510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY

A. 510(k) Number:
k090346

B. Purpose for Submission:
Modification for body fluid application for ADVIA 2120 and the ADVIA 2120i.

C. Manufacturer and Instrument Name:
Siemens Healthcare Diagnostics, Inc.
Body Fluid Application for ADVIA 2120 and the ADVIA 2120i.

D. Type of Test or Tests Performed:
Quantitative, Automated Hematology Analyzer

WBC, NEUT%/#, LYMPH%/#, MONO%/#, EOS%/#, BASO%/#, LUC, RBC, HCT, MCV,RDW, CHCM, HDW, HGB, MCH, MCHC, PLT, MPV, RETIC%/#, MCVg, MCVr, CHCMg, CHCMr. CHg, CHr

Body Fluid Application:
Total nucleated cell (TNC) count and RBC count

E. System Descriptions:

1. Device Description:
   The Body Fluid Application can be run on the ADVIA 2120 and the ADVIA 2120i hematology analyzers. The ADVIA 2120/ADVIA 2120i Body Fluid Application uses the ADVIA 120 CBC TIMEpac and ADVIA 120 Diff TIMEpac reagents to determine the TNC and RBC counts. No additional Body Fluid Application specific reagents are needed.

   The ADVIA 2120 Hematology system consists of the following: an analytical module that aspirates, dilutes, and analyzes whole blood samples; an autosampler that automatically mixes, identifies, and presents the samples for processing; a computer workstation that controls the instrument, provides primary user interface with the instrument and manages the data produced by the instrument; a printer that optionally generates reports based on the instrument results and an autoslide module.

2. Principles of Operation:
   The ADVIA 2120/2120i Body Fluid Application uses the Basophil/Lobularity and RBC/Platelet channels to enumerate the TNC and RBC counts. The TNC count is derived from the Basophil/Lobularity channel. The ADVIA 2120/2120i BASO reagent contains surfactant and phthalic acid which, in the presence of low heat in the Baso channel reaction chamber, lyses RBCs and strips the cytoplasmic membrane from all leukocytes except basophils. This cell suspension is subsequently passed through the flowcell. The cell suspension is intercepted by light from the laser diode where the low-angle light scatter (2° to 3°) and high-angle light scatter (5° to 15°) signals of each cell are counted.

   The RBC count is derived from the RBC/Platelet channel. The ADVIA 2120/2120i

RBC/Platelet reagent uses sodium dodecyl sulfate and glutaraldehyde to sphere and fix the RBCs. This cell suspension uses the same flowcell and low-angle and high-angle light scatter signals as the Baso channel to count the RBCs. The system automatically reports the TNC and RBC counts in conventional or SI units appropriate for body fluid samples.

The Basophil/Lobularity cytochemical reactions consist of two steps:
Step 1 - Red blood cells and platelets are lysed using the ADVIA 120 BASO reagent.
Step 2 - All white blood cells except basophils are stripped of their cytoplasm using the ADVIA 120 BASO reagent and the increased temperature in the reaction chamber.

The Body Fluid Application default Run Screen, displays 3 cytograms: Peroxidase, Basophil/Lobularity and RBC Scatter cytograms. The Basophil/Lobularity and Peroxidase channel cytograms have been modified specifically for the light scatter characteristics of the cells in body fluid samples. The RBC cytogram is the same as the cytogram for whole blood analysis.

The reported TNC count is derived from the Basophil/Lobularity channel. The TNC count is the sum of the events above the Baso Y-channel Noise threshold plus the events left of Baso X-channel 49. The events in the Baso channel Noise region for the Body Fluid Application are excluded from the TNC count.

The WBC count from the Peroxidase channel (WBCP) is compared to the TNC count from the Basophil/Lobularity channel. When the TNC does not agree with the WBCP count within limits specific to the Body Fluid Application, the TNC count is flagged with an asterisk. Determination of the RBC count is the same as for whole blood samples. The Header information includes the body fluid specimen type.

3. Modes of Operation:
   ADVIA 2120/2120i system is a random access analyzer which has three sampling modes: [1] Manual open tube sampler, [2] manual closed tube sampler and [3] automated closed tube sampler (Autosampler).
4. Specimen Identification:
   Sample identification is done by following modes: [1] Manually entered sample ID [2] barcode [3] rack/position numbers (for autosampler only) and [4] instrument automatic incremental numbering.
5. Specimen Sampling and Handling:
   Body fluids samples must be manually mixed by gentle inversion for the manual open and closed tube modes. The automated closed tube sampler mixer rotates first to +45° then to +135°, staying at each orientation for a minimum of 0.6 seconds and a maximum of 15 seconds. A total of 25 mixer-cycles are required before aspiration. Fully mixed samples which have a dwell of over 15 seconds require an additional 20 mixer cycles before aspiration. If a dwell exceeds 5 minutes, the samples must be mixed again for 45 cycles before aspiration.
6. Calibration:
   ADVIA 2120/2120i systems calibration for all reportable CBC, Diff. and retic. (except % retic.) parameters require the ADVIA SETpoint Calibrator and one

whole blood sample.

7. Quality Control:
   It is recommended that the system be controlled using ADVIA TESTpoint or 3•in•1 Hematology Controls. These controls are intended to be integrated into a clinical laboratory's own quality control program and procedures.
8. Software:
   FDA has reviewed applicant's Hazard Analysis and Software Development processes for this line of product types:
   Yes__X___ or No_____

F. Regulatory Information:

1. Regulation section:
   21 CFR 864.5220 - Automated differential cell counter
2. Classification:
   Class II
3 Product code:
   GKZ - Counter, Differential Cell
4. Panel:
   81 Hematology

G. Intended Use:

1. Indication(s) for Use:
   The ADVIA 2120/2120i auto-analyzer Body Fluid Application is an in vitro diagnostic test for the enumeration of the total nucleated cell (TNC) count and RBC count for pleural, peritoneal, and peritoneal dialysis (PD) specimens collected in K2 or K3 EDTA.
2. Special Conditions for Use Statement(s):
   None

H. Substantial Equivalence Information:

1. Predicate Device Name(s) and 510(k) numbers:
   COULTER® LH 750 Body Fluids Application - k030606
2. Comparison with Predicate Device:

Similarities		
Item	Predicate Device	Device
	COULTER® LH 750 Body Fluids Application	Siemens ADVIA 2120/2120i Body Fluids Application
Instrument	Automated Hematology Analyzer	Same
Intended Use	Enumeration of the total nucleated cell (TNC) count and RBC count	Same
Dilution	Automated dilution	Same
Counts	Automated calculation of counts	Same

Differences		
Item	Predicate Device	Device
	COULTER® LH 750 Body Fluids Application	Siemens ADVIA 2120/2120i Body Fluids Application
Sample Type	Pleural, peritoneal, and Peritoneal Dialysis fluids	Cerebrospinal fluid, serous fluid, and synovial fluid
Detection	Impedance technology	Light scatter technology

I. Special Control/Guidance Document Referenced:

1. CLSI Document. EP5-A2. Evaluation of Precision Performance of Clinical Chemistry Devices; Approved Guideline—Second Edition". 2004. CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898.
2. CLSI Document H56-A. "Body Fluid Analysis for Cellular Composition; Approved Guideline". 2006. CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898.
3. CLSI Document EP17-A. "*Protocol for determination of Limits of Detection and Limits of Quantitation*". 2004. CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898.
4. CLSI Document GP10. "*Assessment of the Clinical Accuracy of Laboratory Tests Using Receiver Operating Characteristic (ROC) Plots*". CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898.

J. Performance Characteristics*:*

1. **Analytical Performance:**

a. Accuracy:

The Body Fluid Application TNC count was evaluated by comparing the hemocytometer count to the ADVIA 2120/2120i count for 434 body fluid samples. The Body Fluid Application RBC count was evaluated by comparing the manual chamber count to the ADVIA 2120/2120i count for 78 body fluid samples. Samples were pleural, peritoneal, and peritoneal dialysis specimens collected in EDTA tubes with no visible clotting. The results are considered acceptable if they meet the design input requirements for the method comparison study. The design input requirements are >0.85 for r, 1 +/- 0.15 for slope and 0 +/- 100 for intercept.

Parameter	n	r	Slope	Intercept	95% CI Slope	95% CI Intercept
TNC	434	0.942	0.90	65	0.87 to 0.93	-22 to 153
RBC	78	0.964	0.99	-0.5	0.93 to 1.05	-14.6 to 13.5

Conclusion: The results of the method comparison studies indicated comparable performance relative to the reference method.

b. Precision/Reproducibility:

Precision Studies

A precision study was performed based on the guidelines contained in CLSI EP5-A2. The TNC and RBC pools were assayed over a 20 day period with two runs per day; two replicates per run, for total of 80 replicates for each pool.
Stabilized pools were prepared with target TNC concentrations of 50, 100, 500, 1000, and 10000 cells/uL in plasma diluted 1:2 with phosphate buffer.
Stabilized pools were prepared with target RBC concentrations of 50, 100, 500, and 1000 x 10^3 cells/uL in plasma diluted 1:2 with phosphate buffer.
The results were analyzed using nested analysis of variance consistent with the guidance contained in CLSI EP5-A2.

TNC Precision

Within Run				Between Run		Between Day		Total	
Level	**Mean**	**SD**	**CV**	**SD**	**CV**	**SD**	**CV**	**SD**	**CV**
50	58.6	6.887	11.7%	3.001	5.1%	1.103	1.9%	7.6	12.9%
100	118.33	10.908	9.2%	0	0.0%	3.47	2.9%	11.4	9.7%
500	593.48	26.32	4.4%	0	0.0%	8.304	1.4%	27.6	4.7%
1000	1179.5	35.152	3.0%	17.329	1.5%	19.276	1.6%	43.7	3.7%
10000	116.27	196.994	1.7%	61.866	0.5%	136.538	1.2%	247.542	2.1%

RBC Precision

Within Run				Between Run		Between Day		Total	
Level	**Mean**	**SD**	**CV**	**SD**	**Mean**	**SD**	**CV**	**SD**	**CV**
50	52.664	2.202	4.2%	0	0.0%	1.001	1.9%	2.4	4.6%
100	104.99	2.994	2.8%	0	0.0%	1.4	1.3%	3.3	3.1%
500	518.48	6.405	1.2%	2.04	0.4%	3.497	0.7%	7.6	1.5%
1000	1023.7	13.055	1.3%	6.404	0.6%	0	0.0%	14.5	1.4%

The results are considered acceptable if they meet the design input requirements for within-run precision. The design input requirements are ≤ 15% CV at 500 cells/µL for the TNC count and ≤ 10% CV at 50 x10^3 cells/µL for the RBC count.
Conclusions: The results of the precision studies using stabilized specimens indicated acceptable precision over the time period tested.

Repeatability Testing

Specimens were utilized from the stability study, described below. One specimen of each body fluid type assayed 10 times in one run on a single day. Additional assays were not possible because of sample volume and sample stability limitations. The mean, standard deviation (SD), and coefficient of variation are calculated for each sample included in the study. The results are considered acceptable if they meet the design input requirements for within-run precision. The design input requirements are ≤ 15% CV at 500 cells/µL for the TNC count and ≤ 10% CV at 50 x10^3 cells/µL for the RBC count.

	TNC			RBC		
Fluid	**Mean**	**SD**	**CV**	**Mean**	**SD**	**CV**
Pleural	93492	8749.5	9.4	80	6.9	8.6
Peritoneal	274	22.5	8.2	41	1.6	3.9
PD	64	6.5	10.2	28	1.7	6.1

Conclusion: The results from the repeatability study demonstrate that within-run precision of the body fluid counts meets the design input requirements of ≤ 15% CV at 500 cells/µL for the TNC count and ≤ 10% CV at 50 $x10^3$ cells/µL for the RBC count.

Reproducibility/Specimen Stability

The goal of the testing the Body Fluid Software is to demonstrate stability of fluids under different conditions using the system software. This was accomplished by using remnant body fluid samples collected in K2 or K3 EDTA at two external sites. These sites collected pleural, peritoneal and peritoneal dialysate body fluids. The samples were run on ADVIA 2120/2120i using the body fluid application. These fluid samples were tested within 2-hours to represent the baseline or zero (0) time result, as well at different time points, and different conditions (room temperature and at 2-8°C). Specimens included:

Five (5) samples with RBC counts that fall in the range from ≥ 10,000 cells/uL to 6.76 x 10^6 cells/µL for each type of fluid.
Five (5) samples with TNC counts that fall in the range of 0.030 to 400 x 10^3 cells/µL (30 to 400,000) for each type of fluid.

Conclusion: The observed regression slopes all meet the acceptance criteria of falling within the range of 0.9 to 1.1. It can be concluded that body fluid samples are stable at room temperature for 8 hours and when refrigerated for 24 hours.

c. *Linearity:*

The objective of the linearity protocol was to verify low level linearity of the body fluid TNC and RBC counts. High level linearity of the counting methods has been established previously (k042251) in whole blood sampling modes. Low level pools with a target WBC concentration of 500 cells/uL and an RBC target concentration of 500 thousand cells/uL were created. Serial dilutions of these pools were made with cell free materials (plasma diluted in PBS) over the low level range of the assays. The WBC pools were aspirated 13 times each in body fluid mode, and the RBC pools were aspirated 12 times each.

TNC Linearity

% of High Pool	Observed TNC (cells/uL)	Expected TNC (cells/uL)	Obs vs Exp %	Obs vs Exp Abs
0%	1	0		1
4%	19	20	95.0	-1
8%	41	40	102.5	1
12%	67	60	111.7	7
16%	80	80	100.0	0
20%	105	99	106.1	6
100%	497	497	100.0	0

RBC Linearity

% of High Pool	Observed RBC (10E3cells/uL)	Expected RBC (10E3cells/uL)	Obs vs Exp %	Obs vs Exp Abs
0%	0	0		0
2%	11	10	110.0	1
6%	30	31	96.8	-1
10%	52	52	100.0	0
15%	77	77	100.0	0
20%	106	103	102.9	3
100%	515	515	100.0	0

Conclusion: The results meet the design requirements that the deviations are < 10% or within 10 cells.

d. Carryover:

The carryover protocol was run in all three sampling modes: manual open tube, manual closed tube, and automated sampling. Carryover was tested by four different methods in each sampling mode:

A normal whole blood assayed in CBC/Diff mode followed by three aspirations of a saline sample in body fluid mode.

A manipulated whole blood sample with elevated WBC counts (approximately 30,000 cells/uL) and RBC counts (approximately 6 million cells/uL) assayed in CBC/Diff mode followed by three aspirations of a saline sample in body fluid mode.

A normal whole blood assayed in body fluid mode followed by three aspirations of a saline sample in body fluid mode.

A manipulated whole blood sample with elevated WBC counts (approximately 30,000 cells/uL) and RBC counts (approximately 6 million cells/uL) assayed in body fluid mode followed by three aspirations of a saline sample in body fluid mode.

Five sets of carryover sequences were run for each type of carryover test within each sampling mode. A saline primer was assayed before each body fluid sample when using the Autosampler as instructed in the body fluids instructions for use.

Carryover results are listed in the Tables below. Each Table entry is the average of the 5 carryover sequences run in each configuration.

Open Tube Sampling Carryover

Sample	TNC % Carryover	RBC % Carryover
Normal WBC/RBC CBC/Diff	0.17%	0.11%
Elevated WBC/RBC CBC/Diff	0.16%	0.16%
Normal WBC/RBC BF	0.27%	0.26%
Elevated WBC/RBC BF	0.38%	0.31%

Manual Closed Tube Sampling Carryover

Sample	TNC % Carryover	RBC % Carryover
Normal WBC/RBC CBC/Diff	0.11%	0.11%
Elevated WBC/RBC CBC/Diff	0.14%	0.10%
Normal WBC/RBC BF	0.36%	0.25%
Elevated WBC/RBC BF	0.29%	0.25%

Automated Sampling Carryover

Sample	TNC % Carryover	RBC % Carryover
Normal WBC/RBC CBC/Diff	0.00%	0.01%
Elevated WBC/RBC CBC/Diff	0.00%	0.01%
Normal WBC/RBC BF	0.00%	0.00%
Elevated WBC/RBC BF	0.00%	0.00%

Conclusion: The carryover results meet the specification of <0.4% for the TNC, and <0.3% for the RBC count for Manual Closed Tube Sampler (MCTS) and the Manual Open Tube Sampler (MOTS), and <0.1% for both the TNC and RBC counts for the Autosampler.

e. Interfering Substances:
If TNC and RBC counts from patient samples where the manual differential shows a proportional neutrophil count ≥ 80%, crystals, the presence of lipids and/or chylomicrons, cellular deterioration and disintegration, or the patient is under treatment with a sclerosing agent, the results should be confirmed by an alternate method.

2. Other Supportive Instrument Performance Data Not Covered Above:
The range of reportable values study included a limit of blank (LoB) study, a limit of detection study (LoD), and a limit of quantitation (LoQ) study as defined in CLSI EP17 CLSI EP17-A to determine the lowest concentration of TNC and RBC counts that can be detected with at least 95% probability.

Limit of Blank

Cell free samples were prepared by mixing equal amounts of cell free plasma and PBS. These samples were assayed in body fluid mode over a 4-day period. A total of 80 assays were performed on the blank samples.

The results are analyzed non – parametrically as recommended in section 4.1.1 of CLSI EP17-A. The TNC LoB is four (4) cells, and the RBC LoB is 1,000 cells. The limit of blank was determined to be 4.24 cells / uL for TNC and 1.43 x 10^3 cells/uL for RBC.

Limit of Detection

Limit of detection (LoD) was estimated by preparing pools with a TNC concentration of approximately 20 cells/uL and an RBC concentration of approximately 10 x 10^3 cells/uL. Based on the LoB results, five samples prepared with a TNC concentration of approximately 20 cells/uL were assayed 13 times each day for a total of four days. Likewise, five samples prepared with an RBC concentration of approximately 10 – 11 x10^3 cells/uL were assayed 13 times each day for a total of four days. LoD is calculated as LoD = LoB + (c-beta)x(SD) as recommended in section 4.3.2 of CLSI EP17-A. The limit of detection was determined to be 12.8 cells/uL for TNC and 2.8 x 10^3 cells/uL for RBC.

Limit of Quantitation

Pools were prepared with target TNC concentrations of 0, 20, 40, 60, 80, 100 and 500 cells/uL. Each pool was assayed 13 times each. Pools were prepared with target RBC concentrations of 0, 10, 30, 50, 75, 100, and 500 x10^3 cells/uL. Each pool was assayed 12 times each. LoQ is determined as the analyte level where a 20% total CV is obtained. The LoQ for the TNC count is 27 cells/uL and the LoQ for the RBC count is 4.94 x10^3 cells/uL.

K. Proposed Labeling:

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

L. Conclusion:

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