

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

A. 510(k) Number:

k131515

B. Purpose for Submission:

New reagents (Gamma-Glutamyltransferase, Lipase, Total Thyroxine) added onto ACE *Alera* instrument (k123018)

Addition of lithium heparin plasma samples to already cleared reagents on the ACE (k930104, k981377) and ACE *Axcel* (k113382, k113438, k113437) instruments.

C. Measurand:

Gamma-Glutamyltransferase (γ -GT), Lipase, Total Thyroxine (T4)

D. Type of Test:

Quantitative, photometric/colorimetric methods

E. Applicant:

Alfa Wassermann Diagnostic Technologies, LLC

F. Proprietary and Established Names:

ACE γ -GT Reagent

ACE Lipase Reagent

ACE T4 Reagent

G. Regulatory Information:

Product Code	Classification	Regulation	Panel
JPZ	Class I, meets limitations of exemptions in 862.9(c)(9)	21 CFR 862.1360 Gamma-glutamyl transpeptidase and isoenzymes test system	Clinical Chemistry (75)
CHI	Class I, meets limitations of exemptions in 862.9(c)(9)	21 CFR 862.1465 Lipase test system	Clinical Chemistry (75)
KLI	Class II	21 CFR 862.1700 Total thyroxine test system	Clinical Chemistry (75)

H. Intended Use:

1. Intended use(s):

See Indications for use below.

2. Indication(s) for use:

The ACE γ -GT Reagent is intended for the quantitative determination of gamma-glutamyltransferase activity in serum and lithium heparin plasma using the ACE, ACE *Alera*, and ACE Axcel Clinical Chemistry Systems. Gamma-glutamyltransferase measurements are used in the diagnosis and treatment of liver diseases such as alcoholic cirrhosis and primary and secondary liver tumors. This test is intended for use in clinical laboratories and physician office laboratories. For *in vitro* diagnostic use only.

The ACE Lipase Reagent is intended for the quantitative determination of lipase activity in serum and lithium heparin plasma using the ACE, ACE *Alera*, and ACE Axcel Clinical Chemistry Systems. Lipase measurements are used in diagnosis and treatment of diseases of the pancreas such as acute pancreatitis and obstruction of the pancreatic duct. This test is intended for use in clinical laboratories and physician office laboratories. For *in vitro* diagnostic use only.

The ACE T4 Reagent is intended for the quantitative determination of total thyroxine (T4) in serum and lithium heparin plasma using the ACE, ACE *Alera*, and ACE Axcel Clinical Chemistry Systems. Total thyroxine measurements are used in the diagnosis and treatment of thyroid diseases. This test is intended for use in clinical laboratories and 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 and use in Point-of-Care settings

4. Special instrument requirements:

For use on the ACE, ACE Axcel and ACE *Alera* Clinical Chemistry Systems

I. Device Description:

The ACE γ -GT Reagent consists of two reagent bottles, the γ -GT buffer and γ -GT substrate. The γ -GT substrate reagent contains L- γ -glutamyl-3-carboxy-4-nitroanilide (3.0 mmol/L) and Glycylglycine (150 mmol/L). The γ -GT buffer reagent contains buffer and preservative.

The ACE Lipase Reagent consists of two reagent bottles, the Lipase reagent (lyophilized) and Lipase activator. The lyophilized Lipase reagent is reconstituted by pouring the Lipase reagent solvent in to the Lipase reagent lyophilized bottle. The Lipase reagent (reconstituted) (R1) contains 1,2-Diglyceride (egg) (1.1 mM), Monoglyceride lipase (*Bacillus* sp.) (0.88U/mL), Glycerol kinase (*S. canus*) (<1.34 U/mL), Glycerol-3-phosphate oxidase (*Streptococcus* sp.) (<40 U/mL), TOOS (0.07%), ATP (bacterial) (0.66 mM), Peroxidase (horseradish) (<1.34 U/mL), Colipase (porcine) (<40 U/mL), Human serum albumin (0.27%), Ascorbate oxidase (cucumber, zucchini) (<2.66 U/L), Cholic acid (ox or sheep) (5.3 mM), Buffer (pH 6.8), Stabilizers and Sodium azide (0.05%). The Lipase activator reagent (R2)

contains Deoxycholate (ox or sheep) (36 mM), 4-Aminoantipyrine (0.12%), Buffer (pH 8.7) and Sodium azide (0.05%).

The ACE T4 Reagent consists of two reagent bottles, the antibody/substrate reagent and the enzyme conjugate reagent. The Antibody/Substrate reagent (R1) contains Mouse monoclonal anti-thyroxine antibody, 8-Anilino-1-naphthalene sulfonic acid (ANS), Glucose-6-phosphate (G6P), Nicotinamide adenine dinucleotide (NAD+), Tris buffer and Sodium azide (0.09%). The Sodium nitrite reagent (R2) contains Glucose-6-phosphate dehydrogenase (G6PD) labeled with thyroxine, Tris buffer and Sodium azide (0.09%).

J. Substantial Equivalence Information:

1. Predicate device name(s):

ACE γ -GT Reagent

ACE Lipase Reagent

ACE T4 Reagent

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

ACE γ -GT Reagent: k930104 (ACE)

ACE Lipase Reagent: k930104 (ACE)

ACE T4 Reagent: k981377 (ACE)

3. Comparison with predicate:

ACE γ-GT Reagent: Similarities and Differences		
Item	New Device ACE γ -GT Reagent, on the ACE, ACE <i>Alera</i> , and ACE Axcel Systems	Predicate Device ACE γ -GT Reagent, on the ACE and ACE Axcel Systems; k930104 (ACE)
Intended use	Same	The ACE γ -GT Reagent is intended for the quantitative determination of gamma-glutamyltransferase activity.
Measurand	Same	Gamma-glutamyltransferase activity
Assay method	Same	Photometric
Measuring range	Same	7 to 950 U/L
Expected values	Same	Male: 13 to 68 U/L Female: 11 to 48 U/L
Matrix	Human serum and Li-heparin plasma	Human serum

ACE Lipase Reagent: Similarities and Differences		
Item	New Device ACE Lipase Reagent, on the ACE, ACE <i>Alera</i> , and ACE Axcel Systems	Predicate Device ACE Lipase Reagent, on the ACE and ACE Axcel Systems; k930104 (ACE)
Intended use	Same	The ACE Lipase Reagent is intended for the quantitative determination of Lipase activity.
Measurand	Same	Lipase activity
Assay method	Same	Photometric
Measuring range	Same	15 to 700 U/L
Expected values	Same	Less than 60 U/L
Matrix	Human serum and Li-heparin plasma	Human serum

ACE T4 Reagent: Similarities and Differences		
Item	New Device ACE T4 Reagent, on the ACE, ACE <i>Alera</i> , and ACE Axcel Systems	Predicate Device ACE T4 Reagent, on the ACE and ACE Axcel Systems; k981377 (ACE)
Intended use	Same	The ACE T4 Reagent is intended for the quantitative determination of total thyroxine (T4).
Measurand	Same	Total thyroxine (T4)
Assay method	Same	Photometric
Measuring range	Same	1.3 to 19.6 µg/dL
Expected values	Same	5.0 to 12.0 µg/dL
Matrix	Human serum and Li-heparin plasma	Human serum

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

EP05-A2: Evaluation of Precision Performance of Quantitative Measurement Methods (2004).

EP06-A: Evaluation of the Linearity of Quantitative Measurement Procedures: A statistical approach (2003).

EP07-A2: Interference Testing in Clinical Chemistry (2005).

EP09-A2-IR: Method Comparison and Bias Estimation Using Patient Samples
(Interim revision; 2010)

EP17-A: Evaluation of Detection Capability for Clinical Laboratory Measurement
Procedures (2004)

L. Test Principle:

The ACE γ -GT Reagent is based on a photometric test in which γ -GT in the sample catalyzes the transfer of the γ -glutamyl group from the L- γ -glutamyl-3-carboxy-4-nitroanilide substrate to the glycylglycine product in the reagent. The product, 5-amino-2-nitobenzoate, absorbs strongly at 408 nm. The rate of increase in absorbance is directly proportional to the γ -GT activity in the sample.

The ACE Lipase Reagent is based on a photometric test in which lipase in the sample acts on a natural substrate, 1,2-diglyceride, to liberate 2-monoglyceride. This is hydrolyzed by monoglyceride lipase in to glycerol and free fatty acid. Glycerol kinase acts on glycerol to form glycerol-3-phosphate, which is in turn acted on by glycerol-3-phosphate oxidase to generate hydrogen peroxide. Peroxidase converts the hydrogen peroxide, 4-aminoantipyrine and TOOS (N-ethyl-N-(2-hydroxy-3-sulfopropyl)-m-toluidine) in to a quinone dye. The rate of formation of the dye, determined bichromatically at an absorbance of 573 nm/692 nm, is proportional to the lipase activity in the sample.

The ACE T4 Reagent is a homogenous enzyme immunoassay based on a photometric test in which 8-anilino-1-naphthalene sulfonic acid (ANS) is used to dissociate thyroxine from the plasma binding proteins. Using antibodies specific to thyroxine, this assay is based on the competition of G6PD labeled thyroxine and the dissociated thyroxine in the sample for a fixed number of specific antibody binding sites. In the absence of thyroxine from the sample, the thyroxine labeled G6PD in the second reagent is bound by the specific antibody in the first reagent, inhibiting the enzyme's activity. The enzyme G6PD catalyzes the oxidation of G6P with NAD^+ to form 6-phosphogluconate and reduced NADH. NADH strongly absorbs at 340 nm whereas NAD^+ does not. The rate of conversion, determined by measuring the increase in absorbance at 340 nm during a fixed time interval, is directly proportional to the amount of thyroxine in the sample.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

The precision of the γ -GT, Lipase and T4 reagents on an in-house ACE *Alera* system was evaluated using human serum samples at three concentrations of the respective measurand and one lot of reagents in an at least 20 day precision study. Each study was performed measuring each sample 2 times per run, 2 runs per day for at least 20 days for a total of 80 measurements per sample. The results are provided in the table below.

In-house on ACE Alera Clinical Chemistry System

Measurand	Sample Mean	Within Run		Total	
		SD	%CV	SD	%CV
γ-GT (U/L)	29	1.0	3.4	1.3	4.7
	71	1.4	2.0	2.4	3.4
	105	1.9	1.8	3.6	3.4
Lipase (U/L)	63	6.2	9.8	6.2	9.9
	379	10.5	2.8	15.4	4.1
	657	20.4	3.1	24.4	3.7
T4 (μg/dL)	6.0	0.19	3.1	0.34	5.6
	10.6	0.26	2.4	0.37	3.5
	17.1	0.56	3.3	0.66	3.9

Plasma and Serum In-house Precision Study:

The precision of the γ-GT, Lipase and T4 reagents for matched serum and Lithium heparin plasma samples at 3 concentrations of the respective measurand was evaluated in-house over a period of 5 to 6 days using one reagent lot (2 reagent lots for γ-GT). In this study, matched serum and plasma samples with a low concentration of measurand from a single donor were spiked to a high concentration of measurand with commercially-available analyte, and then the mid-level samples were prepared by mixing equal portions of the low-level and high-level samples. Each sample was measured 2 times per run for 2 runs per day for 5 to 6 days (N=20 for γ-GT and T4; N=24 for Lipase). All samples were tested on the ACE, ACE Alera and ACE Axcel systems. The precision results are summarized in the table below.

γ-GT Reagent (N=20)

ACE Clinical Chemistry System

Sample Mean (U/L)	Sample Type	Within Run		Total	
		SD	%CV	SD	%CV
38	Serum	0.9	2.4	1.2	3.1
38	Plasma	0.4	1.0	0.8	2.2
313	Serum	2.4	0.8	2.8	0.9
316	Plasma	2.4	0.8	3.0	0.9
602	Serum	2.3	0.4	2.6	0.4
605	Plasma	3.6	0.6	4.4	0.7

ACE Alera Clinical Chemistry System

Sample Mean (U/L)	Sample Type	Within Run		Total	
		SD	%CV	SD	%CV
39	Serum	0.9	2.4	1.0	2.5
40	Plasma	0.7	1.9	0.7	1.9
314	Serum	3.9	1.3	4.5	1.4
317	Plasma	1.7	0.5	2.4	0.8
601	Serum	4.2	0.7	6.1	1.0
604	Plasma	4.3	0.7	4.9	0.8

ACE Axcel Clinical Chemistry System

Sample Mean (U/L)	Sample Type	Within Run		Total	
		SD	%CV	SD	%CV
37	Serum	0.7	1.8	0.9	2.3
38	Plasma	0.7	2.0	0.9	2.4
318	Serum	2.0	0.6	2.6	0.8
319	Plasma	2.6	0.8	2.9	0.9
606	Serum	4.2	0.7	5.4	0.9
608	Plasma	4.4	0.7	5.9	1.0

Lipase Reagent (N=24)**ACE Clinical Chemistry System**

Sample Mean (U/L)	Sample Type	Within Run		Total	
		SD	%CV	SD	%CV
47	Serum	1.7	3.6	3.2	6.7
48	Plasma	2.2	4.6	3.2	6.6
283	Serum	5.1	1.8	13.1	4.6
278	Plasma	2.6	0.9	11.5	4.1
545	Serum	3.9	0.7	24.3	4.5
524	Plasma	5.9	1.1	18.9	3.6

ACE Alera Clinical Chemistry System

Sample Mean (U/L)	Sample Type	Within Run		Total	
		SD	%CV	SD	%CV
45	Serum	1.6	3.5	2.9	6.4
47	Plasma	1.5	3.2	3.5	7.4
286	Serum	3.8	1.3	19.1	6.7
278	Plasma	2.2	0.8	20.0	7.2
547	Serum	4.3	0.8	37.5	6.9
528	Plasma	5.0	1.0	31.7	6.0

ACE Axcel Clinical Chemistry System

Sample Mean (U/L)	Sample Type	Within Run		Total	
		SD	%CV	SD	%CV
44	Serum	2.8	6.3	3.0	6.9
48	Plasma	2.6	5.5	3.1	6.4
280	Serum	3.3	1.2	4.0	1.4
272	Plasma	4.8	1.8	6.8	2.5
534	Serum	5.5	1.0	9.5	1.8
518	Plasma	5.8	1.1	10.2	2.0

T4 Reagent (N=20)**ACE Clinical Chemistry System**

Sample Mean (µg/dL)	Sample Type	Within Run		Total	
		SD	%CV	SD	%CV
7.7	Serum	0.17	2.2	0.35	4.5
7.8	Plasma	0.28	3.5	0.29	3.8
12.7	Serum	0.46	3.6	0.63	4.9
13.1	Plasma	0.24	1.8	0.5	3.8
17.3	Serum	0.5	2.9	0.74	4.3
17.6	Plasma	0.76	4.3	0.76	4.3

ACE Alera Clinical Chemistry System

Sample Mean (µg/dL)	Sample Type	Within Run		Total	
		SD	%CV	SD	%CV
7.7	Serum	0.15	2.0	0.19	2.4
7.8	Plasma	0.14	1.9	0.21	2.7
12.5	Serum	0.24	1.9	0.48	3.9
12.9	Plasma	0.28	2.2	0.67	5.2
17.1	Serum	0.27	1.6	0.57	3.3
17.4	Plasma	0.41	2.4	0.44	2.6

ACE Axcel Clinical Chemistry System

Sample Mean (µg/dL)	Sample Type	Within Run		Total	
		SD	%CV	SD	%CV
7.9	Serum	0.18	2.3	0.21	2.6
8.0	Plasma	0.15	1.9	0.21	2.7
12.9	Serum	0.3	2.3	0.43	3.4
13.2	Plasma	0.19	1.5	0.71	5.4
17.5	Serum	0.5	2.9	0.75	4.3
17.6	Plasma	0.6	3.4	0.6	3.4

Serum POL Precision Study:

The precision of the γ -GT, Lipase and T4 reagents at an in-house and 3 physician office laboratories (POLs) using serum samples at 3 concentrations of the respective measurand and one reagent lot (2 reagent lots for γ -GT) was evaluated using a multiple day precision study. In this study, a serum sample with a low concentration of measurand from a single donor was spiked to a high concentration of measurand with commercially-available analyte, and then the mid-level sample was prepared by mixing equal portions of the low-level and high-level samples. The study was performed measuring each sample 2 times per run, 2 runs per day for a total of 5 days (N=20 measurements per sample) on the ACE Alera system. The precision results are provided in the table below.

γ -GT Reagent (N=20)

ACE Alera Clinical Chemistry System

Sample Mean (U/L)	Site	Within Run		Total	
		SD	%CV	SD	%CV
19	In-house	1.4	7.1	1.5	7.9
18	POL-1	0.7	4.2	0.9	5.2
18	POL-2	0.9	4.8	1.0	5.6
18	POL-3	0.9	5.2	1.0	5.3
298	In-house	3.3	1.1	3.7	1.2
287	POL-1	2.2	0.8	2.6	0.9
315	POL-2	1.9	0.6	2.3	0.7
299	POL-3	2.7	0.9	2.8	1.0
524	In-house	2.6	0.5	3.3	0.6
503	POL-1	4.5	0.9	4.5	0.9
561	POL-2	3.5	0.6	3.5	0.6
528	POL-3	3.0	0.6	4.6	0.9

Lipase Reagent (N=20)

ACE Alera Clinical Chemistry System

Sample Mean (U/L)	Site	Within Run		Total	
		SD	%CV	SD	%CV
24	In-house	2.0	8.7	2.0	8.7
23	POL-1	2.2	9.6	2.7	12.0
21	POL-2	1.8	8.5	1.9	8.9
22	POL-3	1.1	5.0	2.3	10.5
158	In-house	2.4	1.5	3.0	1.9
154	POL-1	3.8	2.5	7.7	5.0
154	POL-2	3.8	2.5	3.9	2.5

148	POL-3	1.9	1.3	5.2	3.5
315	In-house	2.8	0.9	11.5	3.7
292	POL-1	9.9	3.4	14.5	5.0
310	POL-2	2.5	0.8	6.3	2.0
293	POL-3	4.2	1.4	12.3	4.2

T4 Reagent (N=20)

ACE Alera Clinical Chemistry System

Sample Mean (µg/dL)	Site	Within Run		Total	
		SD	%CV	SD	%CV
4.1	In-house	0.09	2.2	0.17	4.3
4.3	POL-1	0.09	2.2	0.15	3.5
4.1	POL-2	0.12	2.8	0.18	4.3
3.9	POL-3	0.17	4.4	0.2	5.0
10.1	In-house	0.14	1.4	0.33	3.2
10.3	POL-1	0.41	4.0	0.43	4.2
10.1	POL-2	0.25	2.5	0.29	2.9
10.1	POL-3	0.31	3.1	0.49	4.9
16.0	In-house	0.27	1.7	0.41	2.6
16.4	POL-1	0.58	3.5	0.89	5.4
16.3	POL-2	0.46	2.8	0.76	4.7
17.6	POL-3	0.79	4.5	0.97	5.5

b. Linearity/Assay Reportable Range:

The sponsor assessed linearity on the ACE Alera analyzer with each sample tested in triplicate using one lot of reagents. Nine diluted samples with measurand concentrations evenly distributed were prepared by diluting a high measurand concentration serum pool. This yielded linearity samples with levels that spanned the measuring range of each of the three analytes (γ -GT, Lipase and T4) measured.

The sponsor calculated linear and polynomial regressions from mean observed values versus expected values using weighted regression model. The linear regressions between the expected values and the measured values are found in the table below:

Measurand	Range tested	Slope (95% CI)	Intercept (95% CI)
γ -GT (U/L)	4.0 to 993	1.036 (1.031 to 1.042)	0.8 (-0.8 to 2.4)
Lipase (U/L)	11.0 to 739	0.971 (0.965 to 0.977)	0.2 (-1.0 to 1.5)
T4 (µg/dL)	1.2 to 19.7	1.057 (1.033 to 1.081)	-0.09 (-0.28 to 0.10)

The linearity data provided support the sponsor's claims for the reportable range for the γ -GT reagent of 7.0 to 950 U/L, Lipase reagent of 15 to 700 U/L, and T4 reagent of 1.3 to 19.6 µg/dL.

Auto-dilution Study:

The sponsor provided auto-dilution studies to confirm the auto-dilution function on the ACE Alera analyzer for the ACE γ -GT reagent (1:3 dilution) and ACE Lipase reagent (1:2 dilution) using plasma and/or serum samples. The studies compared the results obtained from auto-dilution of the samples to the manual dilution results. All samples recovered within $\pm 5\%$ bias.

The ACE T4 reagent is not set for automatic dilution on the ACE Alera analyzer.

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

ACE γ -GT Reagent

Traceable to another commercially available FDA cleared assay by method comparison. This is not a calibrated test.

ACE Lipase Reagent

Traceable to another commercially available FDA cleared assay by method comparison. The calibrator was previously cleared under k113438.

ACE T4 Reagent

Traceable to another commercially available FDA cleared assay by method comparison. The calibrator was previously cleared under k113437.

d. *Detection Limit:*

The LoQ studies for each measurand were evaluated based upon CLSI EP17-A guidance document using the ACE Alera analyzer.

LoQ was determined by evaluating five low level samples with eight replicates over five days for a total of 40 measurements per sample. Sponsor defined LoQ as concentration with a %CV of $< 20\%$.

Measurand	LoQ
γ -GT	7.0 U/L
Lipase	13.0 U/L
T4	1.3 $\mu\text{g/dL}$

Based on the LoQ and the linearity studies, the measuring ranges for the three measurand on the ACE Alera instrument are as shown below:

Measurand	Assay Range
γ -GT	7.0 to 950 U/L
Lipase	15 to 700 U/L
T4	1.3 to 19.6 $\mu\text{g/dL}$

e. *Analytical Specificity:*

Interference testing was performed according to CLSI EP07-A2 guidance document for the γ -GT reagent, Lipase reagent and T4 reagent on the ACE Alera analyzer.

Human serum samples were used with two different concentrations (normal and abnormal) of the measurand (γ -GT, lipase and T4). Interference from unconjugated bilirubin, hemolysate, lipemia and ascorbic acid was tested. For lipemia interference studies, avian triglyceride concentrate (γ -GT and lipase) or Intralipid (T4) was used. Each sample level was spiked with increasing amounts of interferent for a total of six samples with interferent. Control samples at each level that were not spiked with the interferents were used to compare with the spiked samples and percent recovery was calculated for each sample. Interference is considered to be significant when the analyte recovery changes by $\pm 10\%$. The results are summarized in the table below.

Interferent	No Significant Interference at or below:		
	γ -GT	Lipase	T4
Icterus	14.2 mg/dL	12.5 mg/dL	47.2 mg/dL
Hemolysis	125 mg/dL	1000 mg/dL	1000 mg/dL
Lipemia	500 mg/dL	803 mg/dL	1000 mg/dL
Ascorbic acid	6.0 mg/dL	6.0 mg/dL	6.0 mg/dL

The following statements are included as limitations in the respective labeling for the three reagents:

Lipase reagent:

“Do not use icteric samples.”

“Use clear, unhemolyzed serum or lithium heparin plasma.”

“A comprehensive list of drugs and other substances which can affect lipase activity in serum is given by Young, et. Al.¹”

γ -GT reagent:

“Use clear, unhemolyzed serum or lithium heparin plasma.”

“Do not use hemolyzed samples. Hemolysis interferes with the γ -GT assay and may cause erroneous results.”

“A comprehensive list of drugs and other substances which can affect lipase activity in serum is given by Young, et. Al.¹”

T4 reagent:

“In rare instances, patients may have autoantibodies that will interfere with this assay and result in low thyroxine results.”

“The performance characteristics of this assay have not been established for neonatal samples.”

“Use clear, unhemolyzed serum or lithium heparin plasma.”

¹ Young, D.S., et.al., Effects of drugs on Clinical Laboratory Tests. 5th Edition, AACC Press, Washington, D.C. (2000).

In addition, the sponsor has provided cross-reactivity studies with the structurally related compounds, 3,3',5,5'-Tetraiodothyroacetic acid, L-Thyroxine, and D-Thyroxine, and interference studies with HAMA and Rheumatoid Factor for their T4 reagent assay on ACE *Alera* Clinical Chemistry. Interference is considered to be significant when the analyte recovery changes by $\pm 10\%$. The results are summarized in the table below.

Cross-Reactivity Study Results At Normal T4 levels:

Cross-Reactant	Concentration Tested ($\mu\text{g/dL}$)*	% Cross-reactivity
3,3',5,5'- Tetraiodothyroacetic acid	5.0	18.4
L-Thyroxine	5.0	91.6
	10.0	92.8
D-Thyroxine	5.0	68.0
	10.0	74.6

* The tested concentrations of the cross-reactant tested greatly exceed the normal serum or plasma concentrations of these compounds.

Interference Study Results:

Interferent	No Significant Interference at or below:
HAMA	800 ng/mL**
Rheumatoid Factor	516 IU/mL**

** These concentrations were the highest available concentration for interference testing.

- f. *Assay cut-off:*
Not applicable.

2. Comparison Studies:

- a. *Method Comparison with Predicate Device:*

In-house Method Comparison Study:

Method comparison study was carried out in accordance with CLSI EP09-A2-IR guidelines. Serum samples obtained from clinical sites or commercial sources were evaluated. In order to span the reportable range of the assay, some of the samples were spiked (γ -GT - 6, Lipase - 5, T4 – 3) and / or diluted (T4 – 2). All samples were run in singlicate in-house on the ACE *Alera* Clinical Chemistry System (y-axis) and the ACE Clinical Chemistry System (x-axis) using at least two lots of each reagent. Deming linear regression analysis results are as shown:

Method Comparison: In-House ACE (x) versus In-House ACE Alera (y)	γ-GT	Lipase	T4
N	51	49	50
Range	17 to 845 U/L	15 to 691 U/L	1.3 to 18.5 μ g/dL
Slope (95% CI)	0.975 (0.972 to 0.979)	1.038 (1.029 to 1.048)	1.004 (0.972 to 1.037)
Intercept (95% CI)	4.3 (3.5 to 5.2)	-4.8 (-6.4 to -3.3)	-0.08 (-0.33 to 0.17)
Correlation Coefficient (R)	0.9999	0.9995	0.9937

Serum POL Method Comparison Study:

Method comparison studies were performed following CLSI document EP9-A2IR. Patient serum samples were run in singlicate at three POL sites on ACE Alera Clinical Chemistry Systems (y-axis) and in-house on the ACE Clinical Chemistry System (x-axis) using at least two lots of each reagent. In order to span the reportable range of the assay, some of the samples were spiked (γ -GT - 5, Lipase - 5, T4 - 3 at all five sites and T4 - 1 at POL3) and / or diluted (T4 - 2). Data were analyzed by Deming linear regression. Results are summarized in the table below:

Reagent	Statistic Parameters	ACE In-House (x) vs. ACE Alera POL 1 (y)	ACE In-House (x) vs. ACE Alera POL 2 (y)	ACE In-House (x) vs. ACE Alera POL 3 (y)
γ-GT	N	51	51	51
	Range	13 to 821 U/L	16 to 880 U/L	16 to 849 U/L
	Slope (95% CI)	0.950 (0.945 to 0.956)	1.028 (1.02 to 1.036)	0.996 (0.990 to 1.003)
	Intercept (95% CI)	1.9 (0.7 to 3.1)	2.9 (1.2 to 4.7)	2.4 (0.9 to 3.9)
	Correlation Coefficient (R)	0.9998	0.9996	0.9997
Lipase	N	51	50	51
	Range	21 to 669 U/L	16 to 647 U/L	14 to 664 U/L
	Slope (95% CI)	1.028 (1.001 to 1.054)	1.017 (0.993 to 1.040)	0.992 (0.978 to 1.006)
	Intercept (95% CI)	3.3 (-1.0 to 7.6)	-3.5 (-7.3 to 0.3)	-2.9 (-5.2 to -0.7)
	Correlation Coefficient (R)	0.9960	0.9969	0.9988
T4	N	50	50	48
	Range	1.7 to 18.6 μ g/dL	1.7 to 19.2 μ g/dL	1.5 to 18.8 μ g/dL
	Slope	1.022	1.048	1.033

	(95% CI)	(0.986 to 1.058)	(1.007 to 1.089)	(0.983 to 1.083)
	Intercept (95% CI)	-0.14 (-0.42 to 0.13)	-0.31 (-0.63 to 0.01)	-0.10 (-0.47 to 0.27)
	Correlation Coefficient (R)	0.9926	0.9909	0.9868

b. *Matrix Comparison:*

To characterize correlation between lithium heparin plasma (y-axis) and serum (x-axis), a matrix comparison study was performed for each measurand using paired samples on the ACE, ACE *Alera* and ACE *Axcel* analyzers. All specimens were run in singlicate using at least two lots of each reagent. In order to span the reportable range of the assay, some of the samples were spiked (γ -GT – 4 on ACE *Axcel*; Lipase – 10 on ACE and ACE *Alera*, and 4 on ACE *Axcel*; T4 – 3 on all) and / or diluted (T4 – 1 on all). Data were analyzed by Deming linear regression. Results are summarized in the table below:

Serum (x-axis) versus Lithium heparin plasma (y-axis):

Reagent	Statistic Parameters	ACE	ACE <i>Alera</i>	ACE <i>Axcel</i>
γ-GT	N	100	97	53
	Range	8 to 861 U/L	11 to 809 U/L	13 to 915 U/L
	Slope (95% CI)	0.972 (0.964 to 0.981)	0.960 (0.951 to 0.969)	0.987 (0.967 to 1.008)
	Intercept (95% CI)	1.5 (-0.3 to 3.3)	2.8 (0.8 to 4.7)	4.0 (-1.8 to 9.8)
	Correlation Coefficient (R)	0.9990	0.9989	0.9973
Lipase	N	42	43	62
	Range	15 to 659 U/L	18 to 662 U/L	15 to 662 U/L
	Slope (95% CI)	1.024 (1.011 to 1.038)	1.022 (1.010 to 1.033)	0.980 (0.954 to 1.007)
	Intercept (95% CI)	-2.5 (-5.0 to -0.1)	-0.9 (-3.0 to 1.2)	-2.0 (-5.9 to 2.0)
	Correlation Coefficient (R)	0.9992	0.9994	0.9947
T4	N	55	55	55
	Range	2.1 to 18.7 μ g/dL	1.9 to 17.9 μ g/dL	2.1 to 18.2 μ g/dL
	Slope (95% CI)	0.963 (0.916 to 1.009)	0.976 (0.933 to 1.019)	1.007 (0.958 to 1.057)
	Intercept (95% CI)	0.35 (-0.03 to 0.73)	0.17 (-0.18 to 0.51)	0.01 (-0.38 to 0.40)
	Correlation Coefficient (R)	0.9847	0.9870	0.9841

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 for Serum/Plasma:

Expected values are based on the literature reference:

γ -GT: Male, 13.0 to 68.0 U/L; Female, 11.0 to 48.0 U/L (Ref. 2)

Lipase: Less than 60.0 U/L (Ref. 3)

T4: 5.0 to 12.0 μ g/dL (Ref. 4)

² Reference Intervals – UNC Hospitals, McLendon Clinical Laboratories

<http://unchealthcare.org/labtestinfo>

³ Wu, A.H.B. (Ed.), Tietz Clinical Guide to Laboratory Tests, 4th edition, Saunders, Elsevier, St. Louis, MO (2006).

⁴ Tietz, et. al., (Ed.), Clinical Guide to laboratory Tests, W.B. Saunders Co., 2nd edition, Philadelphia, PA (1990)

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.