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

A. 510(k) Number:
k132515

B. Purpose for Submission:
New Device

C. Measurand:
Intact Parathyroid Hormone (PTH)

D. Type of Test:
Quantitative, two-site sandwich chemiluminescent immunoassay

E. Applicant:
Diasorin Inc.

F. Proprietary and Established Names:
LIAISON® N-TACT® PTH Gen II
LIAISON® N-TACT® PTH Gen II Control Set
LIAISON® N-TACT® PTH Gen II Calibration Verifiers

G. Regulatory Information:

1. Regulation section:
   21 CFR 862.1545, Parathyroid Hormone Test System
   21 CFR 862.1660, Quality Control Material (assayed and unassayed)

2. Classification:
   Class II
   Class I, reserved

3. Product code:

   CEW
   JJX

4. Panel:

   Clinical Chemistry (75)

H. Intended Use:

1. Intended use(s):

   See Indication(s) for use below.

2. Indication(s) for use:
   The LIAISON® N-TACT® PTH Gen II is an *in vitro* chemiluminescent immunoassay (CLIA) intended for the quantitative determination of intact human parathyroid hormone in serum, EDTA and Lithium Heparin plasma samples. Measurements of parathyroid hormone levels are used in the differential diagnosis of hypercalcemia and hypocalcemia resulting from disorders of calcium metabolism. The test is to be performed on the LIAISON XL analyzer.

   The LIAISON® N-TACT® PTH Gen II Control Set is intended for use as assayed quality control samples to monitor the accuracy and precision of the DiaSorin LIAISON® N-TACT® PTH Gen II assay.

   The LIAISON® N-TACT® PTH Gen II Calibration Verifiers are assayed quality control materials intended for the quantitative verification of calibration and reportable range of the LIAISON® N-TACT® PTH Gen II assay.

3. Special conditions for use statement(s):

   For prescription use only.

4. Special instrument requirements:

   For use on the DiaSorin LIAISON® XL Analyzer

I. Device Description:

The LIAISON® N-TACT® PTH Gen II Assay in an in vitro diagnostic device consisting of reagents provided in individual compartments within a plastic container identified as the LIAISON Reagent Integral. The Reagent Integral contains:

- Magnetic particles coated with goat polyclonal antibody against 39-84 PTH peptide, BSA, phosphate buffer, surfactant, 0.1% ProClin 300 and 0.05% gentamicin sulfate.
- Conjugate: Goat polyclonal antibody against 1-34 PTH peptide conjugated to an isoluminol derivative, in phosphate buffer, BSA, surfactant, 0.1% ProClin 300 and 0.05% gentamicin sulfate.
- Assay buffer: Phosphate buffer, BSA, surfactant, blockers, 0.09% ProClin 300 and 0.045% gentamicin sulfate.

LIAISON® N-TACT® PTH Gen II Calibrators
Two vials each of Calibrator 1 and Calibrator 2 are included in the reagent kit. The calibrators consist of phosphate buffer, 1-84 PTH, surfactants, BSA, EDTA and 0.2% ProClin 300. Calibrator 1 is manufactured to have a target intact PTH level of 15 pg/mL and Calibrator 2 is manufactured to have a target intact PTH level of 1300 pg/mL.

LIAISON® N-TACT® PTH Gen II Control set contains;
2 levels controls containing 80% human plasma spiked with 1-84 PTH, and preservatives; 4 vials each level; lyophilized
Control Level 1 is manufactured to have a target intact PTH level of 20 pg/mL
Control Level 2 is manufactured to have a target intact PTH level of 300 pg/mL

LIAISON® N-TACT® PTH Gen II Calibration Verifier set contains:
4 levels containing 80% human plasma spiked with 1-84 PTH, with preservative, 1 vial each level, lyophilized
The target concentration for Cal verifier A is 10 pg/mL.
The target concentration for Cal verifier B is 150 pg/mL.
The target concentration for Cal verifier C is 650 pg/mL.
The target concentration for Cal verifier D is 1600 pg/mL.

Each serum/plasma donor unit used in the preparation of these products has been tested by an U.S. FDA approved method and found non-reactive for the presence of the antibody to Human Immunodeficiency Virus 1 and 2, the Hepatitis B surface antigen, and the antibody to Hepatitis C.

J. Substantial Equivalence Information:

1. Predicate device name(s):
   Siemens ADVIA CENTAUR INTACT Parathyroid Hormone (iPTH) Assay
   LIAISON® N-TACT® PTH Control Set
   LIAISON® N-TACT® PTH Calibration Verifiers

2. Predicate 510(k) number(s):
   k020217
   k033426
   k093498

3. Comparison with predicate:

Reagent

Similarities		
Item	Device LIAISON® N-TACT® PTH Gen II	Predicate ADVIA Centaur iPTH (K020217)
Intended Use	For in vitro quantitative determination of intact human parathyroid hormone	Same
Calibration	Two-point calibration	Same
Antibody	Goat Polyclonal	Same
Reagent storage	On-board or in refrigerator at 2-8°C	Same

Reagent

Differences		
Item	Device LIAISON® N-TACT® PTH Gen II	Predicate ADVIA Centaur iPTH (K020217)
Measuring Range	3.0 -1900 pg/mL	2.5-1900 pg/mL
Sample Matrix	EDTA Plasma, Serum, SST serum ,and Lithium Heparin Plasma	EDTA Plasma, Serum
Sample Size	150 µL	200 µL
Open Storage on analyzer	56 days	28 days
Calibrators	2 levels, included in kit	2 levels, not included in kit
Calibration interval	28 days	14 days
Reference Range	14.5-87.1 pg/mL	11.1-79.5 pg/mL

Control

Similarities and differences		
Item	Device LIAISON® N-TACT® PTH Gen II Control set	Predicate LIAISON® N-TACT® PTH Control set (k033426)
Intended Use	Quality control materials for the PTH assay	Same
Levels	2 levels: low and high	Same
Storage	Store at 2-8°C until ready to use	Same

Calibration Verifiers

Similarities and differences		
Item	Device LIAISON® N-TACT® PTH Gen II Calibration Verifier set	Predicate LIAISON® N-TACT® PTH Gen II Calibration Verifier (k093498)
Intended Use	Assayed quality control materials for the quantitative verification of the PTH calibration.	Same
Levels	4 levels, lyophilized	Same
Storage	Store at 2-8°C until ready to use	Same
Volume	2.0 mL	Same

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

- EP5-A2, Evaluation of Precision Performance of Quantitative Measurement Methods; Approved CLSI Guideline
- EP6-A, Evaluation of Linearity of Quantitative Analytical Methods; Approved CLSI Guideline
- EP7-A2, Interference Testing in Clinical Chemistry; Approved CLSI Guideline
- EP9-A2-IR, Method Comparison and Bias Estimation Using Patient Samples; Approved CLSI Guideline
- CLSI Guideline EP17-A2 Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures
- CLSI Guideline C28-A3, Defining, Establishing and Verifying Reference Intervals in the Clinical Laboratory

L. Test Principle:

The LIAISON® N-TACT® PTH Gen II assay is a modified two-step, two-site sandwich assay that uses two goat polyclonal antibodies for capture and detection of intact PTH. Results are determined by a 2 point calibration conversion of the master curve to a working curve. The light signal is measured by a photomultiplier as relative light units (RLU) and is proportional to the concentration of intact PTH present in the calibrators, controls or samples.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Precision testing was performed following CLSI Guideline EP5-A2. A coded panel comprised of 7 frozen EDTA plasma samples spanning the assay range and 2 lots of LIAISON® N-TACT® PTH Gen II controls (2 levels) were tested in the study. The precision panel samples and kit controls were tested on two lots of LIAISON® N-TACT® PTH Gen II in two replicates per run, 2 runs per day for 20 operating days for

a total of 160 replicate results per sample. The 20 day results are summarized for the combined reagent lot numbers as sample mean PTH concentration in pg/mL, standard deviations and coefficient of variation (%CV) for between lot and Total across lots. Precision results are summarized in the table below:

Sample ID	n	Mean PTH (pg/mL)	Between-Lot		Total (Across Lots)	
			SD	%CV	SD	%CV
Lot 1 Kit Control1	160	19.3	0.26	1.3%	0.65	3.3%
Lot 1 Kit Control 2	160	250	9.27	3.7%	8.84	3.5%
Lot 2 Kit Control1	160	18.5	0.28	1.5%	0.57	3.1%
Lot 2 Kit Control 2	160	252	10.59	4.2%	9.05	3.6%
EDTA Plasma 1	160	12.6	0.23	1.8%	0.53	4.2%
EDTA Plasma 2	160	34.5	0.70	2.0%	1.38	4.0%
EDTA Plasma 3	160	86.2	1.68	1.9%	3.07	3.6%
EDTA Plasma 4	160	156.1	5.25	3.4%	5.54	3.5%
EDTA Plasma 5	160	605	24.62	4.1%	19.37	3.2%
EDTA Plasma 6	160	1348	46.58	3.5%	44.06	3.3%
EDTA Plasma 7	160	1477	84.06	5.7%	40.62	2.8%

b. Linearity/assay reportable range:

One patient sample for each sample type (Serum, SST serum, EDTA and Lithium Heparin plasma) was spiked with PTH 1-84 to achieve PTH levels above the measuring range of the assay and diluted with a normal PTH patient sample of the same type to achieve PTH levels in the lower portion of the measuring range. In order to achieve PTH values in the very low range, the low normal sample was further diluted with sample diluent (containing no PTH) to levels near the claimed LoQ of the assay. Each of the 18 concentrations for each sample type was tested in triplicate on the LIAISON XL analyzer with one lot of reagent. The resulting regression equations for each sample type are summarized below:

Serum: $y= 0.9767x - 3.624$; $R^2 = 0.9982$
SST Serum: $y=0.9742x + 3.856$; $R^2 = 0.9987$
EDTA plasma: $y=1.012x – 4.127$; $R^2 = 0.9983$
Lithium Heparin plasma: $y=0.9461x + 3.696$; $R^2 = 0.9992$

The linearity results support the claimed measuring range of this device (3.0-1900 pg/mL) for serum, SST serum, EDTA plasma, and Lithium Heparin plasma samples.

<u>Hook Effect Study</u>

The sponsor performed a hook effect study to determine if there is high dose hook effect.

Testing was performed with 1 kit lot of LIASON N-TACT PTH Gen II. A zero sample was spiked with 1-84 PTH (NIBSC 95/646) to achieve PTH concentrations several times above the assay measuring range of 1900 pg/mL. The spiked samples were measured in triplicate. The study demonstrated that no high dose hook effect was observed at values up to 1,000,000 pg/mL of 1-84 PTH.

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

Traceability

The LIAISON PTH N-TACT Calibrators are traceable to master standards which are prepared from the WHO International standard PTH (1-84) human recombinant. (NIBSC 95/646)

Value Assignment

Calibrator: The concentration of the kit calibrators are assigned through an internal procedure. The LIAISON N-TACT PTH Gen II calibrators are tested on a minimum of 3 LIAISON XL Analyzers, in a minimum of 5 assay runs with six replicates per vial resulting in a minimum of 30 individual replicate results used for final value range assignment. The LIAISON N-TACT PTH Gen II kit calibrators have the following target intact PTH values:

Level 1: 15 pg/mL
Level 2: 1300 pg/mL

Control: A minimum of 10 vials of each level of control are used in the final range assignment. The LIAISON N-TACT PTH Gen II Control Set are tested on a minimum of 3 LIAISON XL Analyzers, using 2 different assay kit lots, in a minimum of 5 assay runs. Two vials of each level are tested in each of the 5 separate assay runs with four replicates per vial resulting in a minimum of 40 individual replicate results used for final value assignment. The LIAISON N-TACT PTH Gen II Control Set has the following target intact PTH ranges:
Control Level 1: 20 pg/mL
Control Level 2: 300 pg/mL

Calibration Verifiers: The 4 levels of calibration verifiers (A-D) are intended to span the assay range of the LIAISON N-TACT PTH Gen II assay. The target concentrations for each lot of calibration verifiers are assigned at the manufacturer by testing a minimum of 12 vials of each level of calibration verifier on 2 different LIAISON N-TACT PTH Gen II assay kit lots on a minimum of 4 LIAISON XL Analyzers, in a minimum of 6 runs with 4 replicates per vial resulting in a minimum of 48 individual replicate results per control level for final value assignment. The target concentrations for cal verifiers are:

Cal verifier A: 10 pg/mL
Cal verifier B: 150 pg/mL
Cal verifier C: 650 pg/mL
Cal verifier D:1600 pg/mL

Stability

Reagent: On-board stability for the LIAISON N-TACT PTH Gen II assay was established by real time studies on the LIAISON XL Analyzer. The stability study protocol and the acceptance criteria have been found acceptable. The on-board stability of the reagent is 56 days with a calibration interval of 28 days. The LIAISON N-TACT PTH Gen II assay reagent is stable until the date printed on the label when stored at 2-8˚C.

Control: On-board stability for the LIAISON N-TACT PTH Gen II Control Set and the LIAISON N-TACT PTH Gen II Calibrators was established by real time studies on the LIAISON XL Analyzer. The stability study protocol and the acceptance criteria have been found acceptable. The on-board stability of the reconstituted calibrators is 7 hours. The on-board stability of the reconstituted controls is 8 hours. The reconstituted controls are stable for 7 hours at room temperature or 8 hours at 2-8˚C .The lyophilized LIAISON N-TACT PTH Gen II Control Set is stable until the date printed on the label when stored at 2-8˚C.

Calibrator: On-board stability for the LIAISON N-TACT PTH Gen II Calibrators was established by real time studies on the LIAISON XL Analyzer. The stability study protocol and the acceptance criteria have been found acceptable. The on-board stability of the reconstituted calibrators is 7 hours. The reconstituted calibrators are stable for 48 hours at 2-8˚C.The lyophilized LIAISON N-TACT PTH Gen II Calibrators is stable until the date printed on the label when stored at 2-8˚C.

Calibration Verifiers: Stability for the LIAISON N-TACT PTH Gen II Calibration Verifiers was established by real time studies on the LIAISON XL. The stability study protocol and the acceptance criteria have been found acceptable. The DiaSorin LIAISON N-TACT PTH GEN II Calibration Verifiers are stable until the expiration date on the vials when stored at 2-8˚C. Reconstituted calibration verifiers are stable for 7 hours at room temperature or 8 hours at 2-8 ˚C. Remaining calibration verifier material should be aliquoted and stored frozen at -20 ˚C. Calibration verifiers are stable through 1 freeze thaw cycle.

d. Detection limit:

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined according to CLSI EP17-A2.

Limit of Blank (LoB) : Five blank EDTA plasma samples were tested on two analyzers, with two reagent lots over six runs (2 replicates/sample run) on three days for a total of 60 results per reagent lot.

Limit of Detection (LoD): Four EDTA plasma samples in the range of the mean LoB to 4 times the mean LoB were tested with the LIAISON® N-TACT® PTH Gen II assay on two analyzers with two reagent lots , over six runs (2 replicates/ sample/run) over three days yielding a total of 96 results.

Limit of Quantitation (LoQ): Nine EDTA plasma samples in the range of 0 to 10 pg/mL were tested on two analyzers with two reagent lots over six runs (2 replicates/sample run) and three days, for a total of 144 results. LoQ is defined as the lowest concentration with an inter-assay precision of ≤ 20% CV.

The following detection limits results were determined with the LIAISON® N-TACT® PTH Gen II Assay:

LoB	LoD	LoQ
≤ 0.5 pg/mL	1.5 pg/mL	3.0 pg/mL

The claimed measuring range of the assay is 3.0 -1900 pg/mL

e. Analytical specificity:

Endogenous Interferent Study*:*
The potential interferents listed below were tested at two PTH levels in EDTA Plasma (70 and 150 pg/mL). Each sample was spiked with the respective interferent at the concentrations shown below. The spiked samples were compared against controls containing no interferent. The two sets of matched spiked and unspiked samples containing each interferent were tested in 12 replicates using one reagent lot. The sponsor defines non-significant interference as results within ± 10% of the control value. The substances listed below were found to cause no significant interference in the LIAISON® N-TACT® PTH Gen II at the highest concentration listed below.

Substance	**Highest Concentration at which no significant interference (≥±10%)was observed**
Hemoglobin	500 mg/dL
Bilirubin (conjugated)	40 mg/dL
Bilirubin (unconjugated)	20 mg/dL
Triglycerides	3,000 mg/dL
Cholesterol	500 mg/dL
Albumin	12 g/dL
Rheumatoid Factor (RF)	612 IU/L
Human Anti-mouse antibody (HAMA)	2670 ng/mL

Drug Interference Study:
A drug interference study was performed to determine whether the presence of exogenous substances such as common pharmaceutical compounds may interfere with the LIAISON N-TACT PTH Gen II results. All interferents were tested with two EDTA

plasma samples with PTH target concentrations of approximately 70 pg/mL and 150 pg/mL. Each sample level was spiked with the respective interferent at levels indicated in the table below. The same samples without interferent were tested as controls. The two sets of matched spiked and unspiked samples were tested in 12 replicates on 1 lot of the LIAISON N-TACT PTH Gen II assay. The sponsor defines non-significant interference as results within ± 10% of the control value. The substances listed below were found to cause no significant interference in the LIAISON® N-TACT® PTH Gen II at the highest concentration listed below.

Drug/Substance	Highest Concentration at which no significant interference (≥±10%)was observed
Acetaminophen	0.2 mg/mL
Acetylsalicylic Acid	0.65 mg/mL
Salicylic Acid	0.6 mg/mL
Ibuprofen	0.5 mg/mL
Alendronate	0.08 mg/mL
Etidronate	1.05 mg/mL
Pamidronate	0.18 mg/mL
Risedronate	0.06 mg/mL
Vitamin D2	240 ng/mL
Vitamin D3	240 ng/mL
Calitriol	1 ng/mL
Alfacalcidol	2.5 µg/mL
Biotin	1 µg/mL
Calcium Acetate	0.4 mg/mL
Calcium Citrate	0.4 mg/mL
Magnesium Chloride	0.4 mg/mL
Aluminum Sulfate	0.4 mg/mL
Lanthanum Chloride	0.4 mg/mL

Cross-Reactivity Study*:*

Cross-Reactivity of PTH fragment 7-84 was tested by spiking 3 concentration levels of either lyophilized PTH 7-84 antigen or WHO PTH 1-84 (NIBSC 95/646) into a zero sample and normalizing the recovery of PTH 7-84 to PTH 1-84 (NIBSC 95/646). Both the PTH 7-84 and PTH 1-84 were reconstituted in specimen diluent to a concentration of 10,000 pg/mL and then diluted to three equivalent concentrations across the assay range. Samples were tested in triplicate on one kit lot of the LIAISON® N-TACT® PTH Gen II assay. The results demonstrated that all substances, except for PTH (7-84), showed less than 0.01% cross reactivity.

Cross-Reactant	Spiked Concentration	% Cross Reactivity
PTH (7 – 84)	1200 pg/mL	53%
PTH (1 – 34)	200,000 pg/mL	< 0.01%
PTH (39 - 68)	200,000 pg/mL	< 0.01%
PTH (44 – 68)	200,000 pg/mL	< 0.01%
PTH (39 – 84)	200,000 pg/mL	< 0.01%
PTH (53 – 84)	200,000 pg/mL	< 0.01%
Calcitonin	200,000 pg/mL	< 0.01%
C-Telopeptide (CTX-1)	200,000 pg/mL	< 0.01%
Osteocalcin	200,000 pg/mL	< 0.01%

The sponsor included the following limitation statement in the assay's package insert:

The LIAISON® N-TACT® PTH Gen II assay will detect non-Intact PTH (1-84) such as PTH fragment (7-84).PTH fragment (7-84) may cause falsely elevated Intact PTH results in patients with abnormal renal function because these patients may have various concentrations of PTH fragment (7-84) in their blood. In patients with abnormal renal function please interpret the Intact PTH results with caution and do not make patient management decisions on the Intact PTH result alone.

f. Assay cut-off:
Not Applicable

2. Comparison studies:

a. Method comparison with predicate device:
Following the CLSI EP9-A2 guidance document, the sponsor performed a method comparison study on 199 EDTA plasma samples from various sources. The samples included 127 presumed normal subjects, 43 hyperparathyroid patients, 26 renal dialysis patients and 3 samples that were spiked with PTH in order to span the complete assay range. All the samples were tested in singlicate using the Siemens ADVIA Centaur® Intact PTH (predicate device) and the LIAISON® N-TACT® PTH Gen II (candidate device). The samples tested ranged from 8.0 pg/mL to 1794 pg/mL. Passing & Bablok linear regression analyses were performed and gave the following results:

Passing & Bablok Fit					
n	Slope	95% CI	Intercept pg/mL	95% CI	Correlation coefficient (r)
198	1.010	0.99 to 1.03	-1.5851	-3.11 to -0.44	0.9953

b. Matrix comparison:
Sixty-five (65) matched patient sets of EDTA plasma, serum, SST serum, and Lithium Heparin plasma samples were tested to determine if these sample types provide equivalent results on the LIAISON® N-TACT® PTH Gen II assay. In order to obtain samples that spanned the measuring range of 3.0-1900 pg/mL, some of the samples were spiked. Each sample type was tested in singlicate using 1 lot of reagent. Passing & Bablok linear regression analyses were performed and gave the following results:

EDTA plasma vs.	**Slope**	**95% CI**	**Intercept pg/mL**	**R^2**
Serum	0.97	0.94 to 1.0	-2.45	0.9986
SST Serum	1.01	0.99 to 1.03	-2.25	0.9996
Lithium Heparin	0.98	0.97 to 1.01	-0.01	0.9991

Based on the data, the sponsor claims EDTA plasma, Lithium Heparin plasma, Serum, and SST serum are acceptable for this assay.

In the limitation section of the package insert, sponsor has the following limitation: "During routine monitoring of intact PTH levels, use the same specimen type throughout the monitoring period to avoid bias in the results."

3. 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:
A reference range study was conducted according to the CLSI C28-A3 guideline. EDTA plasma samples from125 apparently healthy adults aged 21 - 70 years of age from mixed ethnic backgrounds (32.5% dark -skinned, 66.7% light-skinned and 0.8% unknown) with normal Total Calcium, TSH, Phosphorus, Magnesium, Creatinine, Alkaline Phosphatase

and 25 OH Vitamin D values from the northern and southern regions of the U.S were used to establish the reference range.

LIAISON® N-TACT® PTH Gen II Reference Range for EDTA samples

Population(n =125)	Median PTH (pg/mL)	Observed Range 2.5^{th} to 97.5^{th} Percentile
United States	34.00	14.5 – 87.1 pg/mL

It is recommended that each laboratory establish its own range of expected values.

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.