510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K211058

B Applicant

SENTINEL CH. S.p.A.

C Proprietary and Established Names

Lp(a) Ultra

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
DFC	Class II	21 CFR 866.5600 - Low-Density Lipoprotein Immunological Test System	IM - Immunology

II Submission/Device Overview:

A Purpose for Submission:

New device

B Measurand:

Lipoprotein (a) [Lp(a)]

C Type of Test:

Quantitative immunoturbidimetric assay

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

The Lp(a) Ultra assay is intended for in vitro diagnostic use in the immunoturbidimetric quantitative determination of lipoprotein (a) [Lp(a)] in human serum and plasma using an automated analyzer. The measurement of Lp(a) is useful in evaluating lipid metabolism disorders and assessing atherosclerotic cardiovascular disease in specific populations, when used in conjunction with clinical evaluation.

For In Vitro Diagnostic use.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

Harmonization efforts for Lp(a) assay methods have suggested an impact of apolipoprotein(a) isoform size (i.e. molecular weight) heterogeneity on some Lp(a) methods. The effects of apolipoprotein(a) heterogeneity have not been assessed for this assay.

D Special Instrument Requirements:

Beckman Coulter AU680 Chemistry Analyzer

IV Device/System Characteristics:

A Device Description:

Lp(a) Ultra assay is comprised of a kit with ready to use liquids for the quantitative immunoturbidimetric determination of lipoprotein (a). The kit contains two reagents sufficient to perform 86 tests on the Beckman Coulter AU680 Chemistry Analyzer. Reagent 1 is comprised of a glycine buffer at pH 9.0 with < 0.1% sodium azide. Reagent 2 is comprised of a 0.4% suspension of anti-Lp(a) polyclonal antibodies latex particles at pH 7.3 and with < 0.1% sodium azide. The Lp(a) Ultra assay is calibrated with the Lp(a) Cal Set consisting of five lyophilized calibrators.

B Principle of Operation:

Lp(a) Ultra assay is a latex-based immunoturbidimetric assay to measure Lp(a) levels in serum and plasma. The immunoturbidimetric assay is constructed with reagents that agglutinate due to antigen-antibody reaction between Lp(a) in a sample and anti-Lp(a) antibody bound to latex particles. This agglutination is detected as an absorbance change, with the magnitude of the change being proportional to the quantity of Lp(a).

V Substantial Equivalence Information:

A Predicate Device Name(s):

Diazyme Lipoprotein (a) Assay

B Predicate 510(k) Number(s):

K180074

C Comparison with Predicate(s):

Device & Predicate Device(s):	K211058	K180074
Device Trade Name	Lp(a) Ultra	Diazyme Lipoprotein(a) Assay
General Device Characteristic Similarities		
Intended Use/Indications For Use	Intended for the quantitation of lipoprotein (a).	Same
General Device Characteristic Differences		
Instrument	Beckman Coulter AU680 Chemistry Analyzer	Beckman Coulter AU400 Chemistry Analyzer
Analytical measuring range	10 - 100 mg/dL	5.4 - 100 mg/dL

VI Standards/Guidance Documents Referenced:

Clinical and Laboratory Standards Institute (CLSI) EP05-A3 Evaluation of Precision of Quantitative Measurement Procedures; Approved Guideline—Third Edition.

CLSI EP06-A2 Evaluation of Linearity of Quantitative Measurement Procedures. 2nd edition.

CLSI EP07-A3 Interference Testing in Clinical Chemistry; Approved Guideline —Third Edition.

CLSI EP14-A3 Guideline Evaluation of Commutability of Processed Samples; Approved Guideline – Third Edition.

CLSI EP17-A2 Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline—Second Edition.

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:
The precision performance of the Lp(a) Ultra assay on the Beckman Coulter AU680 Chemistry Analyzer was established in two studies for within-run and within-laboratory precision.

Study #1 – within-run precision
In the study, five samples (two controls, two serum pools, and one calibrator in serum) were tested in replicates of 20 in one run for assessment of the within-run precision (repeatability). The assessment was conducted on each of three days using one instrument and one reagent lot. The within-run precision (repeatability) was calculated for each sample and for each day:

Day	Sample	Mean, mg/dL	SD	%CV
1	3, serum	10.6	0.4	3.3
	1, control	19.0	0.7	3.5
	4, serum	32.7	0.4	1.3
	2, control	47.0	1.3	2.7
	5, calibrator	100	0.8	0.7
2	3	10.7	0.5	4.8
	1	18.9	0.7	3.7
	4	32.8	0.5	1.5
	2	45.9	0.9	1.9
	5	97.7	0.5	0.5
3	3	11.1	0.5	4.4
	1	19.1	0.5	2.8
	4	33.6	0.5	1.6
	2	46.0	0.5	1.1
	5	99.4	0.8	0.8

Study #2 – within-laboratory
In the study, five samples were tested in replicates of two per run, with two runs per day for 20 days for a total of 80 measurements per sample. The study was performed using one instrument and one reagent lot. The results for each sample presented in the table below were analyzed for variance by an ANOVA method for the factors of within-run, run, and day. The within-laboratory SD and %CV were estimated using the summation of the within-run, between-run, and between-day variance components.

		Within-Run		Between-Run		Between-Day		Within-Laboratory	
Sample	Mean (mg/dL)	SD	%CV	SD	%CV	SD	%CV	SD	%CV
3, serum	9.2	0.42	4.6%	0.00	0.0%	0.25	2.7%	0.49	5.3%
1, control	19.6	0.90	4.6%	0.00	0.0%	0.00	0.0%	0.90	4.6%
4, serum	37.1	0.52	1.4%	0.57	1.5%	0.94	2.5%	1.22	3.3%
2, control	47.4	0.86	1.8%	0.00	0.0%	1.26	2.7%	1.52	3.2%
5, calibrator	99.2	1.26	1.3%	0.00	0.0%	1.94	2.0%	2.31	2.3%

2. Linearity:

The linearity performance of the Lp(a) Ultra assay was evaluated in a study following the recommendations in CLSI EP06 Evaluation of the Linearity of Quantitative Measurement Procedures, 2nd Edition. In the study, a dilution series of 13 samples were prepared by inter-mixing known volumes of a high Lp(a) concentration serum pool and low Lp(a) concentration serum pool. Each sample was measured in triplicate using each of two reagent lots on a Beckman Coulter AU680 Chemistry Analyzer. The data were analyzed by linear regression using the mean of the observed result versus the expected concentration based on the known dilution ratio. Deviations from linearity within the claimed measuring range were never observed to be greater than ±10%. Based on these results, the sponsor concluded that the test system demonstrated a linear response over the claimed measuring range of 10 to 100 mg/dL.

High dose hook effect study
The sponsor provided data to support the claim that no high dose hook effect is observed with this assay for Lp(a) concentrations up to 500 mg/dL.

3. Analytical Specificity/Interference:

The analytical specificity of the Lp(a) Ultra assay on the Beckman Coulter AU680 Chemistry Analyzer was established by conducting interference testing following the recommendations in CLSI EP07 3rd Edition. Interference from certain endogenous and exogenous substances was assessed using two serum pools with low and high Lp(a) concentrations of 30 mg/dL and 50 mg/dL. Each low and high sample was further divided into two aliquots: control (with no added interferent) and test (with added interferent). Each sample was tested in 5 replicates using one lot and one instrument. A substance was identified as an interferent if the difference in the mean between the control and test sample was outside of the predefined allowable error of within ± 10% or ± 3 mg/dL. The observed maximum interference was -5% with absolute difference of -2.2 mg/dL for the following levels of interfering substances:

Substance	Highest concentration which no significant interference was observed
Endogenous	
Triglycerides	715 mg/dL*
Conjugated bilirubin	60 mg/dL
Unconjugated bilirubin	60 mg/dL
Rheumatoid factor	500 IU/mL
Hemoglobin	1000 mg/dL
Exogenous	
Ascorbic acid	180 mg/dL
Intralipid® Intravenous Fat Emulsion	1000 mg/dL

* For those substances that on initial screening were found to interfere, additional testing was conducted to establish the concentration limit below which no significant interference is

expected. Significant interference was found for triglycerides at concentration of 1000 mg/dL. The results of additional testing for triglycerides are given in the table below:

Lp(a) concentration	Substance	Concentration limit with no significant interference
6.4 to 79.2 mg/dL	Triglycerides	715 mg/dL

4. Assay Reportable Range:

   See linearity section VII A.2.

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

   Traceability
   The Lp(a) Ultra assay is metrologically traceable to externally sourced calibrators with assigned values. For each new lot of calibrators, the assigned values are verified by the sponsor to be within an acceptance interval by testing against the previous lot of calibrators.

6. Detection Limit:

   Detection capability studies of the Lp(a) Ultra on the Beckman Coulter AU680 Chemistry Analyzer for limit of blank (LoB), limit of detection (LoD), and limit of quantitation (LoQ) were conducted following the recommendations in CLSI EP17-A2.

   LoB
   The LoB for the Lp(a) Ultra assay was evaluated using four zero-analyte saline samples, and two reagent kit lots and one instrument. Each of the four samples was measured in replicates of 5 with each of the two lots over three days for a total of 60 replicates per lot. The LoB was analyzed by the parametric method described in the guideline and found to be 0.7 mg/dL for both lots.

   LoD
   The LoD was evaluated using four low-level samples prepared from serum pools using 2 reagent lots and one instrument. The samples were tested in replicates of 5 using each of two lots on three different days for a total of 60 measurement per lot. For each lot, the LoD was analyzed using a parametric data analysis which was the lowest concentration at which the analyte can be detected with 95% probability based on n = 60 replicates. The LoD of the assay was selected from the highest value obtained across the two lots; 1.9 mg/dL.

   LoQ
   The LoQ was evaluated using 8 low-level samples prepared from serum pools diluted with saline using two reagent lots and one instrument. Each of the samples was tested in replicates of 10 for each of the two lots on three different days for a total of 30 measurements per sample per lot. The LoQ was defined as the lowest concentration of analyte which has imprecision of less than or equal to 20% CV. The LoQ of the assay was selected from the highest value obtained across the two lots; 3.0 mg/dL.

7. Assay Cut-Off:

Not applicable.

B Comparison Studies:

1. Method Comparison with Predicate Device:
The agreement accuracy of the Lp(a) Ultra assay on the Beckman Coulter AU680 Chemistry Analyzer was establish with a method comparison study to a legally marketed Lp(a) device. In the study, 146 serum samples were tested in duplicate on the candidate device and duplicate on the comparator device. The data was analyzed by Passing-Bablok regression using the first replicate for the candidate device and the average of duplicates for the comparator device. The slope and intercept of the regression line were calculated, with the following results.

N	Concentration range, candidate device (mg/dL)	Concentration range, comparator device (mg/dL)	Regression Equation	R
146	10.0 to 98.9 mg/dL	10.0 to 98.3 mg/dL	y = 0.985x + 0.788	0.995

2. Matrix Comparison:

A matrix equivalency study to serum was conducted to support use of the Lp(a) Ultra assay on the Beckman Coulter AU680 Chemistry Analyzer with additional specimen matrix types claimed in the product labeling: Na-heparin plasma, Li-heparin plasma, K2EDTA plasma, and K3-EDTA plasma. In the study, 57 donor matched venous specimens of the aforementioned were collected. Each specimen was tested in singlicate using one lot of the reagent kit and one instrument, and the results compared to the mean of duplicate serum measurements. The results were analyzed by Passing-Bablok regression with the concentration from the first replicate of each donor's evaluation tube (y-axis) versus the mean concentration of the serum results (x-axis). The slope and intercept of the regression line were calculated, and summarized as follows:

Specimen	N	Min	Max	r	Intercept	Slope
Lithium heparin	57	10.7	98.4	0.997	-0.16	0.99
Sodium heparin	58	11.1	97.0	0.999	0.32	0.99
K2-EDTA plasma	57	10.6	97.8	0.997	0.31	0.97
K3-EDTA plasma	56	10.6	95.0	0.998	-0.29	0.99

C Clinical Studies:

1. Clinical Sensitivity:

Not applicable.

2. Clinical Specificity:

Not applicable.

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

Not applicable.

D Clinical Cut-Off:

Not applicable.

E Expected Values/Reference Range:

Reference values for the expected concentration range of Lp(a) are taken from literature data. This information is provided in the REFERENCE VALUES section of the product labeling as follows:

REFERENCE VALUES
The defined threshold of Lp(a) concentration at which individuals can be classified as being at increased risk varies greatly among studies, ranging from 20 mg/dL to 40 mg/dL.

A Lp(a) concentration of 30 mg/dL corresponding to the 75th percentile in a male Caucasian reference population is widely used as cut-off or threshold value.[1,2] Different Lp(a) levels can be found in different racial/ethnicity groups.[3,4] Increased serum Lp (a) concentrations are associated with increased risk of premature coronary artery disease and stroke. Because Lipoprotein (a) levels are highly heritable, Lp (a) may be an important marker for premature CHD, especially among Caucasians. Although Lipoprotein (a) levels are higher in African Americans than in Caucasians, associated CHD risk appears to be less.[5] It is recommended that each laboratory establish its own range of normal Lp(a) values for the population in their region. For diagnostic purposes, results obtained should always be evaluated considering the patient's history and all other clinical findings.

References
1) Marcovina SM, Koschinsky ML. A Critical Evaluation of the Role of Lp(a) in Cardiovascular Disease: Can Lp(a) Be Useful in Risk Assessment? Semin Vasc Med 2002 Aug;2(3):335-344.

2) Shai I, Rimm EB, Hankinson SE, et al. Lipoprotein (a) and Coronary Heart Disease Among Women: Beyond a Cholesterol Carrier? Eur Heart J 2005;26:1633-1639.

3) Wu HD, Berglund L, Dimayuga C, et al. High Lipoprotein (a) Levels and Small Apolipoprotein (a) Sizes are Associated With Endothelial Dysfunction in a Multiethnic Cohort. J Am Coll Cardiol 2004 May;43(10):1828-1833.

4) Virani SS, Brautbar A, Davis BC, et al. Associations Between Lipoprotein (a) Levels and Cardiovascular Outcomes in Black and White Subjects. The Atherosclerosis Risk in Communities (ARIC) Study. Circulation 2012;125(2):241-249.

5) Wu, Alan HB. Tietz Clinical Guide to Laboratory Tests. 4th edition. 2006; 678-9.

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.