510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY

A. 510(k) Number:

K180762

B. Purpose for Submission:

New Device

C. Measurand:

Hemoglobin A, F, A2, S, C, E, D

D. Type of Test:

Capillary electrophoresis

E. Applicant:

Sebia, Inc.

F. Proprietary and Established Names:

CAPI 3 HEMOGLOBIN(E)

G. Regulatory Information:

1. Regulation section:

   21 CFR 864.7415, Abnormal hemoglobin assay

2. Classification:

   Class II

3. Product code:

   GKA, Abnormal hemoglobin quantiatation

4. Panel:

   Hematology(81)

H. Intended Use:

1. Intended use(s):

   The CAPI 3 HEMOGLOBIN(E) kit is designed for the separation of the normal hemoglobins (A, A2 and F) in human venous blood samples, and for the detection of the major hemoglobin variants (S, C, E and D), by capillary electrophoresis in alkaline buffer (pH 9.4) with the SEBIA CAPILLARYS 3 TERA instrument.

   The CAPILLARYS 3 TERA instrument is an automated analyzer which performs a complete hemoglobin profile for the quantitative analysis of the normal hemoglobin fractions A, A2 and F and for the detection of major hemoglobin variants S, C, E and D. The assay is performed on the hemolysate of whole blood samples collected in tubes containing K2EDTA or K3EDTA as anticoagulant. The CAPI 3 HEMOGLOBIN(E) is intended to be used in conjunction with other laboratory and clinical findings.

   For In Vitro Diagnostic Use.

2. Indication(s) for use:

   Same As Intended Use

3. Special conditions for use statement(s):

   For prescription use only

4. Special instrument requirements:

   CAPILLARYS 3 TERA

I. Device Description:

The CAPI 3 HEMOGLOBIN (E) kit consists of five components: (1) HEMOGLOBIN(E) buffer which is an alkaline buffer (pH 9.4) supplied in 700 mL vials, (2) Hemolyzing Solution, supplied in 700 mL vials, and (3) Filters, four per kit. The CAPI 3 HEMOGLOBIN (E) kit is used in conjuction with the SEBIA CAPILLARYS 3 TERA instrument.

J. Substantial Equivalence Information:

1. Predicate device name(s):

   CAPILLARYS HEMOGLOBIN(E) using the CAPILLARYS 2 FLEX-PIERCING instrument

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

   K112550

3. Comparison with predicate:

Similarities		
Item	New Device CAPI 3 HEMOGLOBIN(E)	Predicate CAPILLARYS HEMOGLOBIN(E) (K112550)
Intended Use	The CAPI 3 HEMOGLOBIN(E) kit is designed for the separation of the normal hemoglobins (A, A2 and F) in human venous blood samples, and for the detection of the major hemoglobin variants (S, C, E and D), by capillary electrophoresis in alkaline buffer (pH 9.4) with the SEBIA CAPILLARYS 3 TERA instrument. The CAPILLARYS 3 TERA instrument is an automated analyzer which performs a complete hemoglobin profile for the quantitative analysis of the normal hemoglobin fractions A, A2 and F and for the detection of major hemoglobin variants S, C, E and D. The assay is performed on the hemolysate of whole blood samples collected in tubes containing K2EDTA or K3EDTA as anticoagulant. The CAPI 3 HEMOGLOBIN(E) is intended to be used in conjunction with other laboratory and clinical findings. For In Vitro Diagnostic Use.	The CAPILLARYS HEMOGLOBIN(E) kit is designed for the separation of the normal hemoglobins (A, A2 and F) in human blood samples, and for the detection of the major hemoglobin variants (S, C, E and D), by capillary electrophoresis in alkaline buffer (pH 9.4) with the SEBIA CAPILLARYS 2 FLEX-PIERCING instrument. The CAPILLARYS HEMOGLOBIN(E) kit is designed for laboratory use. The CAPILLARYS 2 FLEX-PIERCING instrument is an automated analyzer which performs a complete hemoglobin profile for the quantitative analysis of the normal hemoglobin fractions A, A2 and F and for the detection of major hemoglobin variants S, C, E and D. The assay is performed on the hemolysate of whole blood samples collected in tubes containing K2EDTA or K3EDTA as anticoagulant. For In Vitro Diagnostic Use.
Specimen Type	Human whole blood	Same
Technology	Capillary electrophoresis	Same
Detection Absorbance Wavelength	415 nm	Same

Similarities		
Item	New Device CAPI 3 HEMOGLOBIN(E)	Predicate CAPILLARYS HEMOGLOBIN(E) (K112550)
Software	Phoresis	Same
Controls for Migration	Sebia Normal A2 Control	Same
Use of other Controls	SEBIA Normal Hb A2 Control, Hb AFSC Control and Hb AF Control	Same
Reagent Location	On Board	Same

Differences		
Item	New Device CAPI 3 HEMOGLOBIN(E)	Predicate CAPILLARYS HEMOGLOBIN(E) (K112550)
Number of Separation Units (Capillaries)	12 parallel capillaries	8 parallel capillaries
Bottle for Reagents	RFID	None
Reagent cups	Supplied in separate packaging	Supplied in the kit
Wash Solution	Supplied in separate packaging	Supplied in the kit

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

- CLSI EP05-A3: Evaluation of Precision of Quantitative Measurement Procedures, 3rd Edition
- CLSI EP06-A: Evaluation of Linearity of Quantitative Measurement Procedures: A Statistical Approach, Approved Guidelines
- CLSI EP07-A2: Interference Testing in Clinical Chemistry; Approved Guideline. 2nd Edition
- CLSI EP17-A2: CLSI EP05-A2: Evaluation of Precision Performance of Quantitative Measurement Methods, 2nd Edition
- CLSI EP28-A3c: Defining, Establishing, and Verifying Reference Intervals in Clinical Laboratory 3rd Edition
- FDA Guidance for General Pinciples of Software Validation- Guidance for Industry and FDA Staff

L. Test Principle:

The SEBIA CAPI 3 HEMOGLOBIN(E) test in conjunction with the CAPILLARYS 3 TERA instrument utilizes the principle of capillary electrophoresis in free solution. The CAPILLARYS 3 instrument performs fully automated electrophoresis sequencing from the primary whole blood sample by directly sampling from capped collection tubes. Charged molecules are separated by their electrophoretic mobility in an alkaline buffer with a specific

pH. Separation also occurs according to the electrolyte pH and electroosmotic flow. The hemoglobins, separated in silica capillaries, are directly detected at an absorbance wavelength of 415 nm. The hemoglobin (Hb) fractions are separated by absorbance spectrophotometry. The hemoglobins are reported in % units along with an electrophoresis scan. The resulting electrophoregrams are evaluated visually for pattern abnormalities. Direct detection provides accurate relative quantification of individual hemoglobin fractions: A, F, A2, S, C, D, and E. By using alkaline pH buffer, normal and abnormal (or variant) hemoglobins are detected.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

Note: All results below met the manufacturer's pre-specified acceptance criteria.

a. Precision/Reproducibility:

Precision of the CAPI 3 Hemoglobin assay was evaluated according to CLSI guideline EP05-A3.

Repeatability

Five control materials with different levels of hemoglobin variants were run using the CAPI 3 HEMOGLOBIN(E) procedure performed with one CAPILLARYS 3 TERA instrument and one lot of the CAPI 3 HEMOGLOBIN(E) kit. Each sample was analyzed in duplicate on 12 capillaries per run, two runs per day over 20 days yielding a total of 960 results per sample. The results are expressed as coefficient of variation (%CV).

%Hb A

Sample	Mean (%)	Within Capillary	Between Capillary	Between Run	Between Day	Total
1	97.3	0.0%	0.0%	0.0%	0.0%	0.0%
2	93.5	0.1%	0.1%	0.0%	0.1%	0.1%
3	44.2	0.3%	0.4%	0.3%	0.5%	0.8%
4	54.2	0.4%	0.0%	0.1%	0.3%	0.5%
5	67.15	0.5%	0.1%	0.2%	0.3%	0.6%

%Hb A2

Sample	Mean (%)	Within Capillary	Between Capillary	Between Run	Between Day	Total
1	2.7	1.3%	0.9%	0.0%	0.7%	1.8%
2	6.5	0.8%	0.9%	0.0%	0.7%	1.4%
3	2.6	3.1%	4.1%	2.4%	1.3%	6.0%
4	3.0	2.8%	1.1%	0.8%	0.8%	3.2%

%Hb F

Sample	Mean (%)	Within Capillary	Between Capillary	Between Run	Between Day	Total
3	26.8	0.4%	0.1%	0.5%	0.2%	0.7%

%Hb S

Sample	Mean (%)	Within Capillary	Between Capillary	Between Run	Between Day	Total
3	17.5	0.4%	0.5%	0.1%	0.7%	1.0%

%Hb C

Sample	Mean (%)	Within Capillary	Between Capillary	Between Run	Between Day	Total
3	8.9	0.9%	0.8%	0.4%	1.1%	1.7%

%Hb D

Sample	Mean (%)	Within Capillary	Between Capillary	Between Run	Between Day	Total
4	40.6	0.5%	0.2%	0.1%	0.3%	0.6%

%Hb E

Sample	Mean (%)	Within Capillary	Between Capillary	Between Run	Between Day	Total
5	22.6	1.0%	0.1%	0.4%	0.3%	1.1%

Seven native EDTA venous whole blood samples with percentages of Hb A, Hb A2, Hb F, Hb S, Hb C, Hb D and Hb E fractions were analyzed on three CAPILLARYS 3 TERA instruments using one lot of CAPI 3 HEMOGLOBIN(E) kit for 7 days (with the exception of Hb E which has been analyzed for 6x days). Within each run, each sample was analyzed in duplicate on the same capillary. The results are expressed as coefficient of variation (%CV).

%Hb A

Sample	Mean (%)	Within Run	Between Run	Between Day	Between instrument	Total
1	97.5	0.0%	0.0%	0.0%	0.0%	0.0%
2	95.2	0.1%	0.0%	0.0%	0.0%	0.1%
3	56.7	0.3%	0.0%	0.2%	0.2%	0.4%
4	67.6	0.6%	0.2%	1.1%	0.0%	1.2%
5	56.5	0.3%	0.3%	1.3%	0.0%	1.3%
6	62.0	0.2%	0.1%	0.9%	0.0%	1.0%
7	62.0	0.5%	0.4%	0.0%	0.0%	0.7%

%Hb A2

Sample	Mean (%)	Within Run	Between Run	Between Day	Between instrument	Total
1	2.5	1.1%	0.6%	0.0%	0.3%	1.3%
2	4.8	1.4%	0.0%	0.4%	0.5%	1.5%
3	3.2	1.7%	0.0%	0.0%	0.5%	1.8%
4	2.7	5.2%	0.4%	3.8%	0.0%	6.5%
5	2.8	1.4%	0.8%	1.7%	0.0%	2.3%
6	1.9	1.7%	1.6%	2.6%	0.0%	3.5%
7	3.2	3.0%	1.6%	3.3%	0.0%	4.8%

%Hb F

Sample	Mean (%)	Within Run	Between Run	Between Day	Between instrument	Total
6	34.9	0.4%	0.2%	2.4%	0.0%	2.5%

%Hb S

Sample	Mean (%)	Within Run	Between Run	Between Day	Between instrument	Total
3	40.1	0.4%	0.1%	0.2%	0.4%	0.6%

%Hb C

Sample	Mean (%)	Within Run	Between Run	Between Day	Between instrument	Total
7	34.3	1.1%	0.3%	0.8%	0.0%	1.4%

%Hb D

Sample	Mean (%)	Within Run	Between Run	Between Day	Between instrument	Total
5	38.6	0.4%	0.3%	1.3%	0.0%	1.4%

%Hb E

Sample	Mean (%)	Within Run	Between Run	Between Day	Between instrument	Total
4	23.5	0.8%	0.0%	1.4%	0.0%	1.6%

Reproducibility studies were performed using a total of nine EDTA venous whole blood samples that included three frozen and six contrived samples with different percentages of the Hb A, Hb A2, Hb F, Hb S, Hb C, Hb D and Hb E fractions. The samples were analyzed in duplicate on two capillaries per run, two runs per day using three lots on each of three instruments over 6 days yielding a total of 432 per sample results. The results are expressed as coefficient of variation (%CV).

%Hb A

Sample	Mean (%	Within capillary	Between capillary	Between Run	Between Day	Between Lot	Between instrument	Total
1	97.7	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
2	94.8	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.1%
3	98.3	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
4	62.9	0.2%	0.3%	0.0%	0.2%	0.3%	0.0%	0.5%
5	63.0	0.4%	0.1%	0.3%	0.4%	0.2%	0.0%	0.6%
6	74.7	0.2%	0.3%	0.0%	0.1%	0.1%	0.2%	0.4%
7	57.2	0.2%	0.2%	0.0%	0.2%	0.2%	0.0%	0.4%
8	94.2	0.1%	0.1%	0.0%	0.0%	0.0%	0.0%	0.1%
9	28.4	0.5%	0.2%	0.1%	0.4%	0.0%	0.7%	1.0%

%Hb A2

Sample	Mean %	Within capillary	Between capillary	Between Run	Between Day	Between Lot	Between instrument	Total
1	2.3	1.4%	0.6%	0.4%	0.5%	0.4%	0.5%	1.7%
2	5.2	0.8%	0.5%	0.0%	0.7%	0.6%	0.0%	1.3%
3	1.7	1.5%	1.0%	0.0%	0.6%	0.7%	0.0%	2.1%
4	3.2	1.2%	1.1%	0.0%	0.4%	0.6%	0.3%	1.8%
5	3.2	2.2%	1.5%	0.0%	1.8%	0.5%	1.5%	3.6%
6	3.5	1.5%	1.6%	0.0%	1.2%	0.7%	1.7%	3.1%
7	2.6	1.3%	1.2%	0.0%	0.8%	0.7%	0.0%	2.0%
8	2.2	1.3%	0.9%	0.0%	0.5%	0.3%	0.2%	1.7%
9	2.6	1.3%	0.8%	0.4%	0.6%	0.6%	0.6%	1.9%

%Hb F

Sample	Mean %	Within capillary	Between capillary	Between Run	Between Day	Between Lot	Between instrument	Total
8	3.6	1.3%	1.4%	0.0%	1.1%	0.3%	1.2%	2.5%
9	69.0	0.2%	0.1%	0.0%	0.2%	0.0%	0.3%	0.4%

%Hb S

Sample	Mean %	Within capillary	Between capillary	Between Run	Between Day	Between Lot	Between instrument	Total
4	33.9	0.4%	0.4%	0.0%	0.4%	0.4%	0.0%	0.9%

%Hb C

Sample	Mean %	Within capillary	Between capillary	Between Run	Between Day	Between Lot	Between instrument	Total
5	33.5	0.7%	0.3%	0.5%	0.7%	0.3%	0.3%	1.1%

%Hb D

Sample	Mean %	Within capillary	Between capillary	Between Run	Between Day	Between Lot	Between instrument	Total
7	40.2	0.2%	0.2%	0.0%	0.2%	0.2%	0.0%	0.5%

%Hb E

Sample	Mean %	Within capillary	Between capillary	Between Run	Between Day	Between Lot	Between instrument	Total
6	21.8	0.4%	0.8%	0.0%	0.4%	0.3%	0.3%	1.0%

b. Linearity/assay reportable range:

A linearity study was performed per CLSI EP06-A: *Evaluation of Quantitative Measuring Procedures*; A Statistical Approach. Mixtures of two different blood samples were mixed at different proportions and tested in triplicate and analyzed using the CAPI 3 HEMOGLOBIN(E) procedure on the CAPILLARYS 3 instrument. The tests were determined to be linear within the reportable ranges studied for each of the following hemoglobins fractions: Hb A (1.0−97.3%), HbS (1.1−89.7%), HbA2 (0.2−9.1%), HbF (0.5−83.1%), HbC (0.3−82%), Hb D (1.1−43.5%), Hb E (0.3−86.9%).

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

Reagent Stability: The CAPI 3 HEMOGLOBIN(E) reagents used with the CAPILLARYS 3 TERA instrument are the same reagents that were presented in the predicate device, CAPILLARYS HEMOGLOBIN(E) kit using the CAPILLARYS 2 FLEX PIERCING instrument (K112550). The reagent bottles are the same with the exception the CAPI 3 HEMOGLOBIN(E) kit and the CAPI 3 WASH SOLUTION contain an RFID tag on the bottle(s).

Sample Stability:

Storage at refrigerated temperatures: Eleven whole blood samples with different hemoglobin fractions (Hb A, Hb A2, Hb F, Hb S, Hb C and Hb E) were analyzed upon receipt and after one week of storage at 2–8°C with the CAPI 3 HEMOGLOBIN(E) on the CAPILLARYS 3 TERA instrument. The results obtained met the acceptance criteria. Sample stability for the CAPI 3 HEMOGLOBIN(E) kit on the CAPILLARYS 3 TERA instrument was determined to be 7 days at 2–8°C.

Storage at frozen temperatures: Thirteen whole blood samples with different hemoglobin fractions (Hb A, Hb A2, Hb F, Hb S, Hb Cand Hb D) were analyzed upon receipt and after 3 months of storage at−70 or −80°C, with the CAPI 3 HEMOGLOBIN(E) assay performed with the CAPILLARYS 3 TERA instrument. The results obtained met the acceptance criteria. Sample stability for the CAPI 3 HEMOGLOBIN(E) kit on the CAPILLARYS 3 TERA instrument was determined to be 3 months at −70 or −80°C.

d. Detection limit:

The Limit of Blank (LoB), Limit of Detection (LoD), and Limit of Quantitation was performed per CLSI EP17-A2 guideline, *Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures*; Approved Guideline – Second Edition.

LoB: Two lots of the CAPI 3 HEMOGLOBIN(E) kit were used to detect the percentages of the fraction in the designated hemoglobin position. Five blood samples (B1–B5) were analyzed. For each lot of CAPI 3 HEMOGLOBIN(E) kit, each of the five samples was analyzed on 3 days with four replicates per day using one CAPILLARYS 3 TERA instrument in order to obtain 60 data points per lot.

LoD: Two lots of the CAPI 3 HEMOGLOBIN(E) kit were used to detect the percentages of the fraction in the designated hemoglobin position. Five blood samples were analyzed. For each lot of CAPI 3 HEMOGLOBIN(E) kit, each of the five samples was analyzed on 3 days with four replicates per day using one CAPILLARYS 3 TERA instrument in order to obtain 60 data points per lot. The LoD was determined using the sample from which the hemoglobin fraction is always detected.

LoQ: The LoQ was determined using precision profile approach. The LoQ is determined on four fractions representing the four intervals of migration of the Hb fractions of interest: the C/A2/E mobility interval using Hb A2 as the target analyte representing HbA2, Hb C and Hb E fractions; the D/S mobility interval using Hb S as the target analyte representing Hb S and Hb D fractions; Hb F and Hb A. Four different blood samples were tested with two lots of the CAPI 3 HEMOGLOBIN(E) kit performed on three CAPILLARYS 3 TERA instruments. Testing was performed for 5 days, two runs per day. Within each run, each sample was analyzed three times in order to obtain 90 data points per sample and 360 data points per lot.

Hemoglobin	LoB	LoD	LoQ
A	0.1%	1.0%	1.0%
A2	0.1%	0.2%	0.2%
F	0.2%	0.4%	0.5%
S	0.1%	0.9%	1.1%
C	0.1%	0.3%	0.3%
D	0.1%	0.7%	1.1%
E	0.1%	0.3%	0.3%

e. Analytical specificity:

Interference studies were conducted following CLSI EP07-A2, *Interference Testing in Clinical Chemistry*, using six samples (normal Hb A2 level, increased Hb A2 level Hb S, HbC, HbD, HbE). Each sample was analyzed three times.

Interferent	Maximum Concentration
Bilirubin	20.6 mg/dL or 352µmol/L
Triglycerides	2.2 g/dL or 25.1mmol/L
Excess EDTA	7.2mg/mL

f. Assay cut-off:

Not Applicable

2. Comparison studies:

a. Method comparison with predicate device: The method comparison was conducted at two sites which was found to be acceptable given that the CAPILLARYS 3 TERA instrument has been evaluated in a previous clearance (K161928). At site 1, a total of 153 samples including three controls were analyzed. At site 2, a total of 151 samples including one control were analyzed. Regression analysis was not performed for the Hb D fraction in site 2 due to insufficient data.

Site 1

Fraction	N	% Hb range	Correlation coefficient (r)	Slope (95% CI)	Intercept (95% CI)
Hb A	150	16.9−98.7	1.000	1.010 (1.008, 1.012)	-0.993 (-1.174, -0.812)
Hb A2	148	0.5−9.2	0.998	0.986 (0.976, 0.996)	0.005 (-0.026, 0.037)
Hb F	22	0.8−83.1	1.000	1.009 (1.005, 1.012)	-0.008 (-0.145, 0.129)
Hb S	13	1.8−89.7	1.000	1.010 (1.004, 1.015)	-0.025 (-0.240, 0.191)
Hb C	13	2.0−89.5	1.000	1.008 (1.000, 1.015)	0.099 (-0.167, 0.365)
Hb D	9	3.3−43.7	1.000	1.015 (1.009, 1.021)	-0.068 -0.245 to 0.109
Hb E	13	5.0−86.9	1.000	1.001 (0.996, 1.007)	0.183 0.014 to 0.353

Site 2 (U.S.)

Fraction	N	%Hb range	Correlation coefficient (r)	Slope (95% CI)	Intercept (95% CI)
Hb A	148	15.7−98.3	1.000	1.020 (1.017, 1.023)	-1.928 (-2.176, -1.681)
Hb A2	151	0.9−6.1	0.987	1.017 (0.990, 1.043)	-0.005 (-0.084, 0.074)
Hb F	30	0.5−34.3	0.999	1.009 (0.992, 1.026)	-0.127 (-0.298, 0.044)
Hb S	33	25.8−78.3	0.999	1.006 (0.993, 1.018)	0.475 (-0.057, 0.006)
Hb C	11	25.2−37.3	0.997	1.069 (1.011, 1.127)	-1.431 (-3.333, 0.472)
Hb E	4	22.1−91.9	1.000	1.002 (0.987, 1.017)	0.625 (-0.117, 1.367)

b. Matrix comparison:

Not applicable; the CAPI 3 HEMOGLOBIN(E) kit using the CAPILLARYS 3 TERA instrument is intended to be used with venous whole blood only.

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:

Twenty normal samples from the U.S. healthy population were analyzed with the CAPI 3 HEMOGLOBIN(E) on the CAPILLARYS 3 TERA instrument for the reference range verification study according to the CLSI EP28-A3c guideline, *Defining, Establishing,*

and Verifying Reference Intervals in the Clinical Laboratory; Approved Guideline – Third Edition.

The results are as follows:

Hemoglobin A (HbA): 96.7 – 97.8%
Hemoglobin F (HbF): < 0.5%
Hemoglobin A2 (HbA2): 2.2 – 3.2%

Major hemoglobin variants (S, C, E and D) should not be present in normal hemoglobin samples.

N. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Parts 801 and 809, as applicable.

O. Conclusion:

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