers an Auto Retest/Auto Dilution feature. Due to the requirements discussed above, this feature must not be used with this assay.**
- Centrifuged specimens with a lipid layer on top of the liquid must be transferred to a sample cup or secondary tube. Care must be taken to transfer only the clarified specimen and not the lipemic material.
- Specimens from heparinized patients may be incompletely coagulated and inconsistent results could occur due to the presence of fibrin. To prevent partially coagulated specimens, draw the specimen prior to heparin therapy or into a plasma collection tube.
- Specimens may be stored on or off the clot or red blood cells for up to 14 days at 2-8°C prior to testing. If stored for more than 3 days, specimens must be re-centrifuged prior to testing.
- Specimens which are not tested within the specified time period must be stored frozen (-20°C or colder). Specimens must be removed from the clot or red blood cells prior to freezing.
- No qualitative differences were observed between experimental controls and 22 nonreactive and 21 reactive specimens subjected to 6 freeze/thaw cycles; however, multiple freeze/thaw cycles should be avoided. Specimens must be mixed thoroughly after thawing and centrifuged prior to use, as described in this section, to remove particulate matter and to ensure consistency in the results.
- When shipped, specimens must be packaged and labeled in compliance with applicable state, federal, and international regulations covering the transport of clinical specimens and infectious substances. Specimens may be shipped at -20°C or colder, 2-8°C, or 15-30°C. It is recommended to ship specimens off the clot or red blood cells and frozen or refrigerated.
- All samples (patient specimens, controls, and calibrators) must be tested within 3 hours of being placed on-board the AxSYM System. Refer to the AxSYM System Operations Manual, Section 5, for more detailed discussion of on-board sample storage constraints.
- For optimal results, specimens must be free of fibrin, red blood cells, or other particulate matter.
- Specimens with microbial contamination should be avoided.
- Do not use heat-inactivated specimens.
- Performance has not been established using cadaver specimens or body fluids such as urine, saliva, semen, or amniotic fluid.
- No qualitative performance differences were observed between experimental controls and 22 nonreactive or 22 reactive specimens tested with elevated levels of total bilirubin (≤ 20 mg/dL), hemoglobin (≤ 500 mg/dL), triglycerides (≤ 3,000 mg/dL), total protein (≤ 12 g/dL), or red blood cells (≤ 0.4% v/v).

SAMPLE VOLUME

The sample volume required to perform a single AxSYM HAVAB 2.0 test on the AxSYM System varies according to the type of sample container used. For sample cups, the minimum (STAT and ROUTINE) sample volume required is 172 µL. For every additional AxSYM HAVAB 2.0 test performed from the same sample container, an additional 122 µL sample is required.

The sample cup minimum volume for both STAT and ROUTINE tests is calculated by the AxSYM System. They are displayed on the Order Screen at the time the test(s) is (are) ordered and printed in the Orderlist Report. When using Host Order Query, the Order Screen information and Orderlist Report are not available. Refer to the AxSYM System Operations Manual, Section 5, for a description of the Host Query Option.

For sample volume requirements in Primary or Aliquot Tubes, and control volume requirements for multiple AxSYM HAVAB 2.0 reagent lots, refer to the AxSYM System Operations Manual, Section 5.

To obtain the recommended volume requirements for AxSYM HAVAB 2.0 Index Calibrator and Controls, hold the bottles **vertically** and dispense 8 drops of Index Calibrator and 5 drops of Negative or Positive Control into each respective sample cup.

AxSYM HAVAB 2.0 PROCEDURE

Materials Required

- 6C70-20 AxSYM HAVAB 2.0 **REAGENT PACK**
- 8A75-02 100 **REACTION VESSELS**
- 8A73-02 100 **MATRIX CELLS**
- 6C70-10 AxSYM HAVAB 2.0 Controls
- 8A47-04 **SOLUTION 1 MUP**
- 8A81-04 **SOLUTION 3 MATRIX CELL WASH**
- 8A46 **SOLUTION 4 LINE DILUENT**
- 9A35-05 AxSYM **PROBE CLEANING SOLUTION**
- 8A76-01 **SAMPLE CUPS**

Materials Required but Not Provided

- Pipettes and pipette tips

CAUTION:

- When manually dispensing samples, verify that dispensing equipment does not introduce cross contamination, and that it delivers the specified sample volume. Use a separate pipette tip for each sample.
- For optimal performance, it is important to follow the routine maintenance procedures defined in the AxSYM System Operations Manual, Section 9. If your laboratory requires more frequent maintenance, follow those procedures.

Assay Procedure

HAVAB2 and HAV2INH protocols use the AxSYM HAVAB 2.0 Reagent Pack and the following assay procedure. Each protocol requires independent calibration.

CAUTION:

- The System status must be WARMING, PAUSED, READY, or STOPPED before adding or removing sample segments, reagent packs, or reaction vessels.
- **NOTE:** The AxSYM System Auto Retest/Auto Dilution feature must not be used for this assay. Refer to the **SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS** section in this package insert.

1. Check for sufficient on-board inventory of matrix cells and bulk solutions, and sample segment availability.
2. Check for sufficient waste collection capacity.

CAUTION:

- Do not open the Interior Waste Door or the AxSYM Processing Center Cover while any test is in progress. If opened, all processing will stop. All tests will be terminated and must be repeated.

3. Order the AxSYM HAVAB 2.0 Index Calibrator, AxSYM HAVAB 2.0 Controls, and/or patient specimens as required. Assign or modify the sample segment position (S/P) for each sample, as necessary. Refer to the **QUALITY CONTROL PROCEDURES** section in this package insert for calibration and control requirements.

   Index Calibration

   Perform an AxSYM HAVAB 2.0 calibration by testing a minimum of 2 replicates of the Index Calibrator. Dispense at least 8 drops of the Index Calibrator into a sample cup. Do not simultaneously calibrate more than one AxSYM HAVAB 2.0 reagent lot.

   Controls

   Perform quality control by testing Negative and Positive Controls (one test each). Dispense at least 5* drops each of the Negative and Positive Controls into individual sample cups.

   * When more than one AxSYM HAVAB 2.0 reagent lot is on-board the AxSYM System, multiply the control volume by the number of lots.

Patient Specimens

Ensure that sufficient volume is present in the sample cups or tubes. The sample cup minimum volume is 172 µL for the first AxSYM HAVAB 2.0 test plus 122 µL for each additional AxSYM HAVAB 2.0 test. For volume requirements in Primary or Aliquot Tubes, refer to the AxSYM System Operations Manual, Section 5.

NOTE: The operator may obtain an Orderlist Report by pressing PRINT. The printout contains sample placement information and minimum STAT sample cup volume requirements for all tests ordered. When using the Host Order Query, the Orderlist Report is not available. Refer to the AxSYM System Operations Manual, Section 5, for a description of the Host Query Option.

4. Place sample segments containing the ordered samples into the Sample Carousel.
5. Place the AxSYM HAVAB 2.0 Reagent Pack into the Reagent Pack Carousel.
6. Ensure that reaction vessels (RVs) are present on the RV Carousel. Additional RVs may be added as needed.
7. Press RUN. All entries on the Order screen are automatically transferred to the Order Status screen for sample processing.
8. Review the results to determine whether retesting is required.
9. When testing is completed, remove the samples and the AxSYM HAVAB 2.0 Reagent Pack from the Sampling Center. Store at 2-8°C.

NOTE: When using the on-board reagent tracking feature, the operator must perform a reagent pack scan after removing any pack from the system in order to maintain the validity of the reagent stability time.

Refer to Sections 5 and 6 of the AxSYM System Operations Manual for a detailed explanation of performing assay calibration and sample testing procedures.

QUALITY CONTROL PROCEDURES

CALIBRATION

A minimum of two replicates of the AxSYM HAVAB 2.0 Index Calibrator must be tested for an AxSYM HAVAB 2.0 calibration. A single sample of both the Negative and Positive Controls must be tested as a means of evaluating the assay calibration. Once an AxSYM HAVAB 2.0 calibration is accepted and stored, all subsequent samples may be tested without further calibration unless one or more of the following occur:

- A reagent pack with a new lot number is used for an assay.
- Either of the AxSYM HAVAB 2.0 Control values is out of its specified range.
- The MEIA optics verification update has been performed.

Refer to the AxSYM System Operations Manual, Section 6, for additional information.

The operator must verify that the AxSYM HAVAB 2.0 Control values are within the acceptable ranges specified in this package insert. Refer to the CONTROLS section.

The AxSYM System verifies that the results of an assay calibration meet the specifications assigned to selected validity parameters. An error message occurs when the calibration fails to meet a specification. Refer to the AxSYM System Operations Manual, Section 10, for an explanation of the corrective actions for the error code. Refer to the AxSYM System Operations Manual, Appendix E, for an explanation of the calibration validity parameters that may be used by the AxSYM System.

QUALITY CONTROL

The minimum control requirement for an AxSYM HAVAB 2.0 assay is a single sample of each of the Negative and Positive Controls tested once every 24 hours, each day of use for each reagent lot. Controls may be placed in any position in the Sample Carousel.

The AxSYM HAVAB 2.0 Control values must be within the acceptable ranges specified in this package insert. Refer to the CONTROLS section. If a control value is out of its specified range, the associated test results are invalid and the specimens must be retested. Recalibration may be indicated.

If the quality control procedures in your laboratory require more frequent use of controls, follow those procedures.

Fluorescence Background Acceptance Criteria

Quality control with regard to the MUP substrate blank is automatically determined by the instrument and checked under Assay Parameter 64, Max Intercept-Max MUP intercept, each time a test result is calculated. If the MUP intercept value is greater than the maximum allowable value, the result is invalid. The test request will be moved to the Exceptions List where it will appear with the message "1064 Invalid test result, intercept too high" and the calculated intercept value. Refer to the AxSYM System Operations Manual, Section 10, when this error message is obtained.

Refer to the AxSYM System Operations Manual, Section 2, for further information on parameter files.

RESULTS

CALCULATION

AxSYM HAVAB2 Assay 147

The AxSYM System calculates the mean rate of the Index Calibrator replicates and stores the result. The cutoff rate is then determined by dividing the Index Calibrator mean rate by 2.

$$\text{Cutoff Rate (CO)} = \frac{\text{Index Calibrator Mean Rate}}{2}$$

The AxSYM HAVAB2 assay protocol calculates a result based on the ratio of the sample rate (S) to the cutoff rate (CO) for each sample and control.

$$S/CO = \text{Sample Rate}/\text{Cutoff Rate}$$

AxSYM HAV2INH Assay 188

The cutoff value is defined as 50% of the Index Calibrator mean rate. The AxSYM HAV2INH assay protocol calculates percent inhibition of the rate of the sample relative to the Index Calibrator.

$$\%\ \text{Inhibition} = \frac{\text{Index Calibrator Mean Rate - Sample Rate}}{\text{Index Calibrator Mean Rate}} \times 100$$

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 AxSYM System Operations Manual, Sections 1 and 2.

INTERPRETATION OF RESULTS

AxSYM HAVAB2 Assay 147

- Samples with S/CO values in the range of 1.001 to 3.000 are considered nonreactive.
- Samples with S/CO values in the range of 0.000 to 1.000 are considered reactive.
- Samples with S/CO values greater than 3.000 are invalid, and the samples must be retested.

AxSYM HAV2INH Assay 188

- Samples with %INH in the range of -50.00 to 49.99 are considered nonreactive.
- Samples with %INH in the range of 50.00 to 100.00 are considered reactive.
- Samples with %INH values less than -50.00 are invalid, and the samples must be retested.

LIMITATIONS OF THE PROCEDURE

- This assay was designed and validated for use with human serum or plasma from individual patient specimens. Sufficient data are not available to interpret tests performed on pooled blood or processed plasma and products made from such pools. Pooled specimens must not be used.
- For diagnostic purposes, anti-HAV reactivity should be correlated with patient history and other hepatitis markers for diagnosis of past or present infection, or vaccination against HAV.

- Centrifuged specimens with a lipid layer on the top of the liquid must be transferred to a sample cup or secondary tube. Care must be taken to transfer only the clarified specimen and not the lipemic material. Frozen specimens must be mixed thoroughly and clarified by centrifugation prior to testing. Specimens containing red blood cells or particulate matter must be clarified by centrifugation prior to testing. Refer to the **SPECIMEN COLLECTION AND PREPARATION FOR ANALYSIS** section in this package insert.
- Specimens stored for more than 3 days must be re-centrifuged prior to testing.
- Do not use heat-inactivated specimens.
- Performance has not been established using cadaver specimens or body fluids such as urine, saliva, semen, or amniotic fluid.
- The AxSYM System "Automatic Sample Retest" feature must not be used due to the AxSYM HAVAB 2.0 assay requirement to centrifuge all specimens prior to retesting.
- Specimens from heparinized patients may be incompletely coagulated and inconsistent results could occur due to the presence of fibrin. To prevent partially coagulated specimens, draw the specimen prior to heparin treatment or into a plasma collection tube.

SPECIFIC PERFORMANCE CHARACTERISTICS

PRECISION

The precision of AxSYM HAVAB 2.0 was determined during clinical studies using three reagent master lots. A panel composed of four unique members repeated twice within the panel was tested with each reagent master lot twice daily for five days at three sites. Each daily run included two replicates of the AxSYM HAVAB 2.0 Index Calibrator and Controls. The intra-assay and inter-assay standard deviation (SD) and percent coefficient of variation (%CV) were determined with a variance component analysis[10] for a mixed model[11] (Table I).* S/CO is defined as the sample rate divided by the cutoff rate.

Table I
AxSYM HAVAB 2.0 Precision

Panel Member	Total No. Replicates	Grand Mean (S/CO)	Intra-assay SD	Intra-assay %CV	Inter-assay** SD	Inter-assay** %CV
1	180	1.920	0.0686	3.6	0.0895	4.7
2	180	1.800	0.0655	3.6	0.0806	4.5
3	180	0.666	0.0254	3.8	0.0296	4.4
4	180	0.498	0.0214	4.3	0.0231	4.6
CONTROL –	180	1.938	0.0722	3.7	0.0921	4.8
CONTROL +	180	0.447	0.0165	3.7	0.0196	4.4

** Inter-assay variability contains intra-assay variability.

Panel Member	Total No. Replicates	Grand Mean (Rate)	Intra-assay SD	Intra-assay %CV	Inter-assay SD	Inter-assay %CV
INDEX CAL	180	438.29	16.504	3.8	21.767	5.0

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

SPECIFICITY

A total of 585 serum and plasma specimens were evaluated by one site (Table II).*** Of the 299 random volunteer whole blood donor specimens, 42 specimens (14.05%) were repeatedly reactive by AxSYM HAVAB 2.0 and 41 (97.62%) of the 42 specimens were anti-HAV positive. In 198 random hospital patients, 128 specimens (64.65%) were repeatedly reactive, of which 125 (97.66%) of the 128 specimens were anti-HAV positive. In 88 specimens from individuals with disease states other than HAV and specimens containing potentially interfering substances, 35 specimens (39.77%) were repeatedly reactive and all 35 specimens were anti-HAV positive. The specificity of AxSYM HAVAB 2.0 in these populations was estimated to be 98.96% (380/384) with a 95% confidence interval of 97.35% to 99.72%.

Table II
Reactivity of AxSYM HAVAB 2.0 in Specimens from Random Volunteer Whole Blood Donors, Random Hospital Patients, Individuals with Disease States Other than HAV, and Potentially Interfering Substances

Population	Number of Specimens Tested	Initially Reactive (% of Total)	Repeatedly Reactive (% of Total)	Supplemental Assay Reactive (% of Repeatedly Reactive)
Random Volunteer Whole Blood Donors	299	43 (14.38%)	42 (14.05%)	41 (97.62%)
Random Hospital Patients	198	128 (64.65%)	128 (64.65%)	125 (97.66%)
Disease States Other than HAV and Potentially Interfering Substances[a]	88	35 (39.77%)	35 (39.77%)	35 (100.00%)
Total	585	206 (35.21%)	205 (35.04%)	201 (98.05%)

[a] Specimens from individuals with disease states other than HAV and specimens containing potentially interfering substances included the following categories: antibodies to CMV, EBV, HSV, HIV-1, HBV, HCV, Coxsackie virus, and syphilis; and anti-nuclear antibodies, rheumatoid factor, elevated IgG, elevated IgM, HBV vaccine recipients, and non-viral liver diseases.

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

DETECTABILITY

Three sites evaluated a total of 60 IgM anti-HAV reactive and 289 anti-HAV reactive specimens. AxSYM HAVAB 2.0 detected anti-HAV in 99.71% (348/349) of these specimens. One site evaluated 35 specimens from patients diagnosed with acute HAV infection. AxSYM HAVAB 2.0 detected anti-HAV in 100.00% (35/35) of these specimens. The sensitivity of AxSYM HAVAB 2.0 was 99.74% (383/384) with a 95% confidence interval of 98.56% to 99.99% by the binomial distribution.* Three sites evaluated 160 specimens from populations at increased risk for HAV infection. This population included 10 specimens from hemodialysis patients, 50 specimens from intravenous drug users, 50 specimens from men who have had sex with men, and 50 specimens from hemophilia patients. AxSYM HAVAB 2.0 detected anti-HAV in 100 (62.50%) of the 160 specimens.*

Three sites tested 30 serial bleed panels obtained from HAV vaccinees. AxSYM HAVAB 2.0 showed seroconversion within 15 days from vaccination in 21 panels and within 30 days from vaccination in 9 panels.*

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

BIBLIOGRAPHY

1. Sato A. A Clinical Study of Immunoglobulin Class Specific Antibody Response Following Hepatitis A. *Gastroenterol Jpn* 1988;23(2):129-38.
2. Lemon SM, Binn LN. Serum Neutralizing Antibody Response to Hepatitis A Virus. *J Infect Dis* 1983;148(6):1033-9.
3. CDC. Prevention of Hepatitis A Through Active or Passive Immunization. *MMWR* 1996;45:1-30.
4. Yang NY, Yu PH, Mao ZX, *et al.* Inapparent Infection of Hepatitis A Virus. *Am J Epidemiol* 1988;127(3):599-604.
5. Tassopoulos NC, Roumeliotou-Karayannis A, Sakka M, *et al.* An Epidemic of Hepatitis A in an Institution for Young Children. *Am J Epidemiol* 1987;125(2):302-7.
6. Kudesia G and Follett EAC. Hepatitis A in Scotland - Is it a Continuing Problem? *Scot Med J* 1988;33(2):231-3.
7. Tobias MI, Miller JA, Clements CJ, *et al.* The 1985 National Immunisation Survey: Hepatitis A. *NZ Med J* 1988;101(857):771-2.

8. Lemon SM. Type A Viral Hepatitis: Epidemiology, Diagnosis, and Prevention. *Clin Chem* 1997;43(8B):1494-9.
9. Hollinger FB, Ticehurst J. Hepatitis A Virus. In: Fields BN, Knipe DM, *et al*., editors. *Virology,* Second Edition. New York: Raven Press Ltd, 1990:631-67.
10. Box GEP, Hunter WG, Hunter JS. Statistics for Experimenters: An Introduction to Design, Data Analysis, and Model Building. New York: John Wiley & Sons, Inc., 1978:571-83.
11. SAS Technical Report P-229, SAS/STAT Software: Changes and Enhancements, Release 6.07. The Mixed Procedure 1992:289-366.

All trademarks are property of their respective owners.

ABBOTT
Max-Planck-Ring 2
65205 Wiesbaden
Germany
+49-6122-580

September 2008
© 2000, 2008 Abbott Laboratories