

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

A. 510(k) Number:

k102638

B. Purpose for Submission:

New device

C. Measurand:

Calibrator for Homeocysteine Assay

D. Type of Test:

Not Applicable

E. Applicant:

Diazyme Laboratories

F. Proprietary and Established Names:

Diazyme Homocysteine Assay Buffer Based Calibrators

G. Regulatory Information:

1. Regulation section:

21 CFR §862.1150, Calibrator

2. Classification:

Class II

3. Product code:

JIS – Calibrator, Primary

4. Panel:

Clinical Chemistry (75)

H. Intended Use:

1. Intended use(s):

See Indications for Use below.

2. Indications(s) for use:

Diazyme Homocysteine Assay Buffer Based Calibrators are intended for use with the Diazyme Homocysteine Assay Kit only. The calibrators are used to generate a calibration curve to be used in the calculation of homocysteine concentrations in unknown serum and plasma samples. For *in vitro* diagnostic use only.

3. Special conditions for use statement(s):

For prescription use only

4. Special instrument requirements:

Hitachi 917 and Olympus AU Systems

I. Device Description:

Diazyme Homocysteine Assay Buffer Based Calibrators are for use with the Diazyme Homocysteine Assay Kits to generate a calibration curve to calculate the concentration of homocysteine in unknown serum and plasma samples. It is supplied as a ready to use liquid with the active ingredient L-homocysteine, stabilizers and preservatives.

J. Substantial Equivalence Information:

1. Predicate device name(s):

Diazyme Homocysteine Calibrators

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

k042448

3. Comparison with predicate:

Reagent Similarities and Differences		
	Candidate Device Diazyme Homocysteine Assay Buffer Based Calibrators	Predicate Device Diazyme Homocysteine Calibrators (k042448)
Intended Use	Diazyme Homocysteine Assay	Diazyme Homocysteine Assay

Reagent Similarities and Differences		
	Candidate Device Diazyme Homocysteine Assay Buffer Based Calibrators	Predicate Device Diazyme Homocysteine Calibrators (k042448)
	Calibrator set is intended for use with the Diazyme Homocysteine Assay Kit only. The calibrators are used to generate a calibration curve to be used in the calculation of homocysteine concentrations in unknown serum and plasma samples. For <i>in vitro</i> diagnostic use only.	calibrator set is intended for use with the Diazyme Homocysteine Assay Kit only. For <i>in vitro</i> diagnostic use only.
Specimen Matrix	Human serum and plasma	same
Calibrator Ingredients	Human serum, saline preservatives and active ingredient L-homocystine	Aqueous buffered solution, preservative, and active ingredient L-homocystine
Linear Range	Up to 50 $\mu\text{mol/L}$ for both 2-reagent and 3-reagent HCY	same
Precision	3-reagent HCY, 2-point calibrators on Hitachi 917 Intra: 0.82-1.89% 3-reagent HCY, 5-point calibrators on Hitachi 917, Intra: 0.65-3.66% 2-reagent HCY 2 and 3-point calibrators on Olympus AU400 Intra: 1.54-4.35%	3-reagent HCY: Within: 1.8-3.0%
Accuracy	3-reagent HCY, 2-point calibrators on Hitachi 917: Correlation Coefficient: 0.9961 $y=1.0134x + 0.8266$ 3-reagent HCY, 5-point calibrators on Hitachi 917: Correlation Coefficient: 0.9971 $y=0.9357x + 0.5186$ 2-reagent HCY 2 and 3-point calibrators on Olympus AU400 Correlation Coefficient: 0.9927 $y=1.0167x - 0.0535$ 2-reagent HCY 2 and 5-point calibrators on Olympus AU400 Correlation Coefficient: 0.997	3-reagent HCY: Correlation Coefficient: 0.976 $y=0.98x + 0.87$

Reagent Similarities and Differences		
	Candidate Device Diazyme Homocysteine Assay Buffer Based Calibrators	Predicate Device Diazyme Homocysteine Calibrators (k042448)
	$y=0.9601x - 0.0345$	
Linear Range	3-reagent HCY: Up to 50 $\mu\text{mol/L}$ 2-reagent HCY: Up to 50 $\mu\text{mol/L}$	3-reagent HCY: Up to 50 $\mu\text{mol/L}$

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

- Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline - Second Edition - (CLSI EP5-A2).
- Method Comparison and Bias Estimation Using Patient Samples; Approved Guideline-Second Edition - (CLSI EP9-A2).
- Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline - (CLSI EP6-A).
- Interference Testing in Clinical Chemistry; Approved Guideline-Second Edition – (CLSI EP7-A2)

L. Test Principle:

Not applicable

M. Performance Characteristics (if/when applicable):1. Analytical performance:*a. Precision/Reproducibility:*

The sponsor performed intra precision studies with four serum based samples using the Homocysteine (HCY) Enzymatic Assay. The assay was performed within one day with 12 replicates. The acceptance criterion is precision $CV \leq 5\%$. The results are as follows:

Sample Type	Reagent	N	Instrument	Calibrator	Replicates	%CV
Serum	3-Reagent HCY	4	Hitachi 917	5-point Calibrator	12	0.65-3.66%

Serum	3-Reagent HCY	4	Hitachi 917	2-point calibrator	12	0.82-1.89%
Serum	2-Reagent HCY	4	Olympus AU 400	3-point calibrator	21	1.83-4.35%
Serum	2-Reagent HCY	4	Olympus AU 400	5-point calibrator	21	1.53-2.76%

b. Linearity/assay reportable range:

A linearity study was performed based on the CLSI EP6-A guideline. Eleven levels of the linearity set were prepared by diluting a serum sample containing 50 μM HCY with saline. A slope of 1.0 ± 0.1 and R^2 of ≥ 0.95 for expected versus obtained samples were the acceptance criteria. Results are summarized below:

Sample Type	Analyzer	Instrument	Calibrator	Slope	y-intercept	R^2
Serum	2-Reagent HCY	Olympus AU 400	3point calibrator	0.9978	0.7109	.9957
Serum	2-Reagent HCY	Olympus AU 400	5-point calibrator	1.0438	0.2341	.9982
Serum	3-Reagent HCY	Hitachi 917	2-point calibrator	0.9589	0.5811	.9878
Serum	3-Reagent HCY	Hitachi 917	5-point calibrator	1.048	0.2373	.9967

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

Traceability:

The calibrator value assignment includes testing with NIST Standard Reference Material 1995. The HCY calibrator is prepared by spiking a calculated amount of L-homocysteine stock solution to aqueous solution containing preservative to the target homocysteine concentration. The manufacturer then uses selected measurement methods to test and release the production calibrator on specified analyzer. The production calibrator with the final values is assigned and then used to test library samples, proficiency library samples and higher order trueness NIST SRM1955 serum controls.

Stability:

The calibrators are tested for shelf life protections with an accelerated stability protocol. The sponsor places the calibrators at each level in a 37°C incubator and at

the indicated time, test them with the current Diazyme HCY reagent and calibrators for HCY recovery. The acceptance criteria are that the calibrator should be stable for at least 12 months based on the stress test performance (7 days at 37°C and >1 month at 25°C) and the recovery should meet or exceed 90% of initial value. The study was performed on an Olympus AU400 using 2- HCY reagent and 5-point buffered calibrators. The calibrator did not change more than 10% after 14 days of 37°C stress, 60 days of 25°C in open or closed vial accelerated stability tests. The sponsor states that the calibrators are stable for 24 months when stored at 2-8°C based on Arrhenius theory stress models and real-time studies are on-going.

Expected Values:

The sponsor tested the NIST SRM1955 Controls for recovery. They had a within control range acceptance criteria of 15% or 2 µmol/L for NIST SRM1955 controls.

2-Reagent HCY: Results for Olympus AU400 using 2-reagent HCY reagent and 3-point buffer based calibrator

Sample	Value	Recovery	% Recovery
NIST 1	3.98	3.84	96.48%
NIST 2	8.85	9.23	10.29%
NIST 3	17.7	16.32	9.20%

2-Reagent HCY: Results for Olympus AU400 using 2-reagent HCY reagent and 5-point buffer based calibrator

Sample	Value	Recovery	% Recovery
NIST 1	3.98	3.91	98.24%
NIST 2	8.85	8.05	90.96%
NIST 3	17.7	16.14	91.19%

3-Reagent HCY: Results for Hitachi 917 using 3-reagent HCY reagent and 2-point buffer based calibrator

Sample	Value	Recovery	% Recovery
NIST 1	3.98	3.64	91.46%
NIST 2	8.85	9.905	111.92%
NIST 3	17.7	18.835	106.41%

3-Reagent HCY: Results for Hitachi 917 using 3-reagent HCY reagent and 5-point buffer based calibrator

Sample	Value	Recovery	% Recovery
NIST 1	3.98	3.69	92.71%
NIST 2	8.85	8.74	98.76%
NIST 3	17.7	17.87	100.96%

d. Detection limit:

Not applicable

e. Analytical specificity:

The sponsor performed interference studies by testing serum samples at 12 and 29 μM HCY spiked with interference substances to verify tolerance limits. Potential cross-reactive compounds (see table below) were added and tested using the Diazyme HCY Enzymatic Assay. The following substances present in the serum produced less than 10% deviation when tested at the stated concentrations.

Potential Cross-Reactive Agent	Spiking Concentration
Cysteine	1000 μM
Glutathione	500 μM
Cystathionine	100 μM
Hemoglobin	500 mg/dL
Unconjugated Bilirubin	40 mg/dL
Conjugated Bilirubin	40 mg/dL
Ascorbic Acid	100mM
Triglyceride	500 mg/dL

f. Assay cut-off:

Not Applicable

2. Comparison studies:

a. Method comparison with predicate device:

Accuracy

The sponsor performed accuracy studies with forty serum based samples using the Homocysteine (HCY) Enzymatic Assay and comparing the buffer based calibrators with the current serum based calibrators. The assay was performed within one day with 12 replicates and all samples met the acceptance criteria. The results were as follows:

Sample Type	Analyzer	N	Instrument	Calibrator	Slope	R²
Serum	2-Reagent HCY	40	Olympus AU 400	3-point calibrator	1.0167	.9927
Serum	2-Reagent HCY	40	Olympus AU 400	5-point calibrator	0.9601	.997
Serum	3-Reagent HCY	40	Hitachi 917	2-point calibrator	1.0134	.9961
Serum	3-Reagent HCY	40	Hitachi 917	5-point calibrator	0.9357	.9971

b. Matrix comparison:

Not applicable.

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

Not Applicable

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.