

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
DEVICE ONLY TEMPLATE**

A. 510(k) Number:

K032039

B. Analyte:

Immature Granulocyte parameter

C. Type of Test:

Quantitative flow cytometry assay

D. Applicant:

Sysmex America, Inc.

E. Proprietary and Established Names:

Immature Granulocyte (IG) parameter on the Sysmex® XE-2100 Automated Hematology Analyzer

F. Regulatory Information:

1. Regulation section:
21 CFR 864.5220, automated differential cell counter
2. Classification:
Class II
3. Product Code:
GKZ
4. Panel:
Hematology (81)

G. Intended Use:

1. Indication(s) for use:
“The Immature Granulocyte (IG) parameter on the Sysmex XE-2100 is intended for in Vitro Diagnostics to classify and count immature granulocyte cells in EDTA anti-coagulated blood.”
2. Special condition for use statement(s):
For use with EDTA anticoagulated whole blood.
3. Special instrument Requirements:
For use only on the Sysmex XE-2100 Automated Hematology Counter.

H. Device Description:

The XE-2100 automated cell analyzer has a DIFF (Differential) channel, which identifies and enumerates immature granulocyte cells in addition to the traditionally reported parameters of an automated cell differential. The IG count on the XE-2100 is measured in the DIFF channel. Cells in this channel are detected by a combination of lateral scattered light (inner complexity of the cell), forward scattered light (volume), and lateral fluorescent light via a flow cytometry method utilizing a semiconductor laser. A concise image of each cell is detected. Abnormal and immature cells, with their larger nuclear volume show much higher fluorescence intensity than normal cells, and are easily distinguishable in the DIFF scattergram. A well-defined differentiation or clustering of the leukocyte populations is obtained.

I. Substantial Equivalence Information:

1. Predicate device name(s):
Manual microscopic differential cell count
2. Predicate K number(s):
N/A
3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Type of anticoagulant	EDTA	EDTA
Specimen type	Peripheral blood	Peripheral blood
Differences		
Item	Device	Predicate
Intended Use	To classify and count immature granulocyte cells in EDTA anticoagulated blood.	To manually classify and count the white blood cells into sub categories using a microscope.
Methodology	Automated counting of immature granulocytes on an automated hematology analyzer	Manual counting of immature granulocytes using a microscope. Typically the manual method counts 100 cells. The NCCLS H20-A method uses 400-cell differential performed by 2 techs each counting 200 cells.

Accuracy	Comparison to manual count showed good correlation.	Method of counting of cells using a microscope established as the predicate method.
Pro/Con	A large number of cells can be analyzed and several parameters (i.e. forward scatter, side scatter, and fluorescent labels) rather than morphological appearance alone can be used.	Manual method is imprecise due to the small number of cells counted, typically not more than 100 WBC total. It is labor intensive and time consuming.

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

Class II Special Controls Guidance Document: Premarket Notifications for Automated Differential Cell Counters for Immature or Abnormal Blood Cells; Final Guidance for Industry and FDA (12-4-02). Numerous NCCLS documents imbedded in the Guidance are used.

K. Test Principle:

The identification of immature granulocytes is based on the phospholipid content and the nuclear density of the leukocytes. The mature leukocyte has a higher lipid content than the immature cell and is selectively lysed when diluted with Stromatolyser-4DL, which hemolyzes the red blood cells. At the same time, Stromatolyser-4DS is added to further dilute the sample and to stain white blood cells. Mature leukocytes are disrupted leaving the immature cells intact. There is some shrinkage of the immature cells but they do not lyse. In the optical detector block, the sample is analyzed via a flow cytometry method utilizing a semiconductor laser.

L. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

Within Run Precision: Specifications were verified for precision using samples that were run in the open and closed mode ten times consecutively. The mean, standard deviation, and coefficient of variation were calculated. Results were compared to manufacturer specifications. For within run precision in the open tube mode results had CVs ranging from 5.5 to 8.5 %. For within run precision in the closed tube mode results had CVs ranging from 4.8 to 9.3%. In conclusion the within run results met manufacturer specifications.

Between day precision (reproducibility): Specifications were verified for precision using **e-Check™**, for the time period of evaluation on the open manual mode. Imprecision (SF) (within run and total) were

determined using NCCLS EP-15A. Results were checked to manufacturer acceptable ranges. Between day precision for Level 1: QC 30560804, Imprecision (SD) were: within run: 0.4341, total: 0.471. Between day precision for Level 1: QC 3056084, Imprecision (SD) were: within run: 0.0124, total: 0.01. Between day precision for Level 2: QC 3050805, Imprecision (SD) were: within run: 0.361594, total: 0.407858. Between day precision for Level 2: QC 3050805, Imprecision (SD) were: within run: 0.024376, total: 0.029609. Between day precision for Level 3: QC 30560806, Imprecision (SD) were within run: 0.40651, total: 0.0404502. Between day precision for Level 3: QC 30560806, Imprecision (SD) were within run: 0.83352, total: 0.83351. In conclusion results for between day precision were acceptable.

- b. *Linearity/assay reportable range:*
N/A
- c. *Traceability (controls, calibrators, or method):*
N/A
- d. *Detection limit:*
N/A
- e. *Analytical specificity:*
N/A
- f. *Assay cut-off:*
N/A

2. Comparison studies:

- a. *Method comparison with predicate device:*
Studies were done at one clinical site. Manual WBC differential counts were carried out using NCCLS H20A, i.e. 200 cell counts by two different qualified observers. Specimens for the studies included samples with high WBC counts, lipemia, high bilirubin, platelet clumps, NRBCs, atypical lymphocytes, immature granulocytes (metas, myelos, pros), blast forms, left shift, atypical/abnormal lymphocytes, iron deficiency, thalassemia, etc. The flow cytometry procedures followed were those of Sysmex Corp. of Japan using a Beckman Coulter Epics XL, which is an FDA cleared device.

Studies to evaluate accuracy to the manual WBC differential: With an N of 265, the Correlation (r) was 0.83. Other r values from the literature ranged from 0.79 to 0.90. In conclusion the results obtained at the clinical site were consistent with other studies found in the

literature. The XE-2100 IG parameter showed good correlation to the manual differential cell count.

Study to evaluate the XE-2100 IG parameter to flow cytometry:

With an N of 89, the Correlation (r) was 0.92. In conclusion the XE-2100 IG parameter showed excellent correlation to flow cytometry.

Study to evaluate the IG parameter count of flow cytometry to the manual count:

With an N of 89, the Correlation (r) was 0.72. In conclusion the IG parameter of flow cytometry showed moderate correlation to the manual count.

b. Matrix comparison:

N/A

3. Clinical studies:

a. Clinical sensitivity:

N/A

b. Clinical specificity:

N/A

c. Other clinical supportive data (when a and b are not applicable):

Short Term Stability: The within day study consisted of 6 normal samples each in the manual mode; analyzed at baseline (time-zero minutes after drawn), 5, 15, 30, 45, and 60 minutes after being drawn. This was performed to validate the readiness of a sample with high and low IG results to be analyzed from zero to 60 minutes. In conclusion samples with various IG results display no clinically significant difference from zero to 60 minutes.

Long Term Stability: The long term stability study was performed at room temperature and 4° C. samples were run at zero, 4, 8, 12, 16, 24, 36, 48, 56, and 72 hours. In conclusion the variation of IG% from zero hour of collection to 24 hours was within ± 1.5 IG% and displayed no clinically significant difference.

4. Clinical cut-off:

N/A

5. Expected values/Reference range:

The Normal Population Reference Ranges were determined for the IG% and IG# parameters. The range for each parameter is calculated for 95% confidence intervals. N was 60 with an approximate equal number of healthy males and females. The Mean for IG% was 0.215. The Mean for IG# was 0.0138. The SD for IG% was 0.107. The SD for IG# was 0.00086. In

conclusion the reference range for IG% was <0.4% and for IG# was <0.029 x 10³/μL.

M. Conclusion:

The Sysmex America, Inc. Immature Granulocyte parameter on the Sysmex XE-2100 Automated Hematology Analyzer has been shown to be substantially equivalent to the manual method of counting WBCs into subcategories using a microscope.

