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

A. 510(k) Number:

k162275

B. Purpose for Submission:

New device

C. Measurand:

Alkaline phosphatase

D. Type of Test:

Quantitative colorimetric

E. Applicant:

Randox Laboratories Ltd.

F. Proprietary and Established Names:

Randox RX Daytona Plus Alkaline Phosphatase (ALP)

G. Regulatory Information:

Product Code	Classification	Regulation Section	Panel
CJE	II	21 CFR 862.1050, Alkaline phosphatase or isoenzymes test system	Clinical Chemistry (75)

H. Intended Use:

1. Intended use(s):

   See Indications for Use below

2. Indication(s) for use:

   The Randox Daytona Plus Alkaline Phosphatase (ALP) test system is intended for the quantitative in vitro determination of Alkaline phosphatase (ALP) in serum and lithium heparinized plasma. Measurements of ALP are use in the diagnosis, treatment, and investigation of hepatobilary disease and in bone disease.

3. Special conditions for use statement(s):

   This device is intended for prescription use and in vitro diagnostic use only.

4. Special instrument requirements:

   RX Daytona Plus

I. Device Description:

The Randox ALP kit assay consists of two ready to use reagent solutions, R1 and R2.

R1 is a buffer solution containing 0.35 mol/L of 2-amino-2methy-l-propanol and 2.0 mmol/L of magnesium ions.
R2 is a substrate solution containing 10 mmol/L of p-nitrophenylphosphate.

J. Substantial Equivalence Information:

1. Predicate device name(s):

   Siemens Alkaline Phosphatase Assay

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

   k991576

3. Comparison with predicate:

Similarities and Differences		
Item	**Candidate Device** Randox RX Daytona Plus Alkaline Phosphatase (k162275)	**Predicate Device** Siemens Alkaline Phosphatase Assay (k991576)
Intended Use	For the quantitative in vitro determination of Alkaline Phosphatase activity in serum and plasma.	Same
Test Principle	Colorimetric	Same
Reagent Format	Liquid form, ready to use	Same

Similarities and Differences		
Item	**Candidate Device** Randox RX Daytona Plus Alkaline Phosphatase (k162275)	**Predicate Device** Siemens Alkaline Phosphatase Assay (k991576)
Sample Type	Serum, Lithium Heparin Plasma	Same
Assay Range	8 - 918 U/L	0 - 1100 U/L
Calibrator	Randox Calibration Serum Level 3	Fixed system factor value
Calibration frequency	A 2-point calibration is recommended every 7 days or with a change of reagent lot.	Not required

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

CLSI - EP05-A3, Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline-Third Edition.

CLSI - EP06-A, Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline.

CLSI - EP07-A2, Interference Testing in Clinical Chemistry; Approved Guideline-Second Edition.

CLSI - EP9-A3, Method Comparison and Bias Estimation Using Patient Samples; Approved Guideline-Third Edition.

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

CLSI - EP28-A3c, Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory; Approved Guideline –Third Edition.

L. Test Principle:

The substrate p-nitrophenyl phosphate is hydrolyzed by alkaline phosphatase from the sample, in the presence of magnesium ions, to form p-nitrophenol which is yellow in color and can be read at 405 nm. The intensity of color produced is proportional to the alkaline phosphatase activity in the sample.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Precision studies were conducted in accordance with CLSI EP05-A2. Precision studies were performed by two operators on two RX Daytona Plus systems using serum-based control material, altered human serum samples, and unaltered human serum samples. Testing was performed using two reagent lots of Randox RX Daytona Plus Alkaline Phosphatase (one on each analyzer), in two runs per day for 20 non-consecutive days. Two replicates per run were performed for each sample. Both lots yielded similar results. The results of one representative lot are summarized in the following table:

			Within-Run		Total	
Sample	N	ALP Mean (U/L)	SD	%CV	SD	%CV
QC1	80	127.83	0.5	0.4	2.87	2.2
QC2	80	328.94	1.19	0.4	6.53	2.0
QC3	80	353.25	1.26	0.4	7.61	2.2
Serum 1	80	81.44	1.56	1.9	1.89	2.3
Serum 2	80	212.76	3.35	1.6	3.44	1.6
Serum 3	80	244.36	3.63	1.5	3.77	1.5
Serum 4	80	349.39	4.41	1.3	5.31	1.5
Serum 5	80	24.86	0.80	3.2	1.31	5.3
Serum 6	80	480.71	3.65	0.8	15.26	3.2
Serum 7	80	706.24	4.87	0.7	24.48	3.5
Serum 8	80	895.00	4.49	0.5	30.79	3.4

b. Linearity/assay reportable range:

Linearity studies were performed in accordance with CLSI EP06-A. Low and high concentrated alkaline phosphatase pools were mixed to create 11 intermediate levels: 7.94, 99.93, 192.35, 293.09, 389.59, 481.23, 568.36, 658.86, 748.97, 839.03, and 918.42 U/L. Each level was run in replicates of five on two lots of Randox RX Plus Alkaline Phosphatase reagent on one RX Daytona Plus system. The following linear regression equation was obtained:

$y=1.00 x + 6.49$, $R= 1.00$

The results of the linearity study support the sponsor's claimed measuring range of 8 to 918 U/L.

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

Traceability: Refer to k053153 Randox Calibration Serum Level 3 Randox Assayed Multi-sera Level 2 and Level 3. Randox Calibration Serum Level 3 is traceable to ALP internal master reference calibrator.

Stability: Refer to k053153 Randox Calibration Serum Level 3 Randox Assayed Multi-sera Level 2 and Level 3.

d. *Detection limit:*

Detection limit studies were performed in accordance with CLSI EP17-A2. The limit of blank (LoB), limit of detection (LoD) and limit of quantification (LoQ) were determined using two lots of reagents on one RX Daytona Plus system. The LoB was determined by testing 60 samples of artificial serum diluent and was determined to be 0.14 U/L. The LoD for ALP is 1.0 U/L based on 240 determinations with 4 low level serum samples (60 runs per level). The LoQ was determined by running 4 low level serum samples. The sponsor defines LoQ as the lowest concentration which meets an imprecision of <10% bias. The LoQ was determined to be 7.5 U/L

LoB	LoD	LoQ
0.14 U/L	1.0 U/L	7.5 U/L

The claimed measuring range of the assay is 8 to 918 U/L.

e. *Analytical specificity:*

The effects of potential interferents were determined by calculating the mean value of the spiked interferent with the corresponding control sample. The spiked sample results were compared to control samples prepared without the potential interferents. The sponsor defines significant interference as ± ≤ 10% bias from the control result. The following substances were tested up to the levels indicated at alkaline phosphatase concentrations of 80 and 240 U/L:

Interferents	Highest level tested that demonstrated no significant interference (≤ 10% bias)
Hemoglobin	375 mg/dL
Total bilirubin	60 mg/dL
Conjugated bilirubin	60 mg/dL
Ascorbic acid	6 mg/dL
Intralipid	750 mg/dL
Triglycerides	2000 mg/dL

The sponsor includes the following limitations in the labeling:

"Use only non-hemolyzed serum"

"Use only heparinized plasma that is free from hemolysis."

f. Assay cut-off:

Not applicable.

2. Comparison studies:

a. Method comparison with predicate device:

Method comparison studies were performed in accordance with CLSI EP9-A2 to compare the candidate device to the predicate device, the Siemens Alkaline Phosphatase assay. A total of 106 serum patient samples spanning the range of 8 to 894 U/L were tested by one operator using one lot of reagent on one RX Daytona Plus analyzer. Less than 10% of the samples were altered by dilution or spiking in order to cover the claimed measuring range. The test method was compared to the predicate device and the following linear regression equation was obtained:

$y = 1.005 x - 3.95$, $r = 0.999$

b. Matrix comparison:

A matrix comparison study was performed on one RX Daytona plus system using two lots of Randox RX Daytona Plus Alkaline Phosphatase reagent. A total of 46 matched serum and lithium heparin plasma samples were analyzed spanning the result range of 15.3 to 773.1 U/L. The alkaline phosphatase results from the serum samples were compared to the results of the lithium heparin plasma samples and the following linear regression equation was obtained:

$y = 1.00 x - 0.13$, $r = 1.000$

The results of the matrix comparison study support the sponsor claim that serum and lithium heparin plasma samples can be tested with this assay.

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:

Reference intervals cited from literature were verified using the CLSI C28-A3 guideline. Human serum from 30 normal donors were tested in singlicate on the RX Daytona Plus. The sponsor claims that there were no outliers and that the values were compared to the cited ranges for alkaline phosphatase. Results of the study indicate that all values reported in range for healthy individuals. The sponsor states in the labeling that "Each laboratory should establish its own normal range based upon specific population encountered in daily laboratory operation. This is recommended because normal ranges are affected by age, sex, diet, geographical location and other factors."

Alkaline phosphatase reference ranges:
Adults: 30-120 U/L

Reference: Mosby's Manual of Diagnostic and Laboratory Tests. 3rd Edition. Pagana and Pagana, 2006, page 49.

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.