510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY

A. 510(k) Number: K100903

B. Purpose for Submission: Clearance of New Device

C. Measurand: Total Antibody to *Hepatitis A Virus* in serum and *heparinized* plasma

D. Type of Test: Electrochemiluminescence immunoassay (ECLIA)

E. Applicant: Roche Diagnostics

F. Proprietary and Established Names: Elecsys® Anti-HAV Assay
Elecsys® PreciControl anti-HAV Assay

G. Regulatory Information:

1. Regulation section: 21 CFR §866.3310, Hepatitis A virus Serological Assays
   21 CFR §862.1660, Quality Control Material
2. Classification: Class II
3. Product code: LOL (Hepatitis A Test – antibody and IgM Antibody)
   JJX (Quality control material, assayed and unassayed)
4. Panel: Microbiology (83)

H. Intended Use:

1. Intended use(s):

   The Roche Elecsys Anti-HAV immunoassay is used for the in vitro qualitative detection of total antibodies (IgM and IgG) to hepatitis A virus in human serum and plasma (K_2-EDTA). The assay is intended for use as an aid in the laboratory diagnosis of past or acute/recent hepatitis A infection.

   Assay results, in conjunction with other laboratory results and clinical information, may be used to provide presumptive evidence of infection with hepatitis A virus in persons with signs or symptoms of hepatitis and in persons at risk for hepatitis A infection, or used as an aid to determine the presence of antibody response to HAV in vaccine recipients.

   The electrochemiluminescence immunoassay "ECLIA" is intended for use on Elecsys and cobas e immunoassay analyzers.
   Elecsys PreciControl Anti-HAV is used for the quality control of the Elecsys Anti-HAV immunoassay on the Elecsys and cobas e immunoassay analyzers.

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 users are responsible for establishing their own assay performance characteristics in these populations.

Caution: U.S. Federal Law restricts this device to sale by or on the order of a physician.

2. Indication(s) for use:

   Same as Intended Use

3. Special conditions for use statement(s):

   For prescription use only

4. Special instrument requirements:

   Elecsys 2010 and cobas e 411 analyzers; MODULAR ANALYTICS E170 and cobas e 601 analyzers

I. Device Description:

The Elecsys anti-HAV test is a qualitative assay based on electrochemiluminescence immunoassay "ECLIA" technology. The Elecsys anti-HAV test utilizes a competitive immunoassay format in which sample anti-HAV antibody competes with biotinylated and ruthenylated anti-HAV monoclonal antibodies for a limited amount of cell culture-derived HAV antigen. The sample antibody and the HAV antigen react in the first incubation. The biotinylated antibodies and ruthenium complex[a] -labeled antibodies specific for HAV antigen are added in the second incubation together with streptavidin-coated magnetic microparticles. The unbound HAV antigen reacts with the modified antibodies and the resulting immune complexes are bound to the solid phase through a biotin-streptavidin interaction. If all HAV antigens are complexed by sample anti-HAV antibody during the first incubation, no modified/labeled immune complexes are formed and captured during the second incubation.

Following the second incubation, the reaction mixture is aspirated into the measuring cell where the microparticles are magnetically captured onto the surface of the electrode. Unbound substances are removed by elution with ProCell. Application of a voltage to the electrode induces chemiluminescent emission from the captured immune complexes which is measured by a photomultiplier. The level of signal detected by the system decreases as the concentration of the anti-HAV antibody target present in a patient sample increases.

Results are determined via a calibration curve which is generated by 2-point calibration on the instrument and a master curve provided via the reagent barcode. The calibration process converts the output so that low levels of sample anti-HAV antibodies are expressed by low output and high levels of antibody are expressed by high output. These outputs are finally interpreted on a qualitative basis around the established cut-off output.

a) Tris(2,2'-bipyridyl)ruthenium(II)-complex $(Ru(bpy))_3^{2+}$

J. Substantial Equivalence Information:

1. Predicate device name(s): Abbott AxSym HAVAB® 2.0
2. Predicate Numbers (s): P780012/S009
3. Comparison with predicate:

Similarities

Anti-HAV Immunoassay Comparison		
Feature	**Elecsys Anti-HAV Assay**	**Predicate Device Abbott Axsym HAVAB 2.0 Assay (P780012/S009)**
Intended Use	The Roche Elecsys Anti-HAV immunoassay is used for the in vitro qualitative detection of total antibodies (IgM and IgG) to hepatitis A virus in human serum and plasma (K_2-EDTA). The assay is intended for use as an aid in the laboratory diagnosis of past or acute/recent hepatitis A infection. Assay results, in conjunction with other laboratory results and clinical information, may be used to provide presumptive evidence of infection with hepatitis A virus in persons with signs or symptoms of hepatitis and in persons at risk for hepatitis A infection, or used as an aid to determine the presence of antibody response to HAV in vaccine recipients. The electrochemiluminescence immunoassay "ECLIA" is intended for use on Elecsys and cobas e immunoassay analyzers.	Immunoassay for the qualitative detection of total antibody to hepatitis A virus (anti-HAV) in human serum or plasma (potassium EDTA, sodium heparin, sodium citrate, or lithium heparin). A test for anti-HAV is indicated as an aid in the laboratory diagnosis of previous or ongoing hepatitis A viral infection or in the identification of HAV-susceptible individuals for vaccination. Assay results, in conjunction with other laboratory results and clinical information, may be used to provide presumptive evidence of infection with hepatitis A virus in persons with signs or symptoms of hepatitis and in persons at risk for hepatitis A infection.
Indications for Use	Same	Same
Sample Type	Human serum and plasma	Same

Differences

Anti-HAV Immunoassay Comparison		
Features	**Elecsys Anti-HAV Assay**	**Predicate Device** **Abbott Axsym HAVAB 2.0 Assay (P780012/S009)**
Detection Protocol	Electrochemiluminescence immunoassay (ECLIA)	Microparticle Enzyme Immunoassay (MEIA)
Traceability/ Standardization	Standardized against the "Second International Standard for Anti-Hepatitis A, Immunoglobulin, Human, NIBSC code: 97/646" of the NIBSC (National Institute for Biological Standards and Control).	Not Given
Interpretation of Results	≥ 22.0 IU/L Reactive 18.0 ≤ IU/L< 22.0 Equivocal <18.0 IU/L Negative	0.000 to 1.000 Reactive 1.001 to 1.200 Grayzone 1.201 to 3.000 Nonreactive >3.000 Not applicable
Calibration Interval	Once per reagent lot and • After 1 month (28 days) when using the same reagent lot • After 7 days (when using the same reagent kit on the analyzer) • As required: e.g., quality control findings outside the specified limits	A minimum of two replicates of the AxSYM HAFVAB 2.0 Index Calibrator must be tested. A single sample of both the Negative and Positive Controls must be tested as a means of evaluating the assay calibration. Once the 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 • Either of the AxSYM HAVAB 2.0 Control values is out of its specified range • The MEIA Optics Verification Update has been performed
Controls	Elecsys PreciControl Anti-HAV	Abbott AxSYM HAVAB 2.0 Controls

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

CLSI EP5-A2, "Evaluation of Precision Performance of Quantitative Measurement Methods"

CLSI EP17-A, "Protocols for Determination of Limits of Detection"
Class II Special Controls Guidance Document: Hepatitis A Virus Serological Assays
http://www.fda.gov/downloads/MedicalDevices/DeviceRegulationandGuidance/GuidanceDocuments/ucm071055.pdf

L. Test Principle:

The Elecsys Anti-HAV immunoassay utilizes a competitive immunoassay format employing cell culture derived HAV antigen and a monoclonal antibody (modified by biotinylation or ruthenylation) in competition to the polyclonal sample antibody. Capture of formed immune complexes from the reaction mixture is based on biotin in the immune complex binding to streptavidin-coated magnetic microparticles which are collected on a measuring cell electrode. Signal generation is triggered by the application of a voltage to the electrode (electrochemiluminescence technology). The level of signal count detected by the system decreases as the concentration of the anti-HAV total antibody target present in a patient sample increases. Results are determined via a calibration curve which is instrument-specifically generated by 2-point calibration and a master curve provided via the reagent barcode

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:
Precision and Reproducibility of the Elecsys Anti-HAV assay was evaluated on Elecsys 2010 analyzer and MODULAR ANALYTICS E170 analyzer using Elecsys reagents, three human sera and controls, according to the CLSI EP15-A2/EP5-A2.

For precision study, three human sera (high negative, low positive and moderately positive) and controls (PreciControl Anti-HAV 1 and PreciControl Anti-HAV 2) were tested in replicates of 2 in 2 runs per day for 20 days with one reagent lot spanning at least two cycles of calibration using one Elecsys 2010 analyzer and one Modular Analytics E170 analyzer. Results are presented below.

- Precision on Elecsys® 2010

Sample	Mean (IU/L)	Repeatability		Intermediate Precision (within-laboratory precision)		
		SD (IU/L)	CV (%) (UCL 95%)	SD (IU/L)	CV (%) (UCL 95%)	n
PC Anti-HAV 1	12.3	0.221	1.8 (2.2)	0.378	3.1 (3.7)	80
PC Anti-HAV 2	29.2	0.947	3.2 (4.0)	1.325	4.5 (5.3)	80
HSP 1	17.6	0.534	3.0 (3.7)	0.586	3.3 (3.8)	80
HSP 2	21.9	0.538	2.5 (3.0)	0.667	3.0 (3.6)	80
HSP3	49.3	1.655	3.4 (4.1)	2.261	4.6 (5.4)	80

- Precision on MODULAR® ANALYTICS E170

Sample	Mean (IU/L)	Repeatability		Intermediate Precision (within-laboratory precision)		
		SD (IU/L)	CV (%) (UCL 95%)	SD (IU/L)	CV (%) (UCL 95%)	n
PC Anti-HAV 1	12.54	0.188	1.5 (1.8)	0.688	5.5 (7.1)	80
PC Anti-HAV 2	32.12	0.724	2.3 (2.8)	1.849	5.8 (7.4)	80
HSP 1	18.2	0.459	2.5 (3.1)	0.816	4.5 (5.6)	80
HSP 2	23	0.254	1.1 (1.4)	1.016	4.4 (5.9)	80
HSP3	55.37	1.088	2.0 (2.4)	2.374	4.3 (5.3)	80

PC = PreControl
UCL = Upper Confidence Limit

For reproducibility study, three human sera (high negative, low positive and moderately positive) and controls (PreciControl Anti-HAV 1 and PreciControl Anti-HAV 2) were tested in replicates of 3 in 2 runs per day for 5 days with three Elecsys 2010 analyzers and three Modular Analytics E170 analyzers. Results are presented below.

Elecsys anti-HAV: Within Site Reproducibility: Elecsys® 2010 Analyzer

Clinical Site	Sample	N	Mean IU/L	Repeatability		Between Run		Between Day		Within Site Reproducibility	
				SD	% CV	SD	% CV	SD	% CV	SD	% CV
Site 1	HSP 01	30	22.629	0.523	2.3	0.275	1.2	0.176	0.8	0.616	2.7
	HSP 02	30	49.903	2.469	4.9	0.000	0.0	0.123	0.2	2.472	5.0
	HSP 03	30	18.287	0.527	2.9	0.052	0.3	0.264	1.4	0.592	3.2
	PC A-HAV1	30	12.678	0.321	2.5	0.000	0.0	0.201	1.6	0.379	3.0
	PC A-HAV2	30	29.198	0.707	2.4	0.516	1.8	0.505	1.7	1.011	3.5
Site 2	HSP 01	30	23.196	0.302	1.3	0.452	2.0	0.000	0.0	0.544	2.3
	HSP 02	30	50.965	0.688	1.4	0.55	1.1	0.000	0.0	0.881	1.7
	HSP 03	30	18.499	0.417	2.3	0.336	1.8	0.000	0.0	0.535	2.9
	PC A-HAV1	30	12.942	0.278	2.2	0.309	2.4	0.000	0.0	0.416	3.2
	PC A-HAV2	30	30.814	0.376	1.2	0.441	1.4	0.000	0.0	0.58	1.9
Site 3	HSP 01	30	22.634	0.371	1.6	0.000	0.0	0.266	1.2	0.456	2.0
	HSP 02	30	48.03	1.356	2.8	1.451	3	0.000	0.0	1.985	4.1
	HSP 03	30	17.892	0.398	2.2	0.268	1.5	0.292	1.6	0.561	3.1
	PC A-HAV1	30	12.515	0.248	2.0	0.106	0.9	0.372	3.0	0.459	3.7
	PC A-HAV2	30	29.46	1.144	3.9	0.962	3.3	0.503	1.7	1.578	5.4

Elecsys anti-HAV: Within Site Reproducibility: MODULAR® ANALYTICS E170

Clinical Site	Sample	N	Mean IU/L	Repeatability		Between Run		Between Day		Within Site Reproducibility	
				SD	% CV	SD	% CV	SD	% CV	SD	% CV
Site 1	HSP 01	30	23.433	0.237	1.0	0.419	1.8	0.000	0.0	0.481	2.1
	HSP 02	30	51.725	0.454	0.9	0.707	1.4	0.000	0.0	0.84	1.6
	HSP 03	30	18.918	0.158	0.8	0.167	0.9	0.135	0.7	0.266	1.4
	PC A-HAV1	30	13.205	0.197	1.5	0.211	1.6	0.132	1.0	0.317	2.4
	PC A-HAV2	30	31.477	0.339	1.1	0.787	2.5	0.000	0.0	0.857	2.7
Site 2	HSP 01	30	22.541	0.705	3.1	0.090	0.4	0.231	1.0	0.747	3.3
	HSP 02	30	51.871	1.344	2.6	0.000	0.0	0.302	0.6	1.378	2.7
	HSP 03	30	18.455	0.642	3.5	0.000	0.0	0.355	1.9	0.733	4
	PC A-HAV1	30	12.676	0.695	5.5	0.000	0.0	0.627	4.9	0.936	7.4
	PC A-HAV2	30	31.438	0.733	2.3	0.231	0.7	0.000	0.0	0.769	2.4
Site 3	HSP 01	30	23.946	0.288	1.2	0.22	0.9	0.000	0.0	0.363	1.5
	HSP 02	30	52.166	0.299	0.6	0.829	1.6	0.000	0.0	0.881	1.7
	HSP 03	30	19.426	0.203	1.0	0.336	1.7	0.000	0.0	0.392	2.0
	PC A-HAV1	30	12.974	0.205	1.6	0.225	1.7	0.000	0.0	0.304	2.3
	PC A-HAV2	30	31.236	0.241	0.8	1.038	3.3	0.000	0.0	1.066	3.4

Elecsys anti-HAV: Between Site Reproducibility: Elecsys® 2010 Analyzer

Sample	N	Mean IU/L	Repeatability		Between Run		Between Day		Between Site		Reproducibility	
			SD	% CV	SD	% CV	SD	% CV	SD	% CV	SD	% CV
HSP 01	90	22.819	0.409	1.8	0.3	1.3	0.000	0.0	0.304	1.3	0.591	2.6
HSP 02	90	49.633	1.674	3.4	0.697	1.4	0.000	0.0	1.462	2.9	2.33	4.7
HSP 03	90	18.226	0.451	2.5	0.25	1.4	0.166	0.9	0.276	1.5	0.608	3.3
PC A-HAV1	90	12.712	0.284	2.2	0.179	1.4	0.22	1.7	0.176	1.4	0.438	3.4
PC A-HAV2	90	29.824	0.807	2.7	0.68	2.3	0.394	1.3	0.808	2.7	1.386	4.6

Elecsys anti-HAV: Between Site Reproducibility: MODULAR® ANALYTICS E170

Sample	N	Mean IU/L	Repeatability		Between Run		Between Day		Between Site		Reproducibility	
			SD	% CV	SD	% CV	SD	% CV	SD	% CV	SD	% CV
HSP 01	90	23.307	0.46	2	0.278	1.2	0.000[c]	0.0	0.7	3	0.883	3.8
HSP 02	90	51.921	0.837	1.6	0.542	1	0.000[c]	0.0	0.168	0.3	1.011	1.9
HSP 03	90	18.933	0.399	2.1	0.148	0.8	0.17	0.9	0.472	2.5	0.658	3.5
PC A-HAV1	90	12.952	0.433	3.3	0.000[c]	0	0.368	2.8	0.193	1.5	0.6	4.6
PC A-HAV2	90	31.384	0.487	1.6	0.764	2.4	0.000[c]	0	0.000[c]	0	0.906	2.9

b. *Linearity/assay reportable range:*
The qualitative assay has a quantitative measurement. The detectable range is from 6 IU/L to 60 IU/L.

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

Traceability: This method has been standardized against the "Second International Standard for Anti-Hepatitis A, Immunoglobulin, Human, NIBSC code: 97/646" of the NIBSC (National Institute for Biological Standards and Control).

Controls:

PC A-HAV 1: anti-HAV antibodies (human), with approximately 12 IU/L

PC A-HAV 2: anti-HAV antibodies (human), with approximately 30 IU/L

Calibrators:
Calibration of the Elecsys anti-HAV is performed by using the 2 calibrators provided with the reagent kit. Calibrator 1 is an anti-HAV negative serum; Calibrator 2 is an anti-HAV positive serum with approx. 46 IU/L.
A reagent lot specific master standard curve is provided on the barcode of each reagent kit. The calibrator master standard curve is established by diluting an anti-HAV positive human serum with an analyte free human serum matrix. The concentration value was assigned to it by reading the concentration from the reference calibrator curve using four E2010 and four E170 instruments with 2 runs and 2 replicates per run and per instrument. The master curve is stored in the barcode and is automatically read in by the instrument when the barcode is scanned for the new reagent.
The calibration must be performed once per reagent lot. Acceptance criteria for the assigned values are < 20 IU/L for Cal 1 and ≥ 40 IU/L for Cal 2. Re-calibration is recommended if the same reagent lot was used more than 1 month (28 days), or if the same reagent kit has been left on the analyzer for 7 days.

Stability:

Reagents:

unopened at 2-8 °C	up to the stated expiration date
M, R1, R2 after opening at 2-8 °C	8 weeks
on Elecsys® 2010 and cobas e® 411 at 20-25 °C	8 weeks
on MODULAR® ANALYTICS E170 and cobas e® 601 at 20-25 °C	8 weeks
Reconstituted calibrators at -20 °C	12 weeks
Reconstituted calibrators at 2-8 °C	2 weeks
Reconstituted calibrators on Elecsys® 2010 and cobas e® 411 at 20-25 °C	up to 6 hours
Reconstituted calibrators on MODULAR® ANALYTICS E170 and cobas e® 601 at 32°C	up to 2 hours

PreciControls:

unopened at 2-8 °C	up to the stated expiration date
after opening at 2-8 °C	8 weeks
After opening at -20 °C	3 months (freeze only once)
on Elecsys® 2010 and cobas e® 411 at 20-25 °C	up to 7 hours
on MODULAR® ANALYTICS E170 and cobas e® 601 at 20-25 °C	up to 7 hours

Samples:

Sample stability was determined using different sample materials e.g. Serum, Serum with separating gel, K_2-EDTA Plasma. Four negative samples, one sample around cutoff, and three low positive samples were tested and compared with fresh reference materials.

Samples stored at -20°C	up to 3 months
Samples stored at 2-8°C	Up to 7 days
Freeze and thaw cycles	Up to 5 cycles
on Elecsys® 2010 and cobas e® 411 at 20-25 °C	up to 8 hours

d. Detection limits:

Limit of Blank (**LoB)** of the Elecsys® Anti-HAV assay was determined according to CLSI EP17-A. Limit of Blank determines the highest observed measurement values for samples free of analyte. The LoB was determined as the 95th percentile of measurements of blank samples. The distribution of values for five zero-level human serum has been determined on two Elecsys® 2010 / cobas e® 411 Analyzers and on two MODULAR® ANALYTICS E170 / cobas e® 601 Immunoassay Analyzers over 3 days, 2 runs per day. The LoB for Elecsys® 2010 / cobas e® 411 Analyzer is 3.03 IU/L and 2.099 IU/L for MODULAR® ANALYTICS E170 / cobas e® 601 Immunoassay Analyzers.

Limit of detection (LoD) of the Elecsys® Anti-HAV assay was determined according to CLSI EP17-A. The LoD was determined as the lowest amount of analyte in a sample that can be detected with 95% probability. The distribution of values for five low-level human serum samples has been determined on two Elecsys® 2010 / cobas e® 411 Analyzers and on two MODULAR® ANALYTICS E170 / cobas e® 601 Immunoassay Analyzers over 3 days, 2 runs per day. Samples were measured in one-fold determination in each run. 30 measuring points were collected per instrument.

$LOD = LOB + 1.6529 \times SD_{total}$ (of low analyte samples)
The LoD for Elecsys® 2010 / cobas e® 411 Analyzer is 5.155 IU/L, and 2.994 IU/L for MODULAR® ANALYTICS E170 / cobas e® 601 Immunoassay Analyzers.

The LOD is set at 6.00 IU/L for both types of analyzers and is reported in the labeling.

Calibrators: Range for the electrochemiluminescence signals for the calibrators:
Negative calibrator (Cal1): < 20 IU/L (Elecsys® 2010, MODULAR® ANALYTICS E170 and cobas e® analyzers).
Positive calibrator (Cal2): ≥ 40 IU/L (Elecsys® 2010, MODULAR® ANALYTICS E170 and cobas e® analyzers)

e. Analytical specificity:

Cross-reactivity: 177 samples from 15 potentially cross-reactive sub-groups were used for cross-reactivity study on the Elecsys 2010. 174 Samples were found to be nonreactive (negative) in both the Elecsys anti-HAV assay and the predicate AxSym HAVAB 2.0 assay; 3 samples were found to be discordant between the Elecsys anti-HAV assay and the AxSym HAVAB 2.0 assay. The testing results are summarized in the table below:

Category	HAVAB 2.0 Assay									Total
	Reactive			Equivocal			Non-Reactive			
	Elecsys 2010 HAV 2.1 Assay									
	RX[a]	EQ[b]	NR[c]	RX	EQ	NR	RX	EQ	NR	
Anti-nuclear Antibody (ANA)	0	0	0	0	0	0	0	0	13	**13**
Cytomegalovirus (anti-CMV)	0	0	0	0	0	0	0	0	13	**13**
Epstein-Barr Virus (anti-EBV)	0	0	0	0	0	0	0	0	14	**14**
Elevated IgG	0	0	0	0	0	0	0	0	13	**13**
Elevated IgM	0	0	0	0	0	0	0	0	13	**12**
Hepatitis B Virus (HBsAg or HBV-DNA)	0	0	0	0	0	0	1	0	10	**11**
Hepatitis C Virus (anti-HCV)	0	0	0	0	0	0	0	0	12	**12**
Human Immunodeficiency Virus (anti-HIV-1)	0	0	0	0	0	0	2	0	11	**13**
Herpes Simplex Virus (anti-HSV)	0	0	0	0	0	0	0	0	10	**10**
Mumps / Rubeola	0	0	0	0	0	0	0	0	12	**12**
Parvovirus B19 Infection	0	0	0	0	0	0	0	0	11	**11**
Rheumatoid factor	0	0	0	0	0	0	0	0	7	**7**
Rubella (anti-Rubella)	0	0	0	0	0	0	0	0	10	**10**
Toxoplasmosis (anti-Toxo)	0	0	0	0	0	0	0	0	13	**13**
Varicella Zoster (anti-VZV)	0	0	0	0	0	0	0	0	12	**12**
Total	**0**	**0**	**0**	**0**	**0**	**0**	**3**	**0**	**174**	**177**

[a] Number of Reactive specimens
[b] Number of Equivocal or Borderline specimens
[c] Number of Non-Reactive specimens

HAMA (human anti-mouse antibodies) effect was tested by comparing the recovery of 4 human serum samples spiked with different suitable HAMA serum samples versus 4 unspiked aliquots of samples. No HAMA effect was found.

Interference: The impact of endogenous interfering substances on the Elecsys Anti-HAV assay was determined testing native human serum pools on Elecsys® 2010 Immunoassay Analyzer.

The assay is unaffected by icterus (bilirubin < 50 mg/dL), hemolysis (Hb

< 1000 mg/dL), lipemia (Intralipid < 1500 mg/dL), and biotin < 50 ng/mL. Serum pools (high negative, low positive, and high positive) were used to spike with the intereferent. In patients receiving therapy with high biotin doses (i.e. > 5 mg/day), no sample should be taken until at least 8 hours after the last biotin administration. One aliquot of each serum sample was spiked with the interfering substance; another aliquot was spiked with the same volume of isotonic NaCl solution (dilution pool). The interfering pool was then diluted into the dilution pool in 10% increments. The % recovery was determined by dividing the mean value of the measured concentration by the expected concentration.
Criterion: Recovery of positive samples within ± 20 % of initial value.

In vitro tests were performed on 18 commonly used pharmaceuticals (Acetylcysteine, Ampicillin-Na, Ascorbic acid, Ca-Dobesilate, Cyclosporine, Cefoxitin, Heparin, Intralipid, Levodopa, Methyldopa, Metronidazole, Phenylbutazone, Tetracycline, Acetylsalicylic acid, Rifampicin, Acetaminophen, Ibuprofen, and Theophylline) and in addition on folic acid. No interference with the assay was found.

f. *Assay cut-off:*

The cutoff was set at 20 IU/L according to the WHO recommendation for vaccination. It was established by histogram analysis of the predicate and was confirmed by measuring a total of 736 samples from 2 European centers including hospitalized patients, acute HAV infection patient, blood doors, pregnant women and the other samples that suspected for HAV infection. The 20 IU/L in the Elecsys anti-HAV assay corresponds to 1.0 of S/CO in the predicate.

Non-reactive: < 18 IU/L
Border: 18.0 ≤ IU/L < 22.0 IU/L
Reactive: ≥ 22.0 IU/L

Samples with a concentration < 18.0 IU/L are considered non-reactive in the Elecsys Anti-HAV test and no further testing is necessary.

Samples with a concentration ≥ 22.0 IU/L are considered reactive in the Elecsys Anti-HAV test.

Samples with a concentration between 18.0 and < 22.0 are considered border (borderline). The sample should be retested in duplicate.

- If 2 of the 3 results have a concentration < 18.0 IU/L, the result is interpreted as non-reactive and no further testing is necessary.
- If 2 of the 3 results have a concentration between 18.0 and < 22.0 IU/L, the result is interpreted as borderline. It is recommended that a specimen be drawn in two weeks and retested.

- If 2 of the 3 results have a concentration ≥ 22.0 IU/L, the result is interpreted as reactive.

2. Comparison studies:

   a. *Method comparison with predicate device:*
   The performance of the Elecsys anti-HAV assay was determined by percent agreement among negative samples and percent agreement among positive samples, against a consensus comparator method, in specific populations. The main predicate (Abbott AxSym HAV AB- 2.0) was used as sole comparator/reference. Detailed information can be found in clinical performance.

   b. *Matrix comparison:*

   The effect on quantitation of analyte in the presence of anticoagulants with the Elecsys® Anti-HAV Immunoassay was determined on Elecsys 2010 Immunoassay Analyzer by comparing values obtained from native samples (single donors) drawn into Serum, K_2-EDTA-Plasma and Serum-Gel Separation primary tubes. The following tables summarize the results for the comparison between serum and plasma matrices.

Plasma matrix	Number of positive specimens showing recovery to serum within various ranges		
	< 10%	10-15%	> 15%
K_2-EDTA	24	1	0
Serum Gel	23	2	0

Plasma matrix	Number of borderline specimens showing recovery to serum within various ranges		
	< 10%	10-15%	> 15%
K_2-EDTA	3	0	0
Serum Gel	3	0	0

Plasma matrix	Number of negative specimens showing recovery to serum within various ranges		
	< 1.0 IU/L	1.0 -2.0 IU/L	> 2.0 IU/L
K_2-EDTA	29	1	0
Serum Gel	30	0	0

3. Clinical studies:

   a. Clinical Sensitivity: N/A

   b. Clinical specificity: N/A

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

Clinical Performance

Clinical Study Cohorts:

A multi-center study was conducted in the U.S. to characterize the performance of the Elecsys Anti-HAV Immunoassay. All subjects were tested with the Elecsys Anti-HAV assay on the Elecsys 2010 analyzer and with an FDA-cleared reference method in strict accordance with the manufacturer's package insert instructions.
A total of 1050 samples were obtained from multiple specimen sources, representing subjects for whom routine hepatitis A testing had been ordered, hospitalized patients, subjects at increased risk for hepatitis, subjects with signs and symptoms of hepatitis, subjects characterized with acute hepatitis A, and subjects below the age of 21 years (pediatric/adolescents). All samples (prospective and retrospective) are stored frozen before shipment to Roche and to the respective sites for testing. Among the 1050 samples, 487 were (46.38%) were from males and 563 (53.62%) were from females. 0.38% were from American Indian/Alaska Native, 2.76% were Asian, 15.90% were African American /Black, 65.14% were Caucasian/White, 14.95% unknown and 0.86% others.

Among all specimens tested, one specimen subjected to routine Hepatitis A testing was concordantly equivocal, and was excluded from the calculations.

The positive and negative percent agreement for prospectively collect samples is 98.13% (95.96% to 99.31%) and 97.37% (95.06% to 98.79%), respectively.

The positive and negative percent agreement for retrospectively collect samples is 98.11% (94.59% to 99.61%) and 97.37% (94.36% to 99.03%), respectively.

The positive percent agreement and the negative percent agreement results for the different clinical population are presented in the following table:

Percent Agreement for Elecsys and Predicate anti-HAV Assay Results from Symptomatic Individuals

Elecsys 2010 Result	Predicate Result											
	Site 1 BW			Site 2 WU			Site 3 JH			All Sites		
	RX	EQ	NR	RX	EQ	NR	RX	EQ	NR	RX	EQ	NR
Reactive	60	0	2	0	0	0	16	0	0	76	0	2
Equivocal	1	0	2	0	0	0	1	0	0	2	0	2
Negative	0	0	97	0	0	0	0	0	17	0	0	114
Total	61	0	101	0	0	0	17	0	17	78	0	118
PPA	98.36 (60/61)			0.00 (0/0)			94.12 (16/17)			97.44 (76/78)		
95% CI	91.20 to 99.96			0.00 to 100.00			71.31 to 99.85			91.04 to 99.69		
NPA	96.04 (97/101)			0.00 (0/0)			100.00 (17/17)			96.61 (114/118)		
95% CI	90.17 to 98.91			0.00 to 100.00			80.49 to 100.00			91.55 to 99.07		

Percent Agreement for Elecsys and Predicate anti-HAV Assay Results from Specimens Subjected to Routine Hepatitis A Testing

Elecsys 2010 Result	Predicate Result											
	Site 1 BW			Site 2 WU			Site 3 JH			All Sites		
	RX	EQ	NR	RX	EQ	NR	RX	EQ	NR	RX	EQ	NR
Reactive	90	0	2	0	0	0	36	0	1	126	0	3
Equivocal	0	0	1	0	0	0	0	1	1	0	1	2
Negative	2	0	54	0	0	0	0	0	21	2	0	75
Total	92	0	57	0	0	0	36	1	23	128	1	80
PPA	97.83 (90/92)			0.00 (0/0)			100.00 (36/36)			98.44 (126/128)		
95% CI	92.37 to 99.74			0.00 to 100.00			90.26 to 100.00			94.47 to 99.81		
NPA	94.74 (54/57)			0.00 (0/0)			91.30 (21/23)			93.57 (75/80)		

Percent Agreement for Elecsys and Predicate anti-HAV Assay Results from Hospitalized Patients (Retrospective and Prospective)

Elecsys 2010 Result	Predicate Result											
	Site 1 BW			Site 2 WU			Site 3 JH			All Sites		
	RX	EQ	NR	RX	EQ	NR	RX	EQ	NR	RX	EQ	NR
Reactive	42	0	0	0	0	0	28	0	0	70	0	0
Equivocal	0	0	0	0	0	0	1	0	0	1	0	0
Negative	0	0	90	0	0	0	1	0	51	1	0	141

Total	42	0	90	0	0	0	30	0	51	72	0	141
PPA	100.00 (42/42)			0.00 (0/0)			93.33 (28/30)			97.22 (70/72)		
95% CI	91.59 to 100.00			0.00 to 100.00			77.93 to 99.18			90.32 to 99.66		
NPA	100.00 (90/90)			0.00 (0/0)			100.00 (51/51)			100.00 (141/141)		
95% CI	95.98 to 100.00			0.00 to 100.00			93.02 to 100.00			97.42 to 100.00		

Percent Agreement for Elecsys and Predicate anti-HAV Assay Results from Subjects Characterized as Acute Hepatitis A (Retrospective and Prospective)

	Predicate Result											
	Site 1 BW			**Site 2 WU**			**Site 3 JH**			**All Sites**		
Elecsys 2010 Result	**RX**	**EQ**	**NR**	**RX**	**EQ**	**NR**	**RX**	**EQ**	**NR**	**RX**	**EQ**	**NR**
Reactive	81	0	0	0	0	0	50	0	0	131	0	0
Equivocal	0	0	0	0	0	0	0	0	0	0	0	0
Negative	0	0	3	0	0	0	0	0	0	0	0	3
Total	81	0	3	0	0	0	50	0	0	131	0	3
PPA	100.00 (81/81)			0.00 (0/0)			100.00 (50/50)			100.00 (131/131)		
95% CI	95.55 to 100.00			0.00 to 100.00			92.89 to 100.00			97.22 to 100.00		
NPA	100.00 (3/3)			0.00 (0/0)			0.00 (0/0)			100.00 (3/3)		
95% CI	29.24 to 100.00			0.00 to 100.00			0.00 to 100.00			29.24 to 100.00		

Percent Agreement for Elecsys and Predicate anti-HAV Assay Results from Pediatric and Adolescent Subjects

	Predicate Result											
	Site 1 BW			**Site 2 WU**			**Site 3 JH**			**All Sites**		
Elecsys 2010 Result	**RX**	**EQ**	**NR**	**RX**	**EQ**	**NR**	**RX**	**EQ**	**NR**	**RX**	**EQ**	**NR**
Reactive	0	0	0	0	0	0	19	0	2	19	0	2
Equivocal	0	0	0	0	0	0	0	0	0	0	0	0
Negative	0	0	0	0	0	0	0	0	65	0	0	65
Total	0	0	0	0	0	0	19	0	67	19	0	67
PPA	0.00 (0/0)			0.00 (0/0)			100.00 (19/19)			100.00 (19/19)		
95% CI	0.00 to 100.00			0.00 to 100.00			82.35 to 100.00			82.35 to 100.00		
NPA	0.00 (0/0)			0.00 (0/0)			97.01 (65/67)			97.01 (65/67)		
95% CI	0.00 to 100.00			0.00 to 100.00			89.63 to 99.64			89.63 to 99.64		

Agreement for Elecsys and Predicate anti-HAV Assay Results from Subjects at Increased Risk for Hepatitis

Elecsys 2010 Result	Predicate Result											
	Site 1 BW			Site 2 WU			Site 3 JH			All Sites		
	RX	EQ	NR	RX	EQ	NR	RX	EQ	NR	RX	EQ	NR
Reactive	10	0	1	0	0	0	38	0	2	48	0	3
Equivocal	0	0	0	0	0	0	0	0	1	0	0	1
Negative	1	0	5	0	0	0	2	0	152	3	0	157
Total	11	0	6	0	0	0	40	0	155	51	0	161
PPA	90.91 (10/11)			0.00 (0/0)			95.00 (38/40)			94.12 (48/51)		
95% CI	58.72 to 99.77			0.00 to 100.00			83.08 to 99.39			83.76 to 98.77		
NPA	83.33 (5/6)			0.00 (0/0)			98.06 (152/155)			97.52 (157/161)		
95% CI	35.88 to 99.58			0.00 to 100.00			94.45 to 99.60			93.76 to 99.32		

RX – Reactive, positive, EQ – equivocal, borderline, NR – non-reactive, negative, PPA – positive percent agreement, NPA negative percent agreement, 95% CI - 95% Exact confidence interval

Summary of the Percent Agreements for the Various Specimen Cohorts

Cohort	Positive Percent Agreement		Negative Percent Agreement	
	PPA (x/n)	95 % confidence interval	NPA (x/n)	95 % confidence interval
Routine HAV Testing	98.44 (126/128)	94.47 to 99.81	93.75 (75/80)	86.01 to 97.94
Hospitalized	97.22 (70/72)	90.32 to 99.66	100.00 (141/141)	97.42 to 100.00
Signs and Symptoms	97.44 (76/78)	91.04 to 99.69	96.61 (114/118)	91.55 to 99.07
Increased Risk for Hepatitis	94.12 (48/51)	83.76 to 98.77	97.52 (157/161)	93.76 to 99.32
Characterized Acute HAV	100.00 (131/131)	97.22 to 100.00	100.00 (3/3)	29.24 to 100.00
Pediatric/Adolescent	100.00 (19/19)	82.35 to 100.0	97.01 (65/67)	89.63 to 99.64
Overall	98.12 (470/479)	96.46 to 99.14	97.37 (555/570)	95.70 to 98.52

HAV Vaccination:

Fifty-four specimens that were collected both pre- and post-HAV vaccination was evaluated by the Elecsys anti-HAV assay and the predicate. The post-vaccination specimens were obtained at least four weeks but not more than 10 weeks after the completion of the vaccine regimen. Three HAV vaccines which are currently licensed in the US were used in this study: VAQTA (16), HAVRIX (20) and TWINRIX (18). Two vaccination studies are represented: one conducted in the Eastern region of the US and the other in Penzberg Germany. There was no antibody response observed in pre-vaccination specimens with the exception of two

specimens which were just above the equivocal/borderline (- 23.23 IU/L) and just reactive (30.34 IU/L), respectively.

Percent Agreement for Elecsys and Predicate anti-HAV Assays Results from Pre-HAV Vaccination Specimens

				anti-HAV Assay Results								
Elecsys Result				**Predicate Result**								
	HAVRIX			**TWINRIX**			**VAQTA**			**All Vaccines**		
	RX	**EQ**	**NR**	**RX**	**EQ**	**NR**	**RX**	**EQ**	**NR**	**RX**	**EQ**	**NR**
Reactive	**0**	**0**	**0**	**0**	**0**	**1**	**0**	**0**	**0**	**0**	**0**	**1**
Equivocal	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**
Non-Reactive	**0**	**0**	**20**	**0**	**0**	**17**	**0**	**0**	**16**	**0**	**0**	**53**
Total	**0**	**0**	**20**	**0**	**0**	**18**	**0**	**0**	**16**	**0**	**0**	**54**
PPA	0.00 (0/0)			0.00 (0/0)			0.00 (0/0)			0.00 (0/0)		
95% CI*	0.00 to 100.00			0.00 to 100.00			0.00 to 100.00			0.00 to 100.00		
NPA	100.00 (20/20)			94.44 (17/18)			100.00 (16/16)			98.15 (53/54)		
95% CI*	83.16 to 100.00			72.71 to 99.86			79.41 to 100.00			90.11 to 99.95		

Percent Agreement for Elecsys and Predicate anti-HAV Assays Results from Post-HAV Vaccination Specimens

				anti-HAV Assay Results								
Elecsys Result				**Predicate Result**								
	HAVRIX			**TWINRIX**			**VAQTA**			**All Vaccines**		
	RX	**EQ**	**NR**	**RX**	**EQ**	**NR**	**RX**	**EQ**	**NR**	**RX**	**EQ**	**NR**
Reactive	**20**	**0**	**0**	**17**	**0**	**1**	**16**	**0**	**0**	**53**	**0**	**1**
Equivocal	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**
Non-Reactive	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**
Total	**20**	**0**	**0**	**17**	**0**	**1**	**16**	**0**	**0**	**53**	**0**	**1**
PPA	100.00 (20/20)			100.00 (17/17)			100.00 (16/16)			100.00 (53/53)		
95% CI*	83.16 to 100.00			80.49 to 100.00			79.41 to 100.00			93.28 to 100.00		
NPA	0.00 (0/0)			0.00 (0/1)			0.00 (0/0)			0.00 (0/1)		
95% CI*	0.00 to 100.00			0.00 to 97.50			0.00 to 100.00			0.00 to 97.50		

RX – Reactive, positive, EQ – equivocal, borderline, NR – non-reactive, negative, PPA – positive percent agreement, NPA negative percent agreement, 95% CI - 95% Exact confidence interval

Prevalence Studies:

The Elecsys Anti-HAV assay was used to evaluate the prevalence of HAV total antibodies in an apparently healthy population (normal, healthy individuals without symptoms). The prospective study population for the Elecsys Anti-HAV assay consisted of 602 patients. Of these 602 patients, 300 patients were from the high prevalence region, Western states of the U.S. (New Mexico) and 302 patients were from the low risk region Eastern states of the U.S. (Indiana). The prospective study population was 208 (34.6 %) males and 394 (65.4 %) females (total n = 602) including 493 (81.9 %) Caucasians, 32 (5.3 %) African Americans, 6 (1.0 %) Asians, 69 (11.5 %) American Indians and 2 (0.3 %) Others. The results of prevalence population are summarized according to age groups in decades, gender, geographical area and the number of reactive, non-reactive and equivocal results.

Prevalence rate for reactive anti-HAV assay in specimens collected in a low prevalence region, Eastern states of the US (Indiana), was 35.10 %.
Prevalence rate for reactive anti-HAV assay in specimens collected in a high prevalence region, Western states of the US (New Mexico), was 37.33%. The results of prevalence study are summarized according to age groups and gender.

Expected results for the Elecsys Anti-HAV assay in subjects from low prevalence areas for Hepatitis A

Age range (years)	Gender	Elecsys Anti-HAV result						Total
		Reactive		Equivocal		Non-reactive		
		N	Percent	N	Percent	N	Percent	
11 - 20	Female	0	0.0	0	0.0	1	100.0	1
	Male	0	0.0	0	0.0	1	100.0	1
21-30	Female	1	14.3	0	0.0	6	85.7	7
	Male	0	0.0	0	0.0	6	100.0	6
31-40	Female	2	9.5	1	4.8	18	85.7	21
	Male	0	0.0	0	0.0	2	100.0	2
41-50	Female	3	13.6	1	4.6	18	81.8	22
	Male	1	7.7	0	0.0	12	92.3	13
51-60	Female	7	16.7	1	2.4	34	81.0	42
	Male	9	47.4	0	0.0	10	52.6	19
61-70	Female	23	45.1	0	0.0	28	54.9	51
	Male	12	42.9	0	0.0	16	57.1	28
71-80	Female	24	50.0	0	0.0	24	50.0	48
	Male	14	46.7	0	0.0	16	53.3	30
> 80	Female	4	80.0	0	0.0	1	20.0	5
	Male	6	100.0	0	0.0	0	0.0	6
All ages	Female	64	32.5	3	1.5	130	66.0	197
	Male	42	40.0	0	0.0	63	60.0	105
Total		106	35.1	3	1.0	193	63.9	302

Expected results for the Elecsys Anti-HAV assay in subjects from high prevalence areas for Hepatitis A

Age range (years)	Gender	Elecsys Anti-HAV result						Total
		Reactive		Equivocal		Non-reactive		
		N	Percent	N	Percent	N	Percent	
11- 20	Female	4	50.0	0	0	4	50.0	8
	Male	2	40.0	0	0.0	3	60.0	5
21-30	Female	4	23.5	0	0.0	13	76.5	17
	Male	2	18.2	0	0.0	9	81.8	11
31-40	Female	3	11.1	0	0.0	24	88.9	27
	Male	3	23.1	0	0.0	10	76.9	13
41-50	Female	14	26.9	0	0.0	38	73.1	52
	Male	6	33.3	0	0.0	12	66.7	18
51-60	Female	21	38.9	0	0.0	33	61.1	54
	Male	11	45.8	0	0.0	13	54.2	24
61-70	Female	18	72.0	0	0.0	7	28.0	25
	Male	9	36.0	0	0.0	16	64.0	25
71-80	Female	9	75.0	0	0.0	3	25.0	12
	Male	5	71.4	0	0.0	2	28.6	7
> 80	Female	0	0.0	0	0.0	1	100.0	1
	Male	0	0.0	0	0.0	0	0.0	0
Unknown	Female	1	100.0	0	0.0	0	0.0	1
	Male	0	0.0	0	0.0	0	0.0	0
All ages	Female	74	37.6	0	0.0	123	62.4	197
	Male	38	36.9	0	0.0	65	63.1	103
Total		112	37.3	0	0.0	188	62.7	300

Seroconversion Sensitivity:

Seroconversion sensitivity of the Elecsys anti-HAV assay was shown by testing 3 seroconversion panels in comparison to that of the comparator assay (Abbott AxSym HAV AB- 2.0). Seroconversion panel results were also compared with the data provided by the vendor for the Abbott AxSym HAV AB 2.0 assay. The comparator assay and vendor assay are based on the Abbott AxSym HAV AB-2.0. The results are summarized in the following table:

Elecsys anti-HAV assay Seroconversion Panel Results

Panel ID	Elecsys 2010 assay		[1]Comparator anti-HAV assay		[2]Comparator anti-HAV assay	
	Post bleed day of earliest reactive result	Post bleed day of last positive result	Non-Reactive	Reactive	Non-Reactive	Reactive
HAV-01	0	91	0	91	0	91
[3]PHT 902	16	163	Not tested	Not tested	16	163
RP013	6	190	Not tested	Not tested	9	190

[1]The comparator results were shown by Roche using the Abbott AxSym HAV AB-2.0.

[2]The comparator results were provided by Vendor i.e. Abbott AxSym HAV AB-2.0

[3]The panel was not tested with the reference assay due to the limited sample size tested.

Method Comparison studies on two instruments:

The method comparison for the Elecsys anti-HAV assay between the two platforms E2010 and E170 was demonstrated by testing the prevalence and other clinical cohort specimens on MODULAR Analytics E170 and Elecsys 2010 analyzers. 100 non-reactive specimens were obtained from the Prevalence cohort. 53 reactive specimens were collected in the US from various clinical cohorts and another 49 reactive samples were provided by Roche R & D from sample archive. Among those samples, 96 were low negatives; 6 were high negatives; 12 were low reactive and 88 were high reactive samples. The regression analyses for the comparisons using the prevalence cohort and the combined clinical cohorts were carried out using the least squares and Passing-Bablok regression methods. The Pearson's regression correlation for the 202 data pairs was 0.9951, with slope at 1.024 and intercept at -0.555. Positive percent agreement was 91/94 = 96.8 % (95 % exact confidence limits of 91.0 to 99.3). Negative percent agreement was 98/100 = 98.0 % (95 % exact confidence limits of 93.0 to 99.8). 8 specimens were concordantly equivocal.

4. Clinical cut-off**:**

Refer to assay cutoff section above for additional details.

5. Expected values/Reference range:

The Elecsys anti-HAV assay was used to evaluate the prevalence of HAV antibodies in an apparently healthy population (normal, healthy Individuals without symptoms). A statistically significant number of subjects were prospectively collected in a "high prevalence" region, the Western states, and a "low prevalence" region, the Eastern states, and were tested using only the Elecsys anti-HAV assay and were not compared with the predicate device.

Expected Results from low and high Prevalence based on the test device are presented under the prevalence study section. Prevalence rate for reactive anti-HAV total antibody in specimens collected in a high prevalence region, Western states of the US (New Mexico), was 37.33 %. Prevalence rate for reactive anti-HAV total antibody in specimens collected in a low prevalence region, Eastern states of the US (Indiana), was 35.10 %.

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.