

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY**

A. 510(k) Number:

K060678

B. Purpose for Submission:

New Device

C. Measurand:

Total antibody to hepatitis A virus (total anti-HAV)

D. Type of Test:

Immunoassay, qualitative determination of hepatitis A antibodies (total) in human body fluids using antibody class capture technique followed by chemiluminescence detection.

E. Applicant:

Ortho-Clinical Diagnostics, Inc.

F. Proprietary and Established Names:

VITROS Immunodiagnostic Products Anti-HAV Total Reagent Pack,
VITROS Immunodiagnostic Products Anti-HAV Total Calibrators,
VITROS Immunodiagnostic Products Anti-HAV Total Controls

G. Regulatory Information:

1. Regulation section:

21 CFR§866.3310 Hepatitis A virus (HAV) serological assays

2. Classification:

II Special controls

3. Product code:

LOL – Hepatitis A Test (Antibody and IgM Antibody)

4. Panel:

H. Intended Use:

1. Intended use(s):

VITROS Anti-HAV Total Reagent Pack: For the *in vitro* qualitative detection of total antibody (IgG and IgM) to hepatitis A virus (total anti-HAV) in human adult and pediatric serum and plasma (EDTA, heparin or citrate) using the VITROS ECi/ECiQ Immunodiagnostic System.

The assay is indicated, in conjunction with other serological and clinical information, as an aid in the clinical laboratory diagnosis of individuals with acute or past hepatitis A virus infection, or as an aid in the identification of HAV-susceptible individuals prior to HAV vaccination. The detection of HAV-specific antibodies in human serum or plasma is laboratory evidence of acute or recent HAV infection.

Warning: *This assay is not intended for screening blood or solid or soft tissue donors. Assay performance characteristics have not been established for immunocompromised or immunosuppressed patients. The user is responsible for establishing their own assay performance characteristics in these populations.*

VITROS Anti-HAV Total Calibrator: For *in vitro* use in the calibration of the VITROS Immunodiagnostic System for the qualitative detection of antibodies to hepatitis A virus (anti-HAV) in human serum and plasma (EDTA, heparin or citrate).

VITROS Anti-HAV Total Controls: For *in vitro* use in monitoring the performance of the VITROS Immunodiagnostic System when used for the detection of antibodies to Hepatitis A virus (anti-HAV).

2. Indication(s) for use:

The assay is indicated, in conjunction with other serological and clinical information, as an aid in the clinical laboratory diagnosis of individuals with acute or past hepatitis A virus infection, or as an aid in the identification of HAV-susceptible individuals prior to HAV vaccination. The detection of HAV-specific antibodies in human serum or plasma is laboratory evidence of acute or recent HAV infection.

3. Special conditions for use statement(s):

For Prescription Use Only

4. Special instrument requirements:

VITROS Immunodiagnostic System, cleared by separate 510(k) pre-market notification (K962919/S1)

I. Device Description:

The device uses a competitive immunoassay, where the formation of a signal generating immunoassay complex is inhibited by Anti-HAV antibody (AB) from the tested sample. The system is comprised of three main elements: (i) VITROS Anti-HAV Total Reagent Pack, with the VITROS Anti-HAV Total Calibrator and the VITROS Anti-HAV Total Controls, (ii) VITROS Immunodiagnostic System – instrumentation, which provides automated use of the immunoassay kits, and (iii) common reagents used by the VITROS System.

The VITROS Anti-HAV Total Reagent Pack is composed of streptavidin coated wells, HAV antigen in a buffered matrix of BSA and mouse serum with antimicrobial agent, and mouse anti-HAV monoclonal antibody conjugated to biotin and the same mouse anti-HAV monoclonal antibody conjugated to horse radish peroxidase. The VITROS Anti-HAV Total Calibrator is a combination of anti-HAV negative and anti-HAV total positive plasma from donors confirmed negative for hepatitis B surface antigen (HBsAg), antibodies to human immunodeficiency virus (HIV 1+2) and hepatitis C virus (HCV). The VITROS Anti-HAV Total Controls are a negative control (normal human plasma obtained from HBsAg, HIV 1+2 and HCV negative donors), and a positive control (normal human plasma spiked with anti-HAV Total positive plasma, both from HBsAg, HIV 1+2 and HCV negative donors).

J. Substantial Equivalence Information:

1. Predicate device name(s):

For VITROS Anti-HAV Total Reagent Pack: IMX HAVAB 2.0 assay.

For VITROS Immunodiagnostic Products Anti-HAV Total Controls: Blackhawk BioSystems, Inc. Virotrol II

2. Predicate 510(k) number(s):

PMA P780012, BK960085

3. Comparison with predicate:

Comparison of VITROS Anti-HAV Total Reagent Pack to IMX HAVAB 2.0 assay:

Similarities		
Item	Device	Predicates
	VITROS Anti-HAV Total Reagent Pack	IMX HAVAB 2.0 assay
Intended use	For the <i>in vitro</i> qualitative detection of total antibody (IgG and IgM) to hepatitis A virus (anti-HAV) in human adult and pediatric serum and plasma (EDTA, heparin or citrate) and neonate serum using the VITROS ECi/ECiQ Immunodiagnostic System is a qualitative microparticle enzyme immunoassay for the detection of total antibody to hepatitis A virus
Specimen types	Human adult and pediatric serum and plasma (EDTA, heparin or citrate)	Human serum, plasma (heparin, citrate, EDTA)
Assay Technology	Enzyme immunoassay	Enzyme immunoassay
Antigen	Hepatitis A virus	Hepatitis A virus
Instrumentation	Automated analyzer: ECi Immunodiagnostic System	Automated analyzer: IMX System

Differences		
Item	Device	Predicates
	VITROS Anti-HAV Total Reagent Pack	IMX HAVAB 2.0 assay
Antibody	Mouse anti-HAV monoclonal antibody	Human anti-HAV antibody
Tracer	Horseradish Peroxidase	Alkaline Phosphatase
Sample volume	10 µl	150 µl

Comparison of VITROS Anti-HAV Total Controls to Blackhawk BioSystems, Inc. Virotrol II Controls:

Similarities		
Item	Device	Predicates
	VITROS Anti-HAV Total Controls	Blackhawk BioSystems, Inc. Virotrol II
Intended use	For <i>in vitro</i> use in monitoring the performance of the VITROS Immunodiagnostic System when used for the detection of antibodies to Hepatitis A virus (anti-HAV)	For use with assay procedures for the determination of antibodies to Hepatitis A virus (HAV)
Matrix of controls	Human plasma and antimicrobial agents	Human serum with added human proteins and antimicrobial agents
Differences		
Item	Device	Predicates
	VITROS Anti-HAV Total Controls	Blackhawk BioSystems, Inc. Virotrol II
Intended use	Only for the detection of antibodies to HAV	Can be used for the determination of antibodies to hepatitis B surface antigen (HBs)
Control level	Positive and negative	Positive
Expected values	Each control has a quoted mean value derived from a minimum of 10 assays and a standard deviation anticipated for single determinations of each control in a number of different laboratories using different reagent lots. Values are lot specific	There is no assigned value. The VIROTROL II reagents have been designed to produce a positive reaction when used in the proper manner with many commercial test kits.

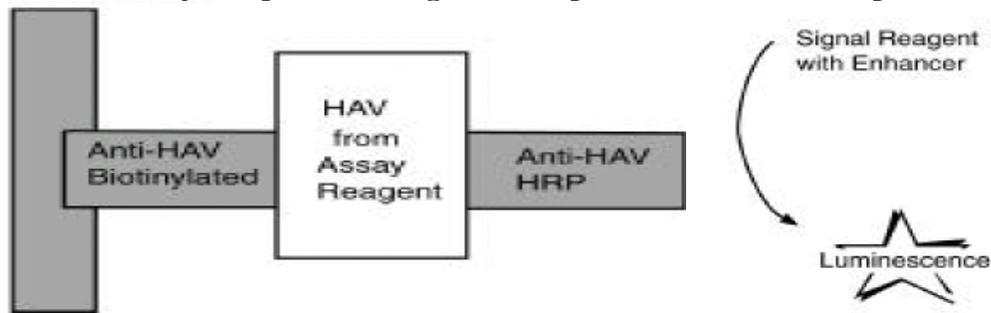
K. Standard/Guidance Document Referenced (if applicable):

Class II Special Control Guidance Document: Hepatitis A Virus Serological Assays, issued February 9, 2006 (<http://www.fda.gov/cdrh/oivd/guidance/1536.pdf>)

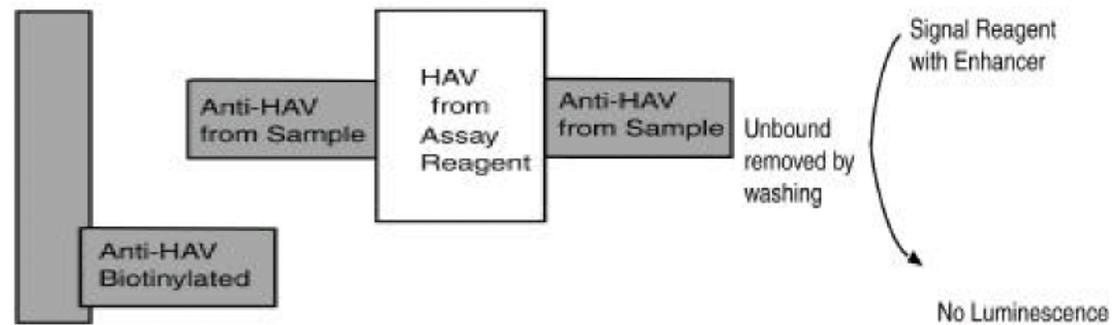
L. Test Principle:

Competitive immunoassay where the formation of an immunoassay complex that generated signal is inhibited by Anti-HAV antibody (AB) from the tested sample. The immunoassay complex: streptavidin plate–biotinylated mouse Anti-HAV AB–HAV antigen–HPR-labeled mouse anti-HAV AB, that generated luminescent signal can not be formed anymore in the presence of Anti-HAV AB from the patient sample.

Immunoassay complex with negative sample (no Anti-HAV AB present in sample):



Immunoassay complex with positive sample (Anti-HAV AB present in sample):



The competitive assay technique uses a pre-incubation of sample (potentially containing anti-HAV AB) with HAV antigen in the assay reagent followed by incubation with a biotinylated mouse monoclonal anti-HAV AB and horseradish peroxidase (HRP)-labeled mouse monoclonal anti-HAV AB. The immune complex of biotinylated mouse Anti-HAV AB – HAV antigen – HPR-labeled mouse anti-HAV AB is captured by streptavidin on the wells, unbound materials are removed by washing. The bound HRP conjugate is measured by a luminescent reaction. The light signals are read by the VITROS System. The amount of HRP conjugate bound is **inversely proportional** to the concentration of anti-HAV AB present. The assay used two controls: negative, anti-HAV negative normal human plasma and positive, normal human plasma spiked with anti-HAV AB total positive plasma.

Assay Type	Assay Time and Temperature	
Competitive	Incubation time:	45 minutes
	Time to first result:	53 minutes
	Temperature:	37° C

Interpretation of the results:

Results are calculated as a normalized signal, relative to a cut-off value (signal/cutoff, s/c). During the calibration process a lot-specific parameter, encoded on the lot calibration card, is used to determine a valid stored cut-off value for the VITROS Immunodiagnostic System. Results are automatically calculated by the VITROS Immunodiagnostic System.

$$\text{Result} = \frac{\text{Signal for test sample}}{\text{Cut-off value}}$$

Patient sample results will be displayed as “Antibody Pos”, “Borderline”, “Antibody Neg”, or “Retest?*” (* indicates the sample will require retesting following the testing algorithm). An initial result labeled with “Borderline” indicates a sample that requires duplicate repeat testing for anti-HAV. An initial result labeled “Retest?” indicates a sample which requires dilution and re-assay.

VITROS Anti-HAV Assay Results	Result Test	Clinical Interpretation if Immune Status
< 0.80	Antibody Pos	Indicates a reactive sample and the presence of anti-HAV. Indicates individual has been previously infected with or is presumed to be immune to HAV infection.
≥ 0.80 to < 1.00	Borderline*	Indicates a borderline sample. It is recommended that a new specimen be collected within 2 – 4- weeks and retested.
≥ 1.00 to < 4.00	Antibody Neg	Indicates a non-reactive sample, negative for anti-HAV. Indicates that the individual has not been infected and is presumed not to be immune to HAV infection.
≥ 4.00	Retest?	Indicates a sample, which requires dilution and retesting.

*The 2/3 rule is applied, if 2/3 tests are borderline again, analysis of follow-up sample (collected within 2-4 weeks) is recommended.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Precision was evaluated based on the Clinical and Laboratory Standards Institute (formerly NCCLS) protocol EP5-A2. The precision panel consisting of 4 samples (a negative, a negative close to the cut-off, a positive close to the cut-off and a positive) was prepared and shipped to 3 different sites. Two replicates of each of 4 panel samples were assayed at each of the 3 different sites once per day for at least 20 different days, over one calibration interval. The experiment was performed using 1 reagent lot on three different VITROS Immunodiagnostic Systems at three different sites. The data presented is a summary of the product performance by site:

Clinical Site	Mean VITROS Anti-HAV Total S/C (Ratio)	Within Day*		Between Day**		Total***		No. of Observ.	No. of Days
		SD	CV (%)	SD	CV (%)	SD	CV (%)		
Site 1	1.74	0.015	0.9	0.041	2.3	0.043	2.5	40	20
	1.08	0.018	1.6	0.026	2.4	0.031	2.9	40	20
	0.73	0.008	1.1	0.019	2.6	0.020	2.8	40	20
	0.52	0.006	1.1	0.012	2.3	0.013	2.6	40	20
Site 2	1.93	0.054	2.8	0.029	1.5	0.061	3.2	40	20
	1.14	0.015	1.3	0.033	2.9	0.036	3.2	40	20
	0.74	0.011	1.5	0.028	3.8	0.030	4.0	40	20
	0.51	0.007	1.3	0.026	5.1	0.027	5.3	40	20
Site 3	1.93	0.029	1.5	0.068	3.5	0.074	3.8	40	20
	1.18	0.030	2.5	0.038	3.2	0.048	4.1	40	20
	0.77	0.013	1.6	0.030	3.8	0.032	4.2	40	20
	0.55	0.012	2.2	0.030	5.4	0.032	5.8	40	20

* **Within Day:** variability of the assay performance from replicate to replicate

** **Between Day:** variability of the assay performance from day to day

*** **Total:** variability of the assay performance combining the effects of within day and between day

b. Linearity/assay reportable range:

The quantitative values of the assay allows a signal to cut-off ratio from >0.0 to maximum 4.0. Sample with the maximum 4.0 S/Co value are required to be diluted and retested.

c. Traceability, Stability, Expected values (controls, calibrators, or methods):

Each quoted mean value is derived from a minimum of 10 assays. The standard deviation is that which would be anticipated for singleton determinations of each control in a number of different laboratories using different reagent lots. Quoted values are lot specific.

Vitros Anti-HAV Controls - Baseline Statistics

Control	Mean	Result	SD
1 (Negative)	2.13	0.228	
2 (Positive)	0.56	0.085	

Calibration is lot specific; reagent packs and calibrators are linked by lot number. A Master Calibration is established for each new reagent lot by performing multiple assays. This is the process by which a lot-specific parameter [a] which links the cut-off value to the calibrator signal is determined.

$$\text{Cut-off value} = (a \times \text{Signal of CAL1})$$

The lot-specific parameter, the expected calibrator signal and the data which enables a System to calculate the cut-off value, are encoded on the lot calibration card. Scanning the lot calibration card loads the encoded data onto the System. When the calibrator is processed the validity of the calibration is assessed against a quality parameter which compares the actual signal of the calibrator with the expected signal. If the calibration is

acceptable the cut-off 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 control values to determine the validity of the calibration. Recalibration is required after a predetermined calibration interval, or when a different reagent lot is loaded.

d. Detection limit:

The limit of detection (LoD) for anti- HAV Total is 2.024, determined consistent with the guidelines in CLSI EP 17-A and with proportions of false positives (α) less than 5% and false negatives (β) less than 5%; based on 136 determinations each, with 1 blank and 3 low-level samples.

The limit of blank (LoB) is 2.097, determined consistent with the guidelines in CLSI EP 17-A and with proportions of false positives (α) less than 5% and false negatives (β) less than 5%; based on 136 determinations of a sample free of anti-HAV.

e. Analytical specificity:

The specificity of the VITROS Anti-HAV Total assay was evaluated by testing 283 samples from the following potentially cross-reacting sub-groups: SLE, anti-HIV, Cirrhosis, Non-viral Liver Disease, anti-HCV, anti-CMV, anti-HSV I & II, anti-EBV, anti-syphilis, anti-Rubella, anti-Toxoplasma, anti-HBs, anti-HTLV, HBsAg, Rheumatoid Factor, Pregnancy (1st, 2nd, 3rd trimester), HAMA, Rubeola, Rubella, Mumps, VZV and ANA. All initially reactive samples were tested with a reference assay for confirmation. Of the 283 samples tested, four (4) were observed to be discordant. The incidence of discordant samples is not significantly different from the claimed sensitivity and specificity.

Summary of Data from Potentially Cross-Reacting Sub-Groups					
Sample Category	No. Samples Tested	VITROS Anti-HAV Total Assay		Reference Assay	
		No. Negatives	No. Initial Reactives/Borderlines	No. Initial Reactives/Borderlines	No. Discordants
Toxo IgM	4	3	1	1	0
Toxo IgG	20	6	14	13	1
Rubella	15	12	3	3	0
RF	50	24	26	25	1
SLE	10	5	5	5	0
HIV	10	7	3	2	1
aHBs	10	2	8	8	0
HCV	10	7	3	3	0
Cirrhosis	10	7	3	3	0
Syphilis	10	9	1	1	0
HTLV	10	8	2	2	0
Non Viral Liver	12	4	8	8	0

HBsAg	10	4	6	6	0
HAMA	9	5	4	4	0
EBV	10	9	1	0	1
1 st Tri	18	10	8	8	0
2 nd Tri	16	7	9	9	0
3 rd Tri	16	7	9	9	0
HSV 1	3	3	0	N/A	N/A
HSV 2	2	2	0	0	0
CMV	8	8	0	N/A	N/A
VZV	5	5	0	0	0
Mumps	5	5	0	0	0
Rubeola	5	3	2	2	0
ANA	5	3	2	2	0
Total	283	165	118	114	4

Substances that do not interfere:

Serial dilutions were made for bilirubin, triolein, hemoglobin and biotin, and point estimates were made for sodium azide and dipyrone. The mean result of 3 determinations of a solution of each test substance was compared with that of a control pool, for both a negative and positive sample. For each substance, the highest concentration which was considered not to impact results, the mean result from the three kit lots and the classification for both positive and negative samples is shown in the table below:

Compound	Compound Concentration	
Bilirubin	0.342 mmol/L	20 mg/dL
Biotin	10 ng/mL	1 µg/dL
Dipyrone	1 mg/mL	10 mg/dL
Hemoglobin	0.078mmol/L	125 mg/dL
Sodium Azide	1 g/dL	1000 mg/dL
Triolein	33.9 mmol/L	3000 mg/dL

f. Assay cut-off:

The cut-off signal was established at light signal that gave the best discrimination between anti-HAV reactive and anti-HAV negative sample populations, to provide optimum specificity and sensitivity for the assay. The cut-off signal level was assigned a result of 1.00. Assay results < 0.80 indicate a reactive sample, positive for anti-HAV. A result of ≥0.8 and <1.0 indicates a borderline sample. A result of ≥1.0 and <4.0 indicates a non reactive sample, negative for anti-HAV. A result of ≥4.0 indicates a sample which requires dilution and re-testing

2. Comparison studies:

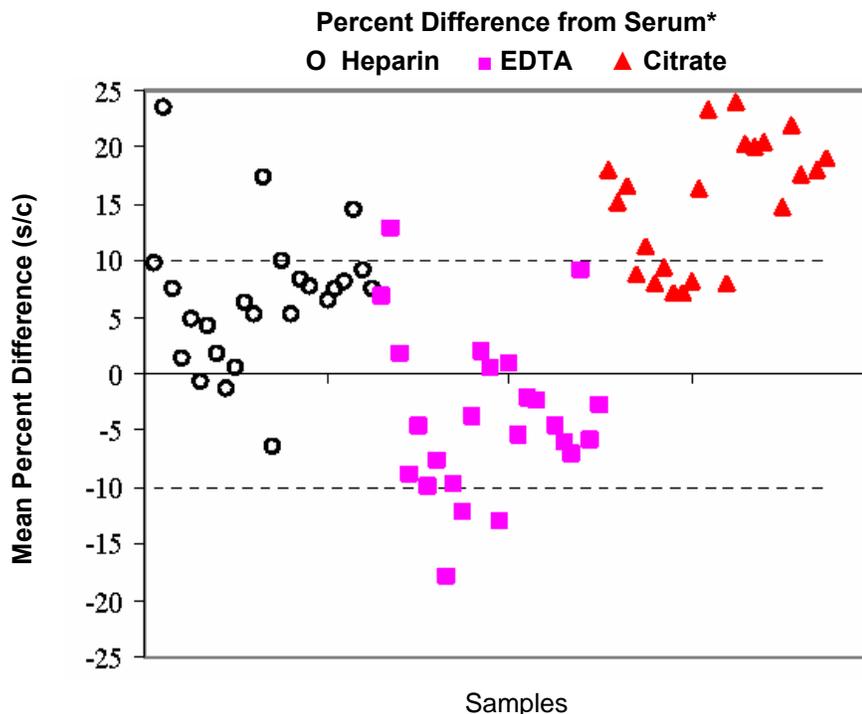
a. *Method comparison with predicate device:*

see section 3c (other clinical supportive data)

b. *Matrix comparison:*

Serum vs. plasma (heparin, EDTA, or citrate)

A total of 25 donors had blood drawn which was spiked with anti-HAV Total positive plasma to a level close to the assay cut-off. The spiked blood was then aliquoted into serum and plasma collection tubes and tested in the VITROS anti-HAV Total assay. The percent difference in the plasma from serum was calculated. Mean percent differences from serum are represented below for each plasma type tested. The differences between serum and citrate samples may be larger than 10% due to the liquid anticoagulant in the tube. There is approximately a 10% dilution of the blood by the liquid anticoagulant in the citrate tubes



***Due to the competitive nature of the VITROS anti-HAV Total assay a positive numerical bias indicates a negative functional bias.**

3. Clinical studies:

a. *Clinical Sensitivity:*

Not applicable

b. *Clinical specificity:*

Not applicable

c. Other clinical supportive data (when a. and b. are not applicable):

Methods comparison with predicate device:

The performance characteristics of the VITROS Anti- HAV Total assay were established in a multi-center prospective among individuals with signs or symptoms or biochemical manifestations (elevated liver function tests) of hepatitis and those at high risk of hepatitis infection due to lifestyle, behavior, occupation, or known exposure events. Specimens were evaluated from 872 subjects prospectively enrolled at three geographically separated collection sites within the United States (Population 1) located in Miami, FL (12.6%), Dallas, TX (37.5%) and Chicago, IL (49.9%). Specimens were also evaluated from 313 subjects with signs and symptoms of viral hepatitis prospectively enrolled in an area in India with a high prevalence of viral hepatitis (Population 2). The subjects in Population 1 were Caucasian (25.6%), African American (53.1%), Hispanic (17.0%), with the remaining 4.3% represented by other ethnic groups. The group was 57.3% male and 42.7% female, and ranged in age from 16 to 81 years. Testing of these specimens with the VITROS Anti - HAV Total assay occurred at diagnostic laboratories located in Miami, FL (12.6%), Port Jefferson, NY (49.9%) and Minneapolis, MN (37.5%). The subjects in Population 2 were Indian (100.0%). The group was 72.8% male and 27.2% female, and ranged in age from 18 to 90 years. Testing of these specimens with the VITROS Anti-HAV Total assay occurred at diagnostic laboratories located in Miami, FL (43.8%), Minneapolis, MN (43.8%) and Port Jefferson, NY (12.5%). Agreement of the VITROS Anti-HAV Total assay was assessed relative to the reference anti-HAV total assay using serum samples from Population 1, Population 2.

Percent Agreement

A comparison of the VITROS Anti-HAV Total assay and the reference anti-HAV Total assay results is presented in the following tables. Data are listed by site and population. Positive and negative percent agreement and 95% exact confidence intervals are also shown:

VITROS and Reference Anti-HAV Total Assay Results in Population 1: Prospective Samples from the U.S. (N=872)								
VITROS Anti-HAV Total Assay Result	Reference Anti-HAV Total Assay Result							
	Site 1		Site 2		Site 3		All Sites	
	Reactive	Negative	Reactive	Negative	Reactive	Negative	Reactive	Negative
Reactive	61	2	267	3	121	4	449	9
Borderline	0	0	0	0	0	0	0	0
Negative	0	47	1	164	1	201	2	412
Total	61	49	268	167	122	205	451	421
Positive Percent Agreement	100% (61/61)		99.63% (267/268)		99.18% (121/122)		99.56% (449/451)	
95% Exact Confidence Interval	94.13% - 100%		97.94% - 99.99%		95.52% - 99.98%		98.41% - 99.95%	
Negative Percent Agreement	95.92% (47/49)		98.20% (164/167)		98.05% (201/205)		97.86% (412/421)	
95% Exact Confidence Interval	86.02% - 99.50%		94.84% - 99.63%		95.08% - 99.47%		95.98% - 99.02%	

VITROS and Reference Anti-HAV Total Assay Results in Population 2: Prospective Samples from India (N=313)								
VITROS Anti-HAV Total Assay Result	Reference Anti-HAV Total Assay Result							
	Site 1		Site 2		Site 3		All Sites	
	Reactive	Negative	Reactive	Negative	Reactive	Negative	Reactive	Negative
Reactive	135	2	39	0	133	4	307	6
Borderline	0	0	0	0	0	0	0	0
Negative	0	0	0	0	0	0	0	0
Total	135	2	39	0	133	4	307	6
Positive Percent Agreement	100% (135/135)		100% (39/39)		100% (133/133)		100% (307/307)	
95% Exact Confidence Interval	97.30% - 100%		90.97% - 100%		97.26% - 100%		98.81% - 100%	
Negative Percent Agreement	0% (0/2)		N/A		0% (0/4)		0% (0/6)	
95% Exact Confidence Interval	0% - 84.19%		N/A		0% - 60.24%		0% - 45.93%	

Performance of the VITROS Anti-HAV Total Assay in Known Anti-HAV IgM Reactive Subjects:

The performance of the VITROS Anti-HAV Total assay was evaluated among serum samples from subjects known to be anti-HAV IgM positive. A total of 77 samples collected in Egypt (N=50) and India (N=27) from subjects with a medical history and laboratory results indicative of acute hepatitis A were tested concurrently with the VITROS and reference anti-HAV IgM and anti-HAV total assays. The VITROS Anti-HAV Total assay was reactive in 100% of the 77 anti-HAV IgM reactive samples. The positive percent agreement of the VITROS Anti-HAV Total assay with anti-HAV IgM reactive status and the 95% exact confidence interval are presented in the following table:

Performance of the VITROS Anti-HAV Total Assay in Known Anti-HAV IgM Reactive Subjects		
Population	Positive Percent Agreement*	95% Exact Confidence Intervals
Anti-HAV IgM Reactive Subjects	96.1% (74/77)	89.0% - 99.2%

*The reference anti-HAV total assay was negative in three subjects, the 3 reactive results with VITROS Anti-HAV Total assay was considered falsely reactive for purposes of the analysis.

Performance of the VITROS Anti-HAV Total Assay in Pediatric Subjects:

The VITROS Anti-HAV Total assay was also evaluated using residual laboratory serum samples from pediatric subjects at low risk for viral hepatitis. The samples were unlinked to the subjects' identities, and were included based on age, gender and available volume remaining after all testing ordered for that sample had been completed. Samples were selected such that the following age ranges (in years) were represented (2-4, 5-9, 10-14, and 15-19). The positive and negative percent agreement of the VITROS Anti-HAV Total assay with the reference anti-HAV total assay, and the 95% exact confidence intervals are presented in the following table. One sample, negative with the reference assay, was Borderline with the VITROS Anti-HAV Total assay and was considered falsely reactive for purposes of the analysis. The positive percent agreement of the VITROS Anti-HAV Total assay and the 95% exact confidence interval are presented in the following table:

Agreement of the VITROS and Reference Anti-HAV Total Assays in Pediatric Subjects				
Population	Positive Percent Agreement	95% Exact Confidence Intervals	Negative Percent Agreement	95% Exact Confidence Intervals
Pediatric Subjects	93.75% (15/16)	69.77% - 99.84%	97.85% (91/93)	92.45% - 99.74%

Seroconversion Study:

Three seroconversion panels each having at least 5 individual samples with a known predetermined result were measured in the VITROS Anti-HAV Total assay and in a reference assay. The results were compared with the published results for the reference assay. The VITROS Anti-HAV Total assay gave seroconversion sensitivity equivalent to a

reference assay in the three panels tested. For one of the 3 seroconversion panels the VITROS Anti-HAV Total assay was slightly more sensitive than the reference assay. The following results were obtained:

Panel ID	VITROS Anti-HAV Total		Anti-HAV Total Reference Assay		Difference in Days to Reactive Result
	Post bleed day of last non-reactive result	Post bleed day of first reactive result.	Post bleed day of last non-reactive result	Post bleed day of first reactive result	
PHT902	3	16	3	16	0
RP-013	6	9	6	9	0
RP-004	P*	1	1	7	6

P* = Positive in first bleed

Cord blood samples:

A total of 20 cord blood (as a surrogate for neonate serum) and 10 adult serum samples were tested in the VITROS Anti-HAV Total assay. None of the samples gave a reactive result in the VITROS Anti-HAV Total Assay. Forty-five (45) µl of anti-HAV positive material was added to 255 µl of cord blood and adult serum. A negative bias* was observed in the cord blood results when compared to the adult serum.

Anti-HAV Total Cord Blood Study			
Sample Type	N	Mean Response (S/C)	St Dev
Cord Blood - Neat	20	2.17	0.205
Cord Blood - Spiked	20	0.89	0.151
Adult Serum - Neat	10	2.19	0.073
Adult Serum- Spiked	10	0.77	0.022

*Due to the competitive nature of the VITROS anti-HAV Total assay a positive numerical bias indicates a negative functional bias.

4. Clinical cut-off:

Not applicable

5. Expected values/Reference range:

HAV Prevalence Population:

The expected results of the VITROS Immunodiagnostic Products Anti-HAV Total assay to detect anti-HAV IgG and IgM were determined in presumably healthy individuals from areas of both high (Western US) and low (Eastern US) HAV disease prevalence in the United States. The population was 50% male and 50% female, with ages that ranged from 18 to 89 years. The majority of the subjects were White/Caucasian (72.0%). Other ethnic groups tested were African American (12.0%), Hispanic/Latino (15.0%) and Asian (1.0%). The expected results

for presumably healthy individuals living in either high or low prevalence areas are presented below:

Expected Results for the VITROS Anti-HAV Total Assay in Subjects From Low Prevalence Areas for Hepatitis A (N=622)

Age Range	Gender	VITROS Anti-HAV Total Result						Total
		Reactive		Borderline		Negative		
		N	Percent	N	Percent	N	Percent	
Age 18 - 20	Female	0	0.0	0	0.0	11	100.0	11
	Male	0	0.0	0	0.0	6	100.0	6
Age 21 - 30	Female	5	18.5	0	0.0	22	81.5	27
	Male	6	15.4	0	0.0	33	84.6	39
Age 31 - 40	Female	5	19.2	0	0.0	21	80.8	26
	Male	5	11.1	1	2.2	39	86.7	45
Age 41 - 50	Female	25	33.3	1	1.3	49	65.3	75
	Male	12	21.8	0	0.0	43	78.2	55
Age 51 - 60	Female	21	20.6	0	0.0	81	79.4	102
	Male	29	25.4	0	0.0	85	74.6	114
Age 61 - 70	Female	12	54.5	0	0.0	10	45.5	22
	Male	19	57.6	0	0.0	14	42.4	33
Age 71 - 80	Female	22	71.0	0	0.0	9	29.0	31
	Male	17	81.0	0	0.0	4	19.0	21
Age 81 - 90	Female	6	85.7	0	0.0	1	14.3	7
	Male	7	87.5	0	0.0	1	12.5	8
Total		191	30.7	2	0.3	429	69.0	622

Expected Results for the VITROS Anti-HAV Total Assay in Subjects From High Prevalence Areas for Hepatitis A (N=378)

Age Range	Gender	VITROS Anti-HAV Total Result						Total
		Reactive		Borderline		Negative		
		N	Percent	N	Percent	N	Percent	
Age 18 - 20	Female	3	100.0	0	0.0	0	0.0	3
	Male	0	0.0	0	0.0	6	100.0	6
Age 21 - 30	Female	23	57.5	1	2.5	16	40.0	40
	Male	17	43.6	0	0.0	22	56.4	39
Age 31 - 40	Female	21	61.8	2	5.9	11	32.4	34
	Male	14	35.9	0	0.0	25	64.1	39
Age 41 - 50	Female	9	50.0	0	0.0	9	50.0	18
	Male	11	45.8	0	0.0	13	54.2	24
Age 51 - 60	Female	25	34.7	0	0.0	47	65.3	72
	Male	13	34.2	0	0.0	25	65.8	38
Age 61 - 70	Female	8	40.0	0	0.0	12	60.0	20
	Male	6	40.0	0	0.0	9	60.0	15
Age 71 - 80	Female	2	28.6	0	0.0	5	71.4	7
	Male	6	46.2	0	0.0	7	53.8	13
Age 81 - 90	Female	5	100.0	0	0.0	0	0.0	5
	Male	3	60.0	0	0.0	2	40.0	5
Total		166	43.9	3	0.8	209	55.3	378

N. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

O. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.