510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE

A. 510(k) Number:

k153651

B. Purpose for Submission:

New Device

C. Measurand:

Thyroid Stimulating Hormone (TSH) in human serum and plasma

D. Type of Test:

Quantitative chemiluminescence immunoassay

E. Applicant:

Beckman Coulter, Inc.

F. Proprietary and Established Names:

Access TSH (3rd IS) Assay and Access TSH (3rd IS) Calibrators on the Access Immunoassay Systems

G. Regulatory Information:

Product Code	Regulation Name	Classification	Regulation Section	Panel
JLW	Thyroid Stimulating Hormone Test System	Class II	21 CFR 862.1690	Clinical Chemistry (75)
JIS	Calibrator	Class II	21 CFR 862.1150	Clinical Chemistry (75)

H. Intended Use:

1. Intended use(s):

   See indication(s) for use below.

2. Indication(s) for use:

   The Access TSH (3rd IS) assay is a paramagnetic particle, chemiluminescent immunoassay for the quantitative determination of human thyroid-stimulating hormone (thyrotropin, TSH, hTSH) levels in human serum and plasma using the Access Immunoassay Systems. This assay is capable of providing 3rd generation TSH results.

   The Access TSH (3rd IS) Calibrators are intended to calibrate the Access TSH (3rd IS) assay for the quantitative determination of human thyroid-stimulating hormone (thyrotropin, TSH, hTSH) levels in human serum and plasma using the Access Immunoassay Systems.

3. Special conditions for use statement(s):

   For prescription use only

4. Special instrument requirements:

   Beckman UniCel Dxl 800 analyzer

I. Device Description:

The Access TSH (3rd IS) assay consists of the reagent pack and calibrators. Other items needed to run the assay include substrate and wash buffer. The Access TSH (3rd IS) assay reagent pack, Access TSH (3rd IS) calibrators, along with the Access wash buffer and substrate are designed for use with the UniCel Dxl 800 analyzer in a clinical laboratory setting.

The Access TSH (3rd IS) reagent pack is ready to use and consists of the following reagents:

- R1a: Paramagnetic particles coated with mouse monoclonal anti- human TSH antibody suspended in TRIS buffered saline, with surfactant, bovine serum albumin (BSA), <0.1% sodium azide, and 0.1% ProClin 300.
- R1b: TRIS buffered saline with surfactant, BSA, protein (murine), < 0.1% sodium azide, and 0.1% ProClin 300.
- R1c: Mouse monoclonal anti-human TSH alkaline phosphatase conjugate in ACES buffered saline, with surfactant, BSA matrix, protein (murine), < 0.1% sodium azide, and 0.25% ProClin 300.
- R1d: Mouse monoclonal anti-human TSH alkaline phosphatase conjugate in ACES buffered saline, with surfactant, BSA matrix, protein (murine), < 0.1% sodium azide, and 0.25% ProClin 300.

Each reagent pack contains enough materials for 100 tests. Reagent packs are stored at 2-10°C.

The calibrator set provides calibrators at six levels; zero and approximately 0.05, 0.30, 3.0, 15.0, and 50.0 µIU/mL. The calibrators are prepared gravimetrically from human thyroid stimulating hormone (hTSH). A description of the calibrators is provided below.

- S0: Buffered bovine serum albumin (BSA) matrix with surfactant, < 0.1% sodium azide and 0.5% ProClin 300. Contains 0 µIU/mL (mIU/L) hTSH.
- S1-S5: Approximately 0.050, 0.30, 3.0, 15.0, and 50.0 µIU/mL (mIU/L) hTSH, respectively, in buffered BSA matrix with surfactant, < 0.1% sodium azide and 0.5% ProClin 300.

The calibrators are contained in 2.5 mL/vials. The calibrator vials are stored at 2-10°C.

The calibration card, included with each kit, contains the bar code that provides the instrument with the individual concentrations for each calibrator level and Assay Protocol File (APF) to run for a particular assay.

The Access Substrate is a dioxetane-based chemiluminescent substrate previously cleared in k922823. No changes are made to the Substrate in this submission.

The Access Wash Buffer II is TRIS buffered saline containing surfactant and preservatives previously cleared in k922823. No change is made to the Wash Buffer in this submission.

Commercially available controls are required but not provided.

J. Substantial Equivalence Information:

1. Predicate device name(s):
   Access HYPERsensitive hTSH assay
   Access HYPERsensitive hTSH Calibrators

2. Predicate 510(k) number(s):
   k954825

3. Comparison with predicate:

Similarities		
Item	Candidate Access TSH (3rd IS) Assay K153651	Predicate Access HYPERsensitive hTSH assay k954825
Intended use	The Access TSH (3rd IS) assay is a paramagnetic particle, chemiluminescent immunoassay for the quantitative determination of human thyroid-stimulating hormone (thyrotropin, TSH, hTSH) levels in human serum and plasma using the Access Immunoassay Systems. This assay is capable of providing 3rd generation TSH results.	The Access HYPERsensitive hTSH assay is a paramagnetic particle, chemiluminescent immunoassay for the quantitative determination of human thyroid-stimulating hormone (thyrotropin, hTSH) levels in human serum and plasma using the Access Immunoassay Systems. This assay is capable of providing 3rd generation (HYPERsensitive hTSH (and/or 2nd generation (Fast hTSH) results.
Analyte Measured	Human thyroid-stimulating hormone (thyrotropin, TSH, hTSH)	Same
Technology	Sandwich immunoassay	Same
Format	Chemiluminescent	Same
Method	Automated	Same
Sample Type	Serum or plasma	Same
Stability	Stable at 2 to 10°C for 28 days	Same

Differences		
Item	Candidate Access TSH (3rd IS) Assay k153651	Predicate Access HYPERsensitive hTSH assay k954825
Standardization	WHO 3rd International Reference Preparation Thyroid Stimulating Hormone, Human (NIBSC Coded 81/565)	WHO 2nd International Reference Preparation Thyroid Stimulating Hormone, Human (NIBSC Coded 80/558)
Measuring Range	0.01 – 50.0 µIU/mL	0.01 – 100 µIU/mL

Similarities		
Item	Candidate Access TSH (3rd IS) Calibrator k153651	Predicate Access HYPERsensitive hTSH Calibrators k954825
Intended use	The Access TSH (3rd IS) Calibrators are intended to calibrate the Access TSH (3rd IS) assay for the quantitative determination of human thyroid-stimulating hormone (thyrotropin, TSH, hTSH) levels in human serum and plasma using the Access Immunoassay Systems.	Same
Calibrator Matrix	Bovine serum albumin	Same
Calibration Curve Stability	28 days	Same

Differences		
Item	Candidate Access TSH (3rd IS) Calibrator k153651	Predicate Access HYPERsensitive hTSH Calibrators k954825
Calibrator Levels	6 levels (0 µIU/mL, and approximately 0.050, 0.30, 3.0, 15.0, and 5.0 µIU/mL)	6 levels (0 µIU/mL, and approximately 0.1, 0.5, 4.0, 10.0, and 100.0 µIU/mL)
Standardization	WHO 3rd International Reference Preparation Thyroid Stimulating Hormone, Human (NIBSC Coded 81/565)	WHO 2nd International Reference Preparation Thyroid Stimulating Hormone, Human (NIBSC Coded 80/558)
Stability	The calibrator shelf life is 180 days when stored unopened at 2-10°C. Vials are stable at 2 to 10°C for 90 days after initial use.	Vials are stable at 2 to 10°C until expiration date stated on the label.

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

CLSI EP05-A3: Evaluation of Precision of Qualitative Measurement Methods Procedures; Approved Guideline

CLSI EP6-A: Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline

CLSI EP7-A2: Interference Testing in Clinical Chemistry; Approved Guideline

CLSI EP09-A3: Measurement Procedure Comparison and Bias Estimation Using Patient Samples; Approved Guideline

CLSI EP17-A2: Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline

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

CLSI EP28-A3c: Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory; Approved Guideline

L. Test Principle:

The Access TSH (3rd IS) assay is a two-site immunoenzymatic ("sandwich") assay. A sample is added to a reaction vessel with mouse anti-hTSH-alkaline phosphatase conjugate, buffered protein solution and paramagnetic particles coated with immobilized mouse monoclonal anti-hTSH antibody. The hTSH binds to the immobilized monoclonal anti-hTSH antibody on the solid phase while the mouse anti-hTSH-alkaline phosphatase conjugate reacts with a different antigenic site on the hTSH. After incubation in a reaction vessel, materials bound to the solid phase are held in a magnetic field while unbound materials are washed away. Then, the chemiluminescent substrate Lumi-Phos 530 is added to the vessel and light generated by the reaction is measured with a luminometer. The light production is directly proportional to the concentration of TSH in the sample. The amount of analyte in the sample is determined from a stored, multi-point calibration curve.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Precision studies were performed following CLSI EP5-A3. Four serum samples with approximate TSH concentrations of 0.02, 0.30, 5.0, and 39.0 µIU/mL were tested. Each sample was assayed in duplicate, in two runs per day, over twenty days for a total of 40 runs and 80 replicates. The study was run on one UniCel Dxl 800 Immunoassay System, using three reagent pack lots and one calibrator lot. The within-run, between-day, between-run, and total imprecision was calculated for each sample, for each instrument and lot combination. Results from multiple lots were similar. Results from one representative lot are provided in the table below:

Sample	Mean (µIU/mL)	Within run		Between run		Between day		Total	
		SD (µIU/mL)	CV (%)	SD (µIU/mL)	CV (%)	SD (µIU/ml)	CV (%)	SD (µIU/mL)	CV (%)
1	0.02	0.0004	1.8	0.0004	4.0	0.0008	3.7	0.0010	4.4
2	0.37	0.006	1.5	0.006	2.0	0.010	2.7	0.013	3.5
3	4.71	0.13	2.7	0.008	0.2	0.11	2.4	0.17	3.6
4	38.76	1.36	3.5	0.49	1.0	1.80	4.6	2.31	5.9

b. Linearity/assay reportable range:

A linearity study was performed following CLSI EP6-A, one high serum sample (spiked with human TSH to approximately 50.0 µIU/mL) and one low serum sample (< 0.01 µIU/mL) were mixed to make nine sample concentrations evenly distributed across the analytical measuring range. Four replicates of the seven mixed samples,

eight replicates of the low sample, and four replicates of the high sample were tested on one UniCel Dxl 800 Immunoassay System using three reagent pack lots and one calibrator lot. Linearity study data were analyzed by weighted linear regression and the observed values (mean of the replicates) were compared to the best fitted straight line (predicted values). The linearity regression results are shown below. Results from other lots were similar.

$$y = 1.007x - 3.24\text{x}10^{-6}$$

The Access TSH (3rd IS) assay claimed measuring range is from 0.01 to 50.0 µIU/mL.

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

Traceability:

The measurand (TSH) in the Access TSH (3rd IS) Calibrator is traceable to WHO 3rd International Reference Preparation, (NIBSC Coded 81/565). The traceability process is based on EN ISO17511.

Stability:

Stability protocols and acceptance criteria for real-time, opened and closed calibrators were reviewed and found to be acceptable.
The calibrator shelf life is 180 days when stored unopened at 2-10°C and the calibrator open vial stability claims is 90 days when stored at 2-10°C.

Value Assignment:

Concentrations of calibrators are assigned through an internal procedure. Primary working calibrators are prepared from WHO 81/565 and secondary working calibrators are value assigned using the primary working calibrators and the Access Immunoassay System. Primary working calibrators and the secondary working calibrators are run together on the Access Immunoassay System in a matched, multi-replicate process. Product (commercial) calibrators are value assigned using the secondary working calibrators on the Access Immunoassay System following a similar value assignment process. The Access TSH (3rd IS) Calibrators are lot specific and provided at six levels; zero and approximately 0.05, 0.30, 3.0, 15.0, and 50.0 µIU/mL (mIU/L).

d. Detection limit:

LoB, LoD, and LoQ below were determined according to CLSI EP17-A2.

i. Limit of Blank (LoB)

The LoB study was run on two UniCel Dxl 800 Immunoassay Systems, using three reagent pack lots and one calibrator lot. One hundred twenty (120) total replicates for each reagent pack lot were tested. Of the total replicates, ninety (90) were comprised of zero level calibrators (30 replicates of each of three lots) and thirty (30) were comprised of Access Wash Buffer II that were measured for the LoB determination. The LoB was determined to be 0.0004 µIU/mL using the 95% non-parametric upper reference limit of the 120 replicates.

ii. Limit of Detection (LoD)

The LoD study was run using five serum samples with low TSH concentrations on two UniCel Dxl 800 Immunoassay Systems, using three reagent pack lots and one calibrator lot. In total, forty-five (45) replicates (nine replicates per day over five days) of each of the five low serum samples (225 replicates per pack lot for a total of 675 replicates) were measured for the LoD determination. The LoD was determined to be 0.001 µIU/mL based on the precision model, multiplied by the 95th percentile of the standard normal distribution and added to the LoB to calculate the LoD.

iii. Limit of Quantitation (LoQ)

The LoQ study was run using eight serum samples with TSH concentrations between LoB and 0.05 µIU/mL. Seven of these samples were used to determine LoQ as the results for the sample at LoB were excluded. Samples were tested on one UniCel Dxl 800 Immunoassay System, using three reagent pack lots and one calibrator lot. Each of the seven samples were run in replicates of nine, in one run per day, for five days on three reagent pack lots (45 replicates of each sample on each reagent lot). In total, one hundred thirty-five (135) replicates of each of the seven serum samples (945 total replicates) were measured for the LoQ determination. The mean concentration and between-run percent coefficient of variation (CV) were calculated for each sample on each day. The concentration that demonstrated 10% between-run CV is the LoQ. Based on this study the LoQ for Access TSH (3rd IS) assay is 0.001µIU/mL.

Analyte	LoB	LoD	LoQ
Access TSH (3rd IS)	0.0004 µIU/mL	0.001 µIU/mL	0.001 µIU/mL

The Access TSH (3rd IS) assay claimed measuring range is 0.01 to 50.0 µIU/mL.

e. Analytical specificity:

i. Interference:

Following CLSI EP7-A2 Interference Testing in Clinical Chemistry-Approved Guideline an interference study was performed assessing common sample abnormalities (including hemolysis, icterus, and lipemia), common prescription drugs, over-the-counter drugs, and medications most often prescribed in the patient population for which the test is ordered. Two serum samples containing hTSH concentrations of approximately 0.30 µIU/mL and 5.0 µIU/mL were spiked with multiple concentrations of the substances below and run on one UniCel Dxl 800 Immunoassay System using three reagent pack lots and one calibrator lot. Five replicates were tested for the control sample and each of the two spiked sample preparations. Interference was determined by testing controls (no interfering substance added) and matched test samples (with interfering substance added). The sponsor defines significant interference as a bias in results of > 10% as compared to the control sample. Of the compounds tested, none were found to cause significant interference using the highest test concentrations indicated in the table below;

Interferent	Interferent Concentration (Low Level)	Interferent Concentration (High Level)
Acetaminophen	30 µg/mL	200 µg/mL
Acetylsalicylic Acid	400 µg/mL	650 µg/mL
Bilirubin (conjugated)	0.3 mg/dL	45 mg/dL
Bilirubin (unconjugated)	0.12 mg/dL	40 mg/dL
Hemoglobin	200 mg/dL	1,000 mg/dL
Heparin (Sodium)	1 U/mL	3 U/mL
Human Growth Hormone (hGH)	20 ng/mL	134 ng/mL
Human Serum Albumin (HSA)	5,100 mg/dL	6,000 mg/dL

Ibuprofen	70 µg/mL	500 µg/mL
Multivitamin (Centrum Liquid)	0.3% (V/V)	0.9% (V/V)
Triglycerides (Intra Lipid)	330 mg/dL	3300 mg/dL

ii. Cross-reactivity:

A cross-reactivity study was performed to evaluate the potential cross-reactivity of the assay with other substances that are similar in structure to hTSH. Serum samples containing hTSH concentrations of approximately 0.30 µIU/mL and 5.0 µIU/mL were spiked with multiple concentrations of the substances below and run on one UniCel Dxl 800 Immunoassay System using three reagent pack lots and one calibrator lot. Five replicates were tested for the control sample and each of the two spiked sample preparations. Results from these spiked samples were evaluated against that of a sample that had been spiked with Tris and/or Phosphate Buffered Saline, defined as the control. The mean dose of the replicates was calculated for the control samples and the spiked sample preparations. The difference was calculated as a percent difference in dose from the expected control dose. The sponsor claims that samples containing substances at the concentrations listed below do not affect the concentration of hTSH reported:

Substance	Highest Concentration Added (mIU/mL)	Cross-reactivity (%)
hCG	1,000,000	< 0.010%
hFSH	1,000	< 0.10%
hLH	3,000	< 0.10%

iii. High Dose Hook Effect:

To determine any hook effect, a known concentration of a WHO reference standard (WHO 81/565) was spiked into synthetic calibrator matrix, and dilutions were prepared from the initial spiked sample to determine the presence of any hook effect. The study was run on one UniCel Dxl 800 Immunoassay System, using three reagent pack lots and one calibrator lot. Five replicates were tested for each sample preparation. The hook effect is defined as the concentration above the highest (S5) calibrator at which sample RLUs drop below the RLUs of the highest (S5) calibrator. No high dose hook effect was observed at concentrations up to 1000 µIU/mL.

iv. HAMA / Heterophile:

Verification studies were performed on characterized human anti- mouse antibody (HAMA) / heterophile samples to evaluate blocker effectiveness in the Access TSH (3rd IS) assay. The Access TSH (3rd IS) reagent pack has been formulated to minimize the effects of HAMA / heterophile interference. While the sponsor states that Access TSH (3rd IS) reagent pack has been formulated to minimize the effects of interference, the study showed interference in some cases. The following cautionary note has been included in the "Limitations of the Procedure" section of the product insert:

"*For assays employing antibodies, the possibility exists for interference by heterophile antibodies in the patient sample. Patients who have been regularly exposed to animals or have received immunotherapy or diagnostic procedures utilizing immunoglobulins or immunoglobulin fragments may produce antibodies, e.g., HAMA, that interfere with immunoassays.*"

f. Assay cut-off:

See detection limit above.

2. Comparison studies:

a. Method comparison with predicate device:

A method comparison study was performed comparing the Access TSH (3rd IS) assay to the Access HYPERsensitive hTSH Assay, using a protocol based on CLSI EP09-A3, Measurement Procedure Comparison and Bias Estimation Using Patient Samples; Approved Guideline – Third Edition. A total of one hundred fifty-five (155) frozen serum samples were run in singleton. The study was run at two external sites on UniCel DxI 800 Immunoassay Systems, using three reagent pack lots and one calibrator lot for the TSH (3rd IS) assay. The Passing-Bablok regression analysis results between Access TSH (3rd IS) values (dependent variable, y) and Access HYPERsensitive hTSH values (x, predicate), are shown below:

Method Comparison Access TSH (3rd IS) on DxI 800
vs. Access HYPERsensitive hTSH

N	Concentration Range (µIU/mL)	Intercept (95% CI) (µIU/mL)	Slope (95% CI)	Correlation Coefficient (r)
155	Test: 0.056 - 42.50 Ref: 0.066 - 47.67	-0.02 (-0.05 to 0.00)	0.940 (0.92 to 0.97)	0.98

b. Matrix comparison:

The sample types for the matrix comparison study were serum (no gel), serum (gel) and lithium- heparin plasma. 79 matched sets of serum and plasma (lithium-heparin) samples were used. Nine (9) altered samples were prepared by spiking known concentrations of human TSH into all three sample types, which had been previously stripped to remove all TSH. These altered samples were prepared to TSH concentrations at a range of approximately 0.01 µIU/mL to 0.2 µIU/mL. The sample concentrations tested were 0.01 to 43.55 mIU/mL for lithium plasma, 0.01 to 39.04 mIU/mL for serum (gel) and 0.01 to 38.85 mIU/mL for serum (no gel). Spiked samples were prepared using human TSH spiked into all three sample types, which had been previously stripped to remove all TSH. The study was run on one instrument using one reagent pack lot and one calibrator lot. One replicate per sample type was analyzed. Linear regression results are shown below:

Sample Type	N	Regression Analysis	Correlation Coefficient (r)
Serum (no gel) vs. serum (gel)	79	y = 1.00x - 0.0002	1.00
Serum (gel) vs. lithium heparin	79	y = 1.00x + 0.0002	0.99
Serum (no gel) vs. lithium heparin plasma	79	y = 1.00x + 0.0004	0.99

Clinical studies:

a. Clinical Sensitivity:

Not applicable

b. Clinical specificity:

Not applicable

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

Not applicable

4. Clinical cut-off:

Not applicable

5. Expected values/Reference range:

The populations used in this study included a general population of approximately equal numbers of males and non-pregnant females between the ages of 21-88, and pregnant female populations, with approximately equal distribution across all three trimesters. Approximately four hundred (367) subjects were enrolled in each population.

After enrollment, the subjects' serum samples were screened for positive thyroid peroxidase antibody (TPOAb) and thyroglobulin antibody (TgAb) using the Beckman Coulter TPO Antibody and Thyroglobulin Antibody II assays prior to reference interval analyses. Samples with positive TPOAb or TgAb results (approximately 10%) were excluded from analysis of the TSH reference intervals. The remaining serum samples were analyzed according to their population. All samples were run in singleton. The study was run at two external sites on UniCel Dxl 800 Immunoassay Systems, using one reagent pack lot and one calibrator lot for the TSH (3rd IS) assay.

The data was analyzed separately for each population using the non-parametric method and following CLSI C28-A3. Additionally, a two-sided non-parametric 95% confidence interval (CI) was used for the analysis. The study was run to establish the central 97.5% reference interval of healthy, normal euthyroid adult and pregnant female populations, as indicated.

Population	Sample Size	Median (µIU/mL)	Range (µIU/mL)	97.5% Reference (µIU/mL)
General Adult Population (non-pregnant females and males) ages 21 and older	367	1.48	0.32 - 7.08	0.45 - 5.33
Pregnant females – 1st Trimester	318	1.13	0.009 - 5.89	0.05 - 3.70
Pregnant females – 2nd Trimester	362	1.47	0.028 - 5.78	0.31 - 4.35
Pregnant females – 3rd Trimester	335	1.61	0.27 - 10.25	0.41 - 5.18

N. Proposed Labeling:

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

O. Conclusion:

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