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

A. 510(k) Number:
   k073191

B. Purpose for Submission:
   New Device

C. Measurand:
   Alkaline Phosphatase

D. Type of Test:
   Colorimetric and Enzymatic

E. Applicant:
   Thermo Fisher Scientific Oy

F. Proprietary and Established Names:
   Alkaline Phosphatase (IFCC) plus, codes 981832 and 981833; Alkaline
   Phosphatase (IFCC)
   eCal, code 981830
   Nortrol, code 981043
   Abtrol, code 981044

G. Regulatory Information:

H.

Product Code	Classification	Regulation Section	Panel
Alk. Phosphatase (CJE)	Class II	21 CFR 862.1050 Alkaline phosphatase Test System	75 Clinical Chemistry(CH)
Product Code	**Classification**	**Regulation Section**	**Panel**
eCal Calibrator, Multi-Analyte Mixture (JIX)	Class II	21 CFR 862.1150 Calibrator	75 Clinical Chemistry(CH)
Product Code	**Classification**	**Regulation Section**	**Panel**
Nortrol, Abtrol Control (JJY)	Class I, reserved	21 CFR§ 862.1660 Quality control material (assayed and unassayed)	75 Clinical Chemistry(CH)

H. Intended Use:

    1. Intended use(s):

Alkaline Phosphatase (IFCC): The Alkaline Phosphatase test system is intended for quantitative *in-vitro* diagnostic determination of the activity of the enzyme Alkaline Phosphatase in serum and plasma on T60 instrument according to the IFCC method. Measurements of alkaline phosphatase are used in the diagnosis and treatment of liver and bone diseases.

For *in vitro* diagnostic use in the quantitative determination of alkaline phosphatase (orthophosphoric - monoester phospho-hydrolase, alkaline optimum, EC 3.1.3.1) activity in human serum or plasma on T60 instruments according to the IFCC method.

eCal
For *in vitro* diagnostic use on T60 instrument. eCal is used as a calibrator for enzyme tests using methods defined by Thermo Fisher Scientific Oy.

Nortrol
For *in vitro* diagnostic use for quantitative testing on T60 instrument. Nortrol is a control serum to monitor trueness and precision of the analytes listed in the separate Nortrol value sheet. The given values are valid for T60 Clinical Chemistry Instruments using methods defined by Thermo Fisher Scientific Oy.

Abtrol
For *in vitro* diagnostic use for quantitative testing on T60 instrument. Abtrol is a control serum to monitor trueness and precision of the analytes listed in the separate Abtrol value sheet. The given values are valid for T60 Clinical Chemistry Instruments using methods defined by Thermo Fisher Scientific Oy.

    2. Indication(s) for use:
See intended use.

    3. Special conditions for use statement(s):
Prescription use only.

    4. Special instrument requirements:
T60 and DPC T60i, DPC T60i Kusti.

I. Device Description:

The Alkaline Phosphatase (IFCC) Plus is supplied as a ready-to-use, IVD reagent kit with reagent A and reagent B (two kit sizes, codes 981832 and 981833). For kit code 981832, 1 mL of reagent B is added to reagent A and the mixture is mixed well before use. For kit code 981833, the entire contents of the reagent B bottle are added to reagent A and the mixture is mixed well before use.

Reagent A contains 2-Amino-2-methyl-1-propanol, (AMP) (pH 10.4, 1.125 mol/l)

Magnesium acetate (2.0 mmol/l), Zinc sulphate (0.5 mmol/l), HEDTA (2.5 mmol/l) and NaN3 (< 0.1 %).

Reagent B contains p-Nitrophenylphosphate (80 mmol/l) and NaN_3 (< 0.1 %).

The calibrator containing alkaline phosphatase from human placenta (recombinant) and controls are separate from the reagents and are provided in individual vials.

eCal calibrator is a lyophilized bovine serum based reference preparation to be used as a multi-analyte calibrator.

Nortrol and Abtrol are freeze-dried products prepared from human serum with added constituents of purified biochemicals (tissue extracts of human and animal origin), chemicals, therapeutic drugs, preservatives and stabilizers. The control is provided in lyophilized form for increased stability.

All human materials included in the calibrators and controls were tested by FDA approved methods and found to be negative for the presence of antibodies to HIV-1, HIV-2, HBsAg, and HCV.

J. Substantial Equivalence Information:
   1. Predicate device name(s):
      Bayer Corporation, model Bayer ADVIA 2400 Chemistry System.
   2. Predicate K number(s):
      k991576
   3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Intended Use	For *in vitro* diagnostic use in the quantitative determination of alkaline phosphatase in human serum or plasma.	Same.
Sample Type	Serum, plasma (Li-heparin)	Serum, plasma (Li-heparin)
Assay Protocol	ALP catalyzes the hydrolysis of p-NPP resulting in the formation of p-nitrophenol in alkaline solution	Same.

Differences		
Item	Device	Predicate
Wavelength	Product formation is followed at 405 nm.	Product formation is followed at 410/478 nm.
Measuring Range	20 – 1000 U/L	0-1100 U/L
Instrument	T60 and DPC T60i, DPC T60i Kusti	ADVIA® 2400 Chemistry system.
Traceability/Standardization	The Alkaline Phosphatase (IFCC) Plus method is traceable to the molar absorbance coefficient of p-nitrophenol.	The ADVIA Alkaline Phosphatase (ALPAMP) method is traceable to the IFCC reference method via patient sample correlation.

K. Standard/Guidance Document Referenced (if applicable):
CLSI EP5-A: Evaluation of Precision Performance of Clinical Chemistry Devices; Approved Guideline. Vol. 19 No.2, 2/1999

CLSI EP6-A: Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline. Vol. 23 No. 16, 4/2003

CLSI EP7-A: Interference Testing in Clinical Chemistry; Approved Guideline. Vol. 22 No. 27, 12/2002

CLSI EP9-A: Method Comparison and Bias estimation Using Patient Samples; Approved Guideline. Vol. 15, No. 17, 12/1995

CLSI EP17-A: Protocols for Determination of Limits of Detection and Limits of Quantitation; Approved Guideline. Vol. 24 No. 34, 10/2004

L. Test Principle:
ALP catalyzes the hydrolysis of p-nitrophenylphosphate (p-NPP). The formation of p-nitrophenol in alkaline solution is followed at a wavelength of 405 nm.

M. Performance Characteristics (if/when applicable):
   1. Analytical performance:
      a. Precision/Reproducibility:
         The precision study was done during 20 days, with two runs per day, two replicates per run, 2 reagent lots, total 80 results per level, including 1 operator, 1 T60 instrument at one site, and 6 calibrations. An additional precision study to cover the lower and higher ends of the measuring range,

was done during 5 days, with two runs per day, two replicates per run, 1 reagent lot,1 instrument and 3 calibrations with the total number of measurements being 20.
Samples:
Three levels of analyte (full precision study, levels 2, 3 and 4).
Two levels of analyte (additional study, levels 1 and 5).
Low level: Normal human sera diluted with 0.9% NaCl
High level: Normal human sera spiked with commercially available ALP preparate.

Results:

Precision Study serum/ plasma application							
Sample	Mean	Within run		Between run		Total	
	U/L	SD	CV%	SD	CV%	SD	CV%
Level 1	41	0.5	1.1	1.4	3.5	1.5	3.7
Level 2	91	0.7	0.7	0.8	0.9	2.9	3.2
Level 3	127	0.9	0.7	1.1	0.9	2.6	2.0
Level 4	374	2.0	0.5	2.0	0.5	8.4	2.3
Level 5	810	6.6	0.8	2.5	0.3	18.5	2.3

b. *Linearity/assay reportable range:*
   Linearity was tested on serum samples. The samples were diluted in 11 steps by mixing samples with each other 1:1. For example, the 50% sample resulted from a 1:1 mixture of the 100% and 0% samples. Four parallel measurements were made in random order for each step using one reagent lot. Eleven samples tested ranging in concentration from 9.4 U/l to 1215 U/L.
   For the extended range linearity study another set of samples were prepared and measured as above with the exception that the samples S5 – S11 were run using an additional secondary dilution. Eleven samples were tested ranging in concentration from 48.8 U/l to 11329.7 U/L.
   Samples:
   Low level: serum pool diluted with 0.9% NaCl while for high level: serum pool was spiked with commercially available alkaline phosphatase of placental origin.
   Results Obtained:
   For Linearity (n=11): Slope = 0.988, Intercept = 0.3702 Obs. Err = 2.7%
   For Ext. Linearity (n=11) Slope = 0.975, Intercept = -1.26, Obs. Err = 5.1%

   The data supported the claimed measuring range of 20 – 1000 U/L, and extended measuring range 20 – 10 000 U/l.

c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*
   The Alkaline Phosphatase (IFCC) Plus method is traceable to the molar absorbance coefficient of p-nitrophenol.
   The sponsor's protocols indicate that calibrator (eCal) is traceable to the

reference procedure given in the value sheet, which is the IFCC reference procedure for 3-reagent method and molar absorbance coefficient of NADH for 1-reagent method. The controls (Nortrol and Abtrol) are traceable to the IFCC procedure. The values are lot-specific and are assigned based on multiple determinations performed using several T60 instruments. The assigned value is the median of all the values generated for each calibrator and control. Additionally, control range is calculated as the target value ± 2 standard deviations. Please refer to the value assignment sheet in the labeling for lot specific values.

To ensure adequate quality control, the sponsor recommends calibrating the test at least every three days and every time a new reagent bottle is used. The sponsor also recommends using quality control samples at least once a day, after each calibration and when a new bottle is used. The sponsor further recommends that all local, state and federal guidelines be followed.

The sponsor claims that all open on-board stability for reagents is 14 days. Reagent in unopened vial is stable at 2-8°C until expiration date on vial label when protected from light.

d. *Detection limit:*
In the Limit of Blank (LOB) study thirty replicates of a blank sample (0.9% NaCl) were run using two T60 instruments and two reagent lots with the total number of measurements being 60. The results support a claimed LOB of 1 U/L.
In the Limit of Detection (LOD) study five low level samples were run in ten replicates with two T60 instruments and two reagent lots during two days with the total number of measurements being 100. The results support a claimed LOD of 5 U/L.

e. *Analytical specificity:*
Interference Studies: For each interfering substance (except for Lipemia, Intralipid®) three parallel measurements of five interfering concentrations (including non-interfered sample) were run in ascending – descending – ascending order. To test the interference by lipemia (Intralipid®), both control and test pools were analyzed, with four replicates from each pool, within one analytical run. There was deemed to be no interference if the observed values were ± 10% or ± 7.5 U/L of initial value.
Lipemia: No interference found up to 1000 mg/dl (10 g/l) of Intralipid.
Hemolysate: No interference found up to 300 mg/dl (3 g/l) of hemoglobin
Bilirubin conjugated: No interference found up to 58 mg/dl (1000 µmol/l) of conjugated bilirubin
Bilirubin unconjugated: No interference found up to 21 mg/dl (360 µmol/l) of unconjugated bilirubin.

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

2. Comparison studies:
   a. *Method comparison with predicate device:*
   One hundred and fifty-nine Li-heparin plasma samples were analyzed using T60i instrument with Alkaline Phosphatase (IFCC) Plus reagents and Bayer Advia 2400 instrument with Alkaline Phosphatase (ALPAMP) reagents to demonstrate the equivalence of the two systems. The samples were split in two and then run either on the predicate or Thermo device. Each individual measurement from the device was compared to one individual measurement result of the predicate.
   Regression analysis results obtained from 148 samples ranging from 21 to 995 U/L were as follows: 95% confidence interval in parenthesis. The r value was 0.997.

	Deming	Regular
Slope	1.003 (0.990 to 1.016)	1.00 (0.987 to 1.013)
Intercept	1.46 (-2.52 to 5.44)	2.06 (-1.92 to 6.04)
Std. Error	19.01	18.99

   b. *Matrix comparison:*
   Fifty-five matched serum and plasma (Li-heparin) samples were run on T60 instrument to demonstrate the use of plasma (Li-heparin) samples. Both serum and plasma samples were run in duplicates but only the first replicate result was used for the calculations. Regression analysis results obtained from 55 samples ranging from 31 to 993 U/L were as follows: 95% confidence interval in parenthesis. The r value was 0.998.

	Deming	Regular
Slope	1.036 (1.018 to 1.055)	1.034 (1.015 to 1.052)
Intercept	-0.576 (-7.609 to 6.456)	-0.018 (-7.046 to 7.011)
Std. Error	19.721	19.709

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:

   Male: 53 - 128 U/l (0.9 - 2.18 µkat/l) at 37 °C
   Female: 42 - 98 U/l (0.71 - 1.67 µkat/l) at 37 °C

Source citation: Burtis, CA and Ashwood, E R (ed.), Tietz Fundamentals of Clinical Chemistry, 5th edition, W B Saunders Company, Philadelphia, 2001, pp. 366-367, 1002.

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.