510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY

I Background Information:

A 510(k) Number

K251074

B Applicant

Truvian Health

C Proprietary and Established Names

Tru Liver Health Test Panel

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
CJE	Class II	862.1050-Alkaline phosphatase or isoenzymes test system	CH - Clinical Chemistry
CIT	Class II, meets the limitations of exemptions in 862.9(c)(9)	862.1100-Aspartate amino transferase (AST/SGOT) test system	CH - Clinical Chemistry
CKA	Class I, meets the limitations of exemptions in 862.9(c)(9)	862.1030-Alanine amino transferase (ALT/SGPT) test system	CH - Clinical Chemistry

II Submission/Device Overview:

A Purpose for Submission:

New device

B Measurand:

Alkaline phosphatase (ALP)
Alanine amino transferase (ALT)
Aspartate amino transferase (AST)

C Type of Test:

Quantitative, Photometric/Colorimetric

III Intended Use/Indications for Use:

A Intended Use(s):
See Indications for Use below.

B Indication(s) for Use:
The Tru Liver Health Test Panel is part of the TruWellness Panel™ and is intended for use on the Tru Analyzer. The Tru Liver Health Test Panel (part of the TruWellness Panel™) is an in vitro diagnostic device and intended to be used for the quantitative determination of Alkaline Phosphatase (ALP), Aspartate Aminotransferase (AST), and Alanine Aminotransferase (ALT) in lithium-heparinized venous whole blood in clinical laboratory or point-of-care settings.

The Tru Liver Health Test Panel (part of the TruWellness Panel™) is an in vitro diagnostic test system that aids the physician in diagnosing the following disorders in adults 18 years of age or older:

Alkaline Phosphatase (ALP): Liver, bone, parathyroid, and intestinal diseases.

Aspartate Aminotransferase (AST): Certain types of Liver and heart diseases.

Alanine Aminotransferase (ALT): Certain Liver (e.g., viral hepatitis and cirrhosis) and heart diseases.

C Special Conditions for Use Statement(s):
Rx - For Prescription Use Only

The alkaline phosphatase (ALP) analyte is sensitive to hemolysis, and the instrument flags samples with hemolysis values of 50 mg/dL or higher. This results in approximately 5% of samples being flagged, preventing result reporting. Therefore, implementing an alternative backup method not subject to hemolysis interference is recommended.

The instrument was validated up to an altitude of 2,000 m (6562 ft). The Tru Liver Health Test Panel should not be used above 2,000 m (6562 ft).

D Special Instrument Requirements:

Tru Analyzer

IV Device/System Characteristics:

A Device Description:

The Tru Liver Health Test Panel contains the following reagents:

Component	Quantity per Kit
2-Oxoglutarate	75.2 µg
Lactate Dehydrogenase	> 48.0 mU
L-Aspartate	1,264.4 µg
Malate Dehydrogenase	> 6.6 mU
Nicotinamide Adenine Dinucleotide (Reduced)	9.2 mg
p-Nitrophenyl Phosphate	8.4 µg
Buffers, Preservatives, Stabilizers	

The single-use consumable kit houses all the components needed to process as well as analyze samples on the Tru Analyzer, including dried reagents, internal process control solutions, barcodes that manage the identity of the kit lot (e.g., Disc and Support Pack ID), calibration information, dilution buffers, and single-use plastic pipette tips. It also serves as a waste container which the user discards of at the end of the run. The Support Pack contains a feature to accept a standard 4 mL blood tube. The Support Pack also houses 22 pipette tips for transferring and mixing samples and reagents, 11 dilution wells to support reagent processing activities within the test system (e.g., sample dilution, reagent dilution, rehydration of dried reagents), and 6 x 2 ml tubes that contain additional wet and dry reagents.

B Principle of Operation:

Alkaline phosphatase (ALP): Alkaline phosphatase hydrolyzes ρ -NPP in a metal-ion buffer and forms ρ -nitrophenol and phosphate. ρ -nitrophenol has an intense color at 405 nm in an alkaline solution. The formation of the color can be followed kinetically, and this rate of change is proportional to the concentration of alkaline phosphatase in the sample.

$$\text{p-NPP} + \text{H2O} \xrightarrow{\text{Alkaline Phosphatase}} \text{Phosphate} + \text{p-NP}$$

Alanine aminotransferase (ALT): ALT catalyzes the transfer of the amino group from alanine to oxoglutarate (α-KG) with the formation of glutamate and pyruvate. This last compound is reduced to lactate in a reaction catalyzed by lactate dehydrogenase (LDH). In this same reaction, an equivalent amount of NADH is oxidized to NAD. The resulting rate of decrease in absorbance at 340 nm is directly proportional to the activity of ALT in the sample.

$$\text{L-alanine} + \text{α-KG} \xrightarrow{\textit{Alanine Aminotransferase}} \text{Pyruvate} + \text{L-glutamate}$$

Aspartate aminotransferase (AST): Aspartate aminotransferase (AST) catalyzes the transfer of the amino group from aspartate to oxoglutarate (α-KG) with the formation of glutamate and

oxaloacetate (OAA). This last compound is reduced to malate in a reaction catalyzed by malate dehydrogenase (MDH). In this same reaction, an equivalent amount of NADH is oxidized to NAD. The resulting decrease in absorbance at 340 nm is followed spectrophotometrically and the rate is directly proportional to the amount of AST present in the sample.

L-aspartate + α-KG *Aspartate Aminotransferase* → OAA + L-glutamate

OAA + NADH *MDH*→ L-malate + NAD

V Substantial Equivalence Information:

A Predicate Device Name(s):

Comprehensive Metabolic Panel

B Predicate 510(k) Number(s):

K171971

C Comparison with Predicate(s):

Device & Predicate Device(s):	K251704	K171971
Device Trade Name	Tru Liver Health Test Panel	Comprehensive Metabolic Panel
General Device Characteristic Similarities		
Intended Use/Indications For Use	In vitro quantitative determination of Alkaline phosphatase (ALP), Aspartate aminotransferase (AST), and Alanine aminotransferase (ALT) concentrations in lithium-heparinized venous whole blood in clinical laboratory or point-of-care settings.	Same
General Device Characteristic Differences		
Specimen Type	Lithium-heparinized venous whole blood	Lithium-heparinized venous whole blood, lithium-heparinized plasma, or serum

Device & Predicate Device(s):	K251704	K171971
Analytical Measuring Range	ALP: 25 – 1200 U/L ALT: 15 – 500 U/L AST: 11 – 700 U/L	ALP: 41 – 1500 U/L ALT: 20 – 500 U/L AST: 20 – 1000 U/L

VI Standards/Guidance Documents Referenced:

• CLSI (Clinical & Laboratory Standards Institute) EP05-A3 - Evaluation of Precision of Quantitative Measurement Procedures; Approved Guideline -Third Edition
• CLSI EP06 - Evaluation of the Linearity of Quantitative Measurement Procedures – Second Edition
• CLSI EP07-A3 - Interference Testing in Clinical Chemistry - Third Edition
• CLSI EP17-A2 - Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline -Second Edition
• CLSI EP37 - Supplemental Tables for Interference Testing in Clinical Chemistry -First Edition

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

A. Reproducibility with Control Materials: This study was designed according to CLSI EP05-A3. The study was conducted at 3 external point-of-care (POC) sites, with 3 analyzers per site and at least 3 POC operators per site. Testing was conducted over 5 days using a single lot of consumables and consisted of 3 levels of control samples at low, medium, and high concentrations, with 6 replicates run per day, 3 run in the morning and 3 run in the afternoon (each replicate is run on an individual instrument). Overall, there were at least 90 replicates for each precision control sample level (3 sites x 1 sample x 3 replicates per run x 2 runs per day x 5 days = 90 replicates). The total precision as well as within run, between day, and between site precision were estimated. Results are displayed below:

				Within-Run		Between-Run		Between-Day		Between-Site		Reproducibility	
Analyte (U/L)	Sample	N	Mean	SD	CV %	SD	CV %	SD	CV	SD	CV %	SD	CV%
ALP	C1	91	71	4	4.9	0	0.0	1	1.4	1	1.9	4	5.5
ALP	C2	91	120	6	4.6	0	0.0	2	1.2	1	0.9	6	4.9
ALP	C3	92	373	8	2.2	2	0.5	0	0.0	7	1.9	11	2.9
ALT	C1	92	62	2	2.9	1	1.1	0	0.0	2	3.2	3	4.5
ALT	C2	91	291	6	1.9	0	0.0	3	0.9	3	1.1	7	2.4
ALT	C3	92	427	5	1.1	2	0.5	3	0.6	4	1.0	7	1.7

				Within-Run		Between-Run		Between-Day		Between-Site		Reproducibility	
Analyte (U/L)	Sample	N	Mean	SD	CV %	SD	CV %	SD	CV	SD	CV %	SD	CV%
AST	C1	92	57	1	1.9	0.3	0.4	0.4	0.7	2	3.6	2	4.1
AST	C2	91	292	6	2.0	0	0.0	3	1.0	4	1.4%	8	2.7%
AST	C3	92	508	7	1.3	3	0.7	4	0.7	5	1.1	10	2.0

B. Whole Blood Precision: This study was evaluated at four sites collecting samples from the intended use population with normal and abnormal analyte levels. At least 20 subjects were enrolled at each site and for each test subject, eight replicates were measured across four instruments and two operators. The SD and CV% precision estimates were calculated by pooling subject standard deviations and/or % CV for predefined low, medium or high sub-intervals selected to represent normal and abnormal regions of the analytical measuring range and encompassing medical decision levels. Pooled imprecision is considered a representative estimate of reproducibility that include variability of sites, instruments, operators, days and repeatability.

Analyte	Range	Subject (N)	Replicate (N)	Mean	SD	CV%
ALP (U/L)	25–130	66	487	79.4	4.3	5.8
ALP (U/L)	130–300	16	122	194.1	5.9	3.1
ALP (U/L)	300–1200	7	47	655.7	20.4	4.2
ALT (U/L)	15–30	39	270	20.9	1.4	6.5
ALT (U/L)	30–100	23	172	47.7	1.8	3.9
ALT (U/L)	100–500	7	56	204.8	9.9	4.4
AST (U/L)	10–30	57	444	19.0	1.2	6.7
AST (U/L)	30–100	22	165	45.4	1.5	3.5
AST (U/L)	100–700	6	44	141.1	5.1	3.7

C. Whole Blood Between-Lot Precision: 22 subjects consisting of 6 healthy individuals and 16 individuals in various disease states were enrolled at a single point-of-care clinical site and tested across 3 lots of the candidate panel on 3 individual Tru Analyzers by 2 operators. A total of 6 replicates per subject participant, 2 replicates per lot, were tested by 1 of the 2 operators. Results below present the number of subjects evaluated, the % difference from the median and pooled %CV by lot and by analyte. The elevated median relative difference percentages observed for AST analyte Lots B and C resulted from values falling near the limit of quantification.

Analyte (U/L)	Subjects per Lot	Median relative difference %			Pooled %CV		
		Lot-A	Lot-B	Lot-C	Lot-A	Lot-B	Lot-C
ALP	22	-0.3 %	-0.6 %	-1.1 %	4.0 %	6.5 %	5.7 %
ALT*	18	-0.1 %	-0.3 %	0.3 %	6.0 %	10.4 %	3.6 %
AST	22	-0.7 %	3.5 %	-4.0 %	5.5 %	5.7 %	6.0 %

**a replicate for ALT Lot-B was identified as a statistical outlier and the table below provides the results without the outlier included.*

Analyte (U/L)	Subjects per Lot	Median relative difference %			Pooled %CV		
		Lot-A	Lot-B	Lot-C	Lot-A	Lot-B	Lot-C
ALT	18	-0.1 %	-0.3 %	0.3 %	6.0 %	6.4 %	3.6 %

D. Control Between-Lot Reproducibility: To evaluate the lot-to-lot reproducibility, 3 control samples (low, medium and high) were tested on 3 Tru Analyzers using 3 lots of the candidate test. Each sample level was tested with at least five (5) replicates on one Tru Analyzer each day with each single-use consumable kit lot over 3 days to achieve a minimum of fifteen (15) replicates per sample level and kit lot. Results are displayed below: -

Assay	Analyte (U/L)	N	Grand Mean	Lot Mean			CV		
				Lot 1	Lot 2	Lot 3	Lot 1	Lot 2	Lot 3
ALP	Low	45	71.1	72.4	70.4	70.4	4.2%	6.3%	5.9%
ALP	Medium	45	118.8	117.4	121.3	117.7	5.7%	3.1%	3.3%
ALP	High	45	379.6	383.0	373.7	382.0	2.0%	3.3%	3.2%
ALT	Low	45	58.5	58.7	58.1	58.8	2.7%	2.7%	2.6%
ALT	Medium	45	288.4	289.6	288.4	287.2	1.8%	1.7%	1.3%
ALT	High	45	421.0	425.5	415.8	421.8	1.9%	2.8%	1.7%
AST	Low	45	56.1	57.3	55.2	55.7	2.40%	2.0%	1.9%
AST	Medium	45	290.7	292.3	290.6	289.4	1.3%	1.2%	1.3%
AST	High	45	504.2	510.1	498.3	504.1	1.6%	2.9%	1.9%

2. Linearity:

The linearity study was conducted in accordance with CLSI EP06-Ed2. Linearity was assessed with multiple panels of samples. The linear range for the ALP, ALT, and AST assays were determined by testing nine (9) samples containing varying concentrations of analyte. Each sample was tested in quadruplicate, using one (1) kit lot, across 4 Tru Analyzers, on a single day. The results of the study supported the claimed linear interval for ALP, ALT, and AST. Within the claimed analytical measuring range, the maximum observed deviation for ALP, ALT, and AST are 7.3%, 1.5% and 5.5%, respectively.

3. Analytical Specificity/Interference:

The sponsor conducted an analytical specificity study according to CLSI EP07-A3 and EP37 guidelines. Testing was performed with contrived lithium heparin whole blood samples from a healthy donor population, with two (2) analyte concentration levels targeted based on recommended levels in CLSI EP07-A3. Potentially interfering substances were prepared in the appropriate diluent and then added to the low and high contrived whole blood samples to create test samples. For any substances identified as an interferent, dose response testing was conducted to assess the highest concentration limit below which no significant interference was observed.
The sponsor defined significant interference as bias >10% between the test and control samples and the results are displayed below:

Substance (mg/dL)	Max Concentration without Interference mg/dL		
	ALP	ALT	AST
Triglycerides	1037	1037	1037
Hemoglobin	48	545	159
Conjugated Bilirubin	22.5	22.5	22.5
Unconjugated Bilirubin	31.5	27.5	14.0

Substance	Max Concentration without Interference			
	ALP	AST	ALT	Units
Acetaminophen	15.6	15.6	15.6	mg/dL
Acetylsalicylic acid	3.0	3.0	3.0	mg/dL
Ampicillin	7.5	7.5	7.5	mg/dL
Cefoxitin	330	660	495	mg/dL
Cyclosporine	0.18	0.18	0.18	mg/dL
Doxycycline	1.35	1.8	1.8	mg/dL
Heparin	3,300	3,300	3,300	U/L
Ibuprofen	21.9	21.9	21.9	mg/dL
Levodopa (L-Dopa)	0.75	0.75	0.75	mg/dL
Methyldopa	2.25	2.25	2.25	mg/dL
Metronidazole	12.3	12.3	12.3	mg/dL
Phenylbutazone	24.1	32.1	32.1	mg/dL
Rifampicin	4.8	4.8	4.8	mg/dL
Theophylline	6.0	6.0	6.0	mg/dL
Acetylcysteine	15	15	15	mg/dL

Substance	Max Concentration without Interference			
	ALP	AST	ALT	Units
Ascorbic Acid	5.25	5.25	5.25	mg/dL
Caffeine	8.1	10.8	10.8	mg/dL
Cephalothin	135	135	180	mg/dL
Cimetidine	3.0	3.0	3.0	mg/dL
Salicylic Acid	2.86	2.86	2.86	mg/dL

The sponsor provided adequate information to support hemolysis, icterus and lipemia flags will detect samples subject to these interferences.

4. Detection Limit and Assay Reportable Range:

The Limit of Blank (LoB) was determined in accordance with the classical approach provided in section 5.3 of CLSI EP17-A2. LoB was determined by running four (4) blank samples on four (4) Tru Analyzers over three (3) days of testing with five (5) replicates per sample for a total of sixty (60) tests for each single-use consumable kit lot.

The LoD (Limit of Detection) and LoQ (Limit of Quantitation) were determined based on recommendations from CLSI EP17-A2. The LoQ corresponds to the lowest concentration of analyte in a sample that had a CV ≤ 20%.

The results of the studies are summarized below and support the claimed measuring ranges for each test.

(U/L)	LoB	LoD	LOQ
ALP	6.15	12	22
AST	2.29	4	11
ALT	5.80	8	11

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

Traceability:

The sponsor claims the following regarding traceability:

Assay	Traceability
ALP	JCCLS standard-CRM001d
AST	Traceable to IFCC through a commercially available method

Assay	Traceability
ALT	Traceable to IFCC through a commercially available method

Sample Stability:

Sample Stability studies performed demonstrate that samples measured on the candidate tests are stable for 60-minutes post-collection.

Altitude Effects:

An altitude study was conducted to evaluate performance of ALP, ALT, and AST at sea-level and an altitude of 2000 meters. This study supports the device performance at altitudes of up to 2000 meters.

6. Assay Cut-Off:

Not Applicable.

B Comparison Studies:

1. Method Comparison with Predicate Device:

A method comparison study was conducted to establish the performance characteristics of the Tru Kidney Liver Test Panel on the Tru Analyzer when used to analyze lithium heparin whole blood clinical specimens. Whole blood samples from apparently healthy patient and from patients with acute or chronic health conditions were collected from 5 point-of-care sites, tested by at least 3 point-of-care operators per site, on at least 4 Tru Analyzers per site, using 5 lots of reagents. Results compared against legally marketed comparators are displayed below. The data demonstrated that accuracy was consistent across sites. Differences at the medical decision points were reviewed and found acceptable.

Analyte (U/L)	N	Range (U/L)	Slope	Intercept	R
ALP	259	30–1185	1.04 (1.02, 1.06)	-3.06 (-4.81, -1.32)	0.997
AST	272	11–673	1.00 (1.00, 1.00)	1.00 (0.50, 1.00)	0.999
ALT	193	15–464	0.91 (0.90, 0.93)	2.62 (2.04, 3.22)	0.999

2. Matrix Comparison:

Not Applicable.

C Clinical Studies:

1. Clinical Sensitivity:

   Not Applicable.

2. Clinical Specificity:

   Not Applicable.

3. Clinical Cut-Off

   Not Applicable.

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

   Not Applicable.

D Expected Values/Reference Range:

The reference ranges were established based on scientific literature. *

Analyte		Reference Interval	Units
ALP		37–108	U/L
AST	Male	< 35	U/L
	Female	< 31	U/L
ALT	Male	< 45	U/L
	Female	< 34	U/L

*C. A. Burtis, E. R. Ashwood, and D. E. Bruns. Tietz Textbook of Clinical Chemistry and Molecular Diagnostics. 4th ed., Elsevier Saunders, St. Louis, (2006).

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.