

**10(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY AND INSTRUMENT COMBINATION TEMPLATE**

A. 510(k) Number:

K082227

B. Purpose for Submission:

New Device

C. Measurand:

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

D. Type of Test:

Quantitative, Qualitative

E. Applicant:

Sebia, Inc.

F. Proprietary and Established Names:

MINICAP 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 quantitation

4. Panel:

Hematology (81)

H. Intended Use:

1. Intended use(s):

The MINICAP HEMOGLOBIN(E) kit is designed for the separation of the normal hemoglobins (A, F and A2) and for the detection of the major hemoglobin variants (especially S, C, E or D), by electrophoresis in alkaline buffer (pH 9.4) with the MINICAP System. The MINICAP performs all sequences automatically to obtain a complete hemoglobin profile for qualitative or quantitative analysis of hemoglobins. The assay is performed on sedimented, centrifuged or washed red blood cells; washing red blood cells is not essential to perform the analysis.

2. Indication(s) for use:

The MINICAP HEMOGLOBIN(E) kit is designed for the separation of the normal hemoglobins (A, F and A2) and for the detection of the major hemoglobin variants (especially S, C, E or D), by electrophoresis in alkaline buffer (pH 9.4) with the MINICAP System. The MINICAP performs all sequences automatically to obtain a complete hemoglobin profile for qualitative or quantitative analysis of hemoglobins. The assay is performed on sedimented, centrifuged or washed red blood cells; washing red blood cells is not essential to perform the analysis.

3. Special conditions for use statement(s):

Not applicable.

4. Special instrument requirements:

Not applicable.

I. Device Description:

The MINICAP HEMOGLOBIN(E) kit consist of five components; (1) MINICAP HEMOGLOBIN(E) buffer contains alkaline buffer (pH 9.4), supplied in 250 mL vials, (2) Hemolyzing Solution, supplied in 225 mL vials, (3) Washing Solution, supplied in 25 mL vials, (4) Reagent cups, supplied in pack of 125, (5) Filters, three per kit. The MINICAP HEMOGLOBIN(E) kit is used in conjunction with a Sebia MINICAP system in silica capillaries are utilized for analyzing. The hemoglobin, separated in capillaries, are directly detected at an absorbance wave length of 415 nm, resulting electrophoregrams. Direct detection provides relative quantification of individual hemoglobin fractions.

J. Substantial Equivalence Information:

1. Predicate device name(s):

CAPILLARYS HEMOGLOBIN(E)

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

K052291

3. Comparison with predicate:

Item	Similarities	
	<i>MINICAP HEMOGLOBIN(E)</i>	<i>CAPILLARYS HEMOGLOBIN(E)</i>
Intended Use	The MINICAP HEMOGLOBIN(E) kit is designed for the separation of the normal hemoglobins (A, F and A2) and for the detection of the major hemoglobin variants (especially S, C, E or D), by electrophoresis in alkaline buffer (pH 9.4) with the MINICAP System. The MINICAP performs all sequences automatically to obtain a complete hemoglobin profile for qualitative or quantitative analysis of hemoglobins. The assay is performed on sedimented, centrifuged or washed red blood cells; washing red blood cells is not essential to perform the analysis.	The CAPILLARYS HEMOGLOBIN(E) kit is designed for the separation of the normal hemoglobins (A, F and A2) and for the detection of the major hemoglobin variants (especially S, C, E or D), by electrophoresis in alkaline buffer (pH 9.4) with the CAPILLARYS System. The CAPILLARYS performs all sequences automatically to obtain a complete hemoglobin profile for qualitative or quantitative analysis of hemoglobins. The assay is performed on sedimented, centrifuged or washed red blood cells; washing red blood cells is not essential to perform the analysis.
Separation System	Capillary electrophoresis	Same
Instrument	MINICAP	CAPILLARYS
Absorbance wave length	415 nm	Same
Sample type	Centrifuged or packed red cells	Same
Sample identification	Yes	Yes
Hemolysis	Performed automatically by the system	Same
Introduction of the sample into the automatic system	Continuous loading	Same
Buffer reagent – vials	Ready to use, 2 vials of 250mL	Ready to use, 2 vials of 700mL
Buffer reagent – chemical composition and storage conditions	Alkaline buffer (pH 9.4) stability: 2 years at 2-8°C)	Same
Hemolysing solution	Ready to use, 1 vial of 225 mL	Ready to use, 1 vial of 440 mL

Similarities		
Item	MINICAP HEMOGLOBIN(E)	CAPILLARYS HEMOGLOBIN(E)
Hemolysing solution – chemical composition and storage conditions	Buffer solution to dilute and hemolyze red blood cells stability: 3 yrs at 2-8°C	Same
Wash solution	Stock solution, 1 vial 25 mL	Stock solution, 1 vial 75 mL
Wash solution – chemical composition and storage	Alkaline solution, stability: 3 years at 2-30°C	Same

Differences		
Item	MINICAP HEMOGLOBIN(E)	CAPILLARYS HEMOGLOBIN(E)
Number of separation units	2 parallel capillaries	7 parallel capillaries
Analyzed samples	2 samples	7 samples
Number of analysis (throughput)	8 sample/ hour	33 sample/ hour

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

Guidance for the Content of Premarket Submissions for Software Contained in Medical Devices - Guidance for Industry and FDA Staff, May 11, 2005.

L. Test Principle:

The MINICAP system uses the principle of capillary electrophoresis in a free solution. With this technique, 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 MINICAP system has capillaries functioning in parallel allowing 2 simultaneous analyses for hemoglobin quantification. A sample dilution with hemolyzing solution is prepared and injected by aspiration at the anodic end of the capillary. A high voltage protein separation is then performed and direct detection of the hemoglobins is made at 415 nm at the cathodic end of the capillary. The resulting electrophoregrams are evaluated visually for pattern abnormalities. By using alkaline pH buffer, normal and abnormal (or variant) hemoglobins are detected.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

Reproducibility **within-run** was determined when seven (7) different blood samples were run using MINICAP HEMOGLOBIN(E) the procedure on 3 MINICAP systems and with 2 lots of MINICAP HEMOGLOBIN(E) kits. Each sample was run 5 times on the 2 capillaries of each MINICAP system. The mean, SD and CV (n=10) were calculated for each sample, each hemoglobin component and each MINICAP system.

Reproducibility (Within-run) results:

Sample #	Hb A	Hb F	Hb S	Hb A2	Hb C	Mean (%)	SD	CV (%)
A	X					97.1/97.23/97.1	0.03/0.0/0.05	0.03/0.0/0.1
				X		2.9/2.8/2.9	0.03/0.0/ 0.05	1.0/0.0/1.7
B	X					62.0/61.2/61.9	0.28/0.41/.028	0.5/0.7/0.5
			X			34.6/35.4/34.7	0.27/0.39/0.27	0.8/1.1/0.8
				X		3.4/3.4/3.4	0.08/0.05/0.05	0.9/1.4/1.4
C	X					93.8/94.0/93.9	0.09/0.10/0.06	0.1/0.1/0.1
		X				3.6/6.5/3.6	0.09/0.08/0.08	2.4/2.2/2.2
				X		2.5/2.5/2.5	0.05/0.05/0.05	1.9/1.8/1.9
E	X					97.4/97.4/97.2	0.04/0.05/0.08	0.05/0.1/0.1
				X		2.6/2.6/2.8	0.04/0.05/0.08	1.7/1.9/3.0
F	X					93.2/93.1/93.0	0.07/0.07/0.19	0.1/0.1/0.2
				X		6.8/6.9/7.0	0.07/0.07/0.19	1.0/1.0/2.7
G	X					49.3/5.02/49.9/	0.60/0.30/0.69	1.2/0.6/1.4
		X				21.3/20.9/21.0	0.28/0.14/0.35	0.3/0.7/1.7
			X			19.9/19.4/19.6	0.31/0.36/0.41	1.6/1.8/2.1
				X		3.0/2.9/2.8	0.11/0.27/0.10	3.7/9.3/3.7
H					X	6.6/6.6/6.8	0.13/0.20/0.16	2.0/3.0/2.3
	X					39.0/39.4/38.6	0.53/0.54/0.62	1.4/1.4/1.6
		X				60.4/6.0/60.8	0.57/0.53/0.63	0.9/0.9/1.0
			X		0.7/0.6/0.6	0.05/0.05/0.04/	7.7/7.7/6.5	

Reproducibility **between-run** was determined when six (6) different blood samples were run ten (10) times using the MINICAP HEMOGLOBIN(E) procedure on 3 MINICAP systems and with 2 lots of MINICAP HEMOGLOBIN(E) kits. The analyzed samples included 2 samples with normal HbA2 level, 1 sample with decreased Ab A2 level and 3 samples with abnormal hemoglobins (Hb F or Hb S or Hb C) and one elevated Hb A2. The mean, SD and CV (n=10) were calculated for each sample for each hemoglobin component and each MINICAP system.

Reproducibility (Between-run) results:

Hb component	Mean (%)	SD	CV (%)
HbA	48.2 – 98.2	0.00 – 0.42	0.0 – 0.9
HbA2	1.3 – 3.3	0.00 – 0.11	0.0 – 3.8
HbF	20.7 – 21.4	0.19 – 0.36	0.9 – 1.7
HbS	19.6 – 42.3	0.14 – 0.34	0.3 – 1.7
HbC	6.7 – 7.2	0.06 – 0.01	0.9 – 1.5

Reproducibility **between-systems and between lots** was determined when six (6) different blood samples were run ten (10) times using the MINICAP HEMOGLOBIN(E) procedure on 3 MINICAP systems and with 2 lots of MINICAP HEMOGLOBIN(E) kits.

Reproducibility (Between-systems) results:

Hb component	Mean (%)	SD	CV (%)
HbA	48.9 – 98.2	0.04 – 0.73	0.5 – 1.4
HbA2	1.8 – 3.2	0.04 – 0.18	1.7 – 6.1
HbF	21.1	0.40	1.9
HbS	20.2 – 41.5	0.43 – 0.68	1.5 – 2.1
HbC	7.0	0.23	3.2

b. Linearity/assay reportable range:

Mixtures of different blood samples or dilutions with saline of blood samples with hemoglobin fractions (HbA, HbA2, HbF, and HbS) were prepared and analyze using the MINICAP HEMOGLOBIN (E) procedure. The tests were determined to be linear within the entire ranges studied for each of the hemoglobin fractions. In addition, three different blood samples were also serially diluted in saline and tested. The tests were determined to be linear within the entire range studied from 1.5 to 21.7 total hemoglobin and hemoglobin fractions percentages were not affected by the hemoglobin concentration of the samples.

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

Not applicable.

d. Detection limit:

Not applicable.

e. Analytical specificity:

Not applicable.

f. Assay cut-off:

Not applicable.

2. Comparison studies:

a. *Method comparison with predicate device:*

Comparison studies were performed at two sites on blood samples (with normal and elevated levels) for Hb A2, Hb F, Hb S and variants (such as hemoglobin S, C, D and E). The blood samples and their diagnostic assessment were provided by hospitals in Europe and the Middle-East.

The **first site** study included 109 blood samples without abnormal hemoglobin (or Hb variants) and 91 blood samples with abnormal hemoglobins (200 total samples).

The results are as follows:

Hb fraction	n	r ²	y-intercept	slope	Range of % values
Hb A2	200	0.992	0.074	0.983	0.0 – 7.0
Hb F	92	1.000	-0.099	1.000	0.0 – 82.7
Hb S	76	0.999	0.144	0.998	37 – 92.4

The hemoglobin variants hemoglobin S, C, D and E detected from the procedure were compared by qualitative analysis. The confirmation of variants was made using HPLC procedure. The results demonstrate a substantial equivalency in the detection of hemoglobin variant by the MINICAP HEMOGLOBIN (E) procedure and the predicate. There was no case observed of false positive (i.e. detection n of abnormal band or abnormal level of a normal band where no such abnormal existed).

The **second site** study included 27 blood samples without abnormal hemoglobin (or Hb variants) and 30 blood samples with abnormal hemoglobins. (57 total samples).

The results are as follows:

Hb fraction	n	r ²	y-intercept	slope	Range of % values
Hb A2	57	0.998	-0.083	1.085	0.0 – 6.1
Hb F	26	0.999	-0.147	1.006	0.2 – 40.7
Hb S	14	1.000	1.249	0.988	3.0 – 89.8

The hemoglobin variants hemoglobin S, C, D and E detected from the procedure were compared by qualitative analysis. The confirmation of variants was made using HPLC procedure. The results demonstrate a substantial equivalency in the detection of hemoglobin variant by the MINICAP HEMOGLOBIN (E) procedure and the predicate. There was no case observed of false positive.

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:

Normal values for individual major electrophoretic hemoglobin zones in the MINICAP system was established from a healthy population of 113 adults (men and women) with normal hemoglobin values using HPLC technique. See below:

Hemoglobin A (Hb A): comprised between 96.8 and 97.8%

Hemoglobin F (Hb F): < 0.5 %

Hemoglobin A2 (Hb A2): comprised between 2.2 and 3.2 %

It is recommended that each laboratory establish its own threshold values.

N. Instrument Name:

Sebia, MINICAP

O. System Descriptions:

1. Modes of Operation:

Open tube batch mode with the following sequence of automated steps:

Bar code reading of sample tubes

Sample hemolysis and dilution from primary tubes

Capillary washing

Injection of hemolyzed samples

Hemoglobin separation and direct detection of the separated hemoglobins on capillaries

2. Software:

The SEBIA MINICAP operating system software is designed to work with the instrumentation, MINICAP. The MINICAP instrumentation directed by the PHORESIS software is fully automated in the performance of the sample identification by barcode labeling, dilution, testing, and calculation of results. The PHORESIS software utilizes *Windows 98 or XP as the operating system with Intel based processors with Visual Basic as the programming language.*

FDA has reviewed applicant's Hazard Analysis and software development processes for this line of product types:

Yes X or No _____

3. Specimen Identification:

Bar Code reader

4. Specimen Sampling and Handling:

Red blood cells are allowed to precipitate at 2° – 8° C, or centrifuged at 5000 rpm for 5 minutes. The maximum volume of plasma is removed and the tube is vortexed for 5 seconds. The open tubes are placed in the sample racks.

5. Calibration:

Not applicable.

6. Quality Control:

It is necessary to run two analysis sequences with the Normal Hb A2 Control (SEBIA) after having changed buffer lot numbers, after a capillary cleaning, and before starting a new analysis sequence. It is also advised to include into each run of samples an assayed blood control (AFSC Control, Normal Hb A2 Control – SEBIA).

P. Other Supportive Instrument Performance Characteristics Data Not Covered In The "Performance Characteristics" Section above:

Q. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

R. Conclusion:

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