510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION DECISION SUMMARY

A. 510(k) Number:

k130321

B. Purpose for Submission:

New Device

C. Measurand:

Aldosterone

D. Type of Test:

Quantitative Chemiluminescent Immunoassay

E. Applicant:

DiaSorin Inc.

F. Proprietary and Established Names:

LIAISON® Aldosterone
LIAISON® Aldosterone Control Set
LIAISON® Aldosterone Calibration Verifiers

G. Regulatory Information:

Product Code	Classification	Regulation Section	Panel
CJM	Class II	21 CFR 862.1045 Aldosterone Test System	Clinical Chemistry (75)
JJX	Class I, reserved	21 CFR 862.1660 Quality Control Material	Clinical Chemistry (75)

H. Intended Use:

1. Intended use(s):

   See indications for use below.

2. Indication(s) for use:

   The LIAISON® Aldosterone assay uses chemiluminescent immunoassay (CLIA)

technology and is intended for the quantitative determination of Aldosterone in human serum, EDTA plasma and urine samples. Aldosterone measurements are intended for use in the diagnosis and treatment of primary aldosteronism (a disorder caused by excessive secretion of aldosterone by the adrenal gland), hypertension caused by primary aldosteronism, selective hypoaldosteronism, edematous states and other conditions of electrolyte imbalance. The test has to be performed on the LIAISON® Analyzer.

The LIAISON® Aldosterone Control Set is intended for use as assayed quality control samples to monitor the accuracy of the LIAISON® Aldosterone assay on the LIAISON® Analyzer.

The LIAISON® Aldosterone Calibration Verifiers are assayed quality control materials intended for the quantitative verification of calibration and reportable range of the LIAISON® Aldosterone assay when performed on the LIAISON® Analyzer.

3. Special conditions for use statement(s):

   For prescription use only

   Urine samples collected for 24 hours must be kept refrigerated during collection. The 24 hours urine samples must be treated per instructions for use before assaying on the analyzer.

4. Special instrument requirements:

   For use on the DiaSorin LIAISON® Analyzer

I. Device Description:

The LIAISON® Aldosterone Assay is an in vitro diagnostic device consisting of reagents provided in individual compartments within a plastic container called the LIAISON® Reagent Integral. Reagent Integral contains:

- Magnetic particles - coated with anti-sheep antibody, sheep anti-aldosterone antibody in buffer containing Phosphate buffer/BSA, <0.1% sodium azide; 2.4 mL
- Conjugate - Proprietary polymer conjugated with aldosterone and an isoluminol derivative, BSA, phosphate buffer/Danazol, with ProClin® 300 and gentamicin sulfate as preservatives.; 4.5 mL
- Assay Buffer - BSA, phosphate buffer, EDTA, tween-20, donkey and sheep serum with ProClin® 300 and gentamicin sulfate as a preservatives, 28 mL

2 Levels LIAISON® Calibrators containing hormone free human serum, aldosterone at 2 different concentrations, stabilizers and preservatives; 2 vials each level, 3 mL. Calibrators are provided ready to use with the LIAISON® Reagent Integral kit (not to be sold separately).

2 levels LIAISON® Controls containing hormone free human serum, aldosterone at 2 different concentrations, stabilizers and preservatives; 2 vials each level, 4.5 mL. Controls are provided ready to use.

4 levels LIAISON® Calibration Verifiers containing hormone free human serum, aldosterone at 4 different concentrations, stabilizers and preservatives; 1 vial each level, 2 mL. Calibration verifiers are provided ready to use.

LIAISON® Aldo Neutralization Buffer consists of two 25 mL-fill vials of phosphate buffer with 0.09% sodium azide as a preservative. LIAISON® Aldo Neutralization Buffer is used to neutralize acid hydrolyzed urine samples before testing on the LIAISON® Aldosterone assay.

Each serum/plasma donor unit used in the preparation of this product 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 (HIV 1/2), the Hepatitis B surface antigen (HBV), and the antibody to Hepatitis C (HCV).

J. Substantial Equivalence Information:

1. Predicate device name(s):

Siemens Coat-a-count No-extraction aldosterone
LIAISON® 25 OH Vitamin D TOTAL Control
LIAISON® 25 OH Vitamin D TOTAL Calibration Verifiers

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

k831178, k071480, k090104

3. Comparison with predicate:

Assay Similarities and Differences		
Item	New Device LIAISON® Aldosterone (k130321)	Predicate Device Siemens Coat-a-count aldosterone (k831178)
Intended Use	For the quantitative determination of Aldosterone in human serum and urine.	Same
Measuring Range	3-100 ng/dL	3-120 ng/dL
Test Principle	Chemiluminescent Immunoassay	^{125}I Radioimmunoassay
Sample size	100 µL	200 µL
Assay time	40 minutes	>18 hours
Sample matrix	Serum, EDTA plasma and 24-hour urine	Serum, 24-hour urine

Urine samples handling and processing time	1. Acid hydrolysis- 18 hrs. 2. Neutralization of urine ~2 minutes	1. Acid hydrolysis - 24 hrs. 2. Ethyl acetate extraction - 1 hr 3. Dry down ~ 15 minutes
Calibration	Two-point calibration by the user. Stable for 14 days.	7 calibrators used to generate assay curve in every assay run

Control similarity and differences		
Item	New Device LIAISON® Aldosterone Control (k130321)	Predicate Device LIAISON® 25 OH Vitamin D TOTAL Control (k071480)
Intended Use	Intended for use as assayed quality control samples to monitor the accuracy of assay.	Same
Analyte	Aldosterone	25 OH vitamin D
Matrix	Liquid Human serum based controls provided in vials with phosphate buffer, ProClin® 300 and Gentamicin.	Liquid human serum-based controls provided in vials with buffer salts and sodium azide.
Levels	Two levels: High and Low	Same
Storage conditions	2-8°C	Same

Calibration Verifier similarity and differences		
Item	New Device LIAISON® Aldosterone Calibration Verifier (k130321)	Predicate Device LIAISON® 25 OH Vitamin D TOTAL Calibration Verifier (k090104)
Intended Use	Assayed quality control materials intended for the quantitative verification of calibration and reportable range of the assay.	Same
Analyte	Aldosterone	25 OH vitamin D
Matrix	Buffered hormone free human serum based matrix (2 mL/vial) with Proclin® 300 as a preservative	Vitamin D free human serum with buffer salts and <0.1% sodium azide
Volume	2 mL	5 mL
Levels	Four levels	Same
Storage conditions	2-8°C	Same

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

- CLSI Guideline EP5-A2, Evaluation of Precision Performance of Quantitative

Measurement Methods;

- CLSI Guideline EP6-A, Evaluation of Linearity of Quantitative Analytical Methods;
- CLSI Guideline EP7-A2, Interference Testing in Clinical Chemistry;
- CLSI Guideline EP9-A2-IR, Method Comparison and Bias Estimation Using Patient Samples;
- CLSI Guideline EP17-A2Evaluation 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 method for quantitative determination of the LIAISON® Aldosterone assay is a direct, competitive, chemiluminescence immunoassay (CLIA). Specific antibody to aldosterone is bound to magnetic particles (solid phase) and aldosterone is linked to an isoluminol derivative. During the first incubation, sample is incubated with a specific anti-Aldosterone monoclonal antibody. Following the 1st incubation, the conjugate is added and competes with Aldosterone for an additional amount of time. After the 2nd incubation, the unbound material is removed with a wash cycle. The starter reagents are then added and a flash chemiluminescent reaction is initiated. The light signal is measured by a photomultiplier as relative light units (RLU) and is inversely proportional to the concentration of Aldosterone present in the calibrators, controls or patient samples.

Preparation of urine samples:
The 24-hour urine samples shall be refrigerated during collection. After measuring and recording the urine volume, add 1 g of boric acid to every 100 mL of urine sample. The borate stabilized urine sample will then be acid hydrolyzed by mixing one part urine with two parts of 0.2M HCl and incubated at 30ºC for 18 hours after thorough mixing. After acid hydrolysis, the urine samples must be neutralized by mixing one part urine with four parts of LIAISON® Aldo Neutralization Buffer. These pre-measurement treatments will cause 15x dilutions to urine samples.

Results of 24 hrs urine samples:
The instrument will measure and report the pretreated (acid hydrolyzed and neutralized) urine aldosterone results. The user must apply the 15x concentration correction factor to obtain the aldosterone concentrations of the urine samples. In addition, the users need to record the total volume of the 24-hour urine samples in order to report the total mass of aldosterone in µg/24 hours.

To calculate urine aldosterone in µg/24 hours, user shall use the following formula:

Corrected aldosterone concentration (result from analyzer in ng/dL x 15) x 24 hours urine volume (in mL) x 10^{-5}

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Following CLSI document EP5-A2, the sponsor evaluated the precision using 2 controls and 9 samples (6 frozen serum samples and 3 frozen urine samples) with concentrations spanning the working range of the assay. The samples were run at two external test sites and one internal test site, using two reagent lots, and 2 runs per day for 20 days (N=480 measurements per sample). Results of within-run and total precision are summarized in the table below.

Sample ID#	N	Mean ng/dL	Within Run		Total across Lots/Across Sites	
			SD	%CV	SD	%CV
QC Level 1	480	6.8	0.24	3.5%	0.65	9.6%
QC Level 2	480	28.8	0.53	1.8%	1.61	5.6%
Serum Sample 1	480	5.9	0.25	4.2%	0.62	10.5%
Serum Sample 2	480	8.8	0.27	3.1%	0.79	9.0%
Serum Sample 3	480	18.5	0.42	2.3%	1.27	6.9%
Serum Sample 4	480	29.8	0.78	2.6%	2.05	6.9%
Serum Sample 5	480	50.4	1.16	2.3%	2.92	5.8%
Serum Sample 6	480	82.6	1.76	2.1%	5.21	6.3%
Urine Sample 1	480	7.4	0.26	3.6%	0.72	9.8%
Urine Sample 2	480	44.1	1.24	2.8%	3.87	8.8%
Urine Sample 3	480	76.3	1.91	2.5%	6.58	8.6%

b. Linearity/assay reportable range:

The sponsor performed linearity studies in accordance with CLSI EP6-A guidelines using three high samples of each sample type (serum, EDTA plasma and urine). High endogenous or spiked samples were diluted to span the working range of the assay. A total of 12 samples (1 high and 11 diluted)

for each linearity sample set were tested in triplicate on the LIAISON® analyzer. Samples tested ranged from 2.2 to 109 ng/dL. The observed values were plotted against the expected values and linear regression was performed. All three sample sets tested with each sample type yielded similar linear regressions. A representative of each sample type is summarized below.

Serum: $y = 0.994x + 0.714$, $R = 1.000$,

EDTA Plasma: $y = 1.006x + 1.425$, $R = 0.998$,

Urine: $y = 0.9963x + 0.694$, $R = 0.999$.

The data support the claimed measuring range of this device, 3.0 to 100 ng/dL for serum, EDTA plasma and urine samples.

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

Traceability: The LIAISON® Aldosterone Calibrators are traceable to aldosterone reference material through gravimetrical preparation and UV spectrophotometric analysis.

Stability: Shelf life stability studies were performed with assay reagents, calibrators, controls and calibration verifiers, and demonstrated that they are stable until the expiration date shown on the product labeling when stored as instructed. Calibrators, controls and calibration verifiers are stable until the expiration date printed on the label when stored as directed. Once opened, calibrators are stable for 6 weeks when properly stored at 2-8°C between uses. Once opened, controls and calibration verifiers are stable for 4 weeks when properly stored at 2-8°C between uses. Calibration curve stability and reagent open vial stability were performed by the sponsor and demonstrated that the calibration curve is stable for 2 weeks and open reagent vials are stable for 6 weeks when stored on board or at 2-8°C. The protocols for stability and acceptance criteria were reviewed and found to be adequate.

Value assignment: concentrations of kit calibrators, controls and calibration verifiers are assigned through an internal procedure. Master calibrators are prepared from a stock solution made from reference material whose concentration is determined spectrophotometrically by the sponsor. The master calibrators are then used to assign values to the kit calibrators, controls and calibration verifiers using a minimum of 3 LIASON analyzers with at least 2 reagent lots. Each lot of calibrators, controls and calibration verifiers were tested over several runs and the mean results are used to determine the target values. The protocols for value assignment and acceptance criteria were reviewed and found to be adequate.

Aldosterone Calibrators have the following target values:
Level 1= 2.5 - 3.5 ng/dL
Level 2= 50 - 60 ng/dL

Aldosterone Controls have the following target ranges:
Level 1= 5.5 - 8.5 ng/dL
Level 2= 25.0 - 35.0 ng/dL

Aldosterone Calibration Verifiers have the following target ranges:
Level 1= 3.4 - 4.6 ng/dL
Level 2= 12.8 - 17.2 ng/dL
Level 2= 38.3 - 51.7 ng/dL
Level 2= 68.0 - 92.0 ng/dL

d. Detection limit:

The Limit of Blank (LoB), Limit of Detection (LoD), and Limit of Quantitation (LoQ) were determined for serum and urine samples in accordance with CLSI document EP17-A2. LoB was calculated using 5 blank serum or urine samples tested on 2 LIAISON® analyzers over three days using 6 runs with two reagent lots. LoD was calculated using 4 low concentration serum or urine samples in the range of LoB to 4xLoB values, tested on 2 LIAISON® analyzers over 3 days using 6 runs and 2 reagent lots. LoQ was calculated using 6 serum samples and 8 urine samples tested on 2 LIAISON® analyzers over 3 days using 6 runs and 2 reagent lots. LoQ was defined as the lowest concentration for which the inter-assay precision %CV is less than 20%.

The LoB, LoD and LoQ results in ng/mL are summarized below:

Sample type	LoB	LoD	LoQ
Serum	0.97	1.45	3.0
Urine	1.26	2.00	2.8

The claimed measuring range of the device is 3.0 to 100 ng/dL for serum, EDTA plasma and urine samples.

e. Analytical specificity:

Interference:

Following CLSI guidance document EP7-A2, interference studies were performed to both serum and pretreated urine samples that contain two different concentrations of aldosterone (12 ng/dL and 30ng/dL for serum samples; 5 ng/dL and 15 ng/dL for pretreated urine samples). Both serum and pretreated urine samples were spiked with a single concentration of 8 different endogenous substances (see below chart) and compared to unspiked control samples. The two sets of matched spiked and control samples containing each interferent were tested on the LIAISON® Aldosterone assay using 12 replicates with 1 reagent lot. The sponsor defines non-significant interference as bias within 10% between the spiked and the control samples. Results of non-significant interference are summarized in the table below:

Substance	Concentration Tested	
	serum	urine
Hemoglobin	600 mg/dL	600 mg/dL
Bilirubin (unconjugated)	40 mg/dL	N/A
Bilirubin (conjugated)	40 mg/dL	40 mg/dL
Triglycerides	3000 mg/dL	3000 mg/dL
Cholesterol	500 mg/dL	500 mg/dL
Total protein	12 g/dL	12 g/dL
Glucose	1 g/dL	1 g/dL
Creatinine	5 mg/dL	500 mg/dL
Urea	N/A	4 g/L

In addition, common pharmaceutical compounds were spiked into serum and pretreated urine samples that contain two different concentrations of aldosterone (12 ng/dL and 30ng/dL for serum samples; 5 ng/dL and 15 ng/dL for pretreated urine samples). Both serum and urine samples were spiked with potential interferents and tested with the LIAISON® Aldosterone assay. The reference sample (control) without interferent was spiked with the respective amount of solvent. Based on the sponsor's definition of non-significant interference (within ± 10% of control value), the sponsor claims no interference for the compounds with concentrations listed in the table below:

Compounds tested	Concentration	
	serum	urine
Amlodipine Besylate	13.9 µg/dL	13.9 µg/dL
Nifedipine	40 µg/dL	43.9 mg/dL
Verapamil	216 µg/dL	237 mg/dL
Furosemide	5.99 mg/dL	5.99 mg/dL
Eplerenone	1.99 mg/dL	1.99 mg/dL
Enalapril	42.4 µg/dL	46.6 mg/dL
Lisinopril	32.7 µg/dL	32.7 µg/dL
Losartan	225 µg/dL	249 mg/dL
Valsartan	1.1 mg/dL	1.1 mg/dL
Hydrochlorothiazide	600 µg/dL	600 µg/dL
Acetylsalicylic Acid	65.2 mg/dL	65.2 mg/dL
Salicylic Acid	59.9 mg/dL	59.9 mg/dL
Valproic Acid	57.6 mg/dL	57.6 mg/dL
Tetracycline	1.51 mg/dL	1.51 mg/dL
Ascorbic Acid	6 mg/dL	200 mg/dL
Acetaminophen	20 mg/dL	20 mg/dL
Metoprolol	1.28 mg/dL	1.28 mg/dL
Spiranolactone	60 µg/dL	60 µg/dL
Triamterene	886 µg/dL	886 µg/dL

Propranolol	230 µg/dL	228 µg/dL
Tartaric Acid	N/A	1 g/dL
Uric Acid	N/A	100 mg/dL
Acetic Acid	N/A	2%
Boric Acid	N/A	2 g/dL

CrossReactivity:

A cross-reactivity study was performed using 3 serum samples (with targeting aldosterone concentrations at 0, 15 and 30 ng/dL) and 3 pretreated urine samples (with targeting aldosterone concentrations at 0.5, 15 and 30 ng/dL). Each sample was spiked with various structurally similar compounds at the concentrations listed in the following table. Spiked and non-spiked samples were tested in triplicate using 1 lot of reagent kit and 1 LIAISON® analyzer. The results demonstrated that all substances showed <0.02% cross reactivity.

Substance	Concentration ng/dL in serum	Concentration ng/dL in urine
Androstendione	10000	100000
Androsterone	100000	1000000
Corticosterone	100000	100000
18-OH-Corticosterone	100000	100000
Cortisol (Hydrocortisone)	100000	200000
Cortisone	200000	200000
21-Hydroxyprogesterone	100000	100000
11-Deoxycortisol	100000	100000
Dexamethasone	200000	200000
DHEA (trans-Dehydroandrosterone)	100000	1000000
Estradiol	100000	100000
Estriol	10000	100000
Estrone	10000	100000
Fludrocortisone	200000	200000
Prazosin HCl	1200000	1200000
Prednisone	100000	100000
Prednisolone	100000	100000
Pregnenolone	100000	100000
Progesterone	100000	100000
17 alpha	100000	100000
Spironolactone	100000	100000
Testosterone	100000	200000

No hook effect was observed for aldosterone concentrations in serum or urine up to 1000 ng/dL.

f. Assay cut-off:

Not applicable

2. Comparisonstudies:

a. Method comparison with predicate device:

Following the CLSI EP9-A2 guidance document, the sponsor performed a method comparison study of the LIAISON® Aldosterone assay versus the predicate device. A total of 144 serum samples (22 of them were aldosterone spiked samples) were compared across methods, following the manufacturers' instructions (samples ranged from 3.02 ng/dL to 97.1 ng/dL on the LIAISON® Aldosterone assay). Singlicate results from the candidate device and mean of duplicate results from the predicate device were used for the linear regression analysis. Passing- Bablok linear regression analysis resulted the following:

y = 0.98x + 1.10; R= 0.988

95% CI for the slope is 0.94 to 1.02 and for the intercept is 0.43 to 1.49 ng/dL.

Additionally, a total of 104 native urine samples were compared across methods, following the manufacturers' instructions. Samples ranged tested from 7.9 ng/dL to 82.8 ng/dL on the LIAISON® Aldosterone assay for the pretreated urine samples, which corresponds to 118.9 ng/dL to 1242 ng/dL for the urine samples after x 15 dilution factor). The predicate device has a dilution factor of x 10 for the pretreated urine samples

Measuring range of the urine samples after multiplying by the dilution factor of 15 was 45 to 1500 ng/dL (3-100 ng/dL x 15).

Singlicate results from the candidate device and mean of duplicate results from the predicate device were used for the linear regression analysis. Passing- Bablok linear regression analysis resulted the following:

y = 0.98x + 34.0; R= 0.948
95% CI for the slope is 0.91 to 1.05 and for the intercept is 11.4 to 56.7 ng/dL.

b. Matrix comparison:

The sponsor performed a matrix comparison using 59 matched patient sets of serum and EDTA plasma samples (7 of them were spiked with additional aldosterone). Samples ranging from 3.59 to 97.3 ng/dL were analyzed in singlicate using one lot of LIAISON® Aldosterone reagent.

Passing-Bablok linear regression analysis resulted the following:

EDTA Plasma (Y) to Serum (X): y=0.964x + 0.033, R=0.997

The sponsor concluded that serum and EDTA plasma samples provided equivalent results on LIAISON® Aldosterone assay.

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:

   Matched serum and EDTA plasma samples drawn from 126 apparently healthy adults were tested to determine the reference range for the LIAISON® Aldosterone assay for serum and plasma samples. Samples were collected from a fasting population with normal blood pressure and normal glucose levels. Exclusion criteria included needs for prescription medications or restricted diets, pregnancy, breast feeding or taking oral contraceptives.

   Upright samples were collected after the individuals have stood for at least 30 minutes. Supine samples were collected after the individuals have laid in supine position for at least 30 minutes. The observed median and central 95% reference intervals for upright and supine positions are listed below.

Population (126)	Median Aldosterone (ng/dL)	Observed Range (ng/dL) 2.5th to 97.5th Percentile
Upright (Serum)	9.80	<3.0 – 39.2
Supine (Serum)	6.76	<3.0 – 23.2
Upright (EDTA plasma)	8.91	<3.0 – 35.3
Supine (EDTA plasma)	6.42	<3.0 – 23.6

24-hour urine samples collected from 91 apparently healthy adults with normal blood pressure were tested to determine the reference range for the LIAISON® Aldosterone assay for 24-hour urine samples. The observed median and central 95% reference intervals for 24-hour urine aldosterone mass are listed below.

Population (91)	**Median Aldosterone (ug/24 hours)**	**Observed Range (ug/24 hours) 2.5th to 97.5th Percentile**
Urine (24-hour)	5.53	1.19 – 28.1

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.