510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K223590

B Applicant

Beckman Coulter, Inc.

C Proprietary and Established Names

Access Folate Assay

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
CGN	Class II	21 CFR 862.1295 - Folic Acid Test System	CH - Clinical Chemistry

II Submission/Device Overview:

A Purpose for Submission:

Modified device

B Measurand:

Folic acid

C Type of Test:

Quantitative; chemiluminescent immunoassay

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

The Access Folate assay is a paramagnetic particle, chemiluminescent immunoassay for the quantitative determination of folic acid levels in human serum, lithium heparin plasma, and red blood cells using the Access Immunoassay Systems.

Folic acid measurements are used in the diagnosis and treatment of megaloblastic anemia.

Folate levels in serum, lithium heparin plasma, and red blood cells are used to assess folate status. The serum folate levels is an indicator of recent folate intake. A low RBC folate value can indicate a prolonged folate deficiency.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

For in vitro diagnostic use

D Special Instrument Requirements:

DxI 9000 Access Immunoassay Analyzer

IV Device/System Characteristics:

A Device Description:

The Access Folate assay consists of the reagent pack and calibrators. Reagent pack is provided in two assay packs containing 50 tests per pack for 100 tests. The Access Folate reagent pack is consists of the following reagents:

- R1a: Mouse monoclonal anti-folate binding protein, paramagnetic particles coated with goat anti-mouse IgG, buffer, human serum albumin (HSA) and 0.1% ProClin* 300.
- R1b: 1.0M Ascorbate, 0.05N HCl, pH 5.5
- R1c: Milk folate binding protein (bovine) in buffer, HSA and 0.1% ProClin 300.
- R1d: Folic acid alkaline phosphate (bovine) conjugate buffer, HSA and 0.1% ProClin 300.
- R1e: 0.6M K_3PO4

Other materials needed to run the assay, but not included in the kit include: Access Folate Calibrators, Quality Control (QC) materials, Lumi-Phos PRO, UniCel DxI Wash Buffer II, Access Red Blood Cell Folate Lysing Agent, and Access Wash Buffer II.

B Principle of Operation:

The Access Folate assay is a competitive binding immunoenzymatic assay. For the assay of folate in serum or lithium heparin plasma, no pre-treatment is required. For the assay of folate in red blood cells, a whole blood sample is first treated off-line with a lysing agent composed of ascorbic acid. This pre-treatment hemolyzes the red blood cells and converts the folate to polyglutamic acid forms present in red cells to the monoglutamic acid form predominant in serum. The sample from the pre-treatment of whole blood is defined as hemolysate.

Fifty-five (55) µL of sample (105 µL when using the automated dilution feature) is added to a reaction vessel with folate binding protein, mouse anti-folate binding protein, folic acid-alkaline phosphatase conjugate, and paramagnetic particles coupled with goat anti-mouse capture antibody. Folate in the sample competes with the folic acid-alkaline phosphatase conjugate for binding sites on a limited amount of folate binding protein. Resulting complexes bind to the solid phase via mouse anti-folate binding protein. After incubation, materials bound to the solid phase are held in a magnetic field while unbound materials are washed away. Then, the chemiluminescent substrate is added to the vessel and light generated by the reaction is measured with a luminometer. The light production is inversely proportional to the concentration of analyte in the sample. The amount of analyte in the sample is determined from a stored, multi-point calibration curve.

Red blood cell (RBC) folate is calculated by multiplying the RBC folate hemolysate result by 21 to correct for the 1:21 dilution that was made during preparation of the hemolysate, and then dividing the result by the patient's hematocrit.

V Substantial Equivalence Information:

A Predicate Device Name(s):
Access Folate on the Access Immunoassay Systems

B Predicate 510(k) Number(s):
K060774

C Comparison with Predicate(s):

Device & Predicate Device(s):	K223590	K060774
Device Trade Name	Access Folate	Access Folate on the Access Immunoassay System
General Device Characteristic Similarities		
Intended Use	For the quantitative determination of folic acid levels in human serum, plasma (heparin), and red	Same

Device & Predicate Device(s):	K223590	K060774
	blood cells	
Assay Technology	Chemiluminescent immunoassay	Same
General Device Characteristic Differences		
Substrate	Lumi-Phos PRO	Access Substrate
Instrument	DxI 9000 Access Immunoassay Analyzer	Access Immunoassay System
Measuring interval	2.0 - 24.8 ng/mL	0.5 - 20 ng/mL (serum)

VI Standards/Guidance Documents Referenced:

CLSI EP05-A3: Evaluation of Precision of Qualitative Measurement Methods Procedures; Approved Guideline – Third Edition

CLSI EP06-Ed2: Evaluation of the Linearity of Quantitative Measurement Procedures – 2nd Edition

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

CLSI EP09c-A3, Method Procedure Comparison and Bias Estimation Using Patient Samples; Approved Guideline – Third Edition

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:
   Precision studies were conducted following the recommendations in the CLSI EP05-A3 guideline.

   Serum: To estimate repeatability and within-laboratory imprecision, five (5) levels of serum were tested using 3 lots of reagent, 3 lots of calibrator, and 3 DxI 9000 Immunoassay analyzers. Each sample was assayed in duplicate, in two runs per day, over 22 days for a total of 44 runs and 88 replicates per instrument. Each instrument was evaluated with one reagent lot. Within-run, between-run, between-day, and total imprecision was calculated. Results from multiple lots were similar. Results from one representative lot are provided in the table below:

Serum										
Folate (ng/mL)			Repeatability (Within-Run)		Between-Run		Between-Day		Within-Laboratory	
Sample	N	Mean	SD	%CV	SD	%CV	SD	%CV	SD	%CV
Sample 1	88	1.7	0.09	5.3	0.1	6.1	0.16	9.7	0.21	12.6
Sample 2	88	4.7	0.1	2.1	0.07	1.4	0.17	3.6	0.21	4.4
Sample 3	88	9.2	0.25	2.7	0.0	0.0	0.27	2.9	0.36	3.9
Sample 4	88	16	0.3	2	0.2	1.2	0.4	2.7	0.6	3.6
Sample 5	88	21	0.4	2.1	0.3	1.3	0.6	2.7	0.8	3.7

Hemolysate: To estimate repeatability and within-laboratory precision, 6 whole blood hemolysate were tested using a single lot of reagent, a single lot of calibrator, and a single DxI 9000 Immunoassay analyzer. Each sample was assayed in duplicate, in 2 runs per day, over 20 days for a total of 80 replicates. Within-run, between-run, between-day, and total imprecision were calculated. Results are provided in the table below. The mean concentration reported below reflects hemolysate sample multiplied by dilution factor (21) and divided by hematocrit value.

Hemolysate										
RBC Folate (ng/mL)			Repeatability (Within-Run)		Between-Run		Between-Day		Within-Laboratory	
Sample	N	Mean	SD	%CV	SD	%CV	SD	%CV	SD	%CV
Sample 1	80	509	14.5	2.8	14.4	2.8	0.0	0.0	20.4	4.0
Sample 2	80	594	26.2	4.4	0.0	0.0	13.1	2.2	29.3	4.9
Sample 6	80	620	26.5	4.3	0.0	0.0	0.0	0.0	26.5	4.3
Sample 3	80	666	12.4	1.9	3.5	0.5	0.0	0.0	12.8	1.9
Sample 4	80	966	20.8	2.2	17.5	1.8	13.4	1.4	30.3	3.1
Sample 5	80	1116	20.1	1.8	12.9	1.2	0.0	0.0	23.9	2.1

2. Linearity:

Serum

A linearity study was conducted according to CLSI EP06-2nd edition. A total of 9 levels of serum samples ranging from 0.480 to 30.848 ng/mL were prepared by mixing different proportions of a serum sample containing a high concentration of folate with a serum sample containing a low concentration of folate. The lowest sample was run in 8 replicates, and all other samples were run in 4 replicates. Samples were tested on 3 DxI 9000 Access Immunoassay Analyzers using 3 reagent lots and three calibrator lots. The data was analyzed using a weighted linear regression model. The deviation from linearity did not exceed 5% within the claimed measuring interval.

Hemolysate
A linearity study was conducted according to CLSI EP06-2nd edition. A total of 9 levels of hemolysate samples ranging from 0.30 to 33.2 ng/mL were prepared by mixing different proportions of a hemolysate sample containing a high concentration of folate with a hemolysate sample containing a low concentration of folate. The lowest sample was run in 8 replicates, and all other samples were run in 4 replicates. Samples were tested on a single DxI 9000 Access Immunoassay Analyzers using a single reagent lot and a single calibrator lot. The data was analyzed using a weighted linear regression model. The deviation from linearity did not exceed 8% within the claimed measuring interval.

The results support the sponsor's claimed measuring interval of 2 – 24.8 ng/mL.

3. Analytical Specificity/Interference:

   Previously established in k060774.

4. Assay Reportable Range:

   See section VII.A.2 Linearity.

5. Traceability, Stability, Expected Values (Controls, Calibrators, or Methods):

   The measurand (Folate) in the Access Folate Calibrator is traceable to WHO International Standard Vitamin B12 and Serum Folate; 03/178.

6. Detection Limit:

   Determination of the limit of blank (LoB), limit of detection (LoD) and limit of quantitation (LoQ) were conducted following the recommendations in the CLSI EP17-A2 guideline. The LoQ was defined as the lowest concentration of analyte which has within-laboratory imprecision less than or equal to 20% CV. Results are summarized below.

Matrix	LoB (ng/mL)	LoD (ng/mL)	LoQ (ng/mL)
Serum	0.8	1.0	2.0
Hemolysate	0.0	0.5	1.0

7. Assay Cut-Off:

   Not applicable.

B Comparison Studies:

1. Method Comparison with Predicate Device:

   A method comparison study was performed comparing the Access Folate assay

on the DxI 9000 analyzer to the comparator device, the Access Folate assay on the Access 2 Immunoassay System. A total of 123 serum samples were evaluated. Samples were tested using three DxI 9000 instruments and three Access 2 instruments with three reagent pack lots and three calibrator lots. The Passing-Bablok regression analysis results between Access Folate DxI 9000 analyzer values and the Access Folate assay on the Access 2 Immunoassay System values are shown below:

Sample Type	N	Concentration Range (ng/mL)*	Slope	Intercept	Correlation Coefficient R
Serum	123	1.4 - 25	1.04	0.081	0.99

**As measured by the comparator method*

The sponsor provided data to demonstrate that results from testing hemolysate samples with the Access Folate assay on the DxI 9000 analyzer are comparable to results generated using the Access Folate assay on the Access 2 Immunoassay System.

2. Matrix Comparison:

Previously established in k060774.

C Clinical Studies:

1. Clinical Sensitivity:

Not applicable

2. Clinical Specificity:

Not applicable

3. Other Clinical Supportive Data (When 1. and 2. Are Not Applicable):

Not applicable

D Clinical Cut-Off:

Not applicable

E Expected Values/Reference Range:

Previously established in k060774.

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

IX Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.