



RETICULOCYTE WORKSHEET 2010

These samples have been tested and found to be homogeneous and stable for the purpose of these exercises, and are manufactured from human red blood cells in a preservative medium.

JUNE	Cycle 10 Run 1	RE10-1-06a	Module 1 - Reticulocyte Count
		RE10-1-06b	Module 1 - Reticulocyte Count
		RE10-2-06a	Module 2 - Reticulocyte Count
		RE10-2-06b	Module 2 - Reticulocyte Count
		RE10-3-06a	Module 3 - Reticulocyte Count
		RE10-3-06b	Module 3 - Reticulocyte Count
DECEMBER	Cycle 10 Run 2	RE10-1-12a	Module 1 - Reticulocyte Count
		RE10-1-12b	Module 1 - Reticulocyte Count
		RE10-2-12a	Module 2 - Reticulocyte Count
		RE10-2-12b	Module 2 - Reticulocyte Count
		RE10-3-12a	Module 3 - Reticulocyte Count
		RE10-3-12b	Module 2 - Reticulocyte Count

Module	Method/Instrument Group
RE1	Manual Method
RE2	Siemens Advia 120/2120, H3 Abbott CD3200/Ruby, CD3500, CD3700, CD4000/Sapphire Sysmex XE2000i, XE2100/XE5000, RAM-1 Beckman Coulter STKS, MAXM, HMX, ACT5 Diff, ABX Pentra 60 C+
RE3	Beckman Coulter Gen.S, LH Series

GENERAL INFORMATION

SAMPLE	Volume Provided	Test	Interpretation	Units
RE10 -1-**a / b	1.0 ml	Reticulocyte Count (In-house + Commercial Stains)	Low / Normal / Raised	%, X10 ⁹ /L
RE10 -2-**a / b	1.0 ml	Reticulocyte Count	Low / Normal / Raised	%, X10 ⁹ /L
RE10 -3-**a / b	2.0 ml	Reticulocyte Count	Low / Normal / Raised	%, X10 ⁹ /L

Any relevant details pertaining to the samples (e.g. age and sex of patient) will be provided on the packing slip accompanying your survey package.

Certain processing instructions are instrument specific. Please read carefully before proceeding.

MODULE 1 (RE1): MANUAL COUNTING

- A vial of commercial Reticulocyte Stain (Retic Stain-A) has been provided to participants who have registered a MANUAL counting method. You are required to perform a **percentage** and **absolute** count using your in-house stain AS WELL AS Retic Stain-A. The results submitted using the in-house stain will assess your laboratory's processes while those using Retic Stain-A will assess the enumeration. Failure to submit both sets of results will produce a "No Results Returned" flag on your report.
- **N.B.** If you use Streck commercial stain as your in-house stain, you are still required to submit both sets of results as the stain QAP provides may have a different lot number to the one you are using.
- The interpretation must be made on the results obtained using Retic Stain-A.

SAMPLE HANDLING AND PROCESSING INFORMATION

A. Survey Samples (RE10-1-**a / **b)

(These instructions are taken directly from the manufacturer's instructions for use).

Use product immediately after removing from refrigerator.

1. Mix by gentle inversion between thumb and index finger until red blood cells are completely resuspended. Do not mix mechanically. Do not rub between palms of hands.
2. Refer to appropriate procedure section below.
3. Wipe threads of vial and cap with clean tissue before replacing cap. Recap vial.
4. Return to refrigerator immediately.

Use product immediately after removing from refrigerator.

5. Mix by gentle inversion between thumb and index finger until red blood cells are completely resuspended. Do not mix mechanically. Do not rub between palms of hands.
6. Process as required.
7. Wipe threads of vial and cap with clean tissue before replacing cap. Recap vial.
8. Return to refrigerator immediately.

B. Retic Stain-A

(These instructions are taken directly from the manufacturer's instructions for use)

1. Mix the survey samples by gentle inversion (See above).
2. Prepare a dilution using equal number of drops each of survey samples and Retic Stain-A.
3. Incubate the tubes at ROOM TEMPERATURE for 20 minutes.

4. Mix well. Prepare a film and allow to dry.
5. Enumerate reticulocytes according to your laboratory's protocol.

C. Calculation of Absolute Reticulocyte Count

A red cell count will also be provided for users of the MANUAL method to convert their percentage count to the absolute count.

Calculations: Absolute Reticulocyte Count = Reticulocytes (%) x Total RBC ($10^{12}/L$)

Example: Reticulocyte Count (%) = 2.2

$$\text{Total RBC Count} = 3.3 \times 10^{12}/L$$

$$\begin{aligned} \text{Absolute Reticulocyte Count} &= \frac{2.2}{100} \times 3.3 \times 10^{12}/L \\ &= 0.0726 \times 10^{12}/L \\ &= 72.6 \times 10^9/L \end{aligned}$$

MODULE 2 (RE2): AUTOMATED COUNTING

SAMPLE HANDLING AND PROCESSING INFORMATION

A. Survey Samples (RE10-2-**a / **b)

(These instructions are taken directly from the manufacturer's instructions for use).

1. Mix by gentle inversion between thumb and index finger until red blood cells are completely resuspended. Do not mix mechanically. Do not rub between palms of hands.
2. Process as required (**Important! See below for specific procedural instructions***).
3. Wipe threads of vial and cap with clean tissue before replacing cap. Recap vial.
4. Return to refrigerator immediately.

AUTOMATED PROCEDURE: The user should follow the instrument manufacturer's instructions for performing automated reticulocytes. If required, transfer the sample into another tube prior to testing.

NB: *When using the sample on the *Abbott CELL-DYN 3500/3700, it is recommended that analysis be performed 30 minutes after sample is added to the reagent.*

When using the sample on the *SYSMEX XE and XT series, it must be processed through the QC mode. (See below).

INSTRUCTIONS FOR QC MODE PROCESSING

A. SYSMEX XE SERIES

Summary: The RCPA specimen needs to be run into one of the Manual E-Check QC files – for example Level 2 Manual. The results are then deleted from the QC files and printed from the Explorer screen. See Operators Manual Chapter 2, Section 3. 3.1

Details:

- Refer to the “Instructions for Use” for the control material.
- For HST sites, set the appropriate conveyor in single mode.
- Press “QC” on the main screen.
- Press “Exec QC” on the QC menu.
- Select one of the manual mode E-Check files – for example “Level 2 Manual”. (Do not use one of the “Other” files).
- Press “Select”.
- Mix the sample as described in the “Processing Instructions ” for the control material.
- Aspirate sample.
- Press “Cancel” – Results do not have to be accepted.
- Go to “Explorer” and select the RCPA results.
- (You may change the ID of the QC specimens that you have just run from “QC-lot number” to an RCPA ID)
- Print the results and delete any results from the QC file that have been accepted.

B. XT SERIES

Summary: The RCPA specimen needs to be run into one of the manual E-Check QC files – for example Level 2 Manual. The results are then printed from the Explorer screen. See Operators Manual Chapter 6, Section 3.1

Details:

- Refer to the “Instructions for Use” for the control material.
- Go to “QC Analysis” on the main menu.
- (If “QC Analysis” is not visible on the main menu then go to “Controller” then “QC Analysis”).
- Select a manual E-Check QC file – for example “Level 2 Manual”. (Do not select one of the “Other” files).
- Mix the sample as described in the “Instructions for Use” for the control material.
- Aspirate sample.
- Press “Cancel”– Results do not have to be accepted.
- Go to “Explorer” and select the RCPA results.
- (You may change the ID of the QC specimens that you have just run from “QC-lot number” to an RCPA ID).
- Print the results and delete any RCPA results from the QC file that have been accepted.

MODULE 3 (RE3): AUTOMATED COUNTING

SAMPLE HANDLING AND PROCESSING INFORMATION (RE10-3-a / **b)**

(These instructions are taken directly from the manufacturer’s instructions for use).

1. Remove vials of control material from the refrigerator. It is not necessary to warm the controls to room temperature before use.
2. To mix: **(Do not mix mechanically)**
 - a. Hold vial horizontally between the palms of the hands and roll the vial back and forth for 20 or 30 seconds.
 - b. Mix by rapid inversion to ensure the cells are suspended.
 - c. Vials stored for an extended period of time may require extra mixing.
 - d. Gently invert the vials 8 to 10 times immediately before sampling.
3. The user should follow the instrument manufacturer’s instructions for performing automated reticulocyte counts.
4. After sampling, return to refrigeration for maximum open-vial stability. If run in the open mode, wipe the threads of both the vial and cap before replacing cap and returning to refrigeration.