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

A. 510(k) Number:

k063701

B. Purpose for Submission:

New device

C. Measurand:

Homocysteine

D. Type of Test:

Quantitative, enzymatic assay

E. Applicant:

Teco Diagnostics

F. Proprietary and Established Names:

Teco Homocysteine Enzymatic Assay

G. Regulatory Information:

1. Regulation section:

   21 CFR 862.1377

2. Classification:

   Class II

3. Product code:

   LPS

4. Panel:

   75 (Chemistry)

H. Intended Use:

1. Intended use(s):

   Teco Enzymatic Homocysteine assay is intended for *in vitro* quantitative determination of total homocysteine in serum or plasma. Homocysteine measurements are used in the diagnostics and treatment of hyperhomocysteinemia.

   > Note: Specimens from patients who are on drug therapy involving S-adenosyl-methionine may show falsely elevated levels of homocysteine. Patients, who are taking methotrexate, carbamazepine, phenytoin, nitrous oxide, anticonvulsants or 6-azuridinetriacetate, may have elevated levels of homocysteine due to their effect on the pathway. Refer to limitations/interfering substance section in this assay package insert.

2. Indication(s) for use:

   Teco Enzymatic Homocysteine assay is intended for *in vitro* quantitative determination of total homocysteine in serum or plasma. Homocysteine measurements are used in the diagnostics and treatment of hyperhomocysteinemia.

3. Special conditions for use statement(s):

   Prescription use only

4. Special instrument requirements:

   Beckman Coulter CX7 or Teco Diagnostics TC 84 Spectrophotometer capable of reading wavelength at 340 nm

I. Device Description:

Teco Homocysteine IVD reagent kit includes the following reagents:

1. Reagent 1 contains Dithiothreitol 0.1 mmol/L (40mL or 60mL bottle).

2. Reagent 2 contains rHCYase 1000U/L (15mL or 20mL bottle).

3. Reagent 3 contains NADH- 1.16 mmol/L; Glutamate Dehydrogenase- 15 KU/L; a-ketoglutarate- 6.0 mmol/L; Pyridoxal-5-phosphate- 20µmol/L (7mL or 20mL bottle).

J. Substantial Equivalence Information:

1. Predicate device name(s):

   Catch Inc. Homogeneous Enzymatic Homocysteine Reagent

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

   k011689

3. Comparison with predicate:

Similarities and Differences		
Item	Teco HCY reagent (candidate device)	Catch HCY reagent (predicate device)
Intended Use	Quantitative *in vitro* diagnostic determination of total L-homocysteine for diagnosis and treatment of hyperhomocysteinemia and homocystinuria.	Quantitative *in vitro* diagnostic determination of total L-homocysteine for diagnosis and treatment of hyperhomocysteinemia and homocystinuria.
Methodology	Enzymatic	Enzymatic
Analyzer	Auto chemistry analyzer/manual	Auto chemistry analyzer
Calibrator	Not provided	Two levels: 1) A: Phosphate buffer 2) B: Gravimetrically prepared S-adenosyl-L-homocysteine (SAH) in phosphate buffer at defined concentrations.
Controls	Not provided	Three levels: L-homocysteine in processed human serum with sodium azide as preservative.
Sample types	Serum, EDTA or Heparin Plasma	Serum, EDTA or Heparin Plasma
Detection Limits	0.5 µmol/L on Synchron CX 7 analyzer, 0.6 µmol/L on TC84	0.54 µmol/L
Reportable range	2.0 to 50.0 µmol/L on Synchron CX 7 analyzer and TC84 analyzer	0.5 to 50 µmol/L

Similarities and Differences		
Item	Teco HCY reagent (candidate device)	Catch HCY reagent (predicate device)
Normal range	3.7-14.6 µmol/L on Synchron CX 7 analyzer	4.0-15.4 µmol/L

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

STANDARDS
Title and Reference Number

1. CLSI Guideline, EP5-A *Evaluation of Precision Performance of Clinical Chemistry Devices*.
2. CLSI Guideline, EP6-A *Evaluation of Linearity of Quantitative Analytical Methods.*
3. CLSI Guideline, EP7-A *Interference Testing in Clinical Chemistry; Approved Guideline*.
4. CLSI Guideline, EP9-A *Method Comparison and Bias Estimation Using Patient Samples.*

L. Test Principle:

The principle of the assay is tHCY being degraded by a recombinant homocysteinase (rHCYase) producing hydrogen sulfide (H_2S), a-ketobutyate and ammonia. The liberated ammonia reacts with a-ketobutyate, in the presence of Glutamate Dehydrogenase and the coenzyme NADH are oxidized to NAD for each mole of urea hydrolyzed. The resulting decrease in absorbance of NADH at 340 nm is proportional to the level of homocysteine in the sample.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

   a. Precision/Reproducibility:

   A precision study was performed using two levels of commercial controls. Samples were run 25 times per day on Synchron CX7 and spectrophotometer TC 84 for within-run precision and five times per day for 5 days for run-to-run precision. Precision study was performed following a modification of CLSI EP5-A. The sponsor's acceptance criteria are ≤ 10 % CV for within-run and run-to-run precision. Mean, SD, and CVs were calculated for within-run and run-to-run precision and are shown in the tables below:

Within-run precision on Synchron CX 7:

	Mean (µmol/L)	SD (µmol/L)	CV% (Within run)
Control Level 1	13	1.05	8.0
Control Level 2	45	1.9	4.0

Within-run precision on TC84:

	Mean (µmol/L)	SD (µmol/L)	CV% (Within run)
Control Level 1	13.2	1.2	9.0
Control Level 2	45.4	1.8	7.0

Run-to-run precision on Synchron CX 7:

	Mean (µmol/L)	SD (µmol/L)	CV% (Run-to-run)
Control Level 1	12.8	1.16	9.0
Control Level 2	45.6	1.15	2.5

Run-to-run precision on TC84:

	Mean (µmol/L)	SD (µmol/L)	CV% (Run-to-run)
Control Level 1	12.8	0.9	7.0
Control Level 2	45.3	1.4	3.0

b. Linearity/assay reportable range:

A linearity study was performed using a commercially available linearity assay kit with 5 different concentrations (2 µmol/L, 7 µmol/L, 12.5 µmol/L, 29.5 µmol/L, and 50 µmol/L). Linearity studies were designed using CLSI EP6-A. Tests were performed on the Synchron CX7 analyzer and on Spectophotometer TC84. Each concentration was tested three times to determine the mean concentration. Values were plotted for the expected concentrations (X) versus the observed concentrations (Y) and an appropriate line fitted by standard linear regressions was calculated as follows:

For Synchron CX 7 analyzer: Y=1.02X + 0.11, r = 0.997

For TC84: Y= 0.96X + 0.94, r = 0.999

The data provided supports the sponsor's claim that the assay has a linearity/reportable range of 2.0- 50.0 µmol/L.

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

No traceability information for the assay was provided. Commercially available calibrator materials (Catch Inc.) are specified in the labeling but not supplied with the assay kit. Calibrators from Catch Inc. were cleared under k011689.

Commercial control materials are required but are not specifically identified in the labeling. In the labeling the sponsor recommends using assayed control material with values for homocysteine in both the normal and abnormal ranges in the labeling according to local, state and federal guidelines.

d. *Detection limit:*

The detection limits were established by reading the change from a saline sample and a known concentration sample on the Synchron CX 7 analyzer and the TC84 spectrophotometer. The sensitivity is defined as the concentration that can be distinguished from zero with 95% confidence. The sponsor obtained the sensitivity for the Synchron CX7 analyzer at 0.5 µmol/L and for the TC84 at 0.6 µmol/L.

e. *Analytical specificity:*

Interference studies were designed using CLSI EP7-A. Interference studies were performed on the Synchron CX 7 analyzer using random samples spiked with the following interferants and tested in triplicate. No significant interference was defined as the observed value within ± 2 µmol/L or ± 10% of the control value. The results obtained indicated that there was no significant interference by the following interferents:

- Hemoglobin up to 1500 mg/dL
- Bilirubin up to 30 mg/dL
- Triglycerides up to 1000 mg/dL
- Glutathione, Methionine, Cysteine, and Pyruvate up to 1000 µmol/L
- S-Adenosyl-L-methionine up to 0.5 mmol /L
- L-Cystathionine up to 2 mmol/L
- Adenosine up to 1 mmol/L
- DL-Homocysteine-thiolactone up to 0.1 mmol/L

The applicant stated the following limitation in the package insert:

"Specimens from patients who are on drug therapy involving S-adenosy-

methionine may show falsely elevated levels of homcysteine. Patients who are taking methotrexate, carbamazepine, phenytoin, nitrous oxide, anticonvulsants, or 6-azuridine triacetate, may have elevated levels of homocysteine due to their effect on the pathway. Refer to the work of Young for a review of drug effects on plasma and serum homocysteine levels."

f. Assay cut-off:

Not applicable

2. Comparison studies:

a. Method comparison with predicate device:

A method comparison study was conducted using 50 paired serum and EDTA plasma samples based on the CLSI EP-9A document. The sponsor used the recommended calibrators (from Catch Inc.) for the method comparison study between the Teco HCY assay (candidate device) and the Catch HCY assay (predicate device). In order to cover the entire analytical range, 4 samples were either diluted or spiked. Testing was performed using the Synchron CX 7 Analyzer verses the Catch Inc. HCY assay.

The results obtained yielded a correlation coefficient and a regression equation as follows:

For serum samples: $Y= 0.99X + 1.03$, $r = 0.98$, samples ranged in Homocysteine values from 3.0 - 48.0 µmol/L.

For EDTA plasma samples: $Y= 0.99X + 0.7$, $r = 0.98$, samples ranged in Homocysteine values from 3.0 - 46.0 µmol/L.

(Y = Teco HCY assay, X = Catch HCY assay)

b. Matrix comparison:

A matrix comparison study was conducted using 10 paired serum, EDTA plasma, and heparinized plasma samples. Testing was performed using the Teco HCY assay on the Synchron CX 7 analyzer. Both plasma samples homocysteine values were plotted against the serum homocysteine values. The acceptance criteria are: Slope = 0.97 – 1.1, $r > 0.95$. Correlation coefficient and a regression equation were calculated as follows:

For EDTA plasma samples: $Y= 0.98X + 0.06$, $r = 0.99$, serum samples ranged in Homocysteine values from 5.0 - 32.0 µmol/L.

For heparinized plasma samples: $Y= 0.97X + 0.5$, $r = 0.96$, serum samples ranged in Homocysteine values from 5.0 - 32.0 µmol/L.

(Y = plasma values, X = serum values)

The sponsor claimed that EDTA plasma and heparinized plasma samples are

recommended for the homocysteine determination. Serum collected in serum separator tubes may also be used.

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:

   A reference range study was conducted using samples from 125 apparently healthy adults (ages from 18 -80) in U.S. Plasma (EDTA) samples were assayed for total homocysteine by the Teco HCY assay on the Synchron CX 7 analyzer. The sponsor established the reference range by using the observed values for males as 5.2 to 14.8 µmol/L and for females as 5.1 to 15.0 µmol/L.

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.