

**SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE**

A. 510(k) Number:

k113262

B. Purpose for Submission:

New device

C. Measurand:

Cholesterol
HDL- Cholesterol
LDL- Cholesterol
Triglycerides

D. Type of Test:

Quantitative, colorimetric assay

E. Applicant:

Alfa Wassermann Diagnostic Technologies, LLC

F. Proprietary and Established Names:

ACE Cholesterol Reagent
ACE HDL-C Reagent
ACE LDL-C Reagent
ACE Triglycerides Reagent

G. Regulatory Information:

1. Regulation section:

21 CFR 862.1175: Cholesterol (total) test system
21 CFR 862.1475: Lipoprotein test system
21 CFR 862.1475: Lipoprotein test system
21 CFR 862.1705: Triglyceride test system

2. Classification:

Class I (for all the four measurands), all meet limitations of exemption per 21 CFR

862.9(c)(4) and 21 CFR 862.9(c)(9)

3. Product code:

CHH: Enzymatic Esterase-Oxidase, Cholesterol

LBS: LDL & VLDL Precipitation, Cholesterol Via Esterase-Oxidase, HDL

MRR: System, Test, Low Density, Lipoprotein

CDT: Lipase Hydrolysis/Glycerol Kinase Enzyme, Triglycerides

4. Panel:

Clinical Chemistry (75)

H. Intended Use:

1. Intended use(s):

See indication for use below

2. Indication(s) for use:

The ACE Cholesterol Reagent is intended for the quantitative determination of cholesterol concentration in serum using the ACE Axcel Clinical Chemistry System. Cholesterol measurements are used in the diagnosis and treatment of disorders involving excess cholesterol in the blood and lipid and lipoprotein metabolism disorders. This test is intended for use in clinical laboratories or physician office laboratories. For in vitro diagnostic use only.

The ACE HDL-C Reagent is intended for the quantitative determination of high density lipoprotein cholesterol (HDL-C) concentration in serum using the ACE Axcel Clinical Chemistry System. Lipoprotein measurements are used in the diagnosis and treatment of lipid disorders (such as diabetes mellitus), atherosclerosis, and various liver and renal diseases. This test is intended for use in clinical laboratories or physician office laboratories. For in vitro diagnostic use only.

The ACE LDL-C Reagent is intended for the quantitative determination of low density lipoprotein cholesterol (LDL-C) concentration in serum using the ACE Axcel Clinical Chemistry System. Lipoprotein measurements are used in the diagnosis and treatment of lipid disorders (such as diabetes mellitus), atherosclerosis, and various liver and renal diseases. This test is intended for use in clinical laboratories or physician office laboratories. For in vitro diagnostic use only.

The ACE Triglycerides Reagent is intended for the quantitative determination of triglyceride concentration in serum using the ACE Axcel Clinical Chemistry System. Triglyceride measurements are used in the diagnosis and treatment of patients with diabetes mellitus, nephrosis, liver obstruction, other diseases involving lipid metabolism

or various endocrine disorders. This test is intended for use in clinical laboratories or physician office laboratories. For in vitro diagnostic use only.

3. Special conditions for use statement(s):

For in vitro diagnostic use only. For prescription use.

4. Special instrument requirements:

ACE Axcel Clinical Chemistry System

I. Device Description:

The ACE Cholesterol Reagent is composed of a single reagent bottle containing 0.5 mmol/L 4-Aminoantipyrine (AAP), 25 mmol/L p-Hydroxybenzoic acid, >150 U/L Cholesterol oxidase (Nocardia), >240 U/L Cholesterol esterase (Porcine pancreas and Pseudomonas) >1600 U/L Peroxidase (Horseradish), stabilizers, preservatives, and fillers.

The ACE HDL-C Reagent is composed of two reagent bottles (Buffer and Color Reagent). The Buffer (R1) contains: Good's Buffer, <1000 U/L Cholesterol oxidase (E. coli), <1300 U/L Peroxidase (Horseradish), <1 mM N,N-bis(4-sulphobutyl)-m-toluidine-disodium salt (DSBmT), <1 mM Accelerator, <0.06% Preservative and <3000 U/L Ascorbic Oxidase (Curcubita sp.). The Color Reagent (R2) contains: <1500 U/L Cholesterol esterase (Pseudomonas sp.), <1 mM 4-Aminoantipyrine (4-AAP), <2% detergent and preservative.

The ACE LDL-C Reagent is composed of two reagent bottles (Buffer and Color Reagent). The Buffer (R1) contains: MES Buffer (pH 6.3), <1.0% Detergent 1, <1500 U/L Cholesterol oxidase (Pseudomonas sp.), <1300 ppg U/L Peroxidase (Horseradish), <0.1% 4-Aminoantipyrine (4-AAP), <3000 U/L Ascorbic acid oxidase (Curcubita sp.), <0.1% Preservative. The Color Reagent (R2) contains: MES Buffer (pH 6.3), <1.0% Detergent 2, <1.0 mmol/L N,N-bis(4-sulphobutyl)-m-toluidine-disodium salt (DSBmT) and <0.1% preservative.

The ACE Triglycerides Reagent is composed of a single reagent bottle. The reagent contains: 0.4 mmol/L 4-Aminoantipyrine (AAP), 2.6 mmol/L Adenosine 5'-triphosphate (ATP), 3.0 mmol/L p-Chlorophenol, >2400 U/L Glycerol phosphate oxidase (GPO) (Microorganism), >1000 U/L Lipase (Pseudomonas), >540 U/L Peroxidase (Horseradish), >400 U/L Glycerol kinase (Cellulomonas), buffer, stabilizers and preservatives.

J. Substantial Equivalence Information:

1. Predicate device name(s):

ACE Cholesterol Reagent
ACE HDL-C Reagent
ACE LDL-C Reagent
ACE Triglycerides Reagent

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

ACE Reagent Kit	Reagent 510(k)
Cholesterol	k931786
HDL-C	k971526
LDL-C	k991733
Triglycerides	k931786

3. Comparison with predicate:

Characteristics	ACE Cholesterol Agent, (Candidate Device)	ACE Cholesterol Reagent (Predicate, k931786)
Indications for Use	The ACE Cholesterol Reagent is intended for the quantitative determination of cholesterol concentration in serum. Cholesterol measurements are used in the diagnosis and treatment of disorders involving excess cholesterol in the blood and lipid and lipoprotein metabolism disorders. For in vitro diagnostic use only.	Same
Instrument/ Platform	ACE Axcel Clinical Chemistry System	ACE Clinical Chemistry System
Sample Type	Serum	Same
Measuring Range	4 - 600 mg/dL	2 - 600 mg/dL
Calibration	GEMCAL	Same
Calibration Stability	30 Days	Same
On-Board Stability	30 Days	Same

Characteristics	ACE HDL-C Agent, (Candidate Device)	ACE HDL-C Reagent (Predicate, k971526)
Indications for Use	The ACE HDL-C Reagent is intended for the quantitative determination of high density lipoprotein cholesterol (HDL-C) concentration in serum. Lipoprotein measurements are used in the diagnosis and treatment of lipid disorders (such as diabetes mellitus), atherosclerosis, and various liver and renal diseases.	Same

Instrument/ Platform	ACE Axcel Clinical Chemistry System	ACE Clinical Chemistry System
Sample Type	Serum	Same
Reportable	8 -125 mg/dL	Same
Calibration	GEMCAL	Same
Calibration Stability	30 Days	Same
On-Board Stability	30 Days	Same

Characteristics	ACE LDL-C Agent (Candidate Device)	ACE LDL-C Reagent (Predicate, k991733)
Indications for Use	The ACE LDL-C Reagent is intended for the quantitative determination of low density lipoprotein cholesterol (LDL-C) concentration in serum. Lipoprotein measurements are used in the diagnosis and treatment of lipid disorders (such as diabetes mellitus), atherosclerosis, and various liver and renal diseases. For in vitro diagnostic use only.	Same
Instrument/ Platforms	ACE Axcel Clinical Chemistry System	ACE Clinical Chemistry Systems
Sample Type	Serum	Same
Reportable Range	4 to 430 mg/dL	3 -500 mg/dL
Calibration	ACE LDL-C Calibrators	Same
Calibration Stability	30 Days	Same
On-Board Stability	30 Days	Same

Characteristics	ACE Triglycerides Agent (Candidate Device)	ACE Triglycerides Reagent (Predicate, k931786)
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Indications for Use	The ACE Triglycerides Reagent is intended for quantitative determination of triglycerides concentration in serum. It is intended for <i>in vitro</i> diagnostic use only.	Same
Instrument/ Platforms	ACE Axcel Clinical Chemistry System	ACE Clinical Chemistry Systems
Sample Type	Serum	Same
Reportable Range	12 -1000 mg/dL	6 -1000 mg/dL
Calibration	GEMCAL	Same
Calibration Stability	30 Days	Same
On-Board Stability	30 Days	Same

K. Standard/Guidance Document Referenced:

- CLSI EP6-A Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach (2003),
- CLSI EP5-A2 Evaluation of Precision Performance of Quantitative Measurement Methods; Second Edition (2004),
- CLSI EP7-A2 Interference Testing in Clinical Chemistry; Approved Guideline; Second Edition (2005),
- CLSI EP9-A2-IR Method Comparison and Bias Estimation Using Patient Samples; Approved Guideline- Second Edition (2002),
- CLSI EP17-A Protocol for Determination of Limits of Detection; Approved Guideline (2004)

L. Test Principle:

The ACE Cholesterol Reagent: Cholesterol esters in serum are completely hydrolyzed by cholesterol esterase to free cholesterol and free fatty acids. The cholesterol liberated by the esterase, plus any endogenous free cholesterol, are oxidized by cholesterol oxidase to yield hydrogen peroxide (H₂O₂), which then reacts oxidatively with p-hydroxybenzoic acid and 4-aminoantipyrine (AAP) in a reaction catalyzed by peroxidase, producing a red colored quinoneimine complex. The amount of chromogen formed, determined by quantitatively measuring the increase in absorbance at 505 nm/647 nm, is directly proportional to the cholesterol concentration in the sample.

The ACE HDL-C Reagent is composed of two bottles: Buffer Reagent (R1) and Color Reagent (R2). R2 contains a unique detergent, which solubilizes only the HDL lipoprotein particles; the detergent also inhibits the reaction of the cholesterol enzymes with LDL and VLDL lipoproteins and chylomicrons by adsorbing to their surfaces. The solubilized HDL cholesterol is released and reacts with cholesterol esterase and cholesterol oxidase and a chromogen 4-Aminoantipyrine to produce a color product measurable by the increase of

absorbance at 505/647 nm, which is directly proportional to the cholesterol concentration in the sample.

The ACE LDL-C Reagent: There are two detergents, Detergent 1 and Detergent 2, in the ACE LDL-C Reagents R1 and R2 respectively. In the first reaction, Detergent 1 solubilizes non-LDL lipoprotein particles (HDL, VLDL and chylomicrons) and releases cholesterol. The cholesterol is consumed by cholesterol esterase and cholesterol oxidase in a non-color forming reaction. In a second reaction, detergent 2 solubilizes the remaining LDL particles and forms peroxide, via the enzymes cholesterol esterase and cholesterol oxidase. The peroxide, in the presence of peroxidase and two peroxidase substrates, 4-aminoantipyrine and DSBmT, results in a purple-red color product measurable by the increase of absorbance at 505/647 nm, which is directly proportional to the cholesterol concentration in the sample.

The ACE Triglycerides Reagent: The ACE Triglycerides Reagent is composed of a single reagent bottle. Triglycerides in serum are hydrolyzed by lipase to form glycerol and free fatty acids. In the presence of adenosine triphosphate (ATP) and glycerol kinase (GK), the glycerol is converted to glycerol-I-phosphate (G-I-P) and the ATP to adenosine diphosphate (ADP). Glycerol-I-phosphate is oxidized by glycerol phosphate oxidase (GPO) to yield hydrogen peroxide (H₂O₂). The hydrogen peroxide then acts to oxidatively couple p-chlorophenol and 4-aminoantipyrine (AAP) in a reaction catalyzed by peroxidase, producing a red colored quinoneimine complex which absorbs strongly at 505 nm.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

In-House: ACE Cholesterol Reagent

Three serum based pools and one normal human serum sample, were tested on one ACE Axcel Clinical Chemistry System two times per run, two runs per day, for a total of 20 or more days. The results are summarized in the table below:

<u>Sample 1</u> Mean 87.7 mg/dL Cholesterol	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	1.4	0.5	0.5	1.6
Coefficient of Variation	1.6%	0.5%	0.6%	1.8%

<u>Sample 2</u> Mean 261.5 mg/dL Cholesterol	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	4.2	0.0	0.8	4.3
Coefficient of Variation	1.6%	0.0%	0.3%	1.6%

<u>Sample 3</u> Mean 430.4 mg/dL Cholesterol	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	8.7	0.0	3.7	9.4
Coefficient of Variation	2.0%	0.0%	0.9%	2.2%

<u>Sample 4</u> Mean 176.4 mg/dL Cholesterol	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	2.2	2.2	0.0	3.1
Coefficient of Variation	1.3%	1.2%	0.0%	1.8%

Point of Care Laboratory: ACE Cholesterol Reagent

Three serum based pools were tested on three ACE Axcel Clinical Chemistry Systems (one at each POL site) at least three times per run, one run per day, for a total of 5 days. The mean, standard deviations and % coefficients of variation (CV) were calculated for each sample.

Cholesterol			Within Run		Total	
Lab	Sample	Mean mg/dL	SD	%CV	SD	%CV
POL 1	1	84.6	1.2	1.4	1.4	1.7
POL 2	1	89.3	1.0	1.1	1.1	1.3
POL 3	1	85.3	1.3	1.5	1.5	1.7
POL 1	2	255.6	2.5	1.0	4.1	1.6
POL 2	2	270.3	2.1	0.8	3.3	1.2
POL 3	2	256.4	2.4	0.9	2.6	1.0
POL 1	3	423.6	6.2	1.5	6.4	1.5
POL 2	3	441.6	5.1	1.2	6.1	1.4
POL 3	3	423.2	3.0	0.7	6.6	1.6

In-House: ACE HDL-C Reagent

Three serum based pools and one normal human serum sample, were tested on one ACE Axcel Clinical Chemistry System two times per run, two runs per day, for a total of 20 or more days. The results are summarized in the table below:

<u>Sample 1</u> Mean 27.8 mg/dL HDL-C	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	0.7	0.6	0.9	1.3
Coefficient of Variation	2.7%	2.3%	3.4%	4.8%

<u>Sample 2</u> Mean 63.0 mg/dL HDL-C	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	1.1	0.0	1.9	2.2
Coefficient of Variation	1.7%	0.0%	3.0%	3.4%

<u>Sample 3</u> Mean 98.3 mg/dL HDL-C	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	1.6	1.3	2.3	3.1
Coefficient of Variation	1.7%	1.3%	2.4%	3.2%

<u>Sample 4</u> Mean 73.9 mg/dL HDL-C	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	1.1	0.9	1.9	2.4
Coefficient of Variation	1.4%	1.2%	2.6%	3.2%

Point of Care Laboratory: ACE HDL-C Reagent

Three serum based pools were tested on three ACE Axcel Clinical Chemistry Systems (one at each POL site) at least three times per run, one run per day, for a total of 5 days. The mean, standard deviations and % coefficients of variation (CV) were calculated for each sample.

HDL-C			Within Run		Total	
Lab	Sample	Mean mg/dL	SD	%CV	SD	%CV
POL 1	1	25.8	0.4	1.6	0.6	2.3
POL 2	1	27.0	0.7	2.6	1.0	3.5
POL 3	1	26.8	0.6	2.2	0.8	3.0
POL 1	2	59.4	0.6	1.1	0.6	1.1
POL 2	2	61.5	1.0	1.6	1.3	2.2

POL 3	2	31.3	0.6	1.0	1.1	1.8
POL 1	3	93.5	1.0	1.1	1.6	1.7
POL 2	3	97.6	1.6	1.6	1.9	1.9
POL 3	3	97.0	0.7	0.7	2.1	2.1

In-House: ACE LDL-C Reagent

Three serum based pools and one normal human serum sample, were tested on one ACE Axcel Clinical Chemistry System two times per run, two runs per day, for a total of 20 or more days. The results are summarized in the table below:

<u>Sample 1</u> Mean 45.1 mg/dL LDL-C	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	2.1	0.5	0.5	2.2
Coefficient of Variation	4.6%	1.2%	1.0%	4.9%

<u>Sample 2</u> Mean 181.7 mg/dL LDL-C	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	4.7	3.2	3.4	6.6
Coefficient of Variation	2.6%	1.8%	1.9%	3.6%

<u>Sample 3</u> Mean 328.5 mg/dL LDL-C	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	8.7	0.0	6.1	10.6
Coefficient of Variation	2.7%	0.0%	1.8%	3.2%

<u>Sample 4</u> Mean 86.3 mg/dL LDL-C	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	2.2	1.9	0.9	3.0
Coefficient of Variation	2.5%	2.2%	1.0%	3.5%

Point of Care Laboratory: ACE LDL-C Reagent

Three serum-based pools were tested on three ACE Axcel Clinical Chemistry Systems (one at each POL site) at least three times per run, one run per day, for a total of 5 days. The mean, standard deviations and % coefficients of variation (CV) were calculated for each sample.

LDL-C			Within Run		Total	
Lab	Sample	Mean mg/dL	SD	%CV	SD	%CV
POL 1	1	45.9	2.0	4.4	2.1	4.6
POL 2	1	41.9	1.3	3.1	2.5	5.9
POL 3	1	46.0	1.9	4.1	2.4	5.2
POL 1	2	185.9	4.9	2.6	7.0	3.8
POL 2	2	164.0	6.2	3.8	8.2	5.0
POL 3	2	180.9	3.7	2.0	4.9	2.7
POL 1	3	336.7	8.0	2.4	8.0	2.4
POL 2	3	307.6	6.9	2.2	10.5	3.4
POL 3	3	328.7	5.7	1.7	8.6	2.6

In-House: ACE Triglycerides Reagent

Three serum-based pools and one normal human serum sample were tested on one ACE Axcel Clinical Chemistry System two times per run, two runs per day, for a total of 20 or more days. The results are summarized in the table below:

<u>Sample 1</u> Mean 73.3 mg/dL Triglycerides	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	1.3	0.8	0.4	1.5
Coefficient of Variation	1.8%	1.0%	0.5%	2.1%

<u>Sample 2</u> Mean 138.9 mg/dL Triglycerides	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	2.5	1.1	0.8	2.9
Coefficient of Variation	1.8%	0.8%	0.6%	2.1%

<u>Sample 3</u> Mean 950.4 mg/dL Triglycerides	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	11.5	7.4	9.6	16.7
Coefficient of Variation	1.2%	0.8%	1.0%	1.8%

<u>Sample 4</u> Mean 58.6 mg/dL Triglycerides	Within Run	Between Run	Between Day	Total
Standard Deviation, mg/dL	1.7	0.7	0.0	1.8
Coefficient of Variation	2.9%	1.3%	0.0%	3.2%

Point of Care Laboratory: ACE Triglycerides Reagent

Four serum-based pools were tested on three ACE Axcel Clinical Chemistry Systems (one at each POL site) at least three times per run, one run per day, for a total of 5 days. The mean, standard deviations and % coefficients of variation (CV) were calculated for each sample.

Triglycerides			Within Run		Total	
Lab	Sample	Mean mg/dL	SD	%CV	SD	%CV
POL 1	1	70.5	1.7	2.3	1.7	2.3
POL 2	1	73.0	1.1	1.5	1.1	1.5
POL 3	1	72.7	1.2	1.7	3.0	4.1
POL 1	2	136.0	2.1	1.5	2.3	1.7
POL 2	2	137.9	2.1	1.5	2.4	1.8
POL 3	2	136.5	1.6	1.2	1.8	1.3
POL 1	3	182.1	3.9	2.1	4.6	2.5
POL 2	3	185.4	2.8	1.5	2.8	1.5
POL 3	3	182.1	1.1	0.6	1.1	0.6
POL 1	4	933.4	4.3	0.5	6.4	0.7
POL 2	4	933.4	8.4	0.9	9.0	1.0
POL 3	4	944.0	7.8	0.8	16.6	1.8

b. Linearity/assay reportable range:

ACE Cholesterol Reagent

Linearity studies were carried out using dilutions of a spiked serum samples. Eleven concentrations were prepared by mixing spiked serum samples in known proportion

with blank samples. All samples were measured in triplicate. The sample range tested was 3 to 966 mg/dL.

Claimed Measuring Range	Intercept	Slope	r ²
4 to 600 mg/dL	-1.1	1.016	0.9993

Based on the linearity data, the measuring range claimed from 4 to 600 mg/dL was supported.

ACE HDL-C Reagent

Linearity studies were carried out using dilutions of a spiked serum samples. Eleven concentrations were prepared by mixing spiked serum samples in known proportion with blank samples. All samples were measured in triplicate. The sample range tested was 7.6 to 137.7 mg/dL.

Claimed Measuring Range	Intercept	Slope	r ²
8 to 125 mg/dL	0.1	1.011	0.9986

Based on the linearity data, the measuring range claimed from 8 to 125 mg/dL was supported.

ACE LDL-C Reagent

Linearity studies were carried out using dilutions of a spiked serum samples. Eleven concentrations were prepared by mixing spiked serum samples in known proportion with blank samples. All samples were measured in triplicate. The sample range tested was 0.3 to 430 mg/dL.

Claimed Measuring Range	Intercept	Slope	r ²
4 to 430 mg/dL	-4.5	0.985	0.9981

Based on the linearity data, the measuring range claimed from 4 to 430 mg/dL was supported.

ACE Triglycerides Reagent

Linearity studies were carried out using dilutions of a spiked serum samples. Eleven concentrations were prepared by mixing spiked serum samples in known proportion with blank samples. All samples were measured in triplicate. The sample range tested was 12.3 to 1249.3 mg/dL.

Claimed Measuring Range	Intercept	Slope	r ²
12 to 1000 mg/dL	8.6	0.9915	0.9995

Based on the linearity data, the measuring range claimed from 12 to 850 mg/dL was supported.

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

Traceability

ACE Cholesterol Reagent is traceable to NIST SRM 1951. The calibrator (GEMCAL) was previously cleared under k931786.

ACE HDL-C Reagent is traceable to a CDC certified method. The calibrator (GEMCAL) was previously cleared under k931786.

ACE LDL-C Reagent is traceable to the CDC LDL cholesterol reference method. The ACE LDL-C Calibrators were previously cleared under k931733.

ACE Triglycerides Reagents is traceable to NIST SRM 1951. The calibrator (GEMCAL) was previously cleared under k931786

On-Board stability:

The stability protocols and acceptance criteria were reviewed and determined to be adequate. All ACE reagent in this submission demonstrated on-board stability of 30 days.

The real-time close-vial stability study protocols and acceptance criteria were reviewed and determined to be adequate. The stability for each reagent is shown in the table below:

Reagent	Closed-Vial Stability (months)
Cholesterol	15
HDL-C	24
LDL-C	24
Triglycerides	15

d. Detection limit:

Protocols for the determination of the limit of the blank (LoB) and the limit of detection (LoD) were performed in accordance with the recommendations in the CLSI Guideline EP17-A. Testing was carried out using true blanks and low level samples (total 60 each) over three days on two ACE Axcel Clinical Chemistry Systems.

The resulting LoB and LoD for the four measurands are summarized below:

Reagent	Limit of Blank (LoB)	Limit of Detection (LoD)
Cholesterol	2.9 mg/dL	3.6 mg/dL
HDL-C	1.0 mg/dL	1.5 mg/dL
LDL-C	2.7 mg/dL	3.9 mg/dL
Triglycerides	10.4mg/dL	11.6 mg/dL

e. Analytical specificity:

ACE Cholesterol Reagent

Interference studies were performed by using serum pools containing Cholesterol with individual interferents at a range of concentrations. The sera were assayed for Cholesterol (n = 3 replicates) and the mean result calculated. Interference was considered to be significant by the sponsor if the analyte recovery changed by $\pm 10\%$. The results reported were obtained on the ACE Axcel Clinical Chemistry System analyzer.

Interferent	Cholesterol Concentrations Tested (mg/dL)	No Significant Interference At or Below:
Unconjugated Bilirubin	Low Pool: 182	30 mg/dL
	High Pool: 405	
Hemoglobin	Low Pool: 159	500 mg/dL
	High Pool: 445	
Triglycerides	Low Pool: 170	2180 mg/dL
	High Pool: 378	
Ascorbic Acid	Low Pool: 185	6 mg/dL
	High Pool: 417	

ACE HDL-C Reagent:

Interference studies were performed by using serum pools containing HDL with individual interferents at a range of concentrations. The sera were assayed for HDL (n = 3 replicates) and the mean result calculated. Interference was considered to be significant by the sponsor if the analyte recovery changed by $\pm 10\%$. The results reported were obtained on the ACE Axcel Clinical Chemistry System analyzer.

Interferent	HDL Concentrations Tested (mg/dL)	No Significant Interference At or Below:
Unconjugated Bilirubin	Low Pool: 43	59 mg/dL
	High Pool: 98	
Hemoglobin	Low Pool: 64	1000 mg/dL
	High Pool: 91	

Triglycerides	Low Pool: 30	652 mg/dL
	High Pool: 101	
Ascorbic Acid	Low Pool: 45	6 mg/dL
	High Pool: 105	

ACE LDL-C Reagent:

Interference studies were performed by using serum pools containing LDL with individual interferents at a range of concentrations. The sera were assayed for LDL (n = 3 replicates) and the mean result calculated. Interference was considered to be significant by the sponsor if the analyte recovery changed by $\pm 10\%$. The results reported were obtained on the ACE Axcel Clinical Chemistry System analyzer.

Interferent	LDL Concentrations Tested (mg/dL)	No Significant Interference At or Below:
Unconjugated Bilirubin	Low Pool: 116	59 mg/dL
	High Pool: 374	
Hemoglobin	Low Pool: 74	1000 mg/dL
	High Pool: 273	
Triglycerides	Low Pool: 113	1233 mg/dL
	High Pool: 352	
Ascorbic Acid	Low Pool: 122	6 mg/dL
	High Pool: 396	

ACE Triglycerides Reagent:

Interference studies were performed by using serum pools containing Triglycerides with individual interferents at a range of concentrations. The sera were assayed for Triglycerides (n = 3 replicates) and the mean result calculated. Interference was considered to be significant by the sponsor if the analyte recovery changed by $\pm 10\%$. The results reported were obtained on the ACE Axcel Clinical Chemistry System analyzer.

Interferent	Triglycerides Concentrations Tested (mg/dL)	No Significant Interference At or Below:
Unconjugated Bilirubin	Low Pool: 78	7 mg/dL
	High Pool: 726	
Hemoglobin	Low Pool: 50	125 mg/dL
	High Pool: 270	
Ascorbic Acid	Low Pool: 75	0.75 mg/dL
	High Pool: 716	

The following limitations are stated in the labeling for ACE Triglyceride Reagent:

“Use clear, unhemolyzed serum. Hemolyzed and icteric samples cannot be used.”

“Advice patients not to take any alcohol or vitamin supplements 24 hours prior to fasting blood work.”

f. Assay cut-off:

Not applicable

g. Cleaning and Disinfection

Not applicable.

2. Comparison studies:

Studies were carried out according to CLSI EP09-A2-IR.

In house: ACE Cholesterol Reagent

One hundred ten serum samples were assayed in parallel by both the test and predicate methods. The results were analyzed by using Deming regression. The range tested was 7 to 527 ng/dL. Altered samples were included in the study (no more than 10%).

The comparison by Deming regression resulted in a slope of 1.012 (95%CI = 0.999 to 1.026), an intercept of -3.3 (95%CI = -6.2 to -0.5), correlation coefficient (R^2) = 0.9977, and a standard error of 5.0.

Point of Care Laboratory: ACE Cholesterol Reagent

Serum samples were assayed in parallel by both the test and predicate methods at three POC sites. The results were analyzed by using Deming regression. (Some altered samples were used among all three sites.)

POL	n	Range	Regression Equation	Correlation Coefficient	Standard Error	Confidence Interval Slope	Confidence Interval Intercept
1	57	22-571	$Y = 1.003x - 2.7$	0.9995	3.0	0.995 to 1.012	-4.6 to -0.7
2	60	17-515	$Y = 1.017x - 1.9$	0.9978	6.9	0.999 to 1.034	-6.1 to 2.3
3	46	62-538	$y = 0.996x - 0.3$	0.9945	8.4	0.964 to 1.028	-7.3 to 6.7

In house: ACE HDL-C Reagent

One hundred eight serum samples were assayed in parallel by both the test and predicate methods. The results were analyzed by using Deming regression. The range tested was 8 to 122 ng/dL. Altered samples were included in the study (no more than 10%).

The comparison by Deming regression resulted in a slope of 0.972 (95%CI = 0.9564 to 0.989), an intercept of 0.5 (95%CI = -0.4 to 1.5), correlation coefficient (R^2) = 0.9957, and a standard error of 1.7.

Point of Care Laboratory: ACE HDL-C Reagent

Serum samples were assayed in parallel by both the test and predicate methods at three POC sites. The results were analyzed by using Deming regression. Some altered samples were used among all three sites.

Lab	n	Range	Regression Equation	Correlation Coefficient	Standard Error	Confidence Interval Slope	Confidence Interval Intercept
POL 1	60	13-110	$y = 0.959x - 2.6$	0.9961	1.8	0.937 to 0.981	-4.0 to -1.3
POL 2	56	20-110	$y = 0.969x - 1.8$	0.9964	1.7	0.947 to 0.991	-3.1 to -0.4
POL 3	43	21-117	$y = 1.015x - 0.5$	0.9898	2.4	0.970 to 1.061	-3.0 to 2.0

In house: ACE LDL-C Reagent

One hundred eight serum samples were assayed in parallel by both the test and predicate methods and the results were analyzed by using Deming regression. The range tested was 32 to 422 ng/dL. Altered samples were included in the study (no more than 10%).

The comparison by Deming regression resulted in a slope of 0.982 (95%CI = 0.968 to 0.996), an intercept of -1.0 (95%CI = -3.1 to 1.0), correlation coefficient (R^2) = 0.9973, and a standard error of 4.7.

Point of Care Laboratory: ACE LDL-C Reagent

Serum samples were assayed in parallel by both the test and predicate methods at three POC sites. The results were analyzed by using Deming regression. Some altered samples were used among all three sites.

POL	N	Range	Regression Equation	Correlation Coefficient	Standard Error	Confidence Interval Slope	Confidence Interval Intercept
1	74	4-395	$y = 1.012x + 1.4$	0.9970	6.0	0.994 to 1.031	-1.3 to 4.0
2	58	9-397	$y = 1.010x - 2.5$	0.9974	5.9	0.991 to 1.030	-5.5 to 0.4
3	47	30-417	$y = 1.037x - 3.2$	0.9940	9.0	1.002 to 1.071	-8.4 to 2.1

In house: ACE Triglycerides Reagent

One hundred eleven serum samples were assayed in parallel by both the test and predicate methods and the results compared by Deming regression. The range tested was 24 to 975 µg/dL. Altered samples were included in the study (no more than 10%).

The comparison by Deming regression resulted in a slope of 1.031 (95%CI = 1.025 to 1.037), an intercept of -0.8 (95%CI = -2.7 to 1.1), correlation coefficient (R^2) = 0.9995, and a standard error of 6.6.

Point of Care Laboratory: ACE Triglycerides Reagent

Serum samples were assayed in parallel by both the test and predicate methods at three POC sites. The results were analyzed by using Deming regression. Some altered samples were used among all three sites.

POL	N	Range	Regression Equation	Correlation Coefficient	Standard Error	Confidence Interval Slope	Confidence Interval Intercept
1	54	23-976	$y = 1.008x - 1.6$	0.9996	6.6	1.000 to 1.016	-3.9 to 0.7
2	60	20-910	$y = 0.997x - 4.0$	0.9996	5.9	0.989 to 1.004	-6.3 to -1.7
3	47	37-978	$y = 1.012x - 2.8$	0.9992	8.4	1.000 to 1.024	-6.3 to 0.6

3. Clinical studies:

Not applicable

4. Clinical cut-off:

Not applicable

5. Expected values/Reference range:

Cholesterol: Desirable: <200 mg/dL, Borderline: 200-239 mg/dL, High: >239 mg/dL)

HDL: <40 mg/dL = Low, \geq 60 mg/dL = High

LDL: Optimal: <100 mg/dL, Near optimal: 100 – 129 mg/dL, Borderline high: 130 – 159 mg/dL, High: 160 – 189, Very high: >189 mg/dL

Triglycerides: <150 = normal, 150 – 199 = borderline high, 200-499 = high, \geq 500 = very high

NCEP, Third Report of National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation and Treatment of High Cholesterol in Adults (Adult Treatment Panel III); Executive Summary. NIH Publication No. 02-5215. National Institutes of Health. Bethesda, Maryland: September 2002.

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.