510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY

I Background Information:

A 510(k) Number

K241427

B Applicant

Beckman Coulter, Inc

C Proprietary and Established Names

Access Syphilis

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
LIP	Class II	21 CFR 866.3830 - Treponema Pallidum Treponemal Test Reagents	MI - Microbiology

II Submission/Device Overview:

A Purpose for Submission:

To establish substantial equivalence to a predicate device and to obtain market clearance for a new assay designed to detect antibodies to *Treponema pallidum* in human serum and plasma.

B Measurand:

Total antibodies to *Treponema pallidum*

C Type of Test:

Chemiluminescent microparticle immunoassay (CMIA)

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

The Access Syphilis assay is a paramagnetic particle, chemiluminescent immunoassay for the qualitative detection of total antibodies to *Treponema pallidum* in human serum and plasma using the Access Immunoassay Systems. It is intended to be used as an aid in the diagnosis of syphilis or in conjunction with a non-treponemal laboratory test and clinical findings to aid in the diagnosis of syphilis infection. The Access Syphilis assay is not intended for blood and tissue donor screening.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

D Special Instrument Requirements:

Access Immunoassay Systems

- Access 2 Immunoassay System
- DxI 9000 Access Immunoassay Analyzer

IV Device/System Characteristics:

A Device Description:

The Access Syphilis assay is a two-step enzyme chemiluminescent microparticle immunoassay. The test is performed by adding a sample to the reaction vessel, along with buffer, paramagnetic particles coated with recombinant *Treponema pallidum* antigens Tp17 and Tp47, and biotinylated Treponema Tp17 and Tp47 antigens. The sample mixture is incubated in the reaction vessel, allowing materials to bind to the solid phase where they are held in a magnetic field. Any unbound materials are washed away.

B Principle of Operation:

The Access Syphilis assay is a paramagnetic particle, chemiluminescent immunoassay for the qualitative detection of total antibodies to *Treponema pallidum* in human serum and plasma using the Access Immunoassay Systems. The Access Immunoassay System is fully automated to allow assay-specific reagents to form immune complexes, immobilize the bound immune complex by magnetic field, and allow the unbound components to be washed away.

The Access Syphilis assay uses the following reagents:

- *Treponema pallidum* antigens labeled with alkaline phosphatase
- Antigen-coated micron-sized paramagnetic particles
- TRIS-buffered saline with biotinylated Tp17 and Tp47 conjugates

The assay is a two-step immunoenzymic “sandwich” assay. The sample is added to the reaction vessel along with buffer, recombinant Tp17- and Tp47-coated paramagnetic particles, and biotinylated Treponema Tp17 and Tp47 antigens. The mixture is incubated to allow Treponemal antibodies that may be present to be captured by the antigens. Following incubation, any unbound materials are washed away, and the materials bound to the solid phase are held in a magnetic field. Alkaline phosphatase conjugates are added that bind to immunoglobulin captured on the particles. A chemiluminescent substrate is added to the vessel and light generated by the reaction is measured with a luminometer. The measured light allows for determination of the

presence of the analyte by comparison with a cut-off value defined during the assay calibration on the instrument.

Test results are determined automatically by the system software. Results expressed as Signal/Cutoff (S/CO) are reported to be "reactive" or "nonreactive" as a function of their relationship with the "cut-off".
The result interpretation for the Access Syphilis assay is as follows:

S/CO < 1.00 Nonreactive
S/CO ≥ 1.00 Reactive

Interpretation of results:
Reactive: Treponemal antibodies detected; indicates active or previous infection with *Treponema pallidum*.
Nonreactive: Treponemal antibodies not detected; indicates no active or previous infection with *Treponema pallidum*.

Test results are intended to aid in diagnosis only. As with all serological tests for syphilis, results should always be interpreted in conjunction with additional treponemal or nontreponemal serologic test results (as appropriate), the patient's clinical symptoms, medical history, and other clinical and/or laboratory findings to produce a diagnosis of syphilis by disease stage.

Access Syphilis Calibrator
Intended for the calibration of the Access Syphilis assay. Provided as one negative and one positive calibrator for *Treponemal pallidum* antibody.

Access Syphilis Controls
Intended for the estimation of test precision and the detection of systematic analytical deviations of the Access Immunoassay System when used for the qualitative detection of antibodies to *Treponemal pallidum* (TP) in human serum and plasma. Provided as three vials each of *Treponema pallidum* antibody positive control and *Treponema pallidum* negative control.

V Substantial Equivalence Information:

A Predicate Device Name(s):
ARCHITECT Syphilis TP Reagent, ARCHITECT Syphilis TP Calibrator, ARCHITECT Syphilis TP Control

B Predicate 510(k) Number(s):
K153730

C Comparison with Predicate(s):

Device & Predicate Device(s):	K241427	K153730
Device Trade Name	Access Syphilis	ARCHITECT Syphilis TP

General Device Characteristic Similarities		
Intended Use/Indications for Use	The Access Syphilis assay is a paramagnetic particle, chemiluminescent immunoassay for the qualitative detection of total antibodies to Treponema pallidum in human serum and plasma using the Access Immunoassay Systems. It is intended to be used as an aid in the diagnosis of syphilis or in conjunction with a non-treponemal laboratory test and clinical findings to aid in the diagnosis of syphilis infection. The Access Syphilis assay is not intended for blood and tissue donor screening.	The ARCHITECT Syphilis TP assay is a chemiluminescent microparticle immunoassay (CMIA) for the qualitative detection of antibodies (IgG and IgM) directed against *Treponema pallidum* (TP) in human serum and plasma. The ARCHITECT Syphilis TP assay is intended to be used as an initial diagnostic test or in conjunction with a nontreponemal laboratory test and clinical findings to aid in the diagnosis of syphilis infection.
Environment of Use	Clinical laboratory	Same
Methodology	Chemiluminescent microparticle immunoassay (CMIA)	Same
Cut-off Index	1.00 S/CO	Same
Assay Type	Two-step sandwich enzyme immunoassay	Same
Detection Method	Automated, chemiluminescence	Same
Controls	2 (Negative and Positive)	Same
General Device Characteristic Differences		
Antigens	Recombinant *Treponema pallidum* antigens Tp17 and Tp47, and biotinylated Treponema Tp17 and Tp47 antigens	Recombinant TP antigens: TpN15, TpN17, and TpN47 (obtained in *E. coli*)
Instrument	Access Immunoassay	ARCHITECT iSystem

	Systems: • Access 2 Immunoassay System • DxI 9000 Access Immunoassay Analyzer	
Components	1. Microparticles – paramagnetic particles coated with recombinant Ag Tp17 and Tp47 in TRIS-buffered saline with surfactant, protein, sodium azide, and ProClin 300. 2. Conjugate – TRIS-buffered saline with biotinylated Tp17 and Tp47 conjugates, surfactant, protein, and sodium azide. 3. Assay Diluent – Phosphate buffer with alkaline phosphatase conjugates, protein, sodium azide, and ProClin 300.	1. Microparticles – TP antigen (*E. coli*, recombinant) coated microparticles in HEPES buffer with detergent. 2. Conjugate – Murine anti-IgG/anti-IgM acridinium-labeled conjugate in MES buffer with protein (bovine) stabilizer. 3. Assay Diluent – Syphilis TP Assay Diluent containing MES buffer with detergent. Preservatives: ProClin 950 and other antimicrobial agents.
Sample Volume	~45 µL	30 µL
Calibrators	1 negative and 1 positive antibody calibrator.	1 positive calibrator, stable up to 30 days.
Compatible Anticoagulants	Human Serum: Serum and serum separator tube Human Plasma: Lithium Heparin Lithium Heparin separator tube Dipotassium (K_2) EDTA	Human Serum: Serum and serum separator tube Human Plasma: Lithium Heparin Lithium Heparin separator tube Dipotassium (K_2) EDTA

	Tripotassium (K_3) EDTA Sodium Citrate Acid Citrate Dextrose (ACD) Citrate Phosphate Dextrose (CPD) Citrate Phosphate Dextrose with Adenine (CPDA)	Tripotassium (K_3) EDTA Sodium Heparin

VI Standards/Guidance Documents Referenced:

CLSI EP05-A3 7-251 Evaluation of Precision of Qualitative Measurement Procedures; Approved Guideline – Third Edition

CLSI EP07 3rd Edition 7-275 Interference Testing in Clinical Chemistry

CLSI EP12-A2 (2008) User Protocol for Evaluation of Qualitative Test Performance; Approved Guideline – Second Edition

CLSI EP24-A2 7-234 Assessment of the Diagnostic Accuracy of Laboratory Tests Using Receiver Operating Characteristic Curves; Approved Guideline – Second Edition

CLSI EP25-A 7-235 Evaluation of Stability of In Vitro Diagnostic Reagents; Approved Guideline

CLSI EP37 1st Edition 7-284 Supplemental Tables for Interference Testing in Clinical Chemistry

CLSI GP44-A4 7-213 Procedures for the Handling and Processing of Blood Specimens for Common Laboratory Tests; Approved Guideline – Fourth Edition

CLSI GP41 7th Edition 7-277 Collection of Diagnostic Venous Blood Specimens

ISO 7000 Sixth edition 2019-07 5-124 Graphical symbols for use on equipment – Registered symbols

ISO 7010 Third edition 2019-07 5-130 Graphical symbols – Safety colors and safety signs – Registered safety signs

ISO 15223-1 Fourth Edition 2021-07 5-134 Medical devices – Symbols to be used with information to be supplied by the manufacturer – Part 1: General requirements

ISO 13485:2016 Medical devices – Quality management systems – Requirements for regulatory purposes

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

Within-lab Precision
The precision of the Access Syphilis assay was evaluated in an internal study using three reagent lots and three calibrator lots. The panel consisted of four samples contrived in undiluted, pooled human serum containing azide and ProClin. Positive specimens were prepared by spiking negative serum with high positive CPD plasma specimens.

Table 1. Precision panel members

Panel Member	Target ratio (S/CO)
Low nonreactive	< 0.1
High nonreactive	0.5 to 1.0
Low reactive	1.0 to 2.0
Reactive	> 5.0

Negative and positive quality controls were also included in each run, with mean S/CO values of 0.10 and 1.56, respectively.

Samples were tested on both Access 2 and DxI 9000 instruments in 2 replicates per run, 2 runs per day for a 20-day period. A total of 80 replicates were tested for each panel member for each lot.

Table 2. Precision study data – DxI 9000 instrument

Precision – DxI 9000 Instrument												
Sample	N	Mean S/CO	Between Lot		Between Day		Between Run		Within Run		Overall	
			SD (S/CO)	CV (%)	SD (S/CO)	CV (%)	SD (S/CO)	CV (%)	SD (S/CO)	CV (%)	SD (S/CO)	CV (%)
Neg Control	238*	0.166	0.064	N/A	0.007	N/A	0.007	N/A	0.008	N/A	0.065	N/A
Pos Control	238*	1.786	0.082	4.6%	0.059	3.3%	0.088	4.9%	0.041	2.3%	0.140	7.8%
Low Negative	240	0.078	0.018	N/A	0.003	N/A	0.003	N/A	0.005	N/A	0.019	N/A
High Negative	239*	0.779	0.065	8.4%	0.021	2.8%	0.023	3.0%	0.019	2.4%	0.075	9.6%
Low Positive	240	1.994	0.078	3.9%	0.048	2.4%	0.072	3.6%	0.046	2.3%	0.125	6.3%
Positive	240	7.598	0.294	3.9%	0.241	3.2%	0.240	3.2%	0.163	2.1%	0.478	6.3%

*A total of five results were flagged during the study due to instrument errors. These specimens were not retested and were excluded from result calculations.

Table 3. Precision study data – Access 2 instrument

Precision – Access 2 Instrument												
Sample	N	Mean S/CO	Between Lot		Between Day		Between Run		Within Run		Overall	
			SD (S/CO)	CV (%)	SD (S/CO)	CV (%)	SD (S/CO)	CV (%)	SD (S/CO)	CV (%)	SD (S/CO)	CV (%)
Neg Control	240	0.107	0.029	N/A	0.009	N/A	0.007	N/A	0.007	N/A	0.032	N/A
Pos Control	240	1.704	0.083	4.9%	0.057	3.4%	0.074	4.3%	0.048	2.8%	0.134	7.9%
Low Negative	240	0.064	0.004	N/A	0.003	N/A	0.004	N/A	0.003	N/A	0.007	N/A
High Negative	240	0.708	0.018	2.6%	0.023	3.2%	0.022	3.2%	0.017	2.5%	0.041	5.8%
Low Positive	240	1.819	0.116	6.4%	0.061	3.4%	0.051	2.8%	0.039	2.2%	0.146	8.0%
Positive	240	6.721	0.491	7.3%	0.216	3.2%	0.203	3.0%	0.179	2.7%	0.601	8.9%

Multi-site Reproducibility

A multicenter reproducibility study was performed at three clinical sites using three lots each of the Access Syphilis assay, three lots of calibrators, and three lots of controls. Each panel consisted of one low negative, one high negative, one low positive, and one positive, as well as two assay controls. Panel members were tested using four replicates per run, two runs per day, for a total of five days to generate 120 data points at each site per platform. Data was analyzed for intra-assay and inter-assay reproducibility.

Table 4. Reproducibility study data – Access 2 instrument

Reproducibility – Access 2 Instrument														
Sample	Mean Value	N	Repeatability		Between-Run		Between-Day		Between-Lot		Between-Site		Reproducibility	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
Neg Control	0.10	360	0.004	N/A	0.005	N/A	0.006	N/A	0.025	N/A	0.005	N/A	0.027	N/A
Pos Control	1.56	360	0.043	2.8%	0.058	3.7%	0.016	1.0%	0.010	0.6%	0.054	3.4%	0.092	5.9%
Low Negative	0.06	360	0.003	N/A	0.003	N/A	0.003	N/A	0.004	N/A	0.005	N/A	0.009	N/A
High Negative	0.75	360	0.019	2.6%	0.018	2.4%	0.018	2.5%	0.013	1.8%	0.005	0.6%	0.035	4.7%
Low Positive	1.89	360	0.042	2.2%	0.065	3.5%	0.037	2.0%	0.058	3.1%	0.000	N/A	0.104	5.5%
Positive	7.19	360	0.223	3.1%	0.197	2.7%	0.000	0.0%	0.247	3.4%	0.068	0.9%	0.393	5.5%

Table 5. Reproducibility study data – DxI 9000 instrument

Reproducibility – DxI 9000 Instrument														
Sample	Mean Value	N	Repeatability		Between-Run		Between-Day		Between-Lot		Between-Site		Reproducibility	
			SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV %	SD	CV%
Neg Control	0.10	360	0.005	N/A	0.003	N/A	0.009	N/A	0.033	N/A	0.010	N/A	0.036	N/A
Pos Control	1.59	360	0.037	2.3	0.050	3.2	0.059	3.7	0.000	N/A	0.019	1.2	0.088	5.5
Low Negative	0.05	360	0.003	N/A	0.001	N/A	0.004	N/A	0.007	N/A	0.005	N/A	0.010	N/A
High Negative	0.75	360	0.017	2.3	0.018	2.4	0.029	3.8	0.018	2.4	0.030	4.0	0.052	6.9
Low Positive	1.94	360	0.044	2.3	0.060	3.1	0.070	3.6	0.053	2.7	0.052	2.7	0.126	6.5
Positive	7.48	360	0.271	3.6	0.271	3.6	0.213	2.8	0.297	4.0	0.387	5.2	0.655	8.8

2. Linearity:

   Not applicable.

3. Analytical Specificity/Interference:

   Interference by Endogenous Substances
   Potential interference with hemoglobin, bilirubin, triolein, protein (albumin), and gamma globulin were evaluated in an analytical study by preparing the panel members at treponemal concentrations targeted at the levels described in the table below.

Table 6. Interference panel members

Panel Member	Target Ratio (S/CO)
Low negative	< 0.1
High negative	0.5 – 1.0
Low positive	1.0 – 2.0
Positive	5.0 – 10.0

Each of the four panel members was split into seven aliquots (for each of the six conditions plus a control). Each aliquot was spiked directly with the substance at the target final concentration. If an effect was observed at a given analyte concentration, a dose effect study was performed to determine the minimum interfering concentration.

Table 7. Interferent concentration levels

Interferent	Interferent Level
Hemoglobin	1000 mg/dL
Conjugated Bilirubin	40 mg/dL
Unconjugated Bilirubin	40 mg/dL
Triolein	36 g/L
Total Protein (Human Albumin)	15 g/dL
Gamma globulin	47.5 g/L*

*Interference was noted at a concentration of 60 g/L. No interference was observed ≤ 47.5 g/L

Interference was observed with the Access Syphilis test in the presence of gamma globulin at a concentration of 60 mg/mL in negative and positive specimens. The highest concentration for which no interference was observed across three lots of the Access Syphilis assay was 47.5 mg/mL. No interference was observed for the other listed substances up to the levels indicated in the table above.

Drug Interference

Potential interference due to the presence of common pharmaceutical compounds was tested in a drug interference study. Panels were prepared as described for the endogenous interference study. Each panel member was divided into ten aliquots (for seven exogenous substances and three control conditions). The aliquots were spiked directly with the drug of interest at the final target concentration listed in the table below. For the control specimens, the same amount of drug diluent (deionized water or ethanol) was spiked-in to avoid bias between test and control specimens. No interference was observed at the levels tested.

Table 8. Interfering substances and concentrations

Substance	Drug class	Target
Acetylsalicylic acid	Anti-inflammatory	3 mg/dL
Ibuprofen	Anti-inflammatory	21.9 mg/dL
Acetaminophen	Antipyretic	15.6 mg/dL
Azithromycin	Antibiotic (macrolides)	1.11 mg/dL
Ceftriaxone sodium	Antibiotic (cephalosporins)	84 mg/dL
Doxycycline hyclate	Antibiotic (tetracyclines)	1.8 mg/dL
Biotin	Vitamin	0.351 mg/dL

Cross-reactivity in Specimens with Medical Conditions not Related to Syphilis

Specimens from individuals diagnosed with other diseases that could cause false positive results were tested in duplicate by the Access Syphilis assay. Discordant results were investigated by additional testing. For each specimen, one replicate was tested on the Access 2 instrument and one replicate was tested with the DxI 9000 instrument.

Table 9. Cross-reactivity panel

Potential Cross-reactant	N	Number of Reactive Samples
Pregnant Multipara	14	0
Hemodialysis	10	0
Transplant	10	0
Rheumatoid Factor (RF)	10	0
Human anti-mouse antibody (HAMA)	10	0
Anti-nuclear antibody (ANA)	10	0
Myeloma	11	1[a]
Flu-vaccinated patients	10	0
Anti-phospholipid	10	0
Lyme Disease (*Borrelia garinii*, *Borrelia afzelii* & *Borrelia burgdorferi* s.s.)	10	0

Leptospirosis	10	0
Systemic Lupus Erythematosus (SLE)	10	0
Hepatitis A Virus (HAV) Total Ab	5	0
Hepatitis A Virus (HAV) IgM	5	0
Hepatitis B Virus (HBV) HBs Ag positive	10	0
Hepatitis B Virus (HBV) anti-HBs positive	22	0
Hepatitis B Virus (HBV) HBc IgM	10	1[a]
Hepatitis B Virus (HBV) HBc Total	20	0
Hepatitis C Virus (HCV) Ig Total Ab	10	0
HTLV-1	10	0
HTLV-2	14	1[b]
Human Immunodeficiency Virus (HIV)-1	10	2[b]
Human Immunodeficiency Virus (HIV)-2	10	1[a]
Herpes Simplex Virus (HSV) 1/2 IgM	15	0
Herpes Simplex Virus (HSV) 1 IgG	10	0
Herpes Simplex Virus (HSV) 2 IgG	2	0
Cytomegalovirus (CMV) IgG	5	0
Cytomegalovirus (CMV) IgM	5	0
Rubella IgG	5	0
Rubella IgM	10	0
Toxoplasma gondii IgG	5	0
Toxoplasma gondii IgM	5	0
Epstein Barr Virus (EBV) IgG	5	0
Epstein Barr Virus (EBV) IgM	5	0
Epstein Barr Virus (EBV) anti-EBNA IgG and anti-VCA IgG	5	0
Epstein Barr Virus (EBV) anti-VCA IgM	5	0
Anti-*Escherichia coli* (*E. coli*)[c]	10	0

[a] Volume insufficient for duplicate testing.
[b] Specimens were determined to be true syphilis positives.
[c] Anti-E. coli specimens were prepared by spiking 10 negative specimens with anti-*E. coli* rabbit antibodies.

4. Assay Reportable Range:
   Not applicable.

5. Traceability, Stability, Expected Values (Controls, Calibrators, or Methods):

Calibrator
The Access Syphilis Calibrators are not provided with the reagent kit. The calibrators are provided as one negative for TP antibody and one positive for TP antibody.

External Controls
The Access Syphilis assay controls are not provided with the reagent kit. The negative control is made from human serum shown to be non-reactive for antibodies to TP. The positive control is made from TP-non-reactive human serum spiked with a pool of anti-TP reactive human serum or plasma.

Sample Stability Studies
This study was conducted using serum and plasma specimens. The stability of TP antibodies in various storage matrices was evaluated for:

- Refrigerated Stability (at 2 to 8˚C) at day 0 and daily at days 5 – 8.
- Room temperature stability (at 20 to 25˚C) at 0 h, 24 h, 48 h, 72 h, and 96 h.
- Frozen stability at -20˚C and -80˚C
    - Non-reactive specimens were evaluated at days 0, 29, 36, 90, and 105.

    - Reactive serum specimens were evaluated at days 0, 29, and 36.
    - Reactive plasma specimens were evaluated at days 0, 29, 36, 90, and 111.
- Freeze/thaw stability for 0 - 6 cycles (at -20 and -80˚C)

Each sample was tested in duplicate with the Access 2 and DxI 9000 instruments at each time point. The evaluation of results was performed separately for each concentration, storage condition, and time point.

The mean S/CO was calculated for each subsequent time point for each panel member, concentration level, storage condition, and tube type. The difference between the mean S/CO at each time point and the corresponding baseline mean S/CO, taken at day zero, was calculated. For negative panel members (S/CO < 1.00), the difference in measured concentration was within ± 0.10 compared to the baseline and the negative agreement was 100% among the different time points. For positive panel members (S/CO ≥ 1.00), the median S/CO percentage bias was within ± 15% versus the baseline and the positive agreement was at least 95% for the low positive specimens (< 2.5x S/CO) and 100% for the positive specimens (≥ 2.5x S/CO).

The study data supported the following storage conditions:
- 2 to 8°C for 7 days
- 20 to 25°C for 72 hours
- -20°C or colder for 30 days
- Samples may be frozen and thawed up to 5 times.

Reagent Stability
To test reagent stability, three lots of Access Syphilis reagent packs were tested in triplicate within a month after production. After the initial time point, testing was conducted in duplicate at 3, 6, 9, 10, 11, and 12 months approximately for each lot, with additional testing for lots one and two at 13 months and testing for lot one through 15 months. The positive calibrator and negative calibrators were tested at each time point to validate the run and calibrate the reagent pack stored at 2-10°C. Four panel members plus a positive and negative control were used to assess real-time reagent stability. The study data from three lots supported stability up to 10.9 months when stored at 2-10°C unopened.

On-board Reagent Stability
To assess the on-board reagent stability of Access Syphilis, ten open kits were tested multiple times between day 0 and day 62. The positive calibrator and negative calibrators were tested at each time point to validate the run and calibrate the reagent pack stored at 2-10°C. Four panel members plus a positive and negative controls were used to assess real-time reagent stability. The study data supported stability up to 56 days after opening when stored at 2-10°C.

Hook Effect
The sponsor conducted a study to demonstrate that the Access Syphilis assay does not produce false non-reactive results when testing samples with high levels of anti-TP antibody. The study utilized five CPD plasma specimens with medium to high levels of anti-TP antibody which were serially diluted with negative serum. Each of the diluted specimens was tested with the Access Syphilis assay. The data from this study showed that there was no high dose hook effect,

suggesting that the Access Syphilis assay is not susceptible to interference from specimens with high levels of anti-syphilis TP antibodies.

6. Detection Limit:
   Not applicable.

7. Assay Cut-Off:
   The Access Syphilis cut-off was evaluated by ROC analysis using 301 antibody-positive and 3047 negative specimens. Specimens were tested in one replicate on the DxI 9000 and Access 2 instruments.

   To determine the cut-off value, a normalized ratio was determined for each specimen by dividing the specimen signal by the signal obtained with the positive calibrator tested in triplicate during the same testing runs. The normalized ratio values were used to build the ROC curve, and the optimal normalized ratio value that gave the best compromise between sensitivity and specificity was determined.

   Result classification:
   S/CO < 1.00 sample is interpreted as "non-reactive"
   S/CO ≥ 1.00 sample is interpreted as "reactive"

B Comparison Studies:

1. Method Comparison with Predicate Device:

   See the "Clinical Studies" section.

2. Matrix Comparison:

   The suitability of various blood collection tube types for use with the Access Syphilis assay was evaluated in an analytical study. Samples were obtained from multiple donors and venous blood was collected in the following tube types:

   - Serum (collected in dry tube) – used as the control
   - Serum separator tubes
   - Citrate phosphate dextrose with adenine (CPDA)
   - Lithium heparin
   - Lithium heparin with gel separator tube
   - Dipotassium (K2) EDTA
   - Tripotassium (K3) EDTA
   - Sodium citrate
   - Citrate phosphate dextrose (CPD)
   - Acid citrate dextrose (ACD)

   A total of 65 paired specimens, 25 non-reactive and 40 reactive, were tested for each matrix. Positive specimens were prepared by spiking negative specimens with highly reactive syphilis samples to targeted concentrations. A total of four levels of reactivity were evaluated:

Table 10. Matrix comparison panel members

Analyte Target Level	Number of Expected Specimens
2.5 – 5x cut-off	10
< 2.5x cut-off*	30
Negative	35
Total	65

*About half of the samples were prepared in the 1.00 – 1.50 S/CO range.

Each sample was tested in duplicate. Percent agreement and bias analysis were calculated in comparison to the control condition (serum collected in a dry tube).

The resulting data demonstrates that serum, serum with gel separator, lithium heparin, lithium heparin with gel separator, dipotassium EDTA, tripotassium EDTA, sodium citrate, citrate phosphate dextrose, acid citrate dextrose, and citrate phosphate dextrose with adenine tubes are acceptable for use with the Access Syphilis assay.

C Clinical Studies:

A multicenter study was conducted on the Access 2 and DxI 9000 instruments between June and September 2023 to evaluate the ability of the Access Syphilis assay to detect antibodies directed against *Treponema pallidum* (TP). A total of 1923 specimens were tested in the Access Syphilis clinical study on both the Access 2 and the DxI 9000 instruments. Of these 1923, 13 were excluded from analysis. The remaining 1910 included 1104 prospectively collected from the intended use population; 50 retrospective samples from a population at high-risk for STIs; 402 retrospective specimens including 22 pregnant women; 204 apparently healthy individuals; and 150 from medically diagnosed individuals with primary, secondary, or latent syphilis. All cohorts were tested at three testing sites in a randomized and blinded fashion.

The clinical performance of the Access Syphilis assay was evaluated by calculating positive percent agreement and negative percent agreement of the assay with the final comparator result based on an algorithm of results from three FDA-cleared syphilis assays: a treponemal chemiluminescent immunoassay (TP-CLIA), a nontreponemal assay (RPR), and a second treponemal assay (TP-PA).

Clinical Performance in Prospectively Collected Specimens in the Intended Use Population

The 1104 prospectively collected specimens in the intended use population, analyzed in the Access Syphilis clinical study consisted of 399 specimens sent for routine syphilis testing (201 females and 198 males, 12 to >89 years old), 405 pregnant females (16 to 46 years old), and 300 HIV-positive specimens (98 females and 202 males, 23 to 77 years old). Of the 1104 prospectively collected specimens, 55 were from patients ≤ 21 years old. Thirteen (13) prospective specimens were excluded from analysis due to system error, duplicate testing, or missing comparator result.

The comparison between the Access Syphilis result and the final comparator result for the prospectively collected specimens in the intended use population is shown in the following table.

Table 11. Percent agreement in the prospective intended use population

Access Syphilis Result Interpretation	Final Comparator Result	
	Positive	Negative
Reactive	184	30
Non-reactive	0	890

Positive percent agreement was 100% (184/184) with a 95% confidence interval of 98.0% to 100.0%. Negative percent agreement was 96.7% (890/920) with a 95% confidence interval of 95.4% to 97.7%.

Table 12. Prospective percent agreement by category

Category	Positive Percent Agreement % (x/n)	95% Confidence Interval (%)	Negative Percent Agreement % (x/n)	95% Confidence Interval (%)
Routine Syphilis	100 (60/60)	94.0 - 100	99.7 (338/339)	98.3 - 99.9
Pregnant	100 (6/6)	61.0 - 100	99.7 (398/399)	98.6 - 100
HIV positive	100 (118/118)	96.8 - 100	84.6[a] (154/182)	78.7 - 89.1

[a] The 28 comparator-negative specimens were tested with an additional TP-CLIA test. Of these, 25 (89.3%) were also reactive.

Clinical Performance in Retrospective Samples

Of the total 1910 specimens analyzed in the Access Syphilis clinical study, 452 were retrospective specimens. The 402 retrospective specimens included 22 specimens from pregnant individuals. An additional 50 retrospective specimens from a population at high risk for contracting STIs were also tested. Each of these specimens was analyzed with the Access Syphilis assay on the Access 2 and DxI 9000 instruments, and comparator assays, employing an algorithm of three syphilis tests to determine the comparator result. The agreements between the Access Syphilis result and the final comparator result are presented below.

Table 13. Percent agreement in retrospective specimens

Category	Positive Percent Agreement % (x/n)	95% Confidence Interval (%)	Negative Percent Agreement % (x/n)	95% Confidence Interval (%)
Retrospective Specimens	100 (378/378)	99.0 - 100.0	0 (2/0)	NA
Retrospective Specimens - Pregnant	100 (20/20)	83.9 - 100.0	50.0 (1/2)[a]	9.4 – 90.6
Retrospective High-risk STI population	100 (20/20)	83.9 - 100.0	80.0 (24/30)[b]	62.7 - 90.5

[a] Specimen reactive by Access Syphilis was also reactive with another FDA-cleared TP-CLIA test.
[b] Of the six specimens reactive by Access Syphilis, one was also reactive with another FDA-cleared TP-CLIA test.
NA = Not applicable

Clinical Performance in Pregnant Females

A total of 427 pregnant female samples were analyzed in the Access Syphilis clinical study; 405 samples were prospectively collected from women coming to the health care facilities at the collection sites and 22 specimens were retrospective. The percent agreement between the Access Syphilis results and the final comparator results for prospectively collected and retrospective specimens from pregnant females is shown below, stratified by pregnancy trimester.

Table 14. Percent agreement in pregnant women

Category	Positive Percent Agreement % (x/n)	95% Confidence Interval (%)	Negative Percent Agreement % (x/n)	95% Confidence Interval (%)
Prospectively Collected	100.0 (6/6)	61.0 – 100.0	99.7 (398/399)	98.6 – 100.0
First Trimester	100.0 (1/1)	20.7 – 100.0	100.0 (59/59)	93.9 – 100.0
Second Trimester	100.0 (1/1)	20.7 – 100.0	100.0 (37/37)	90.6 – 100.0
Third Trimester	100.0 (4/4)	51.0 – 100.0	99.7 (302/303)	98.2 – 99.9
Retrospective	100.0 (20/20)	83.9 – 100.0	50% (1/2)	NA
First Trimester	100.0 (6/6)	61.0 – 100.0	NA	NA
Second Trimester	100.0 (9/9)	70.1 – 100.0	NA	NA
Third Trimester	100.0 (5/5)	56.6 – 100.0	100.0 (1/1)	20.7 – 100.0
Trimester Unknown	NA	NA	0% (0/1)[a]	NA

NA = Not applicable

[a] Specimen tested non-reactive by Access Syphilis and another FDA-cleared TP-CLIA test.

Clinical Performance in Medically Diagnosed Individuals

Samples were tested from 150 individuals medically diagnosed with primary, secondary, or latent syphilis based on standard of care testing and/or physician diagnosis. These specimens included 49 females and 101 males. Results of the Access Syphilis assay for this cohort are summarized below.

Table 15. Reactivity of the Access Syphilis assay in subjects medically diagnosed with syphilis

Medically Diagnosed Individuals			Access Syphilis Result		Percent Agreement with Expected Outcome
Syphilis Stage	**Treatment Status**	**N**	**Reactive**	**Non-reactive**	
Primary	Treated	27	26	1	96.3
	Untreated	49	49	0	100.0
Secondary	Treated	23	23	0	100.0
	Untreated	13	13	0	100.0
Latent	Treated	25	25	0	100.0
	Untreated	13	7	6[a]	53.8
Total		**150**	**143**	**7**	**95.3**

[a] Specimens tested non-reactive with Access Syphilis and another FDA cleared TP-CLIA.

Clinical Performance in Apparently Healthy Individuals

Of the total 1910 specimens analyzed in the Access Syphilis clinical study, 204 specimens were collected from apparently healthy individuals. This included 87 females, 116 males, and one from a person of unknown sex. Of these, 10 specimens from subjects younger than 21 years old were included. The reactivity of the Access Syphilis assay in the apparently healthy population is shown below.

Table 16. Reactivity of the Access Syphilis assay in the apparently healthy population

Category	Access Syphilis Result		
	Number of Reactive (%)	**Number of Nonreactive (%)**	**Total**
Adults	18 (9.3%)	176 (90.7%)	194
< 21 yrs. old	0 (0.0%)	10 (100.0%)	10
Total	18 (8.8%)	186 (91.2%)	204

1. Clinical Sensitivity:
   The clinical sensitivity of the assay was expressed as positive percent agreement of the Access Syphilis assay results with the final comparator result, as explained above.

2. Clinical Specificity:
   The clinical specificity of the assay was expressed as negative percent agreement of the Access Syphilis assay results with the final comparator result, as explained above.

3. Other Clinical Supportive Data (When 1. and 2. Are Not Applicable):
   Not applicable.

D Clinical Cut-Off:

Not applicable.

E Expected Values:

In this clinical study, there were a total of 1104 specimens prospectively collected from the intended use population and tested with the Access Syphilis assay. The Access Syphilis assay was reactive in 214 subjects, for a 19.4% positivity rate of treponemal antibodies in the studied population.

The distribution of the Access Syphilis reactive and non-reactive results by age and gender is summarized below.

Table 17. Distribution of the Access Syphilis reactive and non-reactive results in the intended use population by age and gender.

Age Range (Yr)	Sex	N	Reactive		Non-reactive	
			# Reactive	% Reactive	# Non-reactive	% Non-reactive
12-20	Male	7	0	0.0%	7	100.0%
	Female	48	2	4.2%	46	95.8%
21-30	Male	44	9	20.4%	35	79.6%
	Female	256	8	3.1%	248	96.9%
31-40	Male	72	36	50.0%	36	50.0%
	Female	211	12	5.7%	199	94.3%
41-50	Male	87	38	43.7%	49	56.3%
	Female	68	9	13.2%	59	86.8%
51-60	Male	123	54	43.9%	69	56.1%
	Female	68	17	25.0%	51	75.0%
61-70	Male	54	16	29.6%	38	70.4%
	Female	38	10	26.3%	28	73.7%

71-89[a]	Male	13	2	15.4%	11	84.6%
	Female	15	1	6.7%	14	93.3%
Overall	Male	400	155	38.8%	245	61.3%
	Female	704	59	8.4%	645	91.6%
Total		**1104**	**214**	**19.4%**	**890**	**80.6%**

[a] One (1) subject was >89 Yr.

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

IX Conclusion:

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