SUMMARY OF SAFETY AND EFFECTIVENESS DATA

1. GENERAL INFORMATION

Device Generic Name:	Test, HIV Detection (Human Immunodeficiency Virus (HIV) p24 antigen and Antibodies to HIV Type 1 (HIV-1 group M and group O) and/or Type 2)
Device Trade Name:	LIAISON® XL MUREX HIV Ab/Ag HT LIAISON® XL MUREX Control HIV Ab/Ag HT
Device Product Code:	MZF
Applicant Name and Address:	DiaSorin Inc. 1951 Northwestern Avenue Stillwater, MN 55082
FDA Registration Number:	3008576040
Premarket Approval Application (PMA) Number:	BP190437/0
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:	November, 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 memoranda from the following reviewers were used in developing the SSE:

Discipline Reviewed	Reviewer Names
Scientific Lead	Kavita Singh
Clinical	Pawan Jain
Chemistry/Manufacturing/Controls (CMC)	Iwona Fijalkowska Nitin Verma
Preclinical/Analytical	Mohan Kumar Haleyurgirisetty
Instrumentation and Software	Nicholas Anderson
Statistician	Ho-Hsiang Wu
Bioresearch Monitoring Inspection (BIMO)	Colonious King
DMPQ/Pre-approval Inspection	Deborah Trout Nicole Li David Bailey
Product and Promotional Labeling	Dana C Jones
Scientific and Programmatic Aspects	Pradip Akolkar David Leiby Indira Hewlett Julia Lathrop

2. INTENDED USE

The LIAISON® XL MUREX HIV Ab/Ag HT is an *in vitro* chemiluminescent immunoassay for the simultaneous qualitative detection of HIV p24 antigen and antibodies to HIV-1 (Groups M and O) and HIV-2 in human serum (without or with gel-SST) or plasma (lithium and sodium heparin, sodium citrate, and potassium EDTA), on the LIAISON® XL Analyzer. It is intended to be used as an aid in the diagnosis of HIV-1/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 and/or HIV-2 infection in pediatric subjects (2–21 years) and in pregnant women.

The assay cannot distinguish between the detection of HIV p24 antigen and HIV-1/HIV-2 antibodies.

The LIAISON® XL MUREX HIV Ab/Ag HT assay is not intended for screening donors of blood or blood products, or human cells or tissues or cellular and tissue-based products (HCT/Ps), or organ donors for HIV.

The LIAISON® XL MUREX Control HIV Ab/Ag HT is intended for use as assayed quality control samples to monitor the performance of the LIAISON® XL HIV Ab/Ag HT assay. The performance characteristics of LIAISON® controls have not been established for any other assays or instrument platforms different from LIAISON® XL.

3. DEVICE DESCRIPTION

Assay Principle

The assay simultaneously detects but does not differentiate HIV antibodies and HIV p24 antigen. Qualitative determination of specific antibodies to HIV and HIV p24 antigen is accomplished using a sandwich chemiluminescence immunoassay.

HIV-1 recombinant antigen, HIV-1 group O and HIV-2 biotinylated peptides, and biotinylated monoclonal antibodies to HIV p24 antigen are used for coating magnetic particles (solid phase) and are also linked to isoluminol or fluorescein derivatives (isoluminol-antigen-peptides-monoclonal conjugates and monoclonal anti-HIV p24-fluorescein conjugates).

The LIAISON® XL MUREX HIV Ab/Ag HT assay consists of two incubation phases. In the first incubation phase HIV antibodies present in samples or controls and HIV p24 antigen present in calibrator, samples or controls bind to the solid phase and, for HIV p24 antigen, to monoclonal anti-HIV p24 antigen labelled with fluorescein derivatives.

In the second incubation phase HIV-1 antigen, HIV-1 group O, HIV-2 peptides, monoclonal anti-HIV p24 antigen and monoclonal anti-fluorescein linked to an isoluminol derivative (isoluminol-antigen conjugate) react with HIV antibodies, HIV p24 antigen and monoclonal anti-HIV p24 antigen labelled with fluorescein derivatives already bound to the solid phase.

After each incubation, the unbound material is removed with a wash cycle. Next, the starter reagents are added and a flash chemiluminescence reaction is induced. The light signal, and hence the amount of isoluminol-antigen/peptide/monoclonal conjugate, is measured by a photomultiplier as relative light units (RLU) and is indicative of HIV-1/2/O antibodies or HIV p24 antigen presence in calibrator, samples or controls.

Kit Components

Reagents: The LIAISON® XL MUREX HIV Ab/Ag HT assay is an *in vitro* diagnostic device consisting of four reagents provided in individual compartments within a plastic container called the Reagent Integral. The assay configuration allows performance of 200 tests.

The assay is comprised of the following components as shown in Table 1:

Table 1. LIAISON®XL MUREX HIV Ab/Ag HT Assay - Reagent Integral

MAGNETIC PARTICLES [SORB] 1 vial – 2.5 mL	Magnetic particles coated with HIV-1 gp41 (group M) recombinant antigen (obtained in *E. coli*), HIV-1 group O and HIV-2 biotinylated peptides, biotinylated monoclonal anti-HIV p24 antigen, BSA, PBS buffer, preservatives.
CALIBRATOR 1 [CAL] 1 vial – 2.9 mL	Human serum and/or plasma with low-level reactivity for anti-HIV-2 and anti-HIV-1 (group M), casein, PBS buffer, 0.2% ProClin® 300, an inert yellow dye.
ASSAY BUFFER 1 [BUF1] 1 vial – 10.8 mL	Monoclonal anti-HIV p24 antigen conjugated with fluorescein, bovine serum, casein, BSA, non-specific IgG (mouse polyclonal), HEPES buffer, detergent, EDTA, 0.2% ProClin® 300, preservatives.
CONJUGATE [CONJ] 2 vials – 23 mL	HIV-1 gp41 (group M) recombinant antigen (obtained in *E. coli*), HIV-1 group O, HIV-2 peptides, monoclonal anti-HIV p24 antigen and monoclonal anti-fluorescein derivative, conjugated to an isoluminol derivative, sheep serum, negative human serum, casein, non-specific IgG (mouse polyclonal), BSA, TRIS buffer, 0.2% ProClin® 300, preservatives.

Controls: The LIAISON® XL MUREX Control HIV Ab/Ag HT set consists of five ready-to-use controls. Each control solution allows multiple tests to be performed.

The controls are comprised of the following components as shown below in Table 2:

Table 2: LIAISON® XL MUREX Control HIV Ab/Ag HT (Catalog Number 318291)

NEGATIVE CONTROL 1 vial – 4.5 mL	Human serum non-reactive for HIV antigens and antibodies, 0.2% ProClin® 300, preservatives.
POSITIVE CONTROL (anti-HIV-2) 1 vial – 4.5 mL	Human serum/plasma reactive for HIV-2 antibodies, 0.2% ProClin® 300, preservatives.
POSITIVE CONTROL (anti-HIV-1 O) 1 vial – 4.5 mL	Rabbit polyclonal reactive for HIV-1 O antibodies, human serum, 0.2% ProClin® 300, preservatives.
POSITIVE CONTROL (anti-HIV-1 M) 1 vial – 4.5 mL	Human serum/plasma reactive for HIV-1 M antibodies, 0.2% ProClin® 300, preservatives.
POSITIVE CONTROL (HIV/Ag) 1 vial – 4.5 mL	HIV p24 recombinant antigen (obtained in *E. coli*), stabilized in PBS buffer, bovine aprotinin, casein, 0.2% ProClin® 300.

ProClin® is a trademark of the Dow Chemical Company (Dow) or an affiliated company of Dow. All reagents are supplied ready to use.

Additional materials required but sold separately

The following are required to perform the LIAISON® XL MUREX HIV Ab/Ag HT assay.

- **LIAISON® XL Analyzer** is a fully automated chemiluminescent analyzer, performing the complete sample processing steps of the chemiluminescent assay and generating the final results. Cleared by FDA as a stand-alone analyzer (K103529) and also for use when connected to a previously cleared 3rd party Laboratory Automation System (LAS) (K141116)
- **LIAISON® Wash/System Liquid** (10x) - phosphate buffer solution, < 0.1% sodium azide
- **LIAISON® XL Starter Kit** – catalyst in 4% sodium hydroxide solution and 0.12% hydrogen peroxide solution
- **LIAISON® XL Non reagent accessories:** cuvettes, disposable tips and waste bags

LIAISON® XL MUREX HIV Ab/Ag HT - Reagent Integral Storage And Stability

The Reagent Integral must be stored in an upright position to facilitate resuspension of magnetic particles. When the Reagent Integral is stored sealed and kept upright, the reagents are stable at 2–8°C through the expiration date. Do not freeze. The Reagent Integral should not be used past the expiration date indicated on the kit and Reagent Integral labels. After removing the seals, the Reagent integral is stable for five weeks when stored at 2–8°C or on board the analyzer.

4. INSTRUMENTATION AND SOFTWARE

The following is a summary overview of software, instrumentation and risk management information provided to support a reasonable assurance that the device is safe and effective for its intended use and conditions of use.

Versioning:

The LIAISON® XL Analyzer was cleared with a Moderate Level of Concern (Software version 3.1.4.20) on January 21, 2011 (K103529). Software version 4.2.2.1 was provided to the FDA and cleared in K181464 on August 31, 2018. This PMA submission contains the current LIAISON® XL Software version 4.2.2.2

Risk Management:

The final risk profile of the LIAISON® XL includes all risks (high, middle and low-level) which have been rated and, as appropriate, countermeasures have been effectively implemented. The effectiveness of the countermeasures has been documented and the overall residual risk of the LIAISON® XL is acceptable as per the evidence provided.

Development Management:

The software development activities included establishing detailed software requirements, linking requirements with associate verification tests, verification and validation testing, defect tracking, configuration management and maintenance activities to ensure the software conforms to user needs and intended uses.

5. TEST PROCEDURE

Specimen Collection, Preparation and Storage:

Collection containers were used according to the tube manufacturer's instructions. Blood should be collected aseptically by venipuncture and the serum or plasma (lithium and sodium heparin, sodium citrate and potassium EDTA) separated from clot, red cells or gel separator, after centrifugation. Samples removed from red cells, clot or gel separator having particulate matter, fibrin, turbidity, lipemia, or erythrocyte debris, specimens that have been stored at room temperature (20–25°C), or frozen and thawed, or samples requiring repeat testing, require clarification by further centrifugation (recommended 10,000 g for 10 minutes) before testing, to improve the consistency of results. Grossly hemolyzed or lipemic samples, as well as samples containing particulate matter or exhibiting obvious microbial contamination should not be tested. Check for and remove air bubbles and foam before assaying.

Room temperature storage (between 18 and 30°C) of specimens up to three days does not adversely influence the assay performance. If the assay is performed within seven days of sample collection, the samples may be kept at 2–8°C; otherwise they should be aliquoted and stored frozen (-20°C or below). If samples are stored frozen, mix thawed samples well before testing. Samples are stable through seven freeze/thaw cycles. Self-defrosting freezers are not recommended for sample storage.

The minimum volume required for a single determination is 350 µL specimen (200 µL specimen + 150 µL dead volume. Dead volume is the volume left at the bottom of the tube which the analyzer cannot aspirate).

Calibration:

Assaying of the calibrator contained in the reagent integral allows the analyzer to set the assay cut-off. Recalibration in triplicate is mandatory whenever at least one of the following conditions occurs:

- A new lot of reagent integral or Starter kit is used.
- The previous calibration was performed more than five weeks before.
- The analyzer has been serviced.
- Control values lie outside the expected ranges.

Quality Control:

Quality control must be performed once per day of use or in accordance with local, state, and/or federal regulations or accreditation requirements and your laboratory's

quality control procedures. It is recommended that the user refer to CLSI standard C24-A3 and 42 CFR 493.1256(c) for guidance on appropriate quality control practices. LIAISON® controls should be run in singlicate to monitor the assay performance. If control values lie within the expected ranges, the test is valid. If control values lie outside the expected ranges, the test run is invalid and patient results cannot be reported. Assay calibration should be performed if a control failure is observed and controls and specimens must be retested. The performance of other controls should be evaluated for compatibility with this assay before they are used. Appropriate value ranges should then be established for additional quality control materials.

Interpretation Of Results:

The presence or absence of HIV p24 antigen and/or HIV specific antibodies in the specimens is determined by comparing the chemiluminescence reaction signal to the preliminary cut-off value provided by the assay calibration.
The analyzer automatically calculates the signal-to-cutoff (S/CO) ratios, then interprets the results.
For details, refer to the analyzer operator's manual.

The cutoff discriminating between the reactivity to HIV p24 antigen and/or HIV specific antibodies has a S/CO value of 1.00.

Patient results should be interpreted as follows:

- **Non-Reactive:** Samples with S/CO value of less than 1.00 are considered non-reactive (NR). These samples are considered negative for HIV p24 antigen and HIV specific antibodies and do not require further testing.
- **Reactive:** Samples with S/CO value equal to or greater than 1.00 are considered initially reactive (IR) for HIV p24 antigen and/or HIV-specific antibodies. All initially reactive samples should be repeated in duplicate.
    - If S/CO values are equal to or greater than 1.00 in either or both of the repeat replicates, the samples are considered repeatedly reactive (RR) and must be confirmed with supplemental assays.
    - If S/CO values are less than 1.00 in both of the repeat replicates, the samples are considered non-reactive (NR).

6. WARNINGS AND PRECAUTIONS

- This test kit is intended for use with the approved matrices. Strict adherence to the instructions is necessary to obtain reliable results.
- Caution: All human blood source material used to produce the components provided in this test kit derives from units found to be non-reactive for HBsAg, antibodies to HCV, HIV-1, HIV-2 when tested by an FDA-approved method, except for the positive controls which are reactive for antibodies to HIV-2 HIV-1 group M, or HIV-1 group O. The units positive for HIV antibodies have been obtained from individuals infected with HIV-1 and/or HIV-2. Although these have

been inactivated by heat treatment (56°C for one hour) during the manufacturing process, they should be considered as potentially infectious. Because no test method can offer complete assurance that laboratory specimens are pathogen-free, specimens should be handled at Biosafety Level 2, as recommended for any potentially infectious human serum or blood specimen in the CDCNIH manual, "*Biosafety in Microbiological and Biomedical Laboratories, 5th Edition, Feb. 2007"*, and CLSI Approved Guideline M29-A3, "*Protection of Laboratory Workers from Occupationally Acquired Infections."*

- Observe the normal precautions required for handling all laboratory reagents.
- Do not pipette by mouth. Do not eat, drink, smoke or apply cosmetics in the assay laboratory.
- Avoid direct contact with potentially infected material by wearing laboratory clothing, protective goggles, and disposable gloves.
- Wash hands thoroughly at the end of each assay.
- Avoid splashing or forming an aerosol. All spills of biological material must be cleaned with a sodium hypochlorite solution with 0.5% active chlorine, and the means used must be treated as infected waste.
- All samples and reagents containing biological materials used for the assay must be considered as potentially able to transmit infectious agents. The waste must be handled with care and disposed of in compliance with the laboratory guidelines and the statutory requirements of local and federal agencies.
- Liquid waste must be decontaminated with sodium hypochlorite at a final concentration of 10% for at least half an hour.
- Any materials for reuse must be appropriately sterilized in compliance with the local laws and guidelines. A minimum of one hour at 121°C is usually considered adequate, though the users must check the effectiveness of the sterilization/decontamination cycle by initially validating it and routinely using biological indicators.
- The LIAISON® XL Analyzer should be cleaned and decontaminated on a routine basis. See the relevant Operator's Manual for the procedures.
- Do not use kits or components beyond the expiration date on the label.
- Do not mix reagents from different reagents packs (even for the same reagent).

7. PROCEDURAL LIMITATIONS

- LIAISON® XL MUREX HIV Ab/Ag HT is for *in vitro* diagnostic use only.
- For prescription use only.
- The LIAISON® XL MUREX HIV Ab/Ag HT assay must be used in accordance with the instructions for use in the package insert to obtain accurate results.
- The LIAISON® XL MUREX HIV Ab/Ag HT assay is not intended for the screening donors of blood or blood products, or human cells or tissues or cellular and tissue-based products (HCT/Ps), or organ donors for HIV.

- This assay cannot distinguish between the detection of HIV p24 antigen and HIV-1/HIV-2 antibodies.
- This test is suitable only for testing individual, neat samples, not diluted specimens or sample pools. Bacterial contamination or heat inactivation of the specimens may affect the test results.
- Serum (without or with gel-SST), or plasma derived from lithium and sodium heparin, sodium citrate, or K2-EDTA (ethylenediaminetetraacetate) as anticoagulants may be used with the LIAISON® XL MUREX HIV Ab/Ag HT assay. Using other types of samples may not yield accurate results.
- A non-reactive test result for HIV p24 antigen and/or HIV antibodies does not exclude the possibility of exposure to or infection with HIV. In fact, either the patient may be unable to synthesize HIV specific antibodies or the circulating levels of p24 antigen and/or HIV specific antibodies may be below the assay detection limit. It is recommended that testing be repeated on a fresh specimen after 1–3 months if clinical judgement indicates a suspected false nonreactive result.
- False non-reactive results may be obtained in individuals infected with HIV-1 and/or HIV-2 who are receiving medication for treatment for HIV infection (ART) or prevention of infection (PrEP or PEP).
- Falsely reactive results cannot be ruled out with any test kit, the percentage of which is related to specimen integrity, the specificity of the test kit, and the HIV prevalence in the population being screened.
- A positive LIAISON® XL MUREX HIV Ab/Ag HT assay result interpretation confirms the presence of specific antibodies to HIV-1 and/or HIV-2 in the sample. HIV and AIDS-related conditions are clinical syndromes caused by HIV-1 and HIV-2 and their diagnosis can only be established clinically.
- A person who has antibodies to HIV-1 or HIV-2 is presumed to be infected with the virus, however, false positive results may be obtained if 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. A comprehensive risk history and clinical judgement should be considered before concluding that an individual is not infected with HIV.

8. CONTRAINDICATIONS

There are no contraindications for use for this test.

9. ALTERNATIVE PRACTICES AND PROCEDURES

Several FDA approved *in vitro* diagnostic tests are commercially available for the simultaneous qualitative detection of HIV p24 antigen and antibodies to HIV-1 (Groups M and O) and HIV-2. Assay results in conjunction with other laboratory and clinical

findings may be used as an aid in the diagnosis of HIV-1/HIV-2 infection, including acute or primary HIV-1 infection.

10. POTENTIAL ADVERSE EFFECTS OF THE DEVICE ON HEALTH

Potential adverse effects of the LIAISON® XL MUREX HIV Ab/Ag HT assay relate to the risk of false positive and false negative results. While performance studies indicate that this risk is likely to be minimal, the potential for inaccurate results exist. The risk of incorrect results is minimized by following the procedures and instructions outlined in the Instructions for Use.

11. MARKETING HISTORY

The LIAISON® XL MUREX HIV Ab/Ag HT assay (318290) and LIAISON® XL MUREX Control HIV Ab/Ag HT (318291) are new kits for the United States and have not been marketed in the U.S. or any foreign country.

12. SUMMARY OF PRE-CLINICAL STUDIES

All studies were performed using the LIAISON® XL Analyzer Software version 4.2.2.

12.1 Cut-Off Determination:

(b) (4)

The LIAISON® XL Analyzer automatically calculates the signal-to-cutoff (S/CO) value and interprets the results accordingly. The cut-off discriminating between the reactivity to HIV 24 antigen and/or HIV specific antibodies has a S/CO value of 1.00.
Results were interpreted as follows:

- Samples with S/CO value of less than 1.00 are considered non-reactive.
- Samples with S/CO value equal to or greater than 1.00 are considered reactive for HIV p24 antigen and/or HIV specific antibodies

A Receiver Operating Characteristics (ROC) analysis was performed on the results of the tested specimens to assess the accuracy of the LIAISON® XL MUREX HIV Ab/Ag HT assay to discriminate between non-reactive and reactive results. The optimal values of the cut-off are based on ROC curve, which range between 0.50 and 1.10 S/CO. The cut-off value for this product was initially set at a S/CO of 1.00. The final value of 1.0 was obtained by adjusting with “coefficient a”.

12.2 Cut-Off Change ("coefficient a"):

"Coefficient a" is the factor by which the lot specific cut-off (qualitative determination point between negative and positive results) can be optimized in order to achieve the required balance of specificity and sensitivity. The cut off is a function of the calibrator Relative Light Units (RLU) multiplied by a factor – "coefficient a."

Cut-Off = Calibrator RLU x "coefficient a"

Initial performance evaluation studies for cut-off determination with a limited number of expected positive and negative specimens defined a S/CO value of 1.0 as the cut-off. Based on an evaluation of the preliminary clinical data as well as preliminary analytical sensitivity data, an adjustment of "coefficient a" for the calculation of the cut off by a factor of 1.7 was deemed necessary in order to improve the clinical performance of the clinical populations of the assay. An RLU value that currently provides a S/CO value of 1.7 would provide a S/CO value of 1.0 after the adjustment of "coefficient a."
Testing was performed using the original lot numbers assigned to the kit lots. Data was re-calculated, using the new "coefficient a", under new lot numbers to optimize the kit sensitivity and specificity.

A. ANALYTICAL SENSITIVITY

12.3 Detectable Concentration of HIV Antigen p24 at Cut-Off:

The study was carried out to identify the level of analyte discriminating between presence and absence of the analyte. The analytical sensitivity of the LIAISON® XL MUREX HIV Ab/Ag HT assay was evaluated using serial dilutions prepared from the HIV p24 antigen, NIBSC code 90/636 International Standard to a level below the cut-off value. The standard was diluted with HIV negative serum to obtain samples spanning the assay range. Testing was performed on four LIAISON® XL Analyzers using dilutions in five replicates on three lots of kit reagents and one lot of kit control. The data demonstrate that the sensitivity of the test corresponds to an overall mean of 1.17 IU/mL at the cut off as shown in Table 3.

Table 3: Summary of Detectable Concentration of HIV p24 at Cut-Off

Run	2206000007		2206000004		2206000001		9922100127	
Matrix	**Serum**	**Plasma**	**Serum**	**Plasma**	**Serum**	**Plasma**	**Serum**	**Plasma**
	IU/mL	**IU/mL**	**IU/mL**	**IU/mL**	**IU/mL**	**IU/mL**	**IU/mL**	**IU/mL**
Kit lot #298017	1.19	1.15	1.26	1.25	1.26	1.21	1.14	1.06
Kit lot #298018	1.30	1.31	1.25	1.18	1.29	1.22	1.22	1.20
Kit lot #298016	1.05	1.11	1.12	1.01	1.08	1.06	1.04	1.06

Overall Mean IU/mL	**1.17**

12.4 Detection of Antigen Subtypes:

Fifty cell culture supernatants of different HIV-1 group M subtypes (including CRF), HIV-1 group O and HIV-2 subtypes were tested using LIAISON® XL MUREX HIV Ab/Ag HT assay and a commercially available FDA-approved HIV comparator assay to determine the sensitivity of the assay on the detection of the different subtypes. These results in Table 4, demonstrates equivalent analytical sensitivity of LIAISON® XL MUREX HIV Ab/Ag HT assay to an FDA-approved comparator assay, as all samples were reactive on both.

Table 4. Summary of Detection of Antigen Subtypes

Cell culture supernatants subtype	Number of samples tested	Number of samples detected using LIAISON® XL MUREX HIV Ab/Ag HT	Number of samples detected using comparator assay
HIV-1 Group M, Subtype A	3	3	3
HIV-1 Group M, Subtype AE	10	10	10
HIV-1 Group M, Subtype AG	3	3	3
HIV-1 Group M, Subtype B	10	10	10
HIV-1 Group M, Subtype C	7	7	7
HIV-1 Group M, Subtype CRF06	1	1	1
HIV-1 Group M, Subtype D	3	3	3
HIV-1 Group M, Subtype F	5	5	5
HIV-1 Group M, Subtype G	2	2	2
HIV-1, Subtype O	4	4	4
HIV-2, Subtype A	1	1	1
HIV-2, Subtype B	1	1	1
TOTAL	**50**	**50**	**50**

12.5 Seroconversion Panels:

Thirty-eight commercially available HIV seroconversion panels were tested using LIAISON® XL MUREX HIV Ab/Ag HT assay and a commercially available FDA-

approved HIV comparator assay to determine the sensitivity of the assay. Thirty-six of the thirty-eight panels detected HIV at the same bleed. Two panels were detected one bleed later than the comparator. These results indicate equivalent sensitivity to an FDA-approved comparator assay. The results are summarized in Table 5.

Table 5. Summary of results obtained for Seroconversion Panels

Seroconversion Panel				First Confirmed Positive Bleed		Difference between LIAISON® XL MUREX HIV Ab/Ag HT and Comparator Assay
				LIAISON® XL MUREX HIV Ab/Ag HT	HIV Comparator Assay	
N	PANEL ID	Reactivity	Collection Days	(Day)	(Day)	Difference in First Positive Bleed (Bleed Number)
1	PRB969	Ag/Ab	0,29,48,53,55,61, 63,70,72,77	70	70	0
2	PRB966 (0600-0248)	Ag/Ab	0,2,20,22,30,35, 37,44,48,51	44	44	0
3	PRB968	Ag/Ab	0,3,8,10,15,17, 26,28,33,35	26	26	0
4	PRB971 (0600-0253)	Ag/Ab	0,2,7,11	7	7	0
5	PRB953 (0600-237)	Ag/Ab	0,3,7,10	7	7	0
6	PRB946 (0600-0227)	Ag	0,4,7,11	7	7	0
7	PRB950 (0600-0232)	Ag/Ab	0.18,21,28	18	18	0
8	PRB949 (0600-0230)	Ab	0,6,9,18	18	18	0
9	PRB976 (0600-0261)	Ag	0,2,7,9	7	7	0
10	PRB977 (0600-0262)	Ag/Ab	0,2,13,15	13	13	0
11	PRB956	Ag	0,40,42,47,50	47	47	0
12	PRB955 (0600-0239)	Ag/Ab	0,3,7,12,14	7	3	1
13	PRB975	Ag	0,2,7,9,14	14	14	0
14	PRB962	Ag	0,2,7,9,14,17	14	14	0
15	PRB967	Ag/Ab	0,3,7,17,19,24	17	17	0

Seroconversion Panel				First Confirmed Positive Bleed		Difference between LIAISON® XL MUREX HIV Ab/Ag HT and Comparator Assay
				LIAISON® XL MUREX HIV Ab/Ag HT	HIV Comparator Assay	
N	PANEL ID	Reactivity	Collection Days	(Day)	(Day)	Difference in First Positive Bleed (Bleed Number)
16	PRB963	Ag	0,2,7,9,14,17,21	17	17	0
17	PRB954 (0600-0238)	Ag/Ab	0,2,7,10,14,17,21	17	17	0
18	PRB961	Ag	0,5,7,12,14,19,21, 27,29	27	27	0
19	PRB960	Ag	0,4,7,11,14,18,21, 28,30	28	28	0
20	HIV9081	Ag/Ab	0,24,26,33	24	24	0
21	HIV9011	Ag/Ab	0,4,9,11,16,18,23, 25,30,38,40	38	38	0
22	HIV9012	Ag/Ab	0,2,7,9,14,16,21, 23	16	16	0
23	HIV9013	Ag	0,7,9,14,18,23,25	25	25	0
24	HIV9014	Ab	0,10,22,27,29	10	10	0
25	HIV9019	Ag/Ab	0,3,8	8	8	0
26	HIV9020	Ag/Ab	0,5,7,12,33,35,53, 55,61,63,68,70,75, 77,82,84,89,92,96, 99,103,106	99	99	0
27	HIV9021	Ag/Ab	0,3,7,11,14,18,21, 25,28,32,36,39,43, 47,50,54,57	47	47	0
28	HIV9022	Ag/Ab	0,3,7,10,15,17,23, 25,31	25	25	0
29	HIV9023	Ag	0,2,7,9,14,16,28, 30,35,37,42,44,55, 57,62,64,69,71,76, 78,83,85	83	78	1
30	HIV9025	Ag/Ab	0,7,14,23,28,37,58, 60,65,68,85,91	85	85	0
31	HIV9015	Ag/Ab	0,4,11,14,23,28	23	23	0
32	HIV9016	Ag	0,2,7,9,15,18,23, 27,30,34	30	30	0

Seroconversion Panel				First Confirmed Positive Bleed		Difference between LIAISON® XL MUREX HIV Ab/Ag HT and Comparator Assay
				LIAISON® XL MUREX HIV Ab/Ag HT	HIV Comparator Assay	
N	PANEL ID	Reactivity	Collection Days	(Day)	(Day)	Difference in First Positive Bleed (Bleed Number)
33	HIV9018	Ag/Ab	0,4,7,11,14,18,21, 25,28,32,35	28	28	0
34	HIV9026	Ag/Ab	0,5,13,15,26,31,44	44	44	0
35	HIV9030	Ag/Ab	0,4,7,11,14,18,21, 25,28,33,35,40,42, 47,49,54	47	47	0
36	HIV9033	Ag/Ab	0,11,14,18,21,25, 30,33,40,43,49,53, 60,63,82,84	82	82	0
37	HIV10234	Ag/Ab	0,3,38	38	38	0
38	HIV12007	Ag/Ab	0,2,54,117,119, 124,126,131,133	117	117	0

12.6 HIV Commercial Panels:

Forty-four samples from three commercially available HIV panels were tested to detect reactive samples to assess sensitivity of the LIAISON® XL MUREX HIV Ab/Ag HT assay. Three commercial panels were tested in (b) (4) on one lot of kit reagent, one lot of kit control and FDA-approved HIV comparator assay for comparison. Forty-two out of forty-four samples were reactive and two were non-reactive on both the LIAISON® XL MUREX HIV Ab/Ag HT assay and the FDA-approved comparator assay.
These results demonstrate equivalent sensitivity performance of the LIAISON® XL MUREX HIV Ab/Ag HT assay as compared to the comparator assay.

12.7 Precision:

A precision/reproducibility study was carried out over a twenty-day period on the LIAISON® XL MUREX HIV Ab/Ag HT assay using the LIAISON® XL Analyzer. The CLSI document EP5-A3, *"Evaluation of Precision of Quantitative Measurement Procedures, Third Edition"* was consulted in the preparation of the testing protocol.

Internal Reproducibility 20-day study:

The precision panel consisted of fourteen samples and fifteen controls for a total of 29 members for testing. A coded panel of fourteen serum samples consisted of two

Negative, three HIV-1 group M antibodies, three HIV-1 group O antibodies, three HIV-2 antibodies, and three HIV p24 antigen, which included, high negative, low positive and moderate positive samples. This panel was tested in two replicates per run, two runs per day for twenty days using three different LIAISON® XL MUREX HIV Ab/Ag HT assay reagents and three lots of the LIAISON® XL MUREX Control HIV Ab/Ag HT.

The testing was performed internally at DiaSorin S.p.A. on the LIAISON® XL Analyzer. As shown in Table 6, the Repeatability of the precision panel with the combined 3 lots of LIAISON® XL MUREX HIV Ab/Ag HT ranged from 2.2% to 4.8% and Within Laboratory CV% of the precision panel with the combined 3 lots of LIAISON® XL MUREX HIV Ab/Ag HT ranged from 5.5% to 24.2%.

Among the samples and controls with mean S/CO values between 0.80 and 2.00, there were 2 samples where the overall (within Site) variability exceeded 20% (HIV-1 O Ab Control lot 2 and HIV-1OAbU11) (Table 6). Root-cause analysis determined that the high imprecision was related to lot-to-lot variability specific to the HIV-1 Group O component of the assay. Improvements in the lot-to-lot variability of the HIV-1 Group O component will be achieved through tightening in-process QC control criteria. The impact of the changes will be reported through a post marketing commitment study.

Table 6. LIAISON® XL MUREX HIV Ab/Ag HT 20-Day Combined Lot Precision

Sample ID	N	Mean S/CO	Repeatability		Between Run		Between Day		Between Lot		Total	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
Negative Control lot 1	240	0.27	0.013	4.8	0.010	3.6	0.014	5.1	0.061	22.4	0.064	23.7
Negative Control lot 2	240	0.24	0.011	4.4	0.009	3.8	0.011	4.7	0.053	22.3	0.056	23.5
Negative Control lot 3	240	0.25	0.010	4.1	0.010	3.8	0.013	5.2	0.055	21.8	0.059	23.1
HIV p24 Ag Control lot 1	240	1.67	0.050	3.0	0.023	1.4	0.033	1.9	0.086	5.2	0.107	6.4
HIV p24 Ag Control lot 2	240	1.59	0.039	2.4	0.024	1.5	0.045	2.8	0.080	5.1	0.103	6.5
HIV p24 Ag Control lot 3	240	1.72	0.061	3.5	0.019	1.1	0.032	1.9	0.089	5.2	0.114	6.6
HIV-1 M Ab Control lot 1	240	1.29	0.029	2.2	0.038	3.0	0.034	2.7	0.038	3.0	0.070	5.5
HIV-1 M Ab Control lot 2	240	1.42	0.036	2.6	0.044	3.1	0.050	3.6	0.035	2.5	0.084	5.9
HIV-1 M Ab Control lot 3	240	1.39	0.051	3.7	0.026	1.9	0.027	1.9	0.051	3.7	0.081	5.9
HIV-1 O Ab Control lot 1	240	1.53	0.040	2.6	0.049	3.2	0.026	1.7	0.269	17.6	0.278	18.2

Sample ID	N	Mean S/CO	Repeatability		Between Run		Between Day		Between Lot		Total	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
HIV-1 O Ab Control lot 2	240	1.57	0.037	2.3	0.042	2.7	0.040	2.5	0.330	21.1	0.337	21.5
HIV-1 O Ab Control lot 3	240	1.63	0.063	3.9	0.027	1.6	0.025	1.5	0.315	19.4	0.323	19.9
HIV-2 Ab Control lot 1	240	1.19	0.027	2.3	0.039	3.2	0.016	1.3	0.154	12.9	0.162	13.6
HIV-2 Ab Control lot 2	240	1.11	0.028	2.5	0.027	2.5	0.017	1.6	0.140	12.6	0.146	13.1
HIV-2 Ab Control lot 3	240	1.05	0.035	3.3	0.016	1.5	0.013	1.3	0.140	13.3	0.146	13.9
Negative-U1	240	0.23	0.009	3.7	0.009	3.7	0.011	4.6	0.054	23.2	0.056	24.2
Negative-U2	240	0.24	0.010	4.1	0.006	2.7	0.010	4.3	0.055	23.3	0.057	24.2
HIV-1MAbU3	240	0.48	0.013	2.8	0.013	2.8	0.018	3.8	0.055	11.4	0.061	12.6
HIV-1MAbU4	240	1.10	0.025	2.3	0.032	2.9	0.039	3.5	0.046	4.2	0.073	6.6
HIV-1MAbU5	240	1.96	0.052	2.6	0.052	2.7	0.092	4.7	0.035	1.8	0.123	6.3
HIV-2AbU6	240	0.37	0.010	2.8	0.009	2.5	0.014	3.8	0.068	18.2	0.071	19.0
HIV-2AbU7	240	0.58	0.023	4.0	0.016	2.7	0.024	4.1	0.085	14.6	0.093	15.9
HIV-2AbU8	240	1.27	0.033	2.6	0.043	3.4	0.051	4.1	0.174	13.8	0.189	15.0
HIV-1OAbU9	240	0.37	0.010	2.6	0.010	2.6	0.013	3.6	0.037	9.9	0.042	11.2
HIV-1OAbU10	240	1.05	0.028	2.7	0.021	2.0	0.031	2.9	0.157	14.9	0.163	15.6
HIV-1OAbU11	240	1.76	0.038	2.2	0.047	2.7	0.039	2.2	0.346	19.7	0.354	20.1
HIVp24AgU12	240	0.48	0.013	2.7	0.011	2.3	0.015	3.1	0.055	11.5	0.059	12.4
HIVp24AgU13	240	0.57	0.022	3.9	0.011	2.0	0.017	3.0	0.051	8.9	0.059	10.3
HIVp24AgU14	240	2.14	0.055	2.6	0.040	1.8	0.071	3.3	0.105	4.9	0.144	6.7

External Reproducibility 5-Day Study:

A precision/reproducibility study was conducted for five days at three different sites, two external and one internal with three different lots of LIAISON® XL MUREX HIV Ab/Ag HT assay and one lot of LIAISON® XL MUREX Control HIV Ab/Ag HT which was used in the twenty-day study. The CLSI document EP15-A3, *"User Verification of Precision and Estimation of Bias, Third Edition"* was followed for the testing protocol. The coded panel comprised of the fourteen frozen serum samples described above and the controls were tested, using six replicates per run, in one run per day for five operating days with multiple technicians performing the testing.

The data in Table 7 demonstrate that the total observed (combined lot) reproducibility (%CV) ranged from 5.3–21.2%, which is within the acceptance criteria except for one sample, where it exceeded 20%. The root cause of the high variability in this component is lot-to-lot variability of the HIV-1 Group O component; its mitigation is described above.

Table 7. 5-Day LIAISON® XL MUREX HIV Ab/Ag HT Reproducibility Results (combined 3 sites)

Sample ID	N	Mean S/CO	Repeatability		Between Days/Runs		Within Laboratory Precision		Between Sites/Lots		Reproducibility	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
HIVp24 Ag Control	90	1.760	0.065	3.7	0.056	3.2	0.086	4.9	0.156	8.9	0.178	10.1
HIV-1 M Ab Control	90	1.410	0.063	4.5	0.053	3.8	0.083	5.9	0.127	9.0	0.151	10.7
HIV-1 O Ab Control	90	1.747	0.062	3.5	0.055	3.1	0.083	4.7	0.360	20.6	0.370	21.2
HIV-2 Ab Control	90	1.165	0.045	3.9	0.015	1.3	0.048	4.1	0.144	12.4	0.152	13.1
Negative Control	90	0.255	0.011	4.4	0.014	5.3	0.018	6.9	0.030	11.7	0.035	13.6
Negative-U1	90	0.234	0.010	4.3	0.020	8.7	0.023	9.7	0.040	17.3	0.046	19.8
Negative-U2	90	0.232	0.011	4.5	0.016	6.8	0.019	8.2	0.041	17.6	0.045	19.4
HIV-1MAbU3	90	0.476	0.054	11.4	0.030	6.2	0.062	13.0	0.000	0.0	0.060	12.7
HIV-1MAbU4	90	1.097	0.054	4.9	0.045	4.1	0.070	6.4	0.071	6.4	0.100	9.1
HIV-1MAbU5	90	1.955	0.092	4.7	0.158	8.1	0.183	9.3	0.199	10.2	0.270	13.8
HIV-2AbU6	90	0.402	0.017	4.2	0.004	0.9	0.017	4.3	0.044	11.0	0.048	11.9
HIV-2AbU7	90	0.619	0.032	5.2	0.035	5.6	0.048	7.7	0.064	10.4	0.080	12.9
HIV-2AbU8	90	1.346	0.082	6.1	0.115	8.6	0.142	10.5	0.154	11.4	0.209	15.5
HIV-1OAbU9	90	0.395	0.059	15.0	0.004	0.9	0.059	15.0	0.007	1.8	0.060	15.2
HIV-1OAbU10	90	1.122	0.052	4.6	0.044	4.0	0.068	6.1	0.167	14.9	0.181	16.1
HIV-1OAbU11	90	1.866	0.095	5.1	0.114	6.1	0.149	8.0	0.334	17.9	0.366	19.6
HIVp24AgU12	90	0.478	0.017	3.5	0.016	3.3	0.023	4.8	0.010	2.2	0.025	5.3
HIVp24AgU13	90	0.572	0.023	4.0	0.037	6.5	0.044	7.6	0.013	2.3	0.046	8.0
HIVp24AgU14	90	2.066	0.105	5.1	0.135	6.5	0.171	8.3	0.173	8.4	0.243	11.8

Overall, the data indicate that the precision and reproducibility of the assay is acceptable.

B. ANALYTICAL SPECIFICITY

12.8. Endogenous Interference Study:

Testing was performed in accordance with CLSI Documents EP07-A3, "*Interference Testing in Clinical Chemistry, Third Edition*" and EP7-A3, "*Supplementary Tables for Interference Testing*" to evaluate the interference of the endogenous substances with LIAISON® XL MUREX HIV Ab/Ag HT assay results. Table 8 presents the list of endogenous substances or drugs and the relative concentration used for testing.

In order to evaluate analytical specificity in the presence of endogenous substances, ten negative samples were spiked to reach two levels (high negative and low positive) for each marker (HIV p24 antigen, HIV-1 and HIV-2 antibodies). As a result, two spiked sets of samples were generated for each marker: five high negative samples and five low positive samples. Samples were tested in twenty-six replicates with one lot of kit reagent and one lot of kit control. Each sample was tested in parallel with the reference sample in the same run, in order to exclude the effect of other variables on the results. These results demonstrate no interference at the levels tested with the LIAISON® XL MUREX HIV Ab/Ag HT assay.

Table 8. Summary of Endogenous Substances used in the Interference Study

Substance	Tested Concentrations
Lipids (Glyceryl trioleate)	3000 mg/dL
Lipids (Intralipid)	1000 mg/dL
Hemoglobin	2.5 g/L
Unconjugated bilirubin	40 mg/dL
Conjugated bilirubin	40 mg/dL
Albumin	6 g/dL
Cholesterol total	400 mg/dL
Immunoglobulin G	20 g/L
Biotin (Vitamin H)	3510 ng/mL
Total Protein	150 g/L

12.9 Potential Interfering Common Drugs:

Testing was performed to determine whether the presence of most common used drugs may interfere with assay results. The CLSI Guidance "*Interfering Substances*" (EP7-A3)

and *"Supplementary Tables for Interference Testing in Clinical Chemistry"* (EP37) was followed in the preparation of the testing protocol. Table 9 presents the list of most common used drugs and the relative concentration used for testing.

These results from the studies of potentially interfering substances at two levels (high negative and low positive) for each marker (HIV p24 antigen, HIV-1 group M, HIV-1 group O and HIV-2 antibodies) indicated no interference at the concentration for each substance listed below with the LIAISON® XL MUREX HIV Ab/Ag HT assay.

Table 9. Summary of Potentially interfering substances

Substance	Concentrations tested (mg/dL)
Acetylcysteine	41.5
Ampicillin-Na	100
Ascorbic acid	30
Cyclosporine	0.5
Cefoxitin	660
Heparin	5000 (U/L)
Levodopa	2
Methyldopa+1.5	2.25
Metronidazole	20
Phenylbutazone	40
Doxycycline	5
Acetylsalicylic acid	100
Rifampicin	6
Acetaminophen	20
Ibuprofen	50
Theophylline	10
Tetracycline	5
Ca-Dobesilate	20

12.10 Cross-Reactivity:

The LIAISON® XL MUREX HIV Ab/Ag HT assay was evaluated for analytical specificity study for potentially interfering medical conditions unrelated to HIV infection by spiking antigen and antibody (HIV p24 antigen, HIV-1 Group O and HIV-2 antibodies). All

samples used for this study were negative for HIV p24 antigen and/or HIV antibodies. Each sample was divided in two aliquots: one was spiked with an HIV-positive sample to reach a low level of reactivity and one remained un-spiked.

All un-spiked samples and all spiked samples were tested by LIAISON® XL MUREX HIV Ab/Ag HT assay, and FDA-approved comparator assay in singlicate.

The LIAISON® XL MUREX HIV Ab/Ag HT assay results indicate suitable specificity performance for each of the potentially cross-reactant pathologies, as results were comparable to the results of the comparator assay.

The data presented here demonstrate no interference with the potential cross-reactants at the concentrations tested (Table 10).

Table 10. Cross Reactivity by Unrelated Medical Conditions

Potential Cross Reactant	Spiked samples – N of observed Reactive results with LIAISON® XL MUREX HIV Ab/Ag HT				Samples Non-Reactive by Comparator and LIAISON® XL HIV Ab/Ag
	HIV-1M	HIV-1 O	HIV-2	HIV p24 Ag	
Anti-nuclear antibodies (ANA)	10/10	10/10	10/10	10/10	All
E. Coli (anti-*E.Coli* positive)	9/9	4/4	4/4	4/4	All
CMV (anti-CMV positive IgG and IgM)	10/10	10/10	10/10	10/10	All
Common Cold	9/9	3/3	3/3	3/3	All
Crohn's Disease	10/10	7/7	7/7	7/7	All
C. Trachomatis	10/10	8/8	8/8	8/8	All
EBV (anti-EBV positive IgG and IgM)	10/10	10/10	10/10	10/10	All
Elevated IgG	11/11	10/10	10/10	10/10	All
Elevated IgM	10/10	9/9	9/9	9/9	All
Fungal Infections	6/6	10/10	10/10	10/10	All
Graves Disease	10/10	9/9	9/9	9/9	All
HAMA	10/10	10/10	10/10	10/10	All
Hemodialysis patient	9/9	10/10	10/10	10/10	All
Hepatitis A Virus (anti-HAV positive IgG/IgM)	8/8	10/10	10/10	10/10	All
Hepatitis B Virus (anti-HBV positive PCR)	10/10	10/10	10/10	10/10	All
Hepatitis C Virus (anti-HCV positive)	10/10	10/10	10/10	10/10	All
HSV (anti-HSV positive IgG)	9/9	10/10	10/10	10/10	All
HTLV-1/2 (anti-HTLV positive)	10/10	8/8	8/8	8/8	All

Potential Cross Reactant	Spiked samples – N of observed Reactive results with LIAISON® XL MUREX HIV Ab/Ag HT				Samples Non-Reactive by Comparator and LIAISON® XL HIV Ab/Ag
	HIV-1M	HIV-1 O	HIV-2	HIV p24 Ag	
IgM monoclonal gammopathy	2/2	2/2	2/2	2/2	All
Influenza vaccine recipients	10/10	10/10	10/10	10/10	All
Multiparous pregnancies	10/10	10/10	10/10	10/10	All
Pregnancy 1st trimester	9/9	10/10	10/10	10/10	All
Pregnancy 2nd trimester	8/8	10/10	10/10	10/10	All
Pregnancy 3rd trimester	9/9	9/9	9/9	9/9	All
Rheumatoid Factor	9/9	10/10	10/10	10/10	All
Rubella Virus	10/10	10/10	10/10	10/10	All
Systemic Lupus Erythematosis	9/9	6/6	6/6	6/6	All
T. pallidum (anti-*T.pallidum* positive)	8/8	10/10	10/10	10/10	All
Varicella Zoster Virus (anti-VZV positive IgG)	8/8	10/10	10/10	10/10	All
TOTAL	**263/263**	**255/255**	**255/255**	**255/255**	All

12.11 Sample Equivalence/Matrix Effect:

Thirty paired sets of matched serum (with and without gel SST) and plasma (lithium and sodium heparin, sodium citrate and K2-EDTA) were tested to determine if these sample types provide equivalent results on the LIAISON® XL MUREX HIV Ab/Ag HT assay. Panel was tested for HIV p24 antigen and for HIV antibodies. Each member of the panel was divided into three aliquots. Two sets of aliquots were spiked with an HIV-positive sample to achieve two levels of samples: high negative and low positive samples. The third set of aliquots was un-spiked to serve as control samples. The results of the negative and low positive samples did not change the classification of the expected result. The results in Table 11 with the serum-plasma paired samples indicate that equivalent performance among serum (with and without Gel SST), K2-EDTA, Lithium Heparin and Sodium Heparin and Sodium Citrate plasma matrices.

Table 11. Summarized Results for Sample Equivalence Test Compared to Serum

X reference	Serum without Gel SST				
y exam	**Serum with Gel SST**	**K2 EDTA**	**Na Citrate**	**Li Heparin**	**Na Heparin**
Slope (Passing Bablok fit)	1.000	0.9861	1.011	1.039	0.9958

X reference	Serum without Gel SST				
y exam	**Serum with Gel SST**	**K2 EDTA**	**Na Citrate**	**Li Heparin**	**Na Heparin**
Intercept (Passing Bablok fit)	0.009	0.00898	-0.006915	-0.02276	0.005142
Intercept (95% CI)	-0.03302 to 0.04977	-0.03520 to 0.05594	-0.05083 to 0.02433	-0.07395 to 0.03007	-0.04185 to 0.05387
Correlation	0.934	0.932	0.948	0.932	0.944

12.12 Carry Over Study:

The LIAISON® XL Analyzer uses disposable tips for sample pipetting. A carry-over study was performed to evaluate whether any significant amount of analyte is carried over from one sample reaction cuvette into the subsequent sample reaction cuvettes. Two sample types were used for this evaluation: one anti-HIV negative serum and one high positive anti-HIV serum sample. The negative sample was divided into (b) (4) aliquots and tested in (b) (4) in (b) (4) separate runs.

An additional (b) (4) separate runs of the following sequence were performed:
High Pos, Neg Aliquot 1, High Pos, Neg Aliquot 2, High Pos, Neg Aliquot 3, High Pos, Neg Aliquot 4, High Pos, Neg Aliquot 5.

The data indicates that all acceptance criteria were fulfilled with no detectable level of carry-over/cross contamination.

12.13 Hook Effect:

Three recombinant protein HIV p24 antigen samples, with high analyte levels and three different high antibody samples for each reactivity (HIV-1 group M, HIV-1 group O and HIV-2) were tested neat and after serial dilutions in normal human negative serum. Dilutions were prepared to cover the whole assay range and they were tested in triplicate with one reagent lot, using one sets of kit controls. S/CO values were plotted versus the analyte concentration.

For HIV p24 antigen, a high-dose hook was observed for one sample at a dilution greater than a S/CO value of 1540 and no high-dose hook effect was observed up to a S/CO value up to1540. No reduction of signal was observed for any of the three high analyte level samples tested for each of the HIV antibody reactivities, demonstrating that no high dose hook effect occurs on this assay.

C. STABILITY STUDIES

12.14.1 Sample Stability:

Studies were performed to determine the storage stability of patient serum and plasma samples at storage temperatures of 2–8°C, room temperature (RT), -20°C. A multiple

freeze/thaw (F/T) study was also performed. Serum and plasma samples tested were (b) (4) .

- 2–8°C study – samples were tested unstressed (T=0), and again after 1, 2, 3, 5, 7 (b) (4) days of storage at 2–8°C.
- Room temperature study (RT) - samples were tested immediately after preparation and again after 1, 2, 3, (b) (4) days of storage at RT (b) (4) .
- -20°C study – samples were tested unstressed (T=0) and stored at -20°C or lower (b) (4) .
- Freeze/Thaw (F/T) study – samples were tested unstressed (T=0) and after 1, 2, 3, 4, 5, 6, 7 (b) (4)

The studies demonstrated that serum and plasma samples are stable for:

- 7 days at 2–8°C
- 3 days at room temperature
- (b) (4) at -20°C
- 7 Freeze/Thaw cycles.

12.14.2 Kit Stability:

Real-Time (Shelf-Life):

Stability Studies were performed at t=0 and designated time points with testing session composed of (b) (4) , testing Controls and Stability Sample panel in (b) (4) , using freshly opened integrals, to establish the shelf-life for the LIAISON® XL MUREX HIV Ab/Ag HT assay. Testing was performed on three lots of LIAISON® XL MUREX HIV Ab/Ag HT integrals and using three lots of LIAISON® XL MUREX Control HIV Ab/Ag HT on three LIAISON® XL Analyzer. Each time point was calculated from the manufacturing date of the last manufactured component.

The long-term stability study was targeted for (b) (4) months for all three validation lots. The data evaluation crossed the lower boundary of (b) (4) acceptance criteria between the (b) (4) month and (b) (4) month time point. Shelf life stability for the LIAISON® XL MUREX HIV Ab/Ag HT assay is 13 months.

Reagent On-Board:

The study was carried out to assess the stability of the product by simulating normal conditions of use as specified in the instructions for use (i.e., storage on board the instrument in the refrigerated area). Tests were performed as close as possible to time zero.

Testing was performed on three lots of reagent integrals and two lots of Controls. Enough Reagent Integral were opened at time zero, seals removed: then one kit was placed on board the analyzer and Precision Panel and kit controls (just open) were run

in (b) (4) . The opened Reagent Integrals were then stored at 2–8°C in a refrigerator. Kit performance using the opened Reagent Integral was done (b) (4) with a kit of the same lot but stored closed at the recommended temperature of 2–8°C. The LIAISON® XL MUREX HIV Ab/Ag assay was stored on-board the LIAISON® XL Analyzer throughout the (b) (4) of the study. The LIAISON® XL MUREX Control HIV Ab/Ag HT along with the internal stability panel were tested in (b) (4) .

The data submitted support a claim of On-Board stability for kit LIAISON® XL MUREX HIV Ab/Ag HT of 5 weeks.

Calibration Stability:

A study was conducted to assess the stability of the calibration interval by simulating the normal condition of use as specified in the instruction for use. The Reagent Integral was stored on board the analyzer in the refrigerated reagent bay. Testing was performed on the stability panel and two lots of LIAISON® XL MUREX Control HIV Ab/Ag HT in (b) (4) on three lots of LIAISON® XL MUREX HIV Ab/Ag HT reagents.

Kit performance was evaluated weekly up to (b) (4) . Results were generated using the initial (time zero) assay calibration. To establish calibration stability the LIAISON® XL MUREX Control HIV Ab/Ag HT and the stability panel must be within their established ranges throughout the study.

For all three lots, the claimed calibration stability for the LIAISON® XL MUREX HIV Ab/Ag HT is 5 weeks.

Reagent Open Use Stability:

The aim of this study was to assess the open use stability of the LIAISON® XL MUREX HIV Ab/Ag HT kit reagents by simulating normal conditions of use as specified in the Instructions for Use.

Testing of samples was performed in (b) (4) , with the LIAISON® XL MUREX HIV Ab/Ag HT assay and the LIAISON® XL MUREX Control HIV Ab/Ag HT. Calibration was performed as required according to the defined calibration interval. After testing the opened Reagent Integral was removed from the XL Analyzer and stored at 2–8 °C until the next testing time point. Kit performance using the opened Reagent Integral was evaluated (b) (4) weeks. To establish the open use stability the kit controls and internal stability panel must be within their established ranges throughout the study and deviation must be less than (b) (4) from time zero.

The data provided support the claimed Open kit stability for kit LIAISON® XL MUREX HIV Ab/Ag HT of 5 weeks after opening at 2–8°C.

12.14.3 Kit Control stability:

Real-time (Shelf-Life):

Stability Studies were performed at t=0 and designated time points with testing session composed of (b) (4) , testing Controls and Stability Sample panel in (b) (4) , using freshly opened integrals, to establish the shelf-life for the LIAISON® XL MUREX Control HIV Ab/Ag HT. Testing were performed on three lots of LIAISON® XL MUREX Control HIV Ab/Ag HT and using two lots of LIAISON® XL MUREX HIV Ab/Ag HT integral on three LIAISON® XL platforms. Each time point was calculated from the manufacturing date of the last manufactured component.

The data provided support the shelf life stability claim for the controls of 18 months.

Open use Kit Control:

The aim of this study was to assess stability of the opened Control vials by simulating normal conditions of use, as specified in the instruction for use. Once opened the controls were stored for at least (b) (4) on the LIAISON® XL Analyzer, then returned to 2–8°C until the next testing time point. Three lots of LIAISON® XL MUREX Control HIV Ab/Ag HT were tested in (b) (4) , on two lots of LIAISON® XL MUREX HIV Ab/Ag HT.

The data provided support the claimed Stability of Controls after Opening of 9 weeks.

12.15 Animal Studies:

Not applicable.

12.16 Additional Studies:

Not applicable.

13. SUMMARY OF CLINICAL STUDIES

13.1 Clinical Agreement Study:

A multi-site clinical agreement study was conducted to determine the clinical performance of the LIAISON® XL MUREX HIV Ab/Ag HT assay on samples that would routinely be tested for HIV including pregnant women, samples that were selected from individuals diagnosed with an HIV infection, and samples that were selected from individuals that were presumably HIV antibody and/or antigen positive.

The clinical performance study was conducted using 9,578 samples. Of the 5,759 low risk population, 550 samples were collected fresh. One hundred and fifty of the fresh specimens collected for analysis were plasma (26.8%) and the remaining 400 were serum.

Out of the total samples, 8035 were prospective (Table 12) and 1543 retrospective (Table 13), including pregnant women, and were obtained from eight U.S. commercial vendors. The prospective subjects were 43.8% female (3518), 55.7% male (4474) and

0.5% (43) of unknown gender with an age range of 2–88 yrs. Two specimens from the prospective population “Individuals at for high risk HIV-2” were excluded due to technical error, bringing the total number of prospective samples tested to 8033. The retrospective subjects were 29.0% female (448) and 71.0% male (1095) with an age range of 2-81 years and 11 samples of unknown age. Prespecified inclusion and exclusion criteria were followed and informed consent procedures and IRB approvals were used for the prospective collections.

The samples were collected from 15 different countries including Argentina (n=24; 0.3%), Cameroon (n=47; 0.5%), Colombia (n=72; 0.8%), Congo (n=11; <0.1%), Cote d'Ivoire (n=628; 6.6%), Democratic Republic of Congo (69; 0.7%), Dominican Republic (506; 5.3%), Guinea-Bissau (14; 0.1%), Mexico (1; <0.1%), Nigeria (565; 5.9%), Peru (1777; 18.6%), Sierra Leone (1; <0.1%), South Africa (2; <0.1%), Ukraine (1; <0.1%), and the United States (5760; 60.1%). One hundred (100; 1.0%) AIDS specimens with CDC classification were purchased with unknown country of origin. The specimens collected in the United States were from multiple states including California, Florida, Indiana, New Jersey, New York, Pennsylvania, Texas, and Virginia.

The 8,035 prospective (unselected) adult subjects were of American Indian/Alaskan Native (1.5%), Asian (0.7%), Black/African American (33.1%), Caucasian/White (57.4%), Native Hawaiian or Pacific Islander (<0.1%), Other (1.8%), and Unknown (5.5%) ethnicities and were collected from the low risk, High risk, adults, pediatric and pregnant population, as summarized in Table 12 and 14. Pregnant females less than or equal to 21 years of age were included in both the pediatric prospective population and in the pregnant population, and samples of unknown age are included in the adult population.

The 1,543 retrospective (selected/archived) specimens were of Black/African American (19.0%), Caucasian/White (7.78%), Unknown (68.1%), and Other (5.1%) ethnicities and were collected from the populations summarized in Table 13.

The clinical agreement study was conducted at seven sites. Samples provided to the sites were de-identified/delinked and had a unique barcode identifier used by DiaSorin. The samples were randomly distributed across the testing sites.

Table 12: Characteristics of Prospective Samples

Type of Sample	Total Number of Samples Tested
Individuals at Low-Risk for HIV-1 infection	4,380
Individuals at High-Risk for HIV-1 infection	983
Individuals at High-Risk for HIV-2 infection	500
Pediatrics at Low-Risk for HIV-1 infection	983

Type of Sample	Total Number of Samples Tested
Pediatrics at High-Risk for HIV-1infection	389
High Risk pregnant subjects	404
Low Risk pregnant subjects	396
Total	**8,035***

*Two contaminated samples at high-risk for HIV-2 were discarded

Table 13: Characteristics of Retrospective Samples

Type of Sample	Total Number of Samples Tested
Individuals Diagnosed with AIDS (CDC stage classification)	100
HIV-1 Antigen Positive	50
HIV-1 Antigen Positive with Subtypes	49
HIV-1 Group O Infected	47
HIV-1 Infected Pediatrics	61
HIV-1 Infected Pregnant Subjects	51
HIV-1 Infected Subjects	985
HIV-2 Infected Subjects	200
Total	**1543**

Table 14: Summary of Sample Demographics

Gender	Adult*				Pediatric (2-21)				Unknown Age			
	Prospective		Retrospective		Prospective		Retrospective		Prospective		Retrospective	
	N	%	N	%	N	%	N	%	N	%	N	%
Female	2947	45.4	407	28.1	563	38.0	37	45.1	7	12.5	3	23.1
Male	3549	54.6	1040	71.9	918	62.0	45	54.9	6	10.7	10	76.9
Unknown	0	0.0	0	0.0	0	0.0	0	0.0	43	76.8	0	0.0
Total	**6496**	**100**	**1447**	**100**	**1481**	**100**	**82**	**100**	**56**	**100**	**13**	**100**

*including 2 prospective specimens excluded from all testing due to potential cross-contamination; 1 QNS for complete testing.

13.2 Clinical Agreement Study Result Interpretation:

HIV infection status determination was based on testing all prospective (8,033) and retrospective (1,543) specimens with FDA approved HIV assays according to the CDC recommended laboratory HIV status algorithm for serum or plasma specimens.

The LIAISON® XL MUREX HIV Ab/Ag HT results for the specimens were compared to an FDA-approved confirmatory test to determine the HIV Infection status per the CDC algorithm. Repeatedly reactive samples were investigated further with FDA-approved supplemental assays tests that detect and differentiate HIV-1 and HIV-2 antibodies. Samples that were Indeterminate or Negative for either or both HIV-1 and HIV-2 antibodies were tested with an FDA-approved HIV-1 RNA assay for detection of HIV-1 virus as indicated in the tables when applicable. Samples that were non-reactive with the RNA assay were classified as HIV negative.

The 9,576 samples were tested with the LIAISON® XL MUREX HIV Ab/Ag HT assay on the LIAISON® XL Analyzer. The samples were tested at two different external testing sites and internally at DiaSorin Inc.

13.2.1 Interpretation of Results:

The presence or absence of HIV p24 antigen and/or HIV-1 and/or HIV-2 antibodies in the specimens was determined by comparing the chemiluminescence reaction signal to the preliminary cut-off value provided by the assay calibration. The analyzer automatically calculates the signal-to-cutoff (S/CO) ratios, then interprets the results. The cutoff discriminating between the reactivity to HIV p24 antigen and/or HIV specific antibodies has a S/CO value of 1.00.

Results are interpreted as follows in Table 15 and 16:

Table 15: LIAISON® XL MUREX – Initial Interpretation of Results

Initial Result		
Initial Result (S/CO)	**Instrument Interpretation**	**Retest Procedure**
< 1.00	NONREACTIVE (NR)	No retest required
≥ 1.00	REACTIVE (R)	Retest in duplicate

Note - All specimens that are initially reactive must be centrifuged and retested in duplicate.

Table 16: LIAISON® XL MUREX – Final Interpretation of Results

Final Interpretation			
Initial Result (S/CO)	**Retest Results (S/CO)**	**Final Result**	**Interpretation of Final Result**
NR (<1.00)	No retest required (NA)	NR	HIV p24 Ag and HIV-1/HIV-2 Ab not detected
R (≥ 1.00)	Both tests are NR (<1.00)	NR	HIV p24 Ag and/or HIV-1/HIV-2 Ab not detected
R (≥ 1.00)	One or both tests are R (≥ 1.00)	R	Presumptive evidence of HIV p24 Ag and/or HIV-1/HIV-2 Ab; perform supplemental confirmatory assay(s)

13.3 Clinical Agreement Study Results:

13.3.1 Prospective Adult Population:

Results of Prospective U.S. Adult Subjects:

The performance of the LIAISON® XL MUREX HIV Ab/Ag HT and an FDA approved comparator on the prospectively collected samples from 3767 individuals from US Adult population is presented in Table 17. All confirmed positive samples by the HIV infection status were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay.

Table 17. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity in U.S. Adult Prospective Population and Subjects of Unknown Age

		LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
Specimen Population	**N**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**HIV-1**	**HIV-2**	**UCR**		
Low Risk	2015[A]	2009	6	6	2010	6	5	0	0	0	NA	(2009/2015) 99.7% *99.4%-99.9%*
Low Risk Fresh*	527	525	3	2	526	1	1	0	0	0	NA	(525/527) 99.6% *98.6%-99.9%*
Low Risk Pregnant	367	367	0	0	367	0	0	0	0	0	NA	(367/367) 100.0% *99.0%-100.0%*

		LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
Specimen Population	**N**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**HIV-1**	**HIV-2**	**UCR**		
High Risk	495[B]	478	17	17	475	20	20	16	0	0	(16/16) 100.0% *80.6%-100.0%*	(478/479) 99.8% *98.8%-100.0%*
High Risk Pregnant	363	351	12	12	354	9	9	4	0	0	(4/4) 100.0% *51.0%-100.0%*	(351/359) 97.8% *95.7%-98.9%*
TOTAL	**3767**	**3730**	**38**	**37**	**3732**	**36**	**35**	**20**	**0**	**0**	**(20/20) 100.0%** ***83.9%-100.0%***	**(3730/3747) 99.5%** ***99.3%-99.7%***

[A] 1980 adults and 35 of unknown age
[B] 493 adults and 2 of unknown age
*Samples collected and not frozen prior to testing; UCR = Untypable Cross Reactive

Results of Prospective Non-U.S. Adult Subjects:

The performance of the LIAISON® XL MUREX HIV Ab/Ag HT and an FDA approved comparator on the prospectively collected samples from 2785 individuals from non-US Adult population is presented in Table 18. Confirmed HIV-positive samples were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay, except for two samples.

Table 18. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity in Non-U.S. Adult Prospective Population and Subjects of Unknown Age

		LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			FDA Approved HIV-1 RNA Reactive	LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
Specimen Population	**N**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**HIV-1**	**HIV-2**	**UCR**			
Low Risk	1838[B]	1690	148	148	1701	139	137	92	0	0	12	(104/104) 100% *96.4%-100.0%*	(1690/1734) 97.5% *96.6%-98.1%*
High Risk	488[A]	471	17	17	471	17	17	13	0	0	1*	(13/14) 92.9% *92.9%-98.7%*	(470/474) 99.2% *97.9%-99.7%*
High Risk HIV-2	459	369	90	88	366	93	93	67[†]	2	15	1[**]	(84/85) 98.8% *93.6%-99.8%*	(370/374) 98.9% *97.3%-99.6%*
TOTAL	**2785**	**2530**	**255**	**253**	**2538**	**249**	**247**	**172**	**2**	**15**	**14**	**(201/203) 99.0%** ***96.5%-99.7%***	**(2530/2582) 98.0%** ***97.4%-98.5%***

[A] 448 adults and 11 of unknown age
[B] 1830 adults and 8 of unknown age
UCR = Untypable Cross Reactive
*One High risk sample confirmed positive from Nigeria was negative by LIAISON® XL MUREX HIV Ab/Ag HT
**One High risk HIV-2 sample from Cote d'Ivoire was negative by the LIAISON® XL MUREX HIV Ab/Ag HT assay, positive for HIV-1 on the FDA Approved HIV Ag/Ab Combo Assay as well as the HIV1/2 differentiation assay but was negative on the FDA Approved HIV-1 RNA PCR.

13.3.2 Prospective Pediatric Population:

Results of Pediatric Prospective U.S. Subjects:

The performance of the LIAISON® XL MUREX HIV Ab/Ag HT and an FDA approved comparator on the prospectively collected samples from 946 individuals from the US Pediatric population is presented in Table 19. The one confirmed HIV-positive sample was correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay.

Table 19. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity in U.S. Pediatric Prospective Population

		LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
Specimen Population	**N**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**HIV-1**	**HIV-2**	**UCR**		
Low Risk Pediatric	548	547	1	1	546	3	2	0	0	0	NA	(547/548) 99.8% *99.0%-100.0%*
Low Risk Pediatric Fresh*	23	23	0	0	23	0	0	0	0	0	NA	(23/23) 100.0% *85.7%-100.0%*
Low Risk Pregnant (≤ 21 yrs)	29	29	0	0	29	0	0	0	0	0	NA	(29/29) 100.0% *88.3%-100.0%*
High Risk Pediatric	305	303	3	2	304	1	1	1	0	0	(1/1) 100.0%	(303/304) 99.7% *98.2%-99.9%*
High Risk Pregnant (≤ 21 yrs)	41	40	1	1	41	0	0	0	0	0	NA	(40/41) 97.6% *87.4%-99.6%*
TOTAL	**946**	**942**	**5**	**4**	**943**	**4**	**3**	**1**	**0**	**0**	**(1/1) 100.0%**	**(942/945) 99.7%** ***99.1%-99.9%***

*Samples collected and not frozen prior to testing; UCR = Untypable Cross Reactive

Results of Pediatric Prospective Non-U.S. Subjects:

The performance of the LIAISON® XL MUREX HIV Ab/Ag HT and an FDA approved comparator on the Prospectively collected samples from 535 individuals from non-US Pediatric population is presented in Table 20. All confirmed HIV-positive samples were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay. One high risk pediatric (U.S. population) sample positive by the HIV 1 /2 Differentiation assay was correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay.

Table 20. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity in Non-U.S. Pediatric Prospective Population

		LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
Specimen Population	**N**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**HIV-1**	**HIV-2**	**UCR**		
Low Risk Pediatric	412	408	4	4	410	2	2	0	0	0	NA	(408/412) 99.0% *97.5%-99.6%*
High Risk Pediatric	84	84	0	0	84	0	0	0	0	0	NA	(84/84) 100.0% *95.6%-100.0%*
High Risk HIV-2 Pediatric	39	27	12	12	26	13	13	11	1	0	(12/12)100.0% *75.7%-100.0%*	(27/27) 100.0% *87.5%-100.0%*
TOTAL	**535**	**519**	**16**	**16**	**520**	**15**	**15**	**11**	**1**	**0**	**(12/12)100.0% *77.5%-100.0%***	**(519/523) 99.2% *98.0%-99.7%***

UCR = Untypable Cross Reactive

13.3.3 Retrospective Adult Population:

Results of Retrospective U.S. Adult Subjects:

Reactivity of 1000 individuals from the retrospectively collected US (Adults) population from presumably HIV-1 positive subjects.

The performance of the LIAISON® XL MUREX HIV Ab/Ag HT and an FDA approved comparator on the retrospectively collected US Adult population presumably HIV-1 positive individuals are presented in Table 21. All confirmed HIV-positive samples were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay.

Table 21. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity in the Presumably HIV-1 Positive Retrospective U.S. Adult Population

		LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			FDA Approved HIV-1 RNA Reactive	LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
Specimen Population	**N**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**HIV-1**	**HIV-2**	**UCR**			
HIV-1 Positive	985	0	985	985	0	985	985	983	0	0	1	(984/984) 100.0% *99.6%-100.0%*	(0/1) 0.0% *0.0%-79.3%*
HIV-1 Infected Pregnant	10	0	10	10	0	10	10	10	0	0	0	(10/10) 100.0% *72.2%-100.0%*	NA
HIV-1 Antigen Positive w / Subtype ID	5	0	5	5	0	5	5	5	0	0	0	(5/5)100.0%	NA
TOTAL	**1000**	**0**	**1000**	**1000**	**0**	**1000**	**1000**	**998**	**0**	**0**	**1**	**(999/999)100.0%** ***99.6%-100.0%***	**(0/1) 0.0%** ***0.0%-79.3%***

UCR = Untypable Cross Reactive

Results of Retrospective Non-U.S. Adult Subjects:

Reactivity of 360 individuals from the retrospectively collected non-US (Adults) population from presumably HIV-1 positive subjects.

The performance of the LIAISON® XL MUREX HIV Ab/Ag HT and an FDA approved comparator on the retrospectively collected non-US Adult population presumably HIV-1 positive individuals are presented in Table 22. All confirmed HIV-positive samples were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay

Table 22. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity in the Presumably HIV-1 Positive Retrospective Non-U.S. Adult Population and Subjects of Unknown Age

		LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
Specimen Population	**N**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**HIV-1**	**HIV-2**	**UCR**		
HIV-2 Positive	200[A]	0	200	200	0	200	200	3	67	127	(197/197) 100.0% *98.1%-100.0%*	(0/3) 0.0%

Specimen Population	N	LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
		NR	IR	RR	NR	IR	RR	HIV-1	HIV-2	UCR		
HIV-1 Infected Pregnant	25[B]	0	25	25	0	25	25	25	0	0	(25/25) 100.0% *86.7%-100.0%*	NA
HIV Group O Infected	45[C]	0	45	45	0	45	45	45	0	0	(45/45) 100.0% *92.1%-100.0%*	NA
HIV-1 Antigen Positive	90	0	90	90	0	90	90	88	0	2	(90/90) 100.0% *95.9%-100.0%*	NA
TOTAL	**360**	**0**	**360**	**360**	**0**	**360**	**360**	**161**	**67**	**129**	**(357/357) 100.0% *98.9%-100.0%***	**(0/3) 0.0%**

[A] 190 adults and 10 of unknown age
[B] 24 adults and 1 of unknown age
[C] 43 adults and 2 of unknown age
UCR = Untypable Cross Reactive

13.3.4 Retrospective Pediatric Population:

Results of Retrospective U.S. Pediatric Subjects:

Reactivity of 47 individuals from the retrospectively collected US (pediatric) population from presumably HIV-1 positive subjects.

The performance of the LIAISON® XL MUREX HIV Ab/Ag HT and an FDA approved comparator on the retrospectively collected US pediatric population presumably HIV-1 positive individuals are presented in Table 23. All confirmed HIV-positive US pediatric samples were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay.

Table 23. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity a Presumably HIV-1 Positive Retrospective U.S. Pediatric Population

Specimen Population	N	LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
		NR	IR	RR	NR	IR	RR	HIV-1	HIV-2	UCR		
HIV-1 Positive Pediatrics	44	5	39	39	6	39	38	40	0	0	(36/36) 100.0% *90.4%-100.0%*	(5/8) 62.5% *30.6%-86.3%*

		LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
Specimen Population	**N**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**HIV-1**	**HIV-2**	**UCR**		
HIV-1 Infected Pregnant (≤ 21 yrs)	3	0	3	3	0	3	3	3	0	0	(3/3) 100.0%	NA
TOTAL	**47**	**5**	**42**	**42**	**6**	**42**	**41**	**43**	**0**	**0**	**(39/39)100.0%** ***91.0%-100.0%***	**(5/8) 62.5%** ***30.6%-86.3%***

UCR = Untypable Cross Reactive

Results of Retrospective Non-U.S. Pediatric Subjects:

Reactivity of 35 individuals from the retrospectively collected non-US (pediatric) population from presumably HIV-1 positive subjects.

The performance of the LIAISON® XL MUREX HIV Ab/Ag HT and an FDA approved comparator on the retrospectively collected non-US pediatric population presumably HIV-1 positive individuals are presented in Table 24. All confirmed HIV-positive samples were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay

Table 24. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity a Presumably HIV-1 Positive Retrospective Non-U.S. Pediatric Population

		LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
Specimen Population	**N**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**HIV-1**	**HIV-2**	**UCR**		
HIV-1 Positive Pediatric	17	2	15	15	0	17	17	14	0	0	(14/14) 100.0% *78.5%-100.0%*	(2/3) 66.7% *20.8%-93.9%*
HIV-1 Infected Pregnant (≤ 21 yrs)	13	1	12	12	0	13	13	11	0	0	(11/11) 100.0% *74.1%-100.0%*	(1/2) 50.0% *9.5%-90.5%*
Group O Infected	2	0	2	2	0	2	2	2	0	0	(2/2) 100.0%	NA
HIV-1 Antigen Positive	3	0	3	3	0	3	3	3	0	0	(3/3) 100.0%	NA
TOTAL	**35**	**3**	**32**	**32**	**0**	**35**	**35**	**30**	**0**	**0**	**(30/30) 100.0%** ***88.6%-100.0%***	**(3/5) 60.0%** ***23.1%-88.2%***

UCR = Untypable Cross Reactive

13.3.5 Retrospective HIV-2 Population:

Reactivity of 200 individuals from the retrospectively collected population from non-US presumably HIV-2 positive subjects.

The performance of the LIAISON® XL MUREX HIV Ab/Ag HT and an FDA-approved comparator on the retrospectively collected non-US population presumably HIV-2 positive individuals are presented in Table 25. HIV-2 status of the samples was confirmed with an HIV-2 RNA assay, except for three samples.

Table 25. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity in the HIV-2 Positive Retrospective Population

		LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			HIV-2 RNA reactive		LIAISON® XL Sensitivity 95% CI
Specimen Population	**N**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**HIV-1**	**HIV-2**	**UCR**	**R**	**NR**	
HIV-2 Infected Adults	190	0	190	190	0	190	190	3*	62	122	187	3**	(187/187) 100.0% *98.0%-100.0%*
HIV-2 Infected Unknown Age	10	0	10	10	0	10	10	0	5	5	10	0	(10/10) 100.0% *72.2%-100.0%*
TOTAL	**200**	**0**	**200**	**200**	**0**	**200**	**200**	**3***	**67**	**127**	**197**	**3**	**(197/197)100.0%** ***98.1%-100.0%***

UCR = Untypable Cross Reactive; R= Reactive; NR = Non-Reactive
*Three samples purchased as HIV-2 positive were found to be HIV-1 positive only.
**Three samples reactive on the LIAISON® XL MUREX HIV Ab/Ag HT and FDA Approved HIV Ag/Ab Combo Assay were tested on the HIV1/2 differentiation assay and were Neg (1) or indeterminate (2). All 3 were negative on the RNA PCR test.

13.3.6 Pregnant Population:

The pregnant women samples were prospectively and retrospectively collected from the countries of Argentina, Guinea-Bissau, Peru and the U.S. Subjects ranged in age from 14-47 years old, with one subject ‘s age unknown.

Reactivity of 851 pregnant women including female subjects ≤21 years of age.

The performance of the LIAISON® XL MUREX HIV Ab/Ag HT and an FDA approved comparator including infection status on pregnant women including female subjects ≤21 years of age are presented in Table 26. All confirmed HIV-positive samples were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay.

Table 26. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity in Pregnant Females including ≤ 21 years old

		LIAISON® XL MUREX HIV Ab/Ag HT			FDA Approved HIV Ag/Ab Combo Assay			FDA Approved HIV 1/2 Differentiation Assay Reactive			HIV Infected Status		LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
Specimen Populatio	**N**	**NR**	**IR**	**RR**	**NR**	**IR**	**RR**	**HIV-1**	**HIV-2**	**UCR**	**I**	**NI**		
Low Risk (Healthy) Pregnant	396	396	0	0	396	0	0	0	0	0	0	396	NA	(396/396)100.0% *99.0%-100.0%*
High Risk Pregnant	404	391	13	13	395	9	9	4	0	0	4	400	(4/4) 100.0% *51.0%-100.0%*	(391/400) 97.8% *95.8%-98.8%*
HIV Positive Pregnant	51	1	50	50	0	51	51	49	0	0	49	2	(49/49) 100.0% *92.7%-100.0%*	(1/2) 50.0% *9.5%-90.5%*
TOTAL	**851**	**788**	**63**	**63**	**791**	**60**	**60**	**53**	**0**	**0**	**53**	**798**	**(53/53)100.0% *93.2%-100.0%***	**(788/798) 98.7% *97.7%-99.3%***

UCR = Untypable Cross Reactive; I = HIV Infected; NI = HIV Not-Infected

<u>High Risk Pregnant Population with Trimester</u>

Reactivity of 404 pregnant women including female subjects ≤21 years of age with trimester information are provided in Table 27. All four confirmed HIV-positive samples were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay.

Table 27. LIAISON® XL MUREX HIV Ab/Ag HT assay Reactivity in Pregnant Females at High Risk for HIV Infection by Trimester

HIV Category	Trimester	HIV Infection Status				Total	LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
		HIV Infected		Not Infected				
		LIAISON® XL MUREX HIV Ab/Ag HT		LIAISON® XL MUREX HIV Ab/Ag HT				
		+	-	+	-			
Pregnant Females at High Risk for HIV-1	1	1	0	0	43	44	(1/1) 100.0%	(43/43) 100.0% *91.8%-100.0%*
	2	0	0	5	139	144	NA	(139/144) 96.5% *92.1%-98.5%*
	3	3	0	4	191	198	(3/3) 100%	(191/195) 97.9% *94.8%-99.2%*

HIV Category	Trimester	HIV Infection Status				Total	LIAISON® XL Sensitivity 95% CI	LIAISON® XL Specificity 95% CI
		HIV Infected		Not Infected				
		LIAISON® XL MUREX HIV Ab/Ag HT		LIAISON® XL MUREX HIV Ab/Ag HT				
		+	-	+	-			
	Unknown	0	0	0	18	18	NA	(18/18) 100.0% *82.4%-100.0%*
Total		**4**	**0**	**9**	**391**	**404**	**(4/4) 100.0%** ***51.0%-100.0%***	**(391/400) 97.8%** ***95.3%-98.8%***

13.3.7 HIV-1 Group O Positive Subjects:

Forty-seven (47) samples from HIV-1 Group O positive subjects were tested and results are provided in Table 28. All confirmed HIV-positive samples were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay

Table 28. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity in HIV-1 Group O Positive

Serotype	Number of Specimens tested	LIAISON® XL MUREX HIV Ab/Ag HT	FDA Approved HIV Combo Assay
M and O	9	(9/9)	(9/9)
O	38	(38/38)	(38/38)
Total	**47**	**(47/47)**	**(47/47)**

13.3.8 CDC AIDS Classification Subjects:

One hundred (100) samples from subjects with AIDS as classified by the CDC per the 1993 revised Classification System for HIV Infection and Expanded Surveillance Case Definition for AIDS among Adolescents and Adults were tested as shown in Table 29. All specimens were from an adult population, ages 22 to 72.

Fifty-two (52) category A specimens were further subcategorized as: A1 (n=22), A2 (n=27), and A3 (n=3).

The 30 category B specimens were further subcategorized as: B1 (n=11), B2 (n=16), and B3 (n=3).

The 18 category C specimens were further subcategorized as C1 (n=6), C2 (n=5), and C3 (n=6) one (1) was not further subcategorized. All confirmed positive samples in each HIV infection status category were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay.

Table 29. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity in CDC AIDS Classification Subjects

CDC AIDS Classification	Number of Specimens tested	LIAISON® XL MUREX HIV Ab/Ag HT	FDA Approved HIV Combo Assay
A	52	(52/52)	(52/52)
B	30	(30/30)	(30/30)
C	18	(18/18)	(18/18)
Total	**100**	**(100/100)**	**(100/100)**

13.3.9 HIV-1 Group M Positive with Subtype Identification:

One hundred (100) subjects positive for HIV-1 Group M with subtype were tested. The results are stratified in Table 30. The circulating recombinant form (CRF) HIV-1 Group M subtype consisted of AE (n=4), AG (n=4), BC (n=1), BG (n=2), CPX (n=5). All confirmed positive samples in each HIV infection status subtype were correctly identified by the LIAISON® XL MUREX HIV Ab/Ag HT assay.

Table 30. LIAISON® XL MUREX HIV Ab/Ag HT Reactivity in HIV-1 Group M Positive with Subtype Identification

Subtype	Number of Specimens tested	LIAISON® XL MUREX HIV Ab/Ag HT	FDA Approved HIV Combo Assay
A	6	(6/6)	(6/6)
B	7	(7/7)	(7/7)
C	1	(1/1)	(1/1)
D	3	(3/3)	(3/3)
F	5	(5/5)	(5/5)
G	5	(5/5)	(5/5)
H	4	(4/4)	(4/4)
K	2	(2/2)	(2/2)

Subtype	Number of Specimens tested	LIAISON® XL MUREX HIV Ab/Ag HT	FDA Approved HIV Combo Assay
CRF*	16	(16/16)	(16/16)
TOTAL	**49**	**(49/49)**	**(49/49)**

Summary of the Cumulative Clinical Comparison versus the HIV Infection Status:

The sensitivity and specificity of the LIAISON® XL HIV Ab/Ag HT are summarized in the tables below (Table 31 and Table 32).

Table 31. Clinical Comparison versus the HIV Infection for the U.S. Population (Combined Prospective & Retrospective)

LIAISON® XL HIV Ab/Ag HT	HIV Infection Status		Total
	HIV Infected	Not HIV Infected	
Reactive	1059	23	1082
Non-Reactive	0	4677	4677
TOTAL	**1059**	**4700**	**5759**

Sensitivity: 1059/1059 = 100% 95% CI = 99.6–100%
Specificity: 4677/4700 = 99.5% 95% CI = 99.3–99.7%

Table 32. Clinical Comparison versus the HIV Infection for the Non-U.S. Population (Combined Prospective & Retrospective)

LIAISON® XL HIV Ab/Ag HT	HIV Infection Status		Total
	HIV Infected	Not HIV Infected	
Reactive	600	61	661
Non-Reactive	2	3052	3054
TOTAL	**602**	**3113**	**3715**

Sensitivity: 600/602 = 99.7% 95% CI = 98.8 – 99.9%
Specificity: 3052/3113 = 98.0% 95% CI = 97.5 – 98.5%

Overall, the sensitivity and the specificity of the LIAISON® XL MUREX HIV Ab/Ag HT for the intended use population is acceptable.

14. INSPECTIONS

14.1 Manufacturing Facilities Review/Inspection:

The CMC/Facility information provided in the PMA and the Major Amendment Response was reviewed by CBER and found to be sufficient and acceptable.

Based on the recent inspection history, DMPQ/OCBQ has waived inspection for this PMA.

14.2 Bioresearch Monitoring (BIMO) Inspections:

CBER Bioresearch Monitoring (BIMO) issued two inspection assignments at two testing sites in the United States; however, because of restrictions and contingencies related to the COVID-19 public health emergency, and because review of the clinical data raised no human subject protection concerns, the inspections were cancelled.

15. CONCLUSIONS DRAWN FROM THE STUDIES

15.1 Effectiveness Conclusions:

Multi-center clinical studies were conducted in the U.S. The LIAISON® XL MUREX HIV Ab/Ag HT performed with clinical sensitivity and specificity comparable to an FDA approved assay. Results from the clinical studies indicate that the LIAISON® XL MUREX HIV Ab/Ag HT can be used effectively for the simultaneous qualitative detection of HIV p24 antigen and antibodies to HIV-1 (Groups M and O) and HIV-2 in human serum or plasma. The clinical study results also demonstrate that the LIAISON® XL MUREX HIV Ab/Ag HT assay can be effectively used for qualitative detection in pediatric subjects and in pregnant women.

15.2 Safety Conclusions:

The risk of the device is based on data collected in the clinical study to support PMA approval as described above. Based on the results of the clinical studies, the LIAISON® XL MUREX HIV Ab/Ag HT, when used according to the labeling and in conjunction with other clinical information, is safe to use and poses minimal risk to the patient due to false test results.

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 RNA. 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.

16. BENEFIT-RISK DETERMINATION

The LIAISON® XL MUREX HIV Ab/Ag HT is intended for *in vitro* diagnostic use, and as a result, there is no direct adverse effect on the patient. Adverse reactions may occur during blood donation and may include: mild reactions such as agitation, sweating, pallor, coldness, sense of weakness, or nausea. Some reactions may be more severe, including vomiting, and loss of consciousness such as fainting.

Failure of the product to perform as intended or human error in the use of the test may lead to false results.

A false negative HIV Ab/Ag test result may lead to a non-recognized HIV infection in a patient and the patient not receiving treatment is a serious concern for both the patient and the public as an infected person could spread the disease. This is especially important if the patient is pregnant and the disease may be transferred to the neonate.

A false positive (reactive) HIV Ab/Ag result is not considered a patient or a public risk as all positive HIV Ab/Ag results should be retested in duplicate and followed up with a supplemental antibody immunoassay that differentiates HIV-1 antibodies from HIV-2 antibodies.

17. OVERALL CONCLUSIONS

The data in this application support the reasonable assurance of safety and effectiveness of this device when used in accordance with the indications for use. The data from the nonclinical studies demonstrated acceptable analytical sensitivity, precision, and analytical specificity of the LIAISON® XL MUREX HIV Ab/Ag HT when used according to the instructions for use as stated in the labeling, the warnings and precautions, and limitations sections of the labeling. The clinical studies and the statistical analysis of clinical data in this application has shown that the simultaneous qualitative detection of HIV p24 antigen and antibodies to HIV-1 (Groups M and O) and HIV-2 in human serum or plasma by LIAISON® XL MUREX HIV Ab/Ag HT assay is safe and effective when used according to the directions for use in the labeling.

18. APPROVAL SPECIFICATIONS

- Directions for use: See device labeling.
- Hazards to Health from Use of the Device: See Indications, Contraindications, Warnings, Precautions, and Adverse Events in the device labeling.
- Post-approval Requirements and Restrictions: See approval order.

  Please perform a study to demonstrate precision for HIV-1 group O specimens using at least three lots of reagents produced after the manufacturing process changes are instituted and to submit the data for our review. The data from this

study may be provided per 21 CFR 814.84 in the first Annual Report post approval of the PMA.

19. PANEL RECOMMENDATIONS

Not Applicable – This product was not submitted for review by the Blood Products Advisory Committee.

20. FDA/CBER DECISION

The PMA BP190437/0 is recommended for approval.