SUMMARY OF SAFETY AND EFFECTIVENESS

1. General Information

Device Generic Name:	Elecsys HIV Duo
Device Trade Name:	Elecsys HIV Duo PreciControl HIV Gen II PreciControl HIV; HIV-2 + Grp O
Device Product Code:	MZF
Applicant Name and Address:	Roche Diagnostics 9115 Hague Road Indianapolis, IN 46250 Phone: 317 521 7144 Fax: 317 521 2334
Establishment Registration Number:	1823260
Premarket Approval Application (PMA) Number:	BP190403
Date of Panel Recommendation:	Not Applicable

- □ **I concur with the summary review.**
- □ **I concur with the summary review and include a separate review to add further analysis.**
- □ **I do not concur with the summary review and include a separate review.**

Office's Signatory Authority :	Nicole Verdun, M.D. Director, OBRR/CBER
Date of FDA Notice of Approval :	April 10, 2020

Material Reviewed/Consulted: The PMA, amendments to the PMA, and other specific documentation used in developing the Summary of Safety and Effectiveness (SSE)

Review memos from the following reviewers were used in developing the SSE:

Discipline	Reviewer Names
Product Design	Viswanath Ragupathy
Chemistry / Manufacturing / Controls (CMC)	Viswanath Ragupathy Xue Wang Mohan Kumar HG
Preclinical and Clinical Studies	Viswanath Ragupathy Julia Lathrop Mohan Kumar HG
DMPQ/pre-approval inspection	Lori Peters
Bioresearch Monitoring Inspection (BIMO)	Anthony Hawkins
Statistician	Linye Song
Instrumentation and Software	Lisa Simone Babita Mahajan
Product and Promotional Labeling (OCBQ/DCM/APLB)	Dana Jones Viswanath Ragupathy

2. Intended Use

The Elecsys HIV Duo is an immunoassay intended for the in vitro simultaneous qualitative detection and differentiation of HIV-1 p24 antigen and antibodies to HIV, HIV-1 (groups M and O) and HIV-2, in human serum and plasma. The Elecsys HIV Duo assay is intended to be used as an aid in the diagnosis of HIV-1 and/or HIV-2 infection, including acute or primary HIV-1 infection. The assay may also be used as an aid in the diagnosis of HIV-1/HIV-2 infection in subjects greater than 2 years of age and in pregnant women. Elecsys HIV Duo is not intended for the screening of blood and plasma donors.

Elecsys HIV Duo is an electrochemiluminescence immunoassay “ECLIA” intended for use on the cobas e 801 immunoassay analyzer.

3. Description: Elecsys HIV Duo

3.1 Device Description

The Elecsys HIV Duo is a qualitative serologic sandwich immunoassay intended for the detection and differentiation of HIV-1 p24 antigen and antibodies to HIV-1, HIV-1(group M, and group O) and HIV-2, in human serum and plasma. This assay design consists of two modules, one for detection of HIV-1 p24 antigen using monoclonal antibodies to (HIV-1) p24 and the second for detection of HIV-1 and HIV-2 antibodies using recombinant antigens derived from the Env and Pol-region of HIV-1 (including group O) and HIV-2. The immunoassay is based on the electrochemiluminescence immunoassay (ECLIA) principle.

In the first incubation step for HIV Ag detection (HIVAG), biotinylated monoclonal anti-p24 antibodies and ruthenylated monoclonal anti-p24 antibodies are added to 30µL of specimen, to form a sandwich complex. For anti-HIV detection (AHIV), biotinylated HIV-specific recombinant antigens/peptides and ruthenylated HIV-specific recombinant antigens/peptides are added to a separate 30µL specimen to form a sandwich complex. The incubations are performed in parallel in two separate vessels.

In the second incubation step, the sandwich complex is captured by streptavidin-coated microparticles and the complex becomes bound to the solid phase via interaction of biotin with streptavidin. This reaction mixture is aspirated into the measuring cell of the analyzer where the micro particles are magnetically captured onto the surface of an electrode. Residual unbound substances are removed with ProCell II M. Application of a voltage to the electrode induces electrochemiluminescent emission which is measured by a photomultiplier. The level of signal increases as the concentration of the anti-HIV antibodies and antigens present in a patient specimen increases.

The Elecsys HIV Duo immunoassay requires the use of quality control reagents, the Elecsys PreciControl HIV Gen II and the Elecsys PreciControl HIV; HIV-2+ Grp O. Results are determined automatically by the software by comparing the electrochemiluminescence signal obtained from the specimen with the cutoff value obtained by HIVAG and AHIV embedded calibration. The Elecsys HIV Duo result is calculated automatically based on signal to cutoff ratios (cutoff index, COI) from HIV Ag and anti-HIV.

3.2 cobas e 801 Immunoassay Analyzer

The Elecsys Duo is intended for use with the cobas e 801 Immunoassay analyzer instrument, a fully automated self-contained, immunoassay. The cobas e 801 Immunoassay Analyzer Instrument incorporates a dedicated software package for instrument control, data collection, results analysis, calibration, quality control, and service software.

3.3 Kit Configurations and Components

3.3.1 Reagent Components

The Elecsys HIV Duo immunoassay is available in a 300-test kit size which contains two modules, one module for HIV-1 p24 antigen detection and the second module for detection of the HIV-1 and HIV-2 antibodies. Each module contains three reagent components (M, R1 and R2) supplied by Roche Diagnostics as a bundled reagent pack (cobas e pack), which is placed in the instrument, as a single unit while operational. The calibrators for both the antigen (Cal1 and Cal2) and antibody (Cal3 and Cal4) are provided in lyophilized form and need to be reconstituted before use. All reagent components are labelled with barcodes and radio frequency identification (RFID) chips, that can be read by the analyzer. Tables 1A and 1B show the components for each module supplied by Roche Diagnostics in a single package.

Table 1A. Elecsys HIV Duo Module HIVAG

Name	Description
M	Streptavidin-coated microparticles
R1	Biotinylated monoclonal anti-HIV p24 antibodies (mouse)
R2	Monoclonal anti-HIV p24 antibodies (mouse) labeled with ruthenium complex
HIVDUO Cal1	Negative calibrator 1
HIVDUO Cal2	Positive calibrator 2 (HIV p24 antigen)

Table 1B. Elecsys HIV Duo Module AHIV

Name	Description
M	Streptavidin-coated microparticles
R1	Biotinylated HIV-1/2-specific recombinant antigens (E. coli) Biotinylated HIV-1/2 specific synthetic peptides
R2	HIV-1/2 specific recombinant antigens (E. coli) HIV-1/2 specific synthetic peptides labeled with ruthenium complex
HIVDUO Cal3	Negative calibrator 3
HIVDUO Cal4	Positive calibrator 4 (anti-HIV-1)

3.3.2. PreciControl Components

The Elecsys PreciControl HIV Gen II and Elecsys PreciControl HIV; HIV-2 + Grp O are used for quality control of the Elecsys HIV Duo immunoassay on the cobas e 801 immunoassay analyzer.

The control kit consists of two separate packages containing five lyophilized reagents in total. PreciControl HIV1 to 3 in one package that is labelled as Elecsys PreciControl HIV Gen II, and PreciControl HIV4 and 5 in a separate package labelled as PreciControl HIV; HIV- 2 + Grp O. Control kit components are described in Table 2.

Table 2. PreciControl HIV Gen II and PreciControl HIV; HIV-2 + Grp O Components

Control Kits	Components	Description
PreciControl HIV Gen II	PC HIV1	PreciControl HIV1 contains human serum, negative for HIV (antigen and antibody)
	PC HIV2	PreciControl HIV2 contains inactivated positive human serum for anti-HIV-1 antibodies
	PC HIV3	PreciControl HIV3 contains HIV-1 p24 antigen in human serum
PreciControl HIV; HIV-2 + Grp O	PC HIV4	PreciControl HIV4 contains inactivated positive human serum for anti-HIV-2 antibodies
	PC HIV5	PreciControl HIV5 contains anti-HIV-1 subtype Grp O monoclonal mouse antibodies in human serum

3.3.3. System Reagents for cobas e 801 Analyzer

For processing any Elecsys Immunoassay on the cobas e 801 analyzer, specific system reagents are required, which are packaged and sold separately. The required system reagents are common for use with all marketed Elecsys and cobas e immunoassays.

Table 3. System Reagents Required for Elecsys Immunoassays on the cobas e 801 Analyzer

Catalog Number	Component/ Reagent	Component Volume	Quantity	Type of Material
06908799	ProCell II M	2 L	2	System solution
04880293	Clean Cell M	2 L	2	Measuring cell cleaning solution
11298500	ISE Cleaning Solution/Elecsys SysClean	100 mL	5	System cleaning solution
06908853	PreClean II M	2 L	2	Wash solution

3.3.4. Materials Provided

- 4 bottle labels
- 4 empty labeled snap-cap bottles

3.3.5. Materials required (but not provided)

- PreciControl HIV Gen II, for 6 x 2.0 mL
- PreciControl HIV; HIV 2 + GrpO, for 4 x 2.0 mL
- CalSet Vials, 2 x 56 empty bottles with snap caps
- General laboratory equipment
- cobas e 801 analyzer
- Distilled or deionized water

3.3.6. Accessories for cobas e 801 analyzer:

- ProCell II M, 2 x 2 L system solution
- CleanCell M, 2 x 2 L measuring cell cleaning solution
- Reservoir Cups, 8 cups to supply ProCell II M and CleanCell M
- PreClean II M, 2 x 2 L wash solution
- Assay Tip/Assay Cup tray, 6 magazines x 6 magazine stacks x 105 assay tips and 105 assay cups, 3 waste liners
- Liquid Flow Cleaning Cup, 2 adaptor cups to supply ISE Cleaning Solution/Elecsys SysClean for Liquid Flow Cleaning Detection Unit
- Prewash Liquid Flow Cleaning Cup, 1 adaptor cup to supply ISE Cleaning Solution/Elecsys SysClean for Liquid Flow Cleaning Prewash Unit
- ISE Cleaning Solution/Elecsys SysClean, 5 x 100 mL system cleaning solution

4.1. Test Procedure

4.2. Specimen Collection, Preparation, and Storage

4.1.1. Collecting Specimens

The Elecsys HIV Duo assay can be performed on serum and plasma specimens collected by venipuncture following the instructions provided with the specimen collection device for use and processing. Specimens are processed by centrifugation following standard procedure for separation of serum from the clot. Complete clot formation should take place before centrifugation. The centrifugation step may occur up to (b) (4) post draw. Specimens with obvious microbial contamination should not be used.

The specimen types listed below are acceptable for testing with Elecsys HIV Duo.

- Serum collected using standard sampling tubes or tubes containing separating gel.
- Plasma collected using Li-heparin, K2-EDTA and K3-EDTA plasma tubes as well as Li-heparin plasma tubes containing separating gel.

4.1.2. Storing Specimens

Specimens collected are stable for 7 days at 20-25 °C, 4 weeks at 2-8 °C, 3 months at -20 °C (± 5 °C). Specimens may be frozen and thawed up to 5 times. Specimen collection systems from various manufacturers may contain differing materials which could affect test results in some cases. When processing specimens in primary tubes (specimen collection systems), instructions of the tube manufacturer should be followed. Specimens containing precipitates and frozen/thawed specimens should be centrifuged before performing the assay.

4.2.Calibration

Calibration must be performed once per reagent lot using HIVDUO Cal1, HIVDUO Cal2, HIVDUO Cal3, HIVDUO Cal4 and fresh reagent (i.e. not more than 24 hours since the cobas e pack was registered on the analyzer). Reconstituted calibrators are for single use and should be discarded after use.

4.2.1.Re-calibration is recommended as follows:

- After 12 weeks when using the same reagent lot
- After 28 days when using the same cobas e pack on the analyzer
- As required: e.g. quality control findings with PreciControl HIV Gen II and PreciControl HIV; HIV-2 + Grp O outside the defined limits
- More frequently when this is required by appropriate regulations

4.2.2. Range for the electrochemiluminescence signals (counts) for the calibrators

HIVAG module:

- HIVAG negative calibrator (HIVDUO Cal1): (b) (4)
- HIVAG positive calibrator (HIVDUO Cal2): (b) (4)

AHIV module:

- AHIV negative calibrator (HIVDUO Cal3): (b) (4)
- AHIV positive calibrator (HIVDUO Cal4): (b) (4)

4.3.Quality Control

PreciControl HIV Gen II and PreciControl HIV; HIV-2 + Grp O are used for quality control of the Elecsys Duo assay. All controls should be run individually at least once every 24 hours when the test is performed, once per reagent kit lot, and following each calibration. In addition, the control intervals and limits may be further adapted to each laboratory's individual requirements. Values obtained should fall within the defined limits. Each laboratory should establish corrective measures to be taken if values fall outside defined limits. Follow the applicable government regulations and local guidelines for quality control.

5. Interpretation of Results

Results are determined automatically by the software by comparing the electrochemiluminescence signal obtained from the reaction product of the specimen with the signal of the cutoff value previously obtained by calibration. With the Elecsys HIV Duo assay, HIV-1 p24 antigen (HIVAG), as well as antibodies to HIV-1 and HIV-2 (AHIV) can be detected in parallel as two separate determinations. On the basis of these determinations, the Elecsys HIV Duo main result is subsequently calculated automatically by the analyzer.

The result of a specimen is given either as reactive or non-reactive as well as in the form of a cutoff-index (specimen signal/ cutoff).

Specimens with a HIV DUO cutoff index < 1.00 are non-reactive and these specimens are

considered negative for HIV-1 Ag and HIV-1/-2 specific antibodies and do not need further testing. Specimens with a HIV DUO cutoff index ≥ 1.00 are considered initially reactive. All initially reactive specimens are automatically retested in duplicate. The specimen is considered repeatedly reactive if one or both retests have a cutoff-index (COI) ≥ 1.00. In a rare specimen with a high non-reactive COI (< 1.00) of HIVAG and AHIV, the Elecsys Duo combined main result is reported as reactive with a flag that sub-results of HIVAG and AHIV are non-reactive. A repeatedly reactive results should be confirmed with FDA approved supplemental tests following the recommended HIV testing algorithm.

6. Limitations of the Test

- The Elecsys HIV Duo assay is for in vitro diagnostic use only.
- This assay is not for screening blood or plasma donors.
- Elecsys HIV Duo is limited to the detection of HIV-1 p24 antigen and antibodies to HIV, HIV-1 (Groups M and O) and HIV-2 in human serum and plasma.
- The performance of the Elecsys HIV Duo assay has not been established with cord blood, neonatal specimens, cadaver specimens, heat-inactivated specimens, or body fluids other than serum and plasma such as saliva, urine, and amniotic or pleural fluids
- Heat-inactivated specimens and specimens stabilized with azide should not be used. Specimens should be at 20 - 25 °C prior to use with the assay.
- Due to possible evaporation effects, specimens placed on the analyzers should be analyzed or measured within 2 hours.
- The reported antigen and/or antibody level cannot be correlated to an endpoint titer. The calculated values for anti-HIV and/or p24 antigen in a given specimen as determined by assays from different manufacturers can vary due to differences in assay methods and reagent specificity. Results reported by the laboratory to the physician must include the identity of the assay used. Values obtained with different assay methods cannot be used interchangeably.
- Heterophilic and Human Antibodies to Mouse antigens (HAMA) in human specimens can react with reagent antibodies, interfering with in vitro immunoassays. Patients routinely exposed to animals or animal serum products for diagnosis or therapies can be prone to this interference and anomalous values may be observed. Specimens from patients who have received mouse monoclonal antibodies for diagnosis or therapy may contain human anti-mouse antibodies and may interfere in assays that employ mouse monoclonal antibodies. Additional information may be required for diagnosis.
- A negative test result does not exclude the possibility of exposure to or infection with HIV. HIV antibodies and/or p24 antigen may be undetectable in some stages of the infection and in some clinical conditions.
- A person who has antigen or antibodies to HIV is presumed to be infected with the virus. However, a person who has participated in an HIV vaccine study may develop antibodies to the vaccine and may or may not be infected with HIV.
- The claims, including those pertaining to specimen stability made in the labeling of the cleared/approved reagents of Roche Diagnostics are part of the clearance of the overall IVD test system (assay). Specimen stability was tested only for the temperatures/time frame as claimed by the manufacturer under the conditions claimed in the method sheet. It is the responsibility of the individual laboratory to

use all available references and/or its own studies to determine specific stability criteria for its laboratory.

- This device uses biotin technology for detection of HIV and specimens with elevated levels of biotin may interfere with this assay. The biotin scavenger antibody protects biotin interference and the device is validated to tolerate biotin up to 1200 ng/mL.

7. (b) (4)

(b) (4)) was performed according to (b) (4) for the Elecsys HIV Duo reagents R1, R2 and the controls PC HIV1, PC HIV2, PC HIV3, PC HIV4, and PC HIV5. The calibrators were not tested because they are identical to the controls concerning the ingredients and their concentrations. The Elecsys HIV Duo beads and the PreciControls were not tested because both reagents were identical to previously approved Elecsys Anti HCV PMA((P090007, P090008, and P090009) and the Elecsys HIV combi PT(BP160050) (b) (4)

8. Marketing History

The Elecsys HIV Duo is currently marketed globally in 49 countries since April 2017. The device has not been withdrawn to date from the market in any country for reasons relating to safety and effectiveness of the device.

9. Potential Adverse Effects of the Device on Health

Potential adverse effects of the Elecsys HIV Duo assay relate to the risk of false positive and false negative results. While performance studies indicate that this risk is likely to be very small, the potential for inaccurate results does exist. The risk of an incorrect result is minimized by following the procedures and instructions outlined in the package insert.

10. Summary of Preclinical Studies

10.1. Determination of Cutoff

Studies were performed to establish and verify the cutoff by defining variables of the cutoff formula for the Elecsys HIV Duo. The establishment of the correct cut-off value depends on a balance between sensitivity and specificity. Specificity was evaluated by testing 13,328 negative specimens. Sensitivity was evaluated using WHO standard (traceable to NIBSC90/636) and seroconversion panels. The Elecsys HIV Duo assay sensitivity and specificity was comparable to previously approved combo HIV assays.

Summary: The assay cut-off index was established and set as 1.00. A signal generated from positive or negative specimens and use of pre-established cut-off generates signal to cut-off ratio or cut-off index (COI). Additionally, instrument background signal was calculated and adjusted with the final COI. The assay output result is a qualitative measurement of either reactive or non-reactive. For reactive specimens the COI should be ≥1 and for non-reactive specimens the COI should be ≤1.

- The cutoff sensitivity for the WHO p24 standard (NIBSC code 90/636) is ≤ 1.0 IU/mL and sensitivity of seroconversion panel testing is comparable to approved combo assays.
- The specificity of the Elecsys HIV Duo is comparable to approved combo assays.

This study demonstrated an acceptable level of performance.

10.2. Determination of Limit of Blank and Limit of Detection

The purpose of this study is to determine the capability of the test to differentiate between background signal and signal levels significantly elevated by the presence of analyte. The Limit of Blank (LoB) and Limit of Detection (LoD) for the Elecsys HIV Duo assay were determined in accordance with (b) (4) . LoB is defined as the concentration at which there is a (b) (4) . LoD is defined as the concentration at which there is a (b) (4)

For determination of LoB, (b) (4) cobas e 801 analyzer. A total of (b) (4) measured values for (b) (4) specimens were obtained. Data analysis was based on determination of the (b) (4) measured values. For determination of the LoD, (b) (4) serum specimens with (b) (4) concentration (approximately (b) (4) LoB) were measured using (b) (4) cobas e 801 analyzer. In total, (b) (4) measured values for specimens with (b) (4) concentration were obtained.

Both serum and plasma specimen matrices were evaluated.

Summary :

- LoB was determined as (b) (4)s/co and (b) (4) s/co for serum and (b) (4) s/co and (b) (4) s/co for plasma.

- LoD was determined as (b) (4) s/co and (b) (4) s/co for serum and (b) (4) s/co and (b) (4) s/co for plasma.

10.3 HIV-1 p24 Ag Analytical Sensitivity

Analytical sensitivity of the Elecsys HIV Duo assay for detection of HIV-1 p24 antigen was evaluated using the first International Standard HIV-1 p24 Antigen, NIBSC code 90/636. HIV-1 p24 antigen was diluted with HIV negative serum. Seven dilutions of each standard were prepared and measured. (b) (4) were evaluated to determine Elecsys HIV Duo antigen sensitivity in pg/mL. Measurements on the cobas e 801 analyzer were performed with three lots of the Elecsys HIV Duo assay. Analytical sensitivity was calculated from the S/CO and corresponding antigen standard dilutions tested.

Summary:

The mean results from three kit lots demonstrated antigen sensitivity of 0.392 IU/mL using the NIBSC standard, (b) (4)

10.4. HIV Antigen reactivity (HIV-1 M Subtypes, HIV-1 Group O, and HIV-2)

The purpose of this study was to evaluate reactivity of the Elecsys HIV Duo with 55 HIV antigen lysates from cell culture supernatants of HIV-1 M subtype A, B, C, D, E, F, G, H, CRF01 AE, HIV-1 Group O, unknown HIV-1 subtypes and HIV-2 (Table 4). To determine reactivity, all 55 viral antigen lysates were tested in (b) (4) with the Elecsys HIV Duo.

Table 4. Detection of HIV-1 and HIV-2 Antigens in Culture Supernatants

Subtype	Number of Specimens tested	Elecsys HIV Duo	FDA-approved reference assay
A	4	4/4	4/4
B	6	6/6	6/6
C	4	4/4	4/4
D	5	5/5	5/5
E	4	4/4	4/4
F	1	1/1	1/1
G	2	2/2	2/2
H	2	2/2	2/2
CRF01_AE	1	1/1	1/1
HIV-1 unknown subtype	8	8/8	8/8
HIV-2	6	6/6	6/6
HIV-1 Group O	12	12/12	12/12

Summary: All 55 viral lysates were reactive on the Elecsys HIV Duo assay as shown in the Table 4. The assay demonstrates acceptable performance for the detection of HIV-1 and HIV-2 antigens.

10.5. Seroconversion Panels

Seroconversion sensitivity of the Elecsys HIV Duo was shown by testing 50 commercially-sourced HIV-1 seroconversion panels (Table 5) with a total of 452 specimens and comparing Elecsys HIV Duo results to an FDA approved assay.

Table 5. Elecsys HIV Duo Assay Reactivity in Seroconversion Panels

Specimen ID	Panel/Donor	Difference in days	Elecsys HIV Duo First Reactive Result	FDA approved Reference Assay First Reactive Result
Panel 1	HIV 6243 / 62238	0	Day 25	Day 25
Panel 2	HIV 6247 / 63602	+2	Day 21	Day 23
Panel 3	HIV 6248 / 63331	0	Day 18	Day 18
Panel 4	HIV 9011 / 64954	0	Day 36	Day 36
Panel 5	HIV 9012 / 65389	+2	Day 14	Day 16
Panel 6	HIV 9013 / 65404	+2	Day 23	Day 25
Panel 7	HIV 9014 / 65522	0	Day 10	Day 10
Panel 8	HIV 9016 / 65790	0	Day 30	Day 30
Panel 9	HIV 9017 / 65907	-3	Day 24	Day 21
Panel 10	HIV 9018 / 66575	+3	Day 25	Day 28
Panel 11	HIV 9019 / 65685	0	Day 38	Day 38
Panel 12	HIV 9021 / 67485	0	Day 47	Day 47
Panel 13	HIV 9022 / 64578	0	Day 23	Day 25*
Panel 14	HIV 9023 / 67706	0	Day 78	Day 78
Panel 15	HIV 9024 / 64469	0	Day 53	Day 53
Panel 16	HIV 9025 / 67996	0	Day 85	Day 85
Panel 17	HIV 9026 / 68205	0	Day 44	Day 44
Panel 18	HIV 9027 / 65633	0	Day 14	Day 14
Panel 19	HIV 9028 / 67860	0	Day 53	Day 53
Panel 20	HIV 9029 / 68262	0	Day 45	Day 45
Panel 21	HIV 9030 / 68582	0	Day 47	Day 47
Panel 22	HIV 9032 / 68106	+2	Day 22	Day 24
Panel 23	HIV 9033 / 66686	0	Day 82	Day 82
Panel 24	HIV 9034 / 66632	0	Day 46	Day 46
Panel 25	HIV 9075 / 62216	0	Day 22	Day 22
Panel 26	HIV 9076 / 63753	0	Day 66	Day 66
Panel 27	HIV 9079 / 75062	0	Day 40	Day 40

Specimen ID	Panel/Donor	Difference in days	Elecsys HIV Duo First Reactive Result	FDA approved Reference Assay First Reactive Result
Panel 28	HIV 9081 / 63215	0	Day 24	Day 24
Panel 29	HIV 9089 / 65376	0	Day 16	Day 16
Panel 30	HIV 12007 / 73695	0	Day 117	Day 117
Panel 31	PRB 939-AN	0	Day 16	Day 16
Panel 32	PRB944-AT	+5	Day 2	Day 7
Panel 33	PRB960	0	Day 28	Day 28
Panel 34	PRB961	0	Day 27	Day 27
Panel 35	PRB962	0	Day 14	Day 14
Panel 36	PRB963	0	Day 17	Day 17
Panel 37	PRB964	0	Day 22	Day 22
Panel 38	PRB965	0	Day 5	Day 5
Panel 39	PRB966	0	Day 44	Day 44
Panel 40	PRB967	0	Day 17	Day 17
Panel 41	PRB968	0	Day 26	Day 26
Panel 42	PRB969	0	Day 63	Day 63
Panel 43	PRB971	0	Day 7	Day 7
Panel 44	PRB972	0	Day 18	Day 18
Panel 45	PRB973	0	Day 7	Day 7
Panel 46	PRB974	0	Day 9	Day 9
Panel 47	PRB975	0	Day 14	Day 14
Panel 48	PRB976	+5	Day 2	Day 7
Panel 49	PRB977	0	Day 13	Day 13
Panel 50	PRB978	0	Day 33	Day 33

**QNS on day 23, day 25 reactive for reference method 1*

Summary: Seroconversion testing resulted in positive detection with the Elecsys HIV Duo one bleed earlier in seven panels and one bleed later in one panel compared with the FDA-approved HIV Ag/Ab assay. Equivalent performance was observed in 444 of 452 bleeds tested.

10.6. Hook Effect

The Elecsys HIV Duo was evaluated for high titer hook effects in specimens containing extremely high HIV titers. (b) (4) high titer positive specimens (anti-HIV-1 (COI (b) (4) anti-HIV-2 (COI (b) (4) anti-HIV-1 Group O (COI (b) (4))and HIV-1 p24 antigen (COI (b) (4) were each diluted in HIV negative serum in (b)(4) dilution steps to generate a dilution series that covers the range from negative to high positive s/co values and were measured in (b) (4)

Summary: Results obtained indicated that the Elecsys HIV Duo did not exhibit a high titer

hook effect up to the levels tested. The results demonstrate acceptable performance.

10.7. Effect of Potentially Interfering Substances

To assess the impact of potentially interfering substances on the performance of the Elecsys HIV Duo, serum and plasma from five specimen types that included HIV negative, HIV-1 Group M, HIV-1 Group O, and HIV-2 antibodies and HIV p24 were evaluated. Serum from each specimen type was prepared as unspiked or spiked with interferants (neat and two-fold dilutions). Plasma from each specimen type was prepared as unspiked or spiked with interferants (neat and tenfold serial dilutions). All prepared specimens were tested in (b) (4) Although biotin interference was evaluated up to (b) (4), maximum biotin interference tolerance was demonstrated at 1200ng/mL. Percent mean recovery of s/co values of specimens spiked with interfering substance were calculated against the respective specimens without the interfering substance.

Table 6. Concentration of Interfering Substances

Substance	Concentration tested
Bilirubin	≤ 1129 µmol/L or ≤ 66 mg/dL
Hemoglobin	≤ 0.311 mmol/L or ≤ 500 mg/dL
Intralipid (Lipemia)	≤ 2000 mg/dL
Biotin	≤ 4912 nmol/L or ≤ 1200 ng/mL
Human Serum Albumin	≤ 7.0 g/dL

Summary: None of the potentially interfering substances at the levels tested produced a change in the interpretation of the assay. The results demonstrate acceptable performance at the tested level listed in the above table.

10.8. Interference by Unrelated Medical Conditions

To assess potential interference by unrelated medical conditions, specimens containing potentially interfering factors were tested using the Elecsys HIV Duo. Specimens were tested neat (unspiked) and spiked at levels near COI of anti-HIV-1, anti-HIV-2 and p24 antigen, respectively. Measurements were performed with a single determination using one lot of the Elecsys HIV Duo. The results are presented in Table 7, below:

Table 7. Effect of Potentially Interfering Medical Conditions on sensitivity of Elecsys HIV Duo

Clinical Category	Number Tested	Effect on Elecsys HIV Duo results
ANA	10	0/10
Candida	10	0/10

Clinical Category	Number Tested	Effect on Elecsys HIV Duo results
CMV	10	0/10
EBV	10	0/10
E.coli	10	0/10
Influenza vaccination	10	0/10
HAV	10	0/10
HAVHBV vaccination	8	0/8
HBV	10	0/10
HCV	10	0/10
HSV	10	0/10
HTLV	10	0/10
Lymphoma	10	0/10
Malaria	10	0/10
Monoclonal Gammopathy	10	0/10
Pregnancy 1st Trimester	30	0/30
Pregnancy 2nd Trimester	30	0/30
Pregnancy 3rd Trimester	30	0/30
Rheumatoid Factor	10	0/10
Rubella	10	0/10
Syphilis	10	0/10
Tuberculosis	10	0/10
Rotavirus	10	0/10
Chlamydia	10	0/10
Common Cold	10	0/10
Small Pox	10	0/10
VZV	9	0/9
Multiparous Pregnancies	10	0/10
Graves' Disease	9	0/9
Crohn's Disease	10	0/10

Summary: The presence of potentially interfering substances or medical conditions had no effect on the detection of HIV-1, HIV-2 antibodies, or HIV-1 antigen. No significant effect on background signals in negative specimens (neat specimens). These results demonstrate acceptable performance.

10.9. Drug Interference

To assess potential interference by therapeutic drugs according to CLSI EP07-A2, sixteen common therapeutic drugs were tested. Each drug was separately spiked into a negative, an anti-HIV antibody positive (s/co 1.5 - 4) and an HIV-Ag positive specimen (s/co 1.5 - 4) at a concentration 1-10 times the maximum daily dosage. Testing was done in (b) (4) with comparison of spiked specimen against unspiked serum (reference), by calculating the mean s/co values, absolute deviations and percentage recovery to reference. The concentration of drugs used for testing is shown in Table 8 below:

Table 8. Concentration of Drugs

No.	Compound	Concentration (mg/L)
1	Acetylcysteine	553
2	Ampicillin-Na	1000
3	Ascorbic acid	300
4	Cyclosporine	5
5	Cefoxitin	2500
6	Heparin	5000 U/L
7	Levodopa	20
8	Methyldopa	20
9	Metronidazole	200
10	Phenylbutazone	400
11	Doxycycline	50
12	Acetylsalicylic acid	1000
13	Rifampicin	60
14	Acetaminophen	200
15	Ibuprofen	500
16	Theophylline	100

Summary: Each drug was found to be non-interfering at the concentrations tested. The results demonstrate acceptable performance.

10.10. Serum/Plasma Comparison

The purpose of this study was to examine the suitability of the following types of blood collection tubes for Elecsys HIV Duo assay.

- Serum gel separation tubes (SST)
- Lithium heparin plasma
- Lithium heparin plasma separation tubes (PST)
- Sodium (Na) heparin plasma
- Sodium (Na) citrate plasma
- K2-EDTA plasma

- K3-EDTA plasma
- Acid citrate dextrose (ACD) plasma
- Citrate phosphate dextrose (CPD) plasma
- Potassium oxalate (Oxalate) plasma
- Citrate phosphate dextrose adenine (CPDA) plasma
- Citrate phosphate double dextrose (CP2D) plasma

Additionally, the suitability of serum and plasma from Li-Heparin primary specimen tubes with separating gel was tested by comparing them to Li-Heparin tubes without separating gel. The impact of anticoagulants on the performance of the Elecsys DUO assay was evaluated using matched sets of serum and plasma specimens of all specified types, HIV-1 negative (n=50) or spiked with HIV-1 antigen (n=40), HIV-1 antibody (n=40), and HIV-2 antibody (n=10).

Summary: The results indicate that all HIV-1 negative specimens were non-reactive, and all spiked specimens were reactive in all anticoagulants and tube types. The average recovery for negative specimens were ± 0.2 s/co and spiked HIV positive specimens was 80 - 120 % (s/co) in various anticoagulants and tube types. The average percent recoveries were within acceptable ranges for all anticoagulants tested. The results demonstrate acceptable performance.

10.11. Precision and Reproducibility studies

A Reproducibility study was conducted at three external sites on the cobas e 801 analyzer using three reagent lots and one lot each of the PreciControl HIV Gen II and PreciControl HIV; HIV-2+Group O. For precision testing, eight spiked human serum pools (HSP) and all five levels of the PreciControl material were tested according to CLSI EP05-A3. Specimen and PreciControl pools were evaluated using the experimental de sign consisting of [redacted] aliquots per specimen per run using individual measurement with [redacted]

The following is a description of the specimens used in the evaluation. PC HIV-1 negative for HIV (antigen and antibody), PC HIV2 - positive for anti-HIV-1 antibodies, PC HIV3 - positive for HIV-1 p24 antigen, PC HIV4 - positive for HIV-2 antibodies, PC HIV5 - positive for HIV-1 (Group O) antibodies. HSP 06 - HIV-1 antibody group M low positive sera, HSP 07 - HIV-1 antibody group M high positive sera, HSP 08 - HIV-1 antigen low positive sera, HSP 09 - HIV-1 antigen high positive sera, HSP 10 - HIV-2 antibody low positive sera, HSP 11- HIV-2 antibody high positive sera, HSP 12 - HIV-1 Group O antibody positive sera and HSP 13 - HIV antibody and antigen negative sera.

The summary of precision and reproducibility studies for Elecsys Duo Main result and sub-Results Antigen and Antibody are presented in tables 9A, 9B and 9C.

Table 9A. Overall Repeatability and Reproducibility for Elecsys HIV Duo

			Repeatability		Inter-Run		Inter-Day		Inter-Site*Lot		Inter-Lot		Inter-Site		Reproducibility	
Specimen	Mean	N	SD	CV [%]	SD	CV [%]	SD	CV [%]	SD	CV [%]	SD	CV [%]	SD	CV [%]	SD	CV [%]
HSP 06	3.00	270	0.046	1.5	0.033	1.1	0.030	1.0	0.053	1.8	0.063	2.1	0.029	1.0	0.108	3.6
HSP 07	63.9	270	0.921	1.4	0.555	0.9	0.591	0.9	1.16	1.8	1.56	2.4	1.26	2.0	2.62	4.1

			Repeatability		Inter-Run		Inter-Day		Inter-Site*Lot		Inter-Lot		Inter-Site		Reproducibility	
HSP 08	3.40	270	0.075	2.2	0.036	1.1	0.030	0.9	0.026	0.8	0.043	1.3	0.077	2.3	0.127	3.7
HSP 09	57.8	270	0.852	1.5	0.856	1.5	0.775	1.3	0.334	0.6	0.995	1.7	0.487	0.8	1.84	3.2
HSP 10	4.42	270	0.074	1.7	0.076	1.7	0.050	1.1	0.068	1.5	0.066	1.5	0.000	0.0	0.151	3.4
HSP 11	50.2	270	0.579	1.2	0.668	1.3	0.522	1.0	0.794	1.6	0.000	0.0	0.462	0.9	1.38	2.7
HSP 12	55.6	270	1.00	1.8	0.816	1.5	0.479	0.9	1.01	1.8	0.882	1.6	0.000	0.0	1.93	3.5
HSP 13	0.165	270	0.008	4.7	0.003	2.0	0.004	2.6	0.001	0.6	0.011	6.6	0.000	0.0	0.014	8.8
PC HIV1	0.185	270	0.008	4.3	0.003	1.7	0.005	2.7	0.004	2.1	0.020	10.8	0.000	0.0	0.023	12.2
PC HIV2	2.98	270	0.041	1.4	0.026	0.9	0.026	0.9	0.059	2.0	0.142	4.8	0.029	1.0	0.166	5.6
PC HIV3	7.83	270	0.089	1.1	0.061	0.8	0.073	0.9	0.069	0.9	0.155	2.0	0.000	0.0	0.214	2.7
PC HIV4	3.93	270	0.060	1.5	0.059	1.5	0.066	1.7	0.061	1.6	0.000	0.0	0.037	0.9	0.128	3.3
PC HIV5	5.47	270	0.097	1.8	0.077	1.4	0.089	1.6	0.109	2.0	0.045	0.8	0.055	1.0	0.200	3.7

Table 9B. Overall Repeatability and Reproducibility for Elecsys HIV Duo – HIVAG module

			Repeatability		Inter-Run		Inter-Day		Inter-Site*Lot		Inter-Lot		Inter-Site		Reproducibility	
Specimen	**Mean**	**N**	**SD**	**CV [%]**	**SD**	**CV [%]**	**SD**	**CV [%]**	**SD**	**CV [%]**	**SD**	**CV [%]**	**SD**	**CV [%]**	**SD**	**CV [%]**
HSP 06	0.169	270	0.009	5.0	0.005	3.1	0.002	1.4	0.002	0.9	0.017	10.0	0.002	1.2	0.020	11.8
HSP 07	0.127	270	0.008	6.4	0.003	2.5	0.004	3.4	0.000	0.0	0.008	6.1	0.000	0.0	0.013	9.8
HSP 08	3.40	270	0.075	2.2	0.036	1.1	0.030	0.9	0.026	0.8	0.043	1.3	0.077	2.3	0.127	3.7
HSP 09	57.8	270	0.852	1.5	0.856	1.5	0.775	1.3	0.334	0.6	0.995	1.7	0.487	0.8	1.84	3.2
HSP 10	0.162	270	0.008	4.8	0.006	3.5	0.001	0.8	0.002	1.5	0.014	8.9	0.001	0.8	0.018	10.8
HSP 11	0.165	270	0.074	44.9	0.000	0.0	0.000	0.0	0.002	1.4	0.024	14.5	0.000	0.0	0.078	47.2
HSP 12	0.172	270	0.008	4.6	0.004	2.1	0.005	2.7	0.001	0.7	0.014	8.2	0.000	0.0	0.017	10.1
HSP 13	0.147	270	0.008	5.7	0.004	2.7	0.004	2.8	0.000	0.0	0.010	6.7	0.000	0.0	0.014	9.7
PC HIV1	0.171	270	0.008	4.9	0.004	2.3	0.005	3.0	0.003	1.6	0.020	11.7	0.000	0.0	0.023	13.3
PC HIV2	0.171	270	0.008	4.8	0.003	1.9	0.004	2.3	0.000	0.0	0.019	11.3	0.000	0.0	0.022	12.6
PC HIV3	7.83	270	0.089	1.1	0.061	0.8	0.073	0.9	0.069	0.9	0.155	2.0	0.000	0.0	0.214	2.7
PC HIV4	0.171	270	0.009	5.0	0.002	1.3	0.005	3.0	0.000	0.0	0.020	11.7	0.000	0.0	0.022	13.1
PC HIV5	0.173	270	0.027	15.9	0.000	0.0	0.000	0.0	0.002	1.3	0.017	9.8	0.000	0.0	0.032	18.7

Table 9C. Overall Repeatability and Reproducibility for Elecsys HIV Duo – AHIV module

			Repeatability		Inter-Run		Inter-Day		Inter-Site*Lot		Inter-Lot		Inter-Site		Reproducibility	
Specimen	**Mean**	**N**	**SD**	**CV [%]**	**SD**	**CV [%]**	**SD**	**CV [%]**	**SD**	**CV [%]**	**SD**	**CV [%]**	**SD**	**CV [%]**	**SD**	**CV [%]**
HSP 06	3.00	270	0.046	1.5	0.033	1.1	0.030	1.0	0.053	1.8	0.059	2.0	0.029	1.0	0.106	3.5
HSP 07	63.9	270	0.921	1.4	0.555	0.9	0.591	0.9	1.16	1.8	1.56	2.4	1.26	2.0	2.62	4.1
HSP 08	0.088	270	0.003	3.8	0.001	1.6	0.000	0.0	0.004	4.3	0.003	3.0	0.003	2.9	0.006	7.3
HSP 09	0.077	270	0.003	4.3	0.001	2.0	0.000	0.0	0.003	4.2	0.004	4.7	0.003	3.4	0.007	8.6
HSP 10	4.42	270	0.074	1.7	0.076	1.7	0.050	1.1	0.068	1.5	0.066	1.5	0.000	0.0	0.151	3.4
HSP 11	50.2	270	0.579	1.2	0.668	1.3	0.522	1.0	0.794	1.6	0.000	0.0	0.462	0.9	1.38	2.7
HSP 12	55.6	270	1.00	1.8	0.816	1.5	0.479	0.9	1.01	1.8	0.882	1.6	0.000	0.0	1.93	3.5
HSP 13	0.076	270	0.003	4.5	0.001	2.0	0.000	0.0	0.003	4.0	0.004	5.5	0.003	3.5	0.007	9.1
PC HIV1	0.070	270	0.003	4.5	0.002	2.2	0.000	0.0	0.003	4.5	0.004	5.5	0.002	3.1	0.007	9.2
PC HIV2	2.98	270	0.042	1.4	0.027	0.9	0.025	0.9	0.059	2.0	0.138	4.6	0.029	1.0	0.163	5.5
PC HIV3	0.070	270	0.003	4.6	0.001	0.9	0.001	1.2	0.003	4.5	0.004	5.7	0.003	3.7	0.007	9.5
PC HIV4	3.93	270	0.060	1.5	0.059	1.5	0.066	1.7	0.061	1.6	0.000	0.0	0.037	0.9	0.128	3.3
PC HIV5	5.47	270	0.096	1.8	0.077	1.4	0.089	1.6	0.109	2.0	0.045	0.8	0.055	1.0	0.200	3.7

Summary: The precision and reproducibility of the Elecsys HIV Duo main result and sub-results (antigen and antibody modules) and PreciControls panel members CV is within 10% as shown in above tables. The performance was acceptable, demonstrating only minor variability from run to run, day to day or reagent lot to reagent lot.

10.12. Reagent Stability Studies

10.12.1. Elecsys HIV Duo and PreciControl Reagents

- Reagent stability studies were carried out according to the protocol specifications for the Elecsys HIV Duo reagents, PreciControl HIV Gen II and PreciControl HIV-2 + Group O reagents and calibrators. The (b) (4) months real-time stability data for three lots show that the reagents of the Elecsys HIV Duo and PreciControl HIV Gen II and PreciControl HIV-2 + Group O are stable for the claim of 16 months when stored at 2- 8 °C.
- Temperature Stress Stability testing of the Elecsys HIV Duo reagents was performed after temperature stress. Reagent kits were stored for (b) (4) and at (b) (4) (stressed condition). Results showed that the Elecsys HIV Duo recovery for HIV negative specimen deviations was within (b) (4) and for HIV positive specimen deviations was within (b) (4) of the s/co when stressed for (b) (4)
- Reagent Stability studies were performed to determine the time period over which the reagents (cobas e pack) can be stored at 2 - 8°C once opened. Results showed that the Elecsys HIV Duo reagents were stable for (b) (4) weeks at 2 - 8°C after first opening and the PreciControl reagents were stable for three months at -20°C after opening.
- Reagent On-Board Stability studies were performed to determine the time period in which the reagents can be kept on-board the analyzer once opened. Results showed that the Elecsys HIV Duo reagents were stable for 16 weeks on-board the cobas e 801 analyzer at (b) (4).
- Stability studies were performed to assess in use stability of the PreciControls on the cobas e 801 immunoassay analyzer. Results showed that the PreciControls were stable for up to (b) (4) days at 2 – 8°C, for up to (b) (months at -20°C ± 5°C with up to 3x freeze/thaw cycles.

10.13. Calibrator Stability Studies

- Calibrator Stability after First Opening: To determine Stability after Reconstitution at 2 – 8°C, at -20°C (with repeated freeze/thaw cycles) and On-Board Stability, the calibrators were reconstituted and measured unstressed in duplicate and compared to stressed calibrators. Reconstituted calibrators are stable for 3 days at 2 – 8°C and 16 weeks at -20°C with 3 freeze/thaw cycles. Calibrators may be used only once on board the cobas e 801 analyzer.
- On-Board Stability of Open Calibrators: Studies were performed to assess the on-board stability of the calibrators for the cobas e 801 immunoassay analyzer. Results showed that the Elecsys HIV Duo calibrators were stable for (b) (hours on board the cobas e 801 immunoassay analyzer.

11. Clinical Studies

A multisite clinical study was performed to determine the specificity and sensitivity of Elecsys HIV Duo and compared with an FDA approved HIV assay. Confirmation of repeatedly reactive test results followed CDC recommendations to determine a final HIV status for the specimen tested. Confirmation testing included FDA approved assays that differentiate antigen and HIV-1/2 antibodies as well as HIV-1/2 NAT. Currently, for HIV-2 NAT there are no FDA approved tests that discriminate HIV-1 and HIV-2 available, therefore research use HIV-2 NAT was used to verify the presence of HIV-2 RNA in HIV-2 confirmed positive specimens and specimens from an HIV-2 endemic area.

11.1. Clinical Specificity

Specimens from Low Risk Individuals or HIV Negative Individuals

A multi-site clinical study was performed to determine the specificity of Elecsys HIV Duo. Clinical specificity was determined using 6910 samples from individuals at low risk or negative for HIV infection. Testing of the 6910 samples included a comparison against the FDA-approved HIV-1/HIV-2 antigen and antibody reference assay. Confirmation of repeatedly reactive samples followed CDC recommendations to determine the final HIV status. Confirmatory testing was performed using an FDA-approved HIV-antigen and antibody assay that differentiated HIV-1 and HIV-2 antibodies and two reverse transcription-polymerase chain reaction (RT-PCR) assays.

The 6910 subjects tested included 6108 low-risk adults, 603 low-risk pediatric individuals and 199 pregnant women negative for HIV infection. The low risk cohort tested consisted of 51% plasma (3496/6910) and 49% (3414/6910) serum specimens. 6711 specimens were prospectively collected and 199 were archived or purchased from a vendor. The overall summary of results is presented in Table 10.

Table 10. Elecsys HIV Duo Reactivity with Low Risk or HIV Negative Individuals

		Elecsys HIV Duo			FDA-approved reference assay			Repeatedly Reactive Specimens (Number Reactive / Positive by Method)		
Specimen Population	**Number Tested**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**FDA-approved reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation**	**HIV-1 NAT**	**HIV-2 NAT**
Low Risk Adults	6108	6089	19	19	6091	18	17	14*	0	NT
Low risk pediatrics	603	601	2	2	600	3	3	1	0	NT
Pregnant women negative for HIV	199	199	0	0	199	0	0	NT	NT	NT
Total	6910	6889	21	21	6890	21	20	15*	0	NA

NR= non-reactive, IR= initially reactive, RR= repeatedly reactive, R= Reactive, NT=not tested through algorithm, NA= not applicable
**One subject did not have sufficient volume to complete the testing algorithm and had a final HIV status of "Inconclusive"*

Summary: The specificity of Elecsys HIV Duo was determined using 6,910 subjects from individuals at low risk for HIV or negative for HIV.14/6910 subjects had a final diagnosis of "HIV Positive", 6894/6910 subjects had a final diagnosis of "HIV Negative", and 2 subjects had a final diagnosis of "Inconclusive"(quantity not sufficient to complete confirmation testing algorithm) based on confirmation testing algorithm. These two specimens were excluded from the calculation of specificity for Elecsys Duo, reducing the numerator from 6889 to 6887. Based on the data 6887/6894 were non-reactive on Elecsys HIV Duo and 7 specimens gave false positive results. The specificity of Elecsys HIV Duo in individuals at low risk for HIV or negative for HIV was 99.9% (6887/6894) with a 95% confidence interval of 99.79% to 99.95%.

11.2. Clinical Sensitivity

11.2.1. Elecsys HIV Duo Reactivity in Individuals Known to be Positive for Antibodies to HIV-1

A multi-site clinical study was performed to assess the sensitivity of the Elecsys HIV Duo in 1549 individuals who were known to be HIV-1 antibody positive, including 1249 confirmed positive adults (1,049 U.S. and 200 non-U.S.), 51 HIV-1 confirmed positive pediatric subjects (greater than 2 years of age), 59 HIV-1 confirmed positive pregnant women (49 U.S. and 10 non-U.S.), 90 HIV-1 group M subtypes specimens, 50 HIV-1 group O specimens and 50 HIV-1 p24 antigen/antibody positive subjects. All 1549 specimens were repeatedly reactive using the Elecsys HIV Duo. Table 11 shows the results for subjects from various categories used in the study.

Table 11. Elecsys HIV Duo Reactivity in Individuals Known to be Positive for Antibodies to HIV-1

	Elecsys HIV Duo Assay				FDA-approved reference assay			Repeatedly Reactive Specimens (Number Reactive / Positive by Method)		
Specimen Population	**Number Tested**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**FDA-approved reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation**	**HIV-1 NAT**	**HIV-2 NAT**
HIV-1 Confirmed Positive Adults	1049	0	1049	1049	0	1049	1049	1049	NT	NT
HIV-1 Confirmed Positive Pediatrics	51	0	51	51	1	50	50	51	NT	NT
US HIV Positive Pregnant Women	49	0	49	49	0	49	49	49	NT	NT
Non-US HIV Positive Pregnant Women	10	0	10	10	0	10	10	10	NT	NT
Non-US HIV-1 Confirmed Positive Adults	200	0	200	200	0	200	200	200	NT	NT
HIV-1 Group O	50	0	50	50	0	50	50	49*	NT	NT
HIV-1 Group M Subtypes	90	0	90	90	0	90	90	90	NT	NT

	Elecsys HIV Duo Assay				FDA-approved reference assay			Repeatedly Reactive Specimens (Number Reactive / Positive by Method)		
Specimen Population	**Number Tested**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**FDA-approved reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation**	**HIV-1 NAT**	**HIV-2 NAT**
HIV-1 Antigen Positive/Antibody Positive	50	0	50	50	0	50	50	50	NT	NT
Total	1549	0	1549	1549	1	1548	1548	1548	NA	NA

NR= non-reactive, IR= initially reactive, RR= repeatedly reactive, NT=not tested through algorithm, NA= not applicable
**One subject had insufficient volume for confirmation testing. However, this specimen is congruent reactive for Elecsys and Reference assay.*

Summary: All 1,549 subjects had a final diagnosis of "HIV Positive" based on the confirmation testing algorithm. The sensitivity of the Elecsys HIV Duo in the HIV-1 antibody positive cohort was 100% (1549/1549) with a 95% confidence interval of 99.75% to 100%.

11.2.2. Elecsys HIV Duo Reactivity in Individuals Known to be Positive for Antibodies to HIV-2

The sensitivity of the Elecsys HIV Duo was determined in 200 confirmed positive subjects who were from the Ivory Coast, Cameroon, Senegal or Bolivia, known to be an HIV-2 endemic area. All subjects tested were confirmed to be HIV-2 positive based on a Certificate of Analysis and/or genetic sequencing prior to enrollment. All 200 specimens were confirmed reactive with an HIV-2 FDA-approved reference assay as demonstrated in Table 12.

Table 12. Elecsys HIV Duo Reactivity in Individuals Known to be Positive for Antibodies to HIV-2

	Elecsys HIV Duo				FDA-approved reference assay			Repeatedly Reactive Specimens (Number Reactive / Positive by Method)		
Specimen Population	**Number Tested**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**FDA-approved reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation**	**HIV-1 NAT**	**HIV-2 NAT**
HIV-2 Confirmed Positive	200	0	200	200	0	200	200	200	0	98*
Total	200	0	200	200	0	200	200	200	0	98*

NR= non-reactive, IR= initially reactive, RR= repeatedly reactive
**One subject was QNS for HIV-2 NAT testing.*

Summary: All 200 subjects had a final diagnosis of "HIV Positive" based on confirmation testing algorithm. The sensitivity of the Elecsys HIV Duo in the HIV-2 antibody positive cohort was 100% (200/200) with a 95% confidence interval of 98.12% to 100%. All 200 specimens tested were non-reactive using the HIV-1 NAT assay. Of these 200 specimens 98 were reactive using the HIV-2 NAT assay. Low HIV-2 NAT reactivity may be observed with specimens from HIV positive individuals on anti-retroviral therapy. These results do not have an impact on the performance of the Elecsys HIV Duo assay for HIV detection.

11.2.3. Elecsys HIV Duo Reactivity in Individuals Known to be Positive for HIV-1 Group M subtypes

The sensitivity of Elecsys HIV Duo for the HIV-1 group M subtypes was determined in a multi-site study using a total of 90 specimens. All specimens tested were confirmed to be HIV-1 group M positive based on a Certificate of Analysis and/or genetic sequencing prior to enrollment. The HIV-1 group M cohort contained 6 different subtypes: A, B, C, D, CRF01_AE and CRF02_AG. Each subtype contained 15 specimens each with a total of 90 specimens tested with all 90 subjects having a final diagnosis of HIV positive (Table 13).

Table 13. Elecsys HIV Duo Reactivity in Individuals Known to be Positive for HIV-1 group M subtypes

HIV-1 Group M Subtypes	Number Tested	Repeatedly Reactive on Elecsys HIV Duo	Repeatedly Reactive on FDA-approved reference	Repeatedly Reactive Specimens (Number Reactive / Positive by Method)		
				FDA reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation	HIV-1 NAT	HIV-2 NAT
A	15	15/15	15/15	15	NT	NT
B	15	15/15	15/15	15	NT	NT
C	15	15/15	15/15	15	NT	NT
D	15	15/15	15/15	15	NT	NT
CRF01_AE	15	15/15	15/15	15	NT	NT
CRF02_AG	15	15/15	15/15	15	NT	NT
Total	90	90/90	90/90	90	NA	NA

NT=not tested through algorithm, NA= not applicable

Summary: All 90 specimens had a final diagnosis of “HIV Positive” based on the confirmation testing algorithm.

11.2.4. Elecsys HIV Duo Reactivity in Specimens Positive for Antibodies to HIV-1 Group O

The sensitivity of the Elecsys HIV Duo was determined in a multi-site study with specimens from individuals known to be positive for HIV-1 Group O based on certificate of analysis and/or from genetic sequencing provided prior to enrollment. Table 14 shows the overall number of specimens used in this study and the results obtained.

Table 14. Elecsys HIV Duo Reactivity in Specimens Positive for Antibodies to HIV-1 Group O

		Elecsys HIV Duo			FDA-approved reference			Repeatedly Reactive Specimens (Number Reactive / Positive by Method)		
Specimen Population	**Number Tested**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**FDA-approved reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation**	**HIV-1 NAT**	**HIV-2 NAT**
Group O	50	0	50	50	0	50	50	49*	0	NT
Total	50	0	50	50	0	50	50	49*	0	NA

NR= non-reactive, IR= initially reactive, RR= repeatedly reactive, NT=not tested through algorithm, NA= not applicable

*One subject had insufficient volume for confirmation testing.

Summary: All 50 specimens had a final diagnosis of "HIV Positive" based on confirmation testing algorithm. Non-Reactive HIV-1 NAT results may be observed with specimens from HIV positive individuals on anti-retroviral therapy. These results do not have an impact on the performance of the Elecsys HIV Duo assay for HIV detection.

11.2.5. Elecsys HIV Duo Reactivity with Specimens from High Risk Individuals

The sensitivity and specificity of Elecsys HIV Duo in individuals at high risk for HIV was determined using a total of 1,410 subjects including 506 adults, 200 pediatrics, 204 pregnant women and 500 from an HIV-2 endemic area. The pediatric specimens were collected from subjects ranging in age from 2 to 21 years. The test results are presented in Table 15.

Table 15. Elecsys HIV Duo Reactivity with Specimens from High Risk Individuals

		Elecsys HIV Duo			FDA-approved reference			Repeatedly Reactive Specimens (Number Reactive / Positive by Method)		
Specimen Population	**Number Tested**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**FDA- approved reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation**	**HIV-1 NAT**	**HIV-2 NAT**
Adults	506	494	12	12	494	12	12	12	NT	NT
Pediatrics	200	195	5	5	196	5	4	4	0	NT
Pregnant women	204	188	16	16	189	15	15	15	0	NT
HIV-2 Endemic	500	365	135	135	369	131	131	131	25*	54
Total	1410	1242	168	168	1248	163	162	162	25	54

NR= non-reactive, IR= initially reactive, RR= repeatedly reactive, NT=not tested through algorithm
** All 25 reactive HIV-1 results were from the second PCR method HIV-1 and HIV-2 differentiation NAT.*

Summary: A total of 168/1410 specimens from high risk individuals were repeatedly reactive using the Elecsys HIV Duo assay and,163 specimens from high risk individuals were found to be repeatedly reactive with the FDA approved assay. Final diagnosis based on confirmation testing algorithm indicate that 162 specimens were confirmed to be positive and 1248 subjects were confirmed negative resulting in 6 false positive results for the

Elecsys HIV Duo. Specimens from an HIV-2 endemic region were further tested using an HIV-1/HIV-2 differentiation NAT assay. 54 specimens were found to be HIV-2 NAT only reactive. These results do not have an impact on the performance of the Elecsys HIV Duo assay for HIV detection.

11.3. Elecsys HIV Duo Reactivity in specimens from Pregnant Women

A multi-site clinical study was performed to compare the performance of the Elecsys HIV Duo to an FDA approved HIV assay using specimens from 588 pregnant women including 59 confirmed HIV positive pregnant women (49 US and 10 non US), 204 pregnant women at high risk for HIV infection and 325 pregnant women negative or at low risk for HIV (199 negative for HIV and 126 low risk for HIV). The results are presented in Table 16.

Table 16. Elecsys HIV Duo Reactivity in Pregnant Women

		Elecsys HIV Duo			**FDA-approved reference**			**Repeatedly Reactive Specimens (Number Reactive / Positive by Method)**		
Specimen Population	**Number Tested**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**FDA-approved reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation**	**HIV-1 NAT**	**HIV-2 NAT**
HIV-1-positive pregnant	59	0	59	59	0	59	59	59	NT	NT
High risk pregnant	204	188	16	16	189	15	15	15	0	NT
Pregnant women negative or low risk	325*	324	1	1	324	1	1	1	NT	NT
Total	588	512	76	76	513	75	75	75	0	NA

NR= Non – reactive, IR= initially reactive, RR= repeatedly reactive, NT= not tested through algorithm, NA=not applicable.
**126 were from the low risk population*

Summary: Of 588 subjects tested, 75 subjects had a final diagnosis of “HIV Positive” and 513 subjects had a final diagnosis of “HIV Negative” resulting in 1 false positive result for the Elecsys HIV Duo. Non-Reactive HIV-1 NAT results may be observed with specimens from HIV positive individuals on anti-retroviral therapy. These results do not have an impact on the performance of the Elecsys HIV Duo assay for HIV detection.

11.4. Elecsys HIV Duo Reactivity in specimens from Pregnant Women at High Risk for Infection with HIV

A multi-site clinical study was performed to compare the performance of the Elecsys HIV Duo to an FDA approved HIV assay using specimens from pregnant females at high risk for infection with HIV in different trimesters. A total of 204 specimens from all three trimesters were tested to determine the sensitivity/specificity in this cohort. The results are presented in Table 17.

Table 17. Elecsys HIV Duo Reactivity in specimens from Pregnant Women at High Risk for Infection with HIV

		Elecsys HIV Duo			FDA-approved reference			Repeatedly Reactive Specimens (Number Reactive / Positive by Method)		
Specimen Population	**Number Tested**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**FDA-approved reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation**	**HIV-1 NAT**	**HIV-2 NAT**
First trimester	55	48	7	7	49	6	6	6	0	NT
Second trimester	63	57	6	6	57	6	6	6	NT	NT
Third trimester	86	83	3	3	83	3	3	3	NT	NT
Total	204	188	16	16	189	15	15	15	0	NA

NR= Non – reactive, IR= initially reactive, RR= repeatedly reactive, NT= not tested through algorithm, NA=not applicable

Summary: Of 204 subjects tested, 15 subjects had a final diagnosis of “HIV Positive” and 189 subjects had a final diagnosis of “HIV Negative” resulting in 1 false positive for the Elecsys HIV Duo. Non-Reactive HIV-1 NAT results may be observed with specimens with HIV positive individuals on anti-retroviral therapy. These results do not have an impact on the performance of the Elecsys HIV Duo assay for HIV detection.

11.5. Elecsys HIV Duo Reactivity in Specimens from Pediatric population.

A multi-site clinical study was performed to compare the performance of the Elecsys HIV Duo with an FDA approved HIV assay using specimens from pediatric subjects. A total of 854 pediatric specimens tested included 603 at low risk for HIV infection, 200 at high risk for HIV infection and 51 confirmed HIV positive (34 U.S. and 17 non-U.S.). Specimens collected were in the age range of two to 21 years. The results are presented in Table 18.

Table 18. Elecsys HIV Duo Reactivity with Pediatric specimens

		Elecsys HIV Duo			FDA-approved reference			Repeatedly Reactive Specimens (Number Reactive / Positive by Method)		
Specimen Population	**Number Tested**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**FDA-approved reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation**	**HIV-1 NAT**	**HIV-2 NAT**
Low risk pediatric	603	601	2	2	600	3	3	1	0	NT
High risk pediatric	200	195	5	5	196	5	4	4	0	NT
Pediatric HIV positive	51	0	51	51	1	50	50	51	0	NT
Total	854	796	58	58	797	58	57	56	0	NA

NR= Non – reactive, IR= initially reactive, RR= repeatedly reactive, NT= not tested through algorithm, NA=not applicable

Summary: Of 854 subjects tested, 56 subjects had a final diagnosis of “HIV Positive” and 798 subjects had a final diagnosis of “HIV Negative” resulting in 2 false positive results for

the Elecsys HIV Duo. Non-Reactive HIV-1 NAT may be observed with specimens from HIV positive individuals on anti-retroviral therapy. These results do not have an impact on the performance of the Elecsys HIV Duo assay for HIV detection.

11.6. High Risk Pediatric Specimens Categorized by Age Range and Gender

A multi-site clinical study was performed to compare the performance of the Elecsys HIV Duo with an FDA approved HIV assay using specimens from high risk pediatric subjects. A total of 200 specimens, collected prospectively were tested. Results categorized by age range and gender are shown in Table 19.

Table 19. Distribution of High-Risk Pediatric Specimens Categorized by Age Range and Gender

			Elecsys HIV Duo			FDA-approved reference			Repeatedly Reactive Specimens (Number Reactive / Positive by Method)		
Specimen Population	**Sex**	**Number Tested**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**FDA-approved reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation**	**HIV-1 NAT**	**HIV-2 NAT**
2 - <6 years	Female	0	0	0	0	0	0	0	NT	NT	NT
2 - <6 years	Male	0	0	0	0	0	0	0	NT	NT	NT
6 - <11 years	Female	2	2	0	0	2	0	0	NT	NT	NT
6 - <11 years	Male	2	2	0	0	2	0	0	NT	NT	NT
11 - <16 years	Female	2	2	0	0	2	0	0	NT	NT	NT
11 - <16 years	Male	4	4	0	0	4	0	0	NT	NT	NT
16 - <22 years	Female	99	95	4	4	96	3	3	3	0	NT
16 - <22 years	Male	91	90	1	1	90	2	1	1	NT	NT
Total		200	195	5	5	196	5	4	4	0	N/A

NR= non-reactive, IR= initially reactive, RR= repeatedly reactive, NT= not test through algorithm, NA=not applicable

Summary: Of 200 subjects tested, 4 subjects had a final diagnosis of “HIV Positive” and 196 subjects had a final diagnosis of “HIV Negative” resulting in 1 false positive result for the Elecsys HIV Duo. Non-Reactive HIV-1 NAT results may be observed with specimens from HIV positive individuals on anti-retroviral therapy. These results do not have an impact on the performance of the Elecsys HIV Duo assay for HIV detection.

11.7. Elecsys HIV Duo Reactivity in Individuals Known Positive for HIV-1 Antigen

The sensitivity of the Elecsys HIV Duo was determined in a multi-site study in a total of 102 antigen positive, 50 antibody negative and 52 antigen positive, antibody positive specimens. The cohort consisted of p24 antigen positive specimens based on the certificate of analysis.

A total of 102 specimens were repeatedly reactive using the Elecsys HIV Duo and the FDA approved assay. The results are presented in Table 20.

Table 20. Elecsys HIV Duo Reactivity in Individuals Known Positive for HIV-1 Antigen

		Elecsys HIV Duo			FDA-approved reference assay			Repeatedly Reactive Specimens (Number Reactive / Positive by Method)		
Specimen Population	**Number Tested**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**FDA-approved reference assay HIV-Ag/Ab with HIV-1 and HIV-2 differentiation**	**HIV-1 NAT**	**HIV-2 NAT**
HIV-1 antigen positive /antibody positive	50	0	50	50	0	50	50	50	NT	NT
HIV-1 antigen positive /antibody negative	52	0	52	52	0	52	52	51*	NT	NT
Total	102	0	102	102	0	102	102	101	NA	NA

NR= non-reactive, IR= initially reactive, RR= repeatedly reactive, NT= not test through algorithm, NA=not applicable
**One specimen did not have sufficient volume for testing.*

Summary: 102/102 specimens known to be positive for HIV antigen had a final status of "HIV Positive" and were reactive on both the Elecsys HIV Duo assay and the FDA-approved reference assay.

12. Inspections

12.1. Manufacturing Facilities Review/Inspection

Roche Diagnostic GmbH facilities in (b) (4) Mannheim, Germany have been recently inspected for in-vitro diagnostic products. The registration numbers (b) (4) 9610126 are active for the facility and the FEI # is 9610126 . As the assay will be approved under the regulations for a PMA and not a BLA, no license number is assigned.

FDA inspection in Mannheim/(b) (4) included:

- (b) (4) –premarket approval for Elecsys HBsAg II and post market approval of Elecsys Anti-HCV II. 483 citation.
- (b) (4) – post market approval Elecsys HBsAg II. NAI

Based on the information provided in the PMA submission, and the previous inspection reports supporting the overall compliance status of the manufacturing sites, the review committee recommended waiving the pre-approval inspections for the facilities associated with this PMA.

12.2. Bioresearch Monitoring (BIMO) Inspections

CBER Bioresearch Monitoring (BIMO) issued high-priority inspection assignments at the one testing site in the United States. These inspections did not reveal significant problems that impact the data submitted in this PMA. The inspections were classified as No Action Indicated (NAI).

13. Conclusions Drawn from the Studies

13.1. Risk/Benefit Analysis

As a diagnostic test the Roche Elecsys HIV Duo assay involves removal of blood from an individual for testing purposes. This test presents no more of a safety hazard to an individual than is presented to an individual who is having their blood drawn for any other diagnostic evaluation. The benefit to HIV-1 or HIV-2 infected individuals tested by this assay outweighs any potential adverse event or risk to the patient or user due to assay malfunction or operator error. The potential risks encountered with this in vitro diagnostic test are not unusual in the clinical laboratory setting. Appropriate warnings for these risks are contained in the labeling and package inserts for these devices. Standard good laboratory practices are considered sufficient to mitigate the risks to the end user. Potential adverse effects of the Roche Elecsys HIV Duo assay relate to the risk of false positive and false negative results. While performance studies indicate that this risk is likely to be very small, the potential for inaccurate results exists. The risk of incorrect results is minimized by following the procedures and instructions outlined in the Package Insert.

13.2. Safety and Effectiveness

Multi-center clinical studies were conducted in the U.S. The Roche Elecsys HIV Duo Assay performed with clinical sensitivity and specificity comparable to an FDA approved HIV Ag/Ab combo assay.

Results from the clinical studies indicate that the Roche Elecsys HIV Duo assay, together with supplemental testing, can be used safely and effectively for the simultaneous qualitative detection and differentiation of HIV-1 p24 antigen and antibodies to HIV-1 (groups M and O) and HIV-2 in human serum and plasma. Reactive specimens must be investigated by additional, more sensitive NAT, or supplemental tests. Confirmation of the test result on a freshly drawn specimen and counseling are considered an important part of testing for HIV antigen and antibody to HIV-1 and HIV-2. A negative test result at any point in the investigation of individual subjects does not preclude the possibility of exposure to or infection with HIV-1 and/or HIV-2. Negative results can occur if the quantity of marker present in the specimen is below the detection limit of the assay, or if the marker is not present during the stage of disease in which a specimen is collected.

The safety and effectiveness of the Roche Elecsys HIV Duo assay has been shown in the clinical and non-clinical studies performed. The assay has been shown to be an effective tool in detecting infection with HIV-1 or HIV-2.

14. Panel Recommendations

Not Applicable

15. CBER Decision

The PMA BP190403 is recommended for approval.