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

A. 510(k) Number:

k063206

B. Purpose for Submission:

New device

C. Measurand:

Homocysteine

D. Type of Test:

Quantitative, competitive immunoassay using particle-enhanced nephelometry

E. Applicant:

Dade Behring, Inc.

F. Proprietary and Established Names:

Dimension Vista® HCYS Flex® reagent cartridge, Dimension Vista® Protein 1 Calibrator, Dimension Vista® Protein 1 Control L, M, H.

G. Regulatory Information:

Product Code	Classification	Regulation Section	Panel
Urinary Homocysteine (Nonquantitative) Test System (LPS)	Class II	21 CFR 862.1377, Urinary homocysteine (nonquantitative) test system.	75 Clinical Chemistry (CH)
Product Code	**Classification**	**Regulation Section**	**Panel**
Calibrators, Multi-Analyte Mixture (JIX)	Class II	21 CFR 862.1150, Calibrator	75 Clinical Chemistry (CH)
Product Code	**Classification**	**Regulation Section**	**Panel**
Multi-Analyte controls, All kinds (Assayed and Unassayed) (JJY)	Class I	21 CFR 862.1660, Quality Control Material (assayed and unassayed)	75 Clinical Chemistry (CH)

H. Intended Use:

1. Intended use(s):

   See Indications for use below.

2. Indication(s) for use:

   Dimension Vista® HCYS Flex® reagent cartridge: The HCYS method is an *in vitro* diagnostic test for the quantitative determination of total homocysteine in human serum, heparinized and EDTA plasma. Measurements of homocysteine aid in the diagnosis and treatment of patients suspected of having hyperhomocysteinemia and homocystinuria.

   Dimension Vista® Protein 1 Calibrator: PROT1 CAL is an *in vitro* diagnostic product for the calibration of the C3 Complement (C3), C4 Complement (C4), Homocysteine (HCYS), Immunoglobulin A (IGA), Immunoglobulin G (IGG), Immunoglobulin M (IGM) and prealbumin/Transthyretin (PREALB) methods on the Dimension Vista® System.

   Dimension Vista® Protein 1 Control L, M, H: PROT1 CON L, M, H are assayed intra-laboratory quality controls for assessment of precision and analytical bias in the determination of C3 Complement (C3), C4 Complement (C4), Homocysteine (HCYS), Immunoglobulin A (IGA), Immunoglobulin G (IGG), Immunoglobulin M (IGM) and prealbumin/Transthyretin (PREALB) methods on the Dimension Vista® System.

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

4. Special instrument requirements:

   For use on the Dimension Vista® System only.

I. Device Description:

1. Dimension Vista® HCYS Flex® reagent cartridge contains 12 wells with different kinds of reagents: Wells 1-4 contain diluent, wells 5-6 contain HCYS Reduction reagents (0.35 g/L Dithiothreitol and 0.0075 g/L Adenosine), wells 7-8 contain HCYS supplement reagent 1 (0.02 g/L conjugate of S-Adenosyl-Cysteine/Porcine Thyreoglobulin), wells 9-10 contain HCYS supplement reagent 2 (8,100 Units/L recombinant S-Adenosyl-L-Homocysteine hydrolase), wells 11-12 contain 1.25 g/L of polystyrene particles and 0.038 g/L mouse monoclonal antibodies to S-Adenosyl-Homocysteine).

2. Dimension Vista® Protein 1 Calibrator is a multi-analyte, liquid, human serum based product containing C3, C4, Homocysteine, IGA, IGG, IGM, and prealbumin/Transthyretin (PREALB). Each carton contains 6 vials with 6 different concentrations of analytes, each vial contains 2.0 mL of ready to use material.

3. Dimension Vista® Protein 1 Control L(low), M(medium), H(high) are multi-analyte, liquid, human serum based products containing C3, C4, Homocysteine, IGA, IGG, IGM, and prealbumin/Transthyretin (PREALB). Each carton contains 6 vials with 2.0 mL of ready to use material per vial.

All human source materials were tested and found to be negative for syphilis, HIV 1/2, HBsAg, and HCV.

J. Substantial Equivalence Information:

1. Predicate device name(s):
   Dade Behring N Latex HCY
   Dade Behring N Protein Standard SL
   Dade Behring N/T Protein Control SL

2. Predicate 510(k) number(s):
   k052788, k012470, k01268

3. Comparison with predicate:

Dimension Vista® HCYS Flex® reagent cartridge:

Similarities		
Item	Device	Predicate
Intended Use	Quantitative *in vitro* diagnostic determination of total homocysteine for diagnosis and treatment of hyperhomocysteinemia and homocystinuria.	Quantitative *in vitro* diagnostic determination of total homocysteine for diagnosis and treatment of hyperhomocysteinemia and homocystinuria.
Methodology	Particle Enhanced nephelometry	Particle Enhanced nephelometry
Standardization	Traceable to purified S-adenosyl-homocysteine.	Traceable to purified S-adenosyl-homocysteine.
Antibody	Mouse monoclonal	Mouse monoclonal
Sample types	Serum or Plasma	Serum or Plasma

Differences		
Item	Device	Predicate
Instrument	Dade Behring Dimension Vista System	Dade Behring BN II and BN ProSpec System
Detection Limits	0.2 µmol/L	2.0 µmol/L
Reportable range	2.0 to 57 µmol/L	2.0 to 64 µmol/L
Normal range	5.0 - 15.0 µmol/L From scientific literatures	3.2 - 10.7 µmol/L From scientific literatures

Dimension Vista® Protein 1 Calibrator:

Similarities and differences		
Item	Device	Predicate
Form	Liquid, human serum	Liquid, human serum
Constituents	C3, C4, homocysteine, IGA, IGG, IGM and prealbumin/Transthyretin	IgG, IgG1, IgG2, IgG3, IgG4, IgA, homocysteine, IgE, C3,C4, transferrin, albumin, α1-antitrypsin, α2-macroglobulin, haptoglobin, α1-acid glycoprotein, prealbumin, hemopexin, ceruloplasmin, retinal binding protein, Ig light-chain kappa, Ig light-chain lambda, soluble transferring receptor, ferritin, β2-microglobulin, total protein
Traceable	Traceable to purified S-adenosyl-homocysteine.	Traceable to purified S-adenosyl-homocysteine.

Dimension Vista® Protein 1 Control L, M, H:

Similarities and differences		
Item	Device	Predicate
Form	Liquid, human serum	Liquid, human serum
Constituents	C3, C4, homocysteine, IGA, IGG, IGM and prealbumin/Transthyretin	IgG, IgG1, IgG2, IgG3, IgG4, IgA, homocysteine, IgE, C3,C4, transferrin, albumin, α1-antitrypsin, α2-macroglobulin, haptoglobin, α1-acid glycoprotein, prealbumin, hemopexin, ceruloplasmin, retinal binding protein, Ig light-chain kappa, Ig light-chain lambda, soluble transferring receptor, ferritin, β2-microglobulin, total protein
Traceable	Purified S-adenosyl-homocysteine.	Purified S-adenosyl-homocysteine.

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

STANDARDS
Title and Reference Number

CSLI Guideline, EP5-A2 *Evaluation of Precision Performance of Clinical Chemistry Devices*; *Approved Guideline Second edition*
CSLI Guideline, EP7-A2 *Interference Testing in Clinical Chemistry; Approved Guideline Second edition*
CSLI Guideline, EP9-A2 *Method Comparison and Bias Estimation Using Patient Samples; Approved Guideline Second edition*

L. Test Principle:

Bound homocysteine in the sample is reduced to free homocysteine by the action of dithiothreitol, and converted enzymatically to S-adenosyl-homocysteine (SAH) in the next step. Conjugated S-adenosyl-cysteine (SAC) added at the onset of the reaction competes with the SAH in the sample for bonding by anti-SAH antibodies bound to polystyrene particles. In the presence of SAH there is either no aggregation or a weaker aggregation of the polystyrene particles. In the absence of SAH in the sample an aggregation of the polystyrene particles by conjugated SAC occurs. The higher the SAH content of the reaction mixture, the smaller the scattered light signal. The result is evaluated by comparison with a standard of known concentration.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Precision was evaluated with quality control materials (L, M, H), a plasma pool and two serum pool on the Dimension Vista System following CLSI EP5-A2 guideline. Samples were analyzed in duplicate, twice a day, for 20 days. The repeatability and within-lab SD and % CV were calculated by the analysis of variance method. The data are summarized in the table below:

Material	Mean µmol/L	Repeatability SD (%CV)	Within-Lab SD (%CV)
PROT1 CON L	8.90	0.297 (3.3)	0.618 (7.0)
PROT1 CON M	14.06	0.442 (3.2)	1.014 (7.2)
PROT1 CON H	28.63	0.829 (2.9)	1.833 (6.4)
Serum pool	1.26	0.332 (3.0)	0.749 (6.7)
Serum pool	53.24	1.034 (1.9)	1.447 (6.7)
Plasma pool	5.42	0.104 (1.9)	0.445 (8.2)

b. *Linearity/assay reportable range:*

Linearity across the assay range was determined by testing a calibrator and a high patient serum with a high concentration of homocysteine. The samples were serially diluted with system diluent and each dilution was tested with replicates of three. Data were analyzed using linear regression analysis. The acceptance criteria of slope between 0.9 and 1.1 and correlation coefficient ≥ 0.95 was met for the study.

The reportable range for the assay is 2 – 57 µmol/L.

The sponsor recommends a dilution of 1:10 when patient result falls outside the upper measuring range of 57µmol/L. A dilution study for homocysteine was performed on 8 different patient serum spiked with homocysteine on the Dimension vista system. Each sample was then diluted with system diluent at 1:10 dilution by the analyzer or manually. Each diluted sample was run in replicates of 5. The % recovery of the Dimension vista system for homocysteine assay ranged from 91% to 100.7 % with a mean % recovery of 94%.

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

Traceability and value assignments:

The PROT1 calibrators and PROT1 controls are calibrated against an internal master calibrator, which is calibrated against a commercially available reference preparation. Specifically, homocysteine is calibrated against a commercially available preparation of purified S-adenosyl-homocysteine. The value assignment process for the standard and control are by an internal protocol using two different instruments and by performing multiple determinations using the Dimension Vista Systems and the Dimension HCYS Reagent. The PROT1 calibrators and PROT1 controls are traceable to purified S-adenosyl-homocysteine.

Stability:

To support a 24 month shelf-life for unopened Dimension Vista HCYS flex reagent, PROT1 calibrators and PROT1 controls store at 2-8^{o}C, testing was done on day 0 and after 12, 18, and 24 months. The acceptance criteria are: results obtained must be within a range of 85% to 115% of the control assigned value.

To support an open-vial stability at 2-8^{o}C for the Dimension Vista HCYS flex reagent, PROT1 calibrators and PROT1 controls, vials are stored on board the instrument and contents are tested in duplicates on day 0, 4, 7, 9, 11, 14. The acceptance criteria are: results obtained must not deviate more than ± 10% compared to day 0 results. The sponsor claims the on-board stability of 9 days for the calibrators and controls once the vial is opened. The open well stability of the HCYS Flex reagent cartridge is 21 days for wells 1-12. Sealed wells on the instrument are stable for 30 days.

d. Detection limit:

The analytical sensitivity is defined as the minimal detectable level of analyte, which can be distinguished from zero. The value was calculated as the mean value of twenty replicates of system diluent plus two standard deviations. The sponsor claims the analytical sensitivity to be 0.2 µmol/L.

The reportable range for the assay is 2 – 57 µmol/L.

e. Analytical specificity:

The HCYS method was evaluated for interference according to CLSI/NCCLS EP7-A2. Bias is the difference in the results between the control sample (without the interferent) and the test sample (contains the interferent) expressed in percent. Bias exceeding 10% is considered interference.

Substance tested	**Substance Concentration**	**S. I. Units**	**Homocysteine Concentration µmol/L**	**Bias* %**
Hemoglobin (hemolysate)	1000 mg/dL	0.62 mmol/L	13.22	±0
Bilirubin (unconjugated)	60 mg/dL	1026 µmol/L	12.54	±0
Bilirubin (conjugated)	60 mg/dL	1026 µmol/L	13.28	+6
Lipemia (Triglycerides)	510 mg/dL	5.7 mmol/L	11.83	-7

*** Analyte results should not be corrected based on this bias.**

Non Interfering Substances
The following substances do not interfere with the HCYS method when present in serum and plasma at the concentrations indicated. Inaccuracies (biases) due to these substances are less than 10% at homocysteine concentrations of 3.50 µmol/L to 28.88 µmol/L.

Substance	**Test Concentration**	**S. I. Units**
Acetaminophen	20 mg/dL	1328 µmol/L
Adenosine	27 mg/dL	0.01 mmol/dL
Amikacin	15 mg/dL	256 µmol/L
Aminophyllin	4 mg/dL	0.0095 mmol/dL
Ammonium heparin	3 U/mL	3000 U/L

Ampicillin	5.3 mg/dL	152 µmol/L
Ascorbic acid	5 mg/dL	227 µmol/L
Caffeine	6 mg/dL	308 µmol/L
Carbamazepine	3 mg/dL	127 µmol/L
Chloramphenicol	5 mg/dL	155 µmol/L
Chlordiazepoxide	1 mg/dL	33.3 µmol/L
Chlorpromazine	0.2 mg/dL	6.27 µmol/L
Cholesterol	500 mg/dL	12.9 mmol/L
Cimetidine	2 mg/dL	79.2 µmol/L
Creatinine	30 mg/dL	2652 µmol/L
Dextran 40	6000 mg/dL	1500 µmol/L
Diazepam	0.5 mg/dL	17.6 µmol/L
Digoxin	5 ng/mL	6.15 nmol/L
Erythromycin	6 mg/dL	81.6 µmol/L
Ethanol	400 mg/dL	86.8 mmol/L
Ethosuximide	25 mg/dL	1770 µmol/L
Furosemide	6 mg/dL	181 µmol/L
Gentamicin	12 mg/dL	251 µmol/L
Ibuprofen	50 mg/dL	2425 µmol/L
Immunoglobulin G (IgG)	5 g/dL	50 g/L
L-Cystathion	2.23 mg/dL	0.25 mmol/dL
L-Cysteine	30 mg/dL	0.01 mmol/dL
L-Glutathione	2.142 mg/dL	3.5 mmol/dL
L-Homocysteine-thiolactone	0.0775 mg/dL	0.5 µmol/dL
Lidocaine	1.2 mg/dL	51.2 µmol/L
Lithium chloride	2.3 mg/dL	3.2 mmol/L
Lithium heparin	3 U/mL	3000 U/L
L-Methionine	6 mg/dL	0.04 mmol/dL
Nicotine	0.1 mg/dL	6.2 µmol/L
Penicillin G	25 U/mL	25000 U/L
Pentobarbital	8 mg/dL	354 µmol/L
Phenobarbital	10 mg/dL	431 µmol/L
Phenytoin	5 mg/dL	198 µmol/L
Primidone	4 mg/dL	183 µmol/L
Propoxyphene	0.2 mg/dL	4.91 µmol/L
Protein Albumin	0.6 g/dL	6 g/L
Protein Total	12 g/dL	120g/L
Rheumatoid Factor	500 U/mL	500 U/mL
S-Adenosyl-L-methionine	20 mg/dL	0.05 mmol/dL
Salicylic acid	60 mg/dL	4.34 mmol/L
Sodium heparin	3 U/mL	3000 U/L
Urea	500 mg/dL	83.3 mmol/L
Uric acid	20 mg/dL	1190 µmol/L
Valproic acid	50 mg/dL	3467 µmol/L

A serum sample above the assay range was analyzed on both the BN ProSpec System and the Dimension Vista System to show that the HCYS assay has no hook effect with concentration up to 199.3 µmol/L.

As limitations, the applicant stated the followings in the package insert:

1. Specimens from patients who are on drug therapy involving S-adenosy-methionine may show falsely elevated levels of homocysteine. Certain drugs, such as antiepileptic, antifolates, nitrous oxide anesthesia and antagonists of vitamin B6 are known to elevate the homocysteine concentration in human blood.

2. Patient samples may contain heterophilic antibodies that could react in immunoassays to give falsely elevated or depressed results. This assay has been designed to minimize interference from heterophilic antibodies. Nevertheless, complete elimination of this interference from all patient specimens cannot be guaranteed.

f. Assay cut-off:

None

2. Comparison studies:

a. Method comparison with predicate device:

A method comparison study was performed on 215 matched samples of serum, EDTA plasma, and heparized plasma based on the CLSI EP9-A2 guideline on the Dimension Vista system and the BN ProSpec system. Samples concentration range from 3.43 to 56.52 µmol/L. The method used to fit the linear regression line was Passing Bablok. Regression analysis of these results yielded the following equation: Y = 1.056 X + 0.239, r= 0.995, N=215. Y = Dimension Vista HCYS assay, and X = BN ProSpec N Latex HCY assay.

A summary of the regression statistics for the different matrices is provided below:

Method Comparison Study

Comparative Method/Specimen type	n	Slope	Intercept	Correlation Coefficient
HCY on the BN ProSpec®/EDTA plasma	73	1.076	0.350	0.996
HCY on the BN ProSpec®/ lithium heparin plasma	70	1.075	-0.085	0.997
HCY on the BN ProSpec®/serum	72	1.024	0.716	0.996

b. Matrix comparison:

Matrix comparison studies were performed by collecting fresh whole blood from a total of 10 donors. Samples were collected into lithium-heparin tube, sodium-heparin tube, EDTA tube, and serum separator tubes. A comparison was performed with matched specimens of serum, EDTA, and lithium and sodium heparin plasma on the Dimension Vista system. Linear regression analysis showed correlations of slopes of 0.9 to 1.10 and $r \geq 0.95$ was met when different matrices were compare to the serum sample.

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):

None

4. Clinical cut-off:

None

5. Expected values/Reference range:
The reference interval is based on a report from the NHANES 1999-2000 study with approximately 7300 participants, which yielded a central 90% reference range of 3.2 – 10.7 µmol/L for vitamin replete adult US males and females.

Literature cited: "Pfeiffer CM et al. Biochemical indicators of B vitamin status in the US population after folic acid fortification: results from the National Health and Nutrition Examination Survey 1999-2000. Am J Clin Nutr 2005;82(2):442-450."

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.