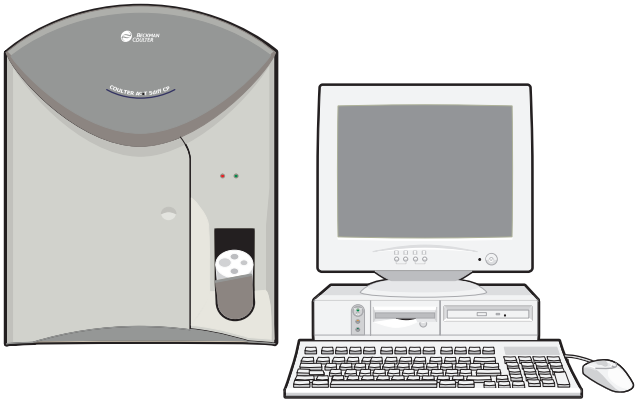
COULTER® A^{C}•T™ 5diff Cap Pierce Hematology Analyzer

Operator's Training Guide



PN 4277205BA (June 2010)

Manufactured for
Beckman Coulter, Inc.
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Brea, CA 92821

BECKMAN COULTER

WARNINGS AND PRECAUTIONS

READ ALL PRODUCT MANUALS AND CONSULT WITH BECKMAN COULTER-TRAINED PERSONNEL BEFORE ATTEMPTING TO OPERATE INSTRUMENT. DO NOT ATTEMPT TO PERFORM ANY PROCEDURE BEFORE CAREFULLY READING ALL INSTRUCTIONS. ALWAYS FOLLOW PRODUCT LABELING AND MANUFACTURER'S RECOMMENDATIONS. IF IN DOUBT AS TO HOW TO PROCEED IN ANY SITUATION, CONTACT YOUR BECKMAN COULTER REPRESENTATIVE.

HAZARDS AND OPERATIONAL PRECAUTIONS AND LIMITATIONS

WARNINGS, CAUTIONS, and IMPORTANTS alert you as follows:

WARNING - Can cause injury.
CAUTION - Can cause damage to the instrument.
IMPORTANT - Can cause misleading results.

BECKMAN COULTER, INC. URGES ITS CUSTOMERS TO COMPLY WITH ALL NATIONAL HEALTH AND SAFETY STANDARDS SUCH AS THE USE OF BARRIER PROTECTION. THIS MAY INCLUDE, BUT IT IS NOT LIMITED TO, PROTECTIVE EYEWEAR, GLOVES, AND SUITABLE LABORATORY ATTIRE WHEN OPERATING OR MAINTAINING THIS OR ANY OTHER AUTOMATED LABORATORY ANALYZER.

WARNING Risk of operator injury if:

- All doors, covers and panels are not closed and secured in place prior to and during instrument operation.
- The integrity of safety interlocks and sensors is compromised.
- Instrument alarms and error messages are not acknowledged and acted upon.
- You contact moving parts.
- You mishandle broken parts.
- Doors, covers and panels are not opened, closed, removed and/or replaced with care.
- Improper tools are used for troubleshooting.

To avoid injury:

- Keep doors, covers and panels closed and secured in place while the instrument is in use.
- Take full advantage of the safety features of the instrument. Do not defeat safety interlocks and sensors.
- Acknowledge and act upon instrument alarms and error messages.
- Keep away from moving parts.
- Report any broken parts to your Beckman Coulter Representative.
- Open/remove and close/replace doors, covers and panels with care.
- Use the proper tools when troubleshooting.

CAUTION System integrity might be compromised and operational failures might occur if:

- This equipment is used in a manner other than specified. Operate the instrument as instructed in the Product Manuals.
- You introduce software that is not authorized by Beckman Coulter into your computer. Only operate your system's computer with software authorized by Beckman Coulter.
- You install software that is not an original copyrighted version. Only use software that is an original copyrighted version to prevent virus contamination.

IMPORTANT If you purchased this product from anyone other than Beckman Coulter or an authorized Beckman Coulter distributor, and, if it is not presently under a Beckman Coulter service maintenance agreement, Beckman Coulter cannot guarantee that the product is fitted with the most current mandatory engineering revisions or that you will receive the most current information bulletins concerning the product. If you purchased this product from a third party and would like further information concerning this topic, call your Beckman Coulter Representative.

REVISION STATUS

Initial Issue, 6/01
Software version 1.03

Issue B, 10/04
Software Version 2.00
Added information about the new features provided by software version 2.00. Updated illustrations.

Issue BA, 06/10
Software Version 2.00.

Updates were made to the company corporate address.

Note: Changes that are part of the most recent revision are indicated in text by a bar in the margin of the amended page.

This document applies to the latest software listed and higher versions. When a subsequent software version changes the information in this document, a new issue will be released to the Beckman Coulter website. For labeling updates, go to www.beckmancoulter.com and download the most recent manual or system help for your instrument.

REVISION STATUS

CONTENTS

CONTENTS

PURPOSE OF THIS DOCUMENT

To provide an authorized trainer with corporately accepted training materials for instructing a new operator in the proper use and operation of a COULTER® A^{C}•T™ 5diff Cap Pierce (CP) hematology analyzer.

DOCUMENTATION

Several documents are available with your new instrument.

- **Instructions For Use manual**, PN 624021, provides information about:
  - getting started,
  - running of your instrument,
  - reviewing results,
  - performing special procedures, such as cleaning, replacing, or adjusting an instrument component,
  - troubleshooting problems,
  - determining what the instrument does,
  - understanding how to safely operate the instrument,
  - powering up the instrument,
  - customizing the setup, and
  - running controls and samples.

  The information contained in the Instructions For Use manual can also be accessed using the A^{C}•T 5diff CP online Help system. The printed version is available by order. Contact your Beckman Coulter Representative if you want to order a printed copy of this manual.
- **Host Transmission Specification**, PN 4277065, defines the information needed to program the transmission interface between the A^{C}•T 5diff CP hematology analyzer and the laboratory's host computer. The printed version is available by order. Contact your Beckman Coulter Representative if you want to order a printed copy of this manual.
- **Operator Manuals CD-ROM**, PN RAX015, provides the operator manuals in two formats: one for viewing and one for printing. Use this CD-ROM at any compatible desktop PC to access the same Operator's Guide information that is accessible through the Help menu on the A^{C}•T 5diff CP Workstation and the same Host Transmission Specification information that is accessible in the printed manual.

  Do not use the Operator Manuals CD-ROM at your A^{C}•T 5diff CP Workstation. This CD-ROM is optimized for use on a desktop home or office PC, not the A^{C}•T 5diff CP Workstation.

ABOUT THIS TRAINING GUIDE

Scope

This training guide is designed to support A^{C}•T 5diff CP hematology analyzer training. This document provides the trainer with the opportunity to customize the training experience to best fit the individual trainee and laboratory. Since trainee experience levels and laboratory needs differ, this training outline is designed with flexibility in mind.

Topics

The materials in this guide support the training of eight different topics:

- Topic 1 GETTING TO KNOW YOUR INSTRUMENT
- Topic 2 STARTUP / SHUTDOWN
- Topic 3 SET UP OPTIONS
- Topic 4 QUALITY ASSURANCE
- Topic 5 CALIBRATION
- Topic 6 SAMPLE ANALYSIS
- Topic 7 DATA REVIEW
- Topic 8 BASIC TROUBLESHOOTING

Each topic consists of several related subjects that may be used, as needed, in your training.

Organization

The eight individual topics contained in the Operator's Training Guide are part of a larger organization that includes four basic types of information:

Introduction	Explains how to use this training guide
Topics	Heart of this document, each topic consists of multiple subjects and operational summaries relating to the overall topic
Summary Pages	Master copy for use by the Key Operator in their laboratory once training is complete
Training Checklist	Method for documenting the specific training an operator receives

Summary Pages

Routine operational tasks detailed in the Operator's Guide are abbreviated in this training document. These abbreviated instructions are referred to as either summary pages or operational summaries. Each summary page is an individual, stand-alone document that is designed for use in your laboratory as a training aid and/or as a reference tool for a trained operator. A master copy of each summary page is located behind the Summary Masters tab.

These summary pages contain essential operational instructions. As a result, it is important that you become proficient in the use of this tool during your training. To eliminate the need to flip back and forth between the topic and the summary masters, the appropriate summaries are inserted within each topic.

How to Identify an Inserted Summary

When a summary is inserted in the training material, a rotated chevron (➤) appears to the left of the Summary Page title. For example, at the point of insertion, the Startup summary would begin with

➤ **Startup Summary**

Since this is a stand-alone summary for use after training is complete, some of the instructions discussed in the training topic may be repeated in the summary. Two rotated chevrons (➤ ➤) mark the end of the inserted summary.

Daily Operations Quick Reference

Daily operational tasks detailed in the summary page format are further abbreviated in this Quick Reference trifold (PN 4277315). An experienced operator may use these abbreviated instructions to perform those routine operations completed on a daily basis, including:

- Startup when the power is already turned on
- Running COULTER® A^{C}•T™ 5diff Control Plus
- Running patient samples (with or without using the Worklist)
- Shutdown

This Quick Reference tool is **not** designed to teach an operator how to run the instrument. It is designed for use by a trained, experienced operator who is already familiar with the information on the corresponding Summary pages.

Training Checklist

This checklist provides the trainer with an official method to document the completion of training on the A^{C}•T 5diff CP hematology analyzer.

When the training process is complete, we recommend that both the trainer and trainee sign the Training Checklist to show agreement that the trainee is sufficiently trained and able to meet the objectives listed for each topic.

RESPONSIBILITIES

Beckman Coulter's Responsibility

Beckman Coulter is responsible for instructing a Key Operator in the proper use and operation of the A^{C}•T 5diff CP hematology analyzer.

Our goal is to provide your laboratory with high quality training that best fits your laboratory's needs. This means the emphasis given to each topic may differ, with some topics needing to be covered more thoroughly than others.

Key Operator's Responsibility

It is the Key Operator's responsibility to train all other operators in their laboratory. To assist you in this process, a master copy of each operational summary is located at the end of the Operator's Training Guide. Once trained by an authorized Beckman Coulter trainer, the Key Operator is authorized to copy these masters for use in training other laboratory personnel.

CONVENTIONS

This training guide uses the following conventions:

- A primary window is the window displayed after you select one of the following tabs: **Worklist**, **Run**, **Results**, **Quality Assurance**, or **Analyzer / Logs**.
- Instructions assume operator is using the mouse for navigation.
- Tabs are the major navigation tool.
- Black text on a tab indicates the name of the window currently displayed. Although text is greyed-out on the other tabs, these tabs are still active and may be selected as desired.
- If an icon is active, it appears in color; if icon is inactive, the coloring is removed (greyed-out).
- Software navigation to access a needed function or screen appears as a sequence of menu items separated by double arrow heads. For example, the path to the command for priming the reagents is **Diagnostics ▸▸ Operator ▸▸ Diluter Systems ▸▸ Prime Reagents**.
- **Bold** indicates a menu item such as **File** or a tab such as **Worklist**.
- *Italics font* indicates screen text and/or messages displayed by the instrument such as *Calibration Passed*.
- Information that is to be typed is in Courier font such as type the password `123`.
- Instrument or system is sometimes used to refer to the A^{C}•T 5diff Cap Pierce hematology analyzer (meaning both Analyzer and Workstation).
- CP refers to cap pierce.
- **Note** (in bold font) prefaces supplemental information.
- An ATTENTION contains information that is important to remember or helpful when performing a procedure.
- Main card refers to the main circuit board (card) in the instrument.
- RBC bath is sometimes referred to as RBC/Plt bath.
- The terms “screen” and “window’ are used interchangeably.
- A^{C}•T 5diff Rinse reagent is sometimes referred to as Rinse.
- A^{C}•T 5diff Fix reagent is sometimes referred to as Fix.
- A^{C}•T 5diff Hgb Lyse reagent is sometimes referred to as Hgb Lyse.
- A^{C}•T 5diff WBC Lyse reagent is sometimes referred to as WBC Lyse.
- A^{C}•T 5diff Diluent reagent is sometimes referred to as Diluent.
- ▢ indicates a key such as Enter.
- Tab indicates the operator needs to press and release the Tab key.
- ▢ + ▢ indicates to press and hold the first key listed, then press the next key.
- A rotated chevron (➤) placed to the left of a Summary Page title identifies an operational summary inserted within the training materials. For example,

  ### ➤ Changing Reagent Summary

- Two rotated chevrons (➤ ➤) designate the end of an operational summary.

SAFETY SYMBOLS

Safety symbols alert you to potentially dangerous conditions. These symbols, together with text, apply to specific procedures and appear as needed in the operational summary pages throughout this training guide.

Symbol	Warning Condition	Action
	**Biohazard**. Consider all materials (specimens, reagents, controls, and calibrators, and so forth) and areas these materials come into contact with as being potentially infectious.	Wear standard laboratory attire and follow safe laboratory procedures when handling any material in the laboratory.
	**Probe hazard.** The probe is sharp and may contain biohazardous materials, such as controls and calibrators.	Avoid any unnecessary contact with the probe and probe area.

GRAPHICS

All graphics, including screens and printouts, are for illustration purposes only and must not be used for any other purposes.

OBJECTIVES

When the subject is complete, you will be able to . . .

Workstation

- Locate and name the three major components of the computer system.
- Locate and explain the function of specific keys on the keyboard.
- Log on the Workstation using the proper User name and password.
- Use the mouse to navigate through the software and select desired items or functions.
- Locate the menu bar and access the pull-down text menus using the mouse.
- Locate the tool bar and identify each icon and its purpose.
- Quickly access a primary window by selecting its corresponding tab.
- Use the buttons, fields, boxes, scrollable lists, and scroll bars as needed.
- Explain the significance of the green, red, and yellow background colors.
- Access and use the online Help system as needed.
- Load paper in the printer and verify the printer is ready to print.

Main Unit

- Locate and name the hardware controls and indicators on an A^{C}•T 5diff Cap Pierce (CP) hematology analyzer.
- Given an instrument that appears to be without power,
  - Tell whether it is powered down or in Standby.
  - Bring the instrument back into operation.
- Reset the system.
- Locate the cap pierce module and the two tube holders.
- Explain how to select the proper tube holder and sample position.
- Demonstrate how to properly change the tube holder.

Single Point of Aspiration

- Explain why the A^{C}•T 5diff CP hematology analyzer is referred to as a single point of aspiration system.
- Briefly overview closed-vial operation versus open-vial operation.
- List four situations when open-vial operation may be necessary.

Reagents

- Locate the reagent compartment.
- Name the reagents used on the A^{C}•T 5diff CP hematology analyzer.
- State if the reagent is used for CBC analysis, DIFF analysis, or both.
- State the open container stability of each reagent used on an A^{C}•T 5diff instrument.
- Explain how to change reagents.
- Explain how to handle and replace a waste container, if applicable.

NOTES

WORKSTATION

Functions of the A^{C}•T 5diff CP Workstation

- Controls and monitors instrument operation
- Displays, stores, outputs, and allows recall of sample results
- Stores and graphs control results for the Quality Assurance program
- Automatically calculates new calibration factors
- Allows bidirectional communication with a host computer
- Consists of three major components: the computer, the monitor, and the printer

Major Components

Computer

- **Must not use this system as a personal computer**
- Consists of hardware components and software
- Hardware is the term used to describe the physical components of a computer system
  - Locate the following hardware components:
    - Power ON/OFF button
    - Power ON/OFF indicator light
    - Hard drive indicator and symbol
    - CD-ROM drive
    - Diskette drive
- Software refers to a sequence of detailed instructions (called a program) that directs the computer to perform certain actions

Monitor

- Computer uses the monitor to visually display information
- Locate the Power ON/OFF switch and the indicator that lights when power is on
- Locate screen adjustment controls

Printer

- Used to either print a copy of the screen displayed on the monitor or of data stored in the computer's database
- Locate the Power ON/OFF switch
- Explain use of the ONLINE/OFFLINE button
- Demonstrate how to load the paper
- Demonstrate how to change the ink cartridge

Bar-code Reader (Optional)

- Hand-held scanner for reading the bar-code label on a specimen tube before sampling
- Operator must press button on the bar-code reader before scanning a label
- An audible beep indicates barcode was read successfully

Keyboard

- Direct line of communication between the operator and the computer containing the Workstation software program

Layout

- Alphanumeric keys:
  - Configured in a standard typewriter keyboard layout
  - Consists of all standard alphanumeric keys plus a handful of special computer keys and symbols
- Tab
  - May be used to move from field to field on some windows
- Caps Lock
  - Works like the Shift Lock key on a typewriter
  - With Caps Lock on,
    - Pressing any alphabet key on the keyboard produces an uppercase letter
    - Pressing the Shift key plus a letter produces a lowercase letter
    - Only shifts the letter keys, all other keys on the keyboard remain the same
- Numeric keypad:
  - Keypad contains the numbers 0 through 9, plus the period, an Enter key, and various mathematical symbols
  - Can expedite manual entry of patient ID numbers and certain quality control entries
- Num Lock
  - Controls operation of the numeric keypad
  - Locate light that illuminates when Num Lock is turned on
  - With Num Lock turned on, numeric keypad produces numbers
  - Workstation keyboard is designed to be used with Num Lock turned on
  - With Num Lock turned off, numeric keypad on a typical keyboard doubles as a cursor keypad
- Cursor control keys:
  - ← ↑ ↓ →
    - Also called arrow keys
    - Directional keys for moving the cursor
    - Used to highlight menu items, scroll up and down screens, or move to a field on a screen to enter or edit data
  - Page Up and Page Down may be used to page through stored sample and control data
  - Home and End may be used to locate the beginning or the end of stored data
- Keys not used with the Workstation: Insert, F1 through F12, Print Screen, Esc, and Spacebar

- Enter
  - Mainly used to select a highlighted menu or submenu option which then activates a change in the screen display or initiates a function
  - Do not use Enter to bring up the Primary Window as pressing this key may activate a command
  - Do not use Enter to save new information on any setup screen
  - Keyboard has two Enter keys; both work identically
- Ctrl and Alt
  - Modifier keys that have no action by themselves but can be used in conjunction with other keys to generate special operations
  - Must simultaneously press Ctrl + Alt + Delete to log onto the Workstation

Software

- Workstation software is based on Windows-NT
- Familiarity with Windows conventions and use is beneficial but not required

Initial Windows-NT Logon

- When the software initially loads, a Windows-NT logon window appears
- Logon Information box is where you enter user name and password information

User Names and Passwords

- When logging on the system, required to type a user name
- Type one of the following:
  - BCI (for normal operation), or
  - Admin (for special procedures)
- For normal operation (which is the majority of the time):
  - Log on using BCI as the User name
  - 123 is password for BCI User name
  - Once operator enters the password for BCI, the system prompts for the Operator ID
- For special procedures and only when instructed to by a Beckman Coulter Representative:
  - Log on using the Admin User name provided to you at the time it is needed

Operator ID

- A 3-character (alphanumeric) ID of your choice which uniquely identifies you as the current operator
- Enter your Operator ID after logging on. The system then automatically loads the Workstation application software.

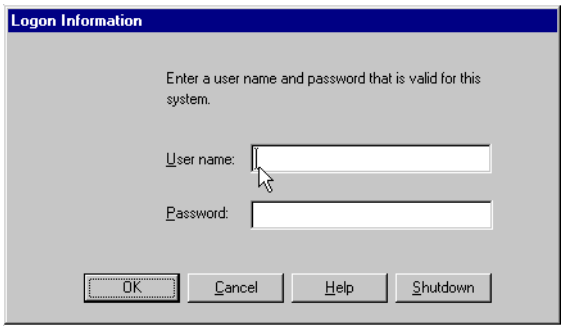
Software Passwords

- Various Workstation software areas, such as Setup and Calibration, require an “Administrator” password before permitting access

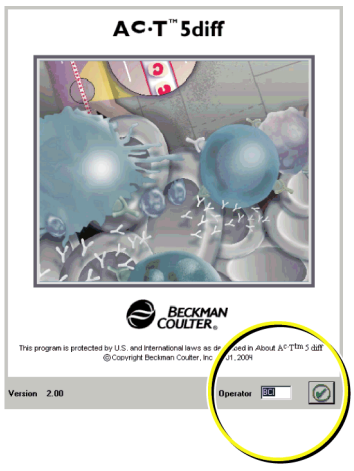
- When "Administrator" password is requested, type `123` to gain access to that particular software screen
- Other Workstation software areas, such as service diagnostics, requires a "Service" password before permitting access; "Service" password is for use only by Beckman Coulter Representatives

Logging On the Workstation

- When the Begin Logon box is displayed on the Workstation screen, operator must follow the instructions on the screen and simultaneously press Ctrl + Alt + Delete
- When Logon Information box appears:



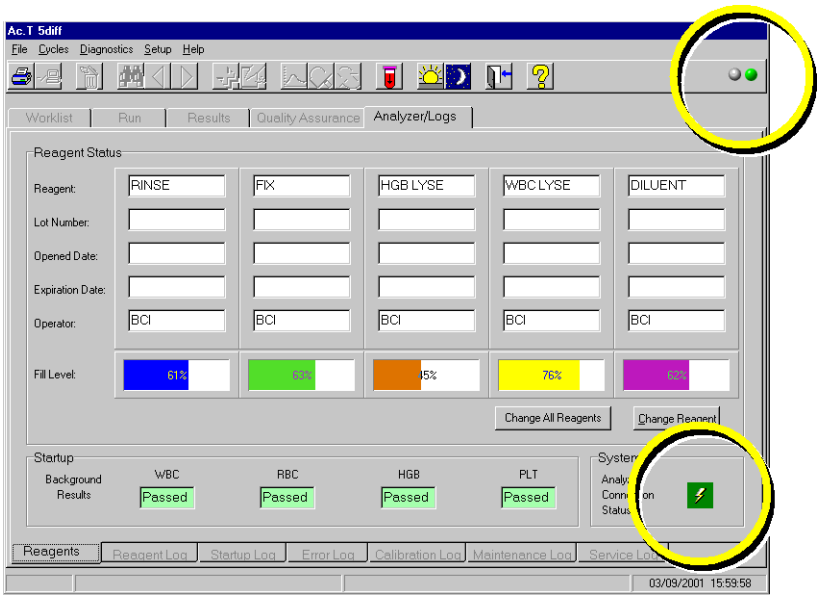
  - Type `BCI` as the User name (User name for normal operation)
  - Press Tab
  - Type `123` (Password for BCI User name)
  - Press Enter or click **OK** to log on
- When the following window appears:



  - Type your 3-character Operator ID
  - Press Enter or click 
- Wait for Analyzer / Logs window to appear

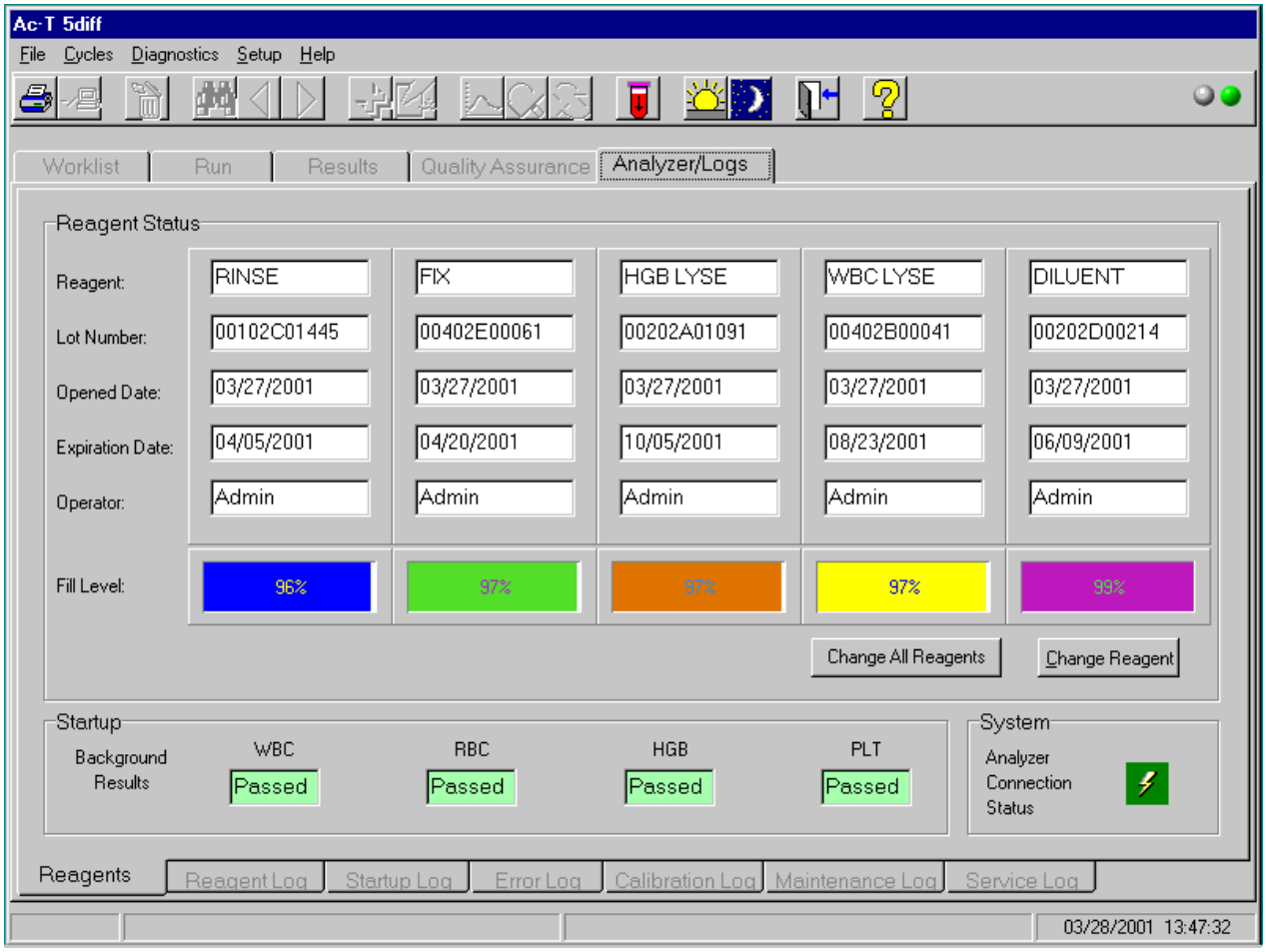
- Verify background behind  (lightening bolt) is green

  Note: Analyzer and Workstation should begin communicating within 30 seconds. In the upper right corner, the System State indicator on the right should also be green.



Primary Windows

Primary Window (Analyzer / Logs)



- First window to appear after log on
- May also be referred to as Reagents window
- Primary naviagation window
  - Provides operator with quick access to other primary windows via tabs
  - Use the mouse to select the desired tab
  - Each primary window can be reached within two tab selections

Tab Selections

- Five main tab options appear across the top of the Analyzer / Logs window

  From left to right:

  - **Worklist**
  - **Run**
  - **Results**
  - **Quality Assurance**
  - **Analyzer/Logs**
- **Analyzer/Logs** is the selected main tab option after log on
- Sub-options appear across the bottom of the window when a main tab is selected
- **Reagents**, **Reagent Log**, **Startup Log**, **Error Log**, **Calibration Log**, **Maintenance Log**, and **Service Log** are the sub-option tabs appearing at the bottom of the Analyzer / Logs window
- **Analyzer/Logs** and **Reagents** appear in black print to identify the current primary window being displayed
- Any window accessed using a tab is considered a primary window
- All other tabs are functional even though they appear greyed-out
- Any other tab may be selected any time, as desired

Table of Tab Options and Sub-Options

- Following table provides an overview of the various displays that can be accessed from the primary window
  - Five main tab options are in bold
  - Associated sub-options are indented under each main tab, as applicable
- Descriptions are not inclusive
- Additional information concerning these selections is detailed in other sections of this training guide as applicable

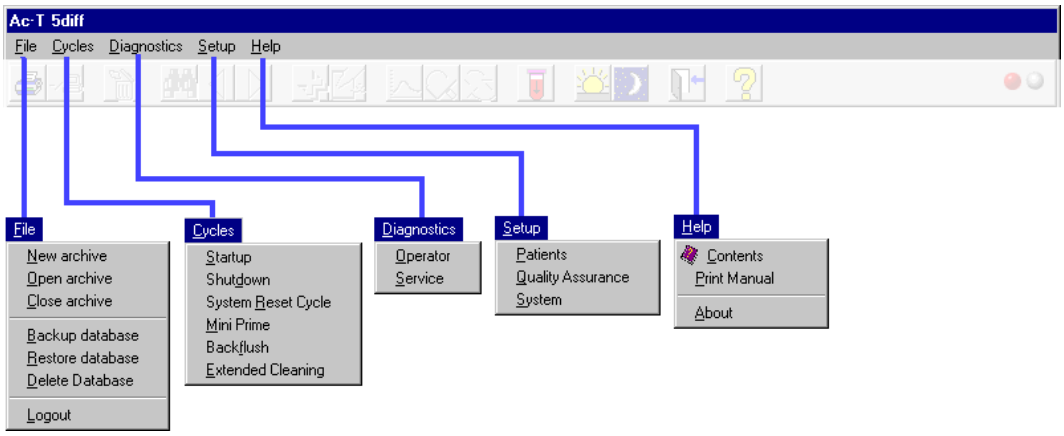
Tab Name	Purpose
Analyzer / Logs	• Provides access to various logs via the tabs at the bottom of the window.
Reagents	• Displays current reagent status and the background status of the last completed Startup. • This is the default display.
Reagent Log	• Tab used to access the Reagent Log display. • Maintains most recent 50 entries displaying the most recent log entry at the top of the log and the oldest entry at the bottom of the log. • Rollover is first in, first out when the 51st entry is added. • Additional information about reagents and the Reagent Log is located under the REAGENTS heading towards the end of this topic.
Startup Log	• Tab used to access the Startup Log display. • Maintains most recent 50 entries displaying the most recent log entry at the top of the log and the oldest entry at the bottom of the log. • Rollover is first in, first out when the 51st entry is added. • Additional information about Startup and the Startup Log is located in Topic 2 (Startup / Shutdown).
Error Log	• Tab used to access the Error Log display. • Maintains most recent 100 entries displaying the most recent log entry at the top of the log and the oldest entry at the bottom of the log. • Rollover is first in, first out when the 101st entry is added. • Additional information about the Error Log is located under the BASIC TROUBLESHOOTING TECHNIQUES heading in Topic 8 (Basic Troubleshooting).
Calibration Log	• Tab used to access the Calibration Log display. • Maintains the most recent 50 entries displaying the most recent log entry at the top of the log and the oldest entry at the bottom of the log. • Rollover is first in, first out when the 51st entry is added. • Additional information about calibration and the Calibration Log is located in Topic 5 (Calibration).
Maintenance Log	• Tab used to access the Maintenance display. • Maintains the most recent 100 entries displaying the most recent log entry at the top of the log and the oldest entry at the bottom of the log. • Rollover is first in, first out when the 101st entry is added. • Additional information about preventive maintenance and the Maintenance Log is located under the PREVENTIVE MAINTENANCE heading in Topic 8 (Basic Troubleshooting).
Service Log	• Tab used to access the Service Log display. • Must use the Service Password to access. • Function is similar to that of the Maintenance Log. • Used by your Beckman Coulter Service Representative to maintain a record of the service performed on A^{C}•T 5diff CP hematology analyzer in your laboratory. • Maintains the most recent 100 entries displaying the most recent log entry at the top of the log and the oldest entry at the bottom of the log. • Rollover is first in, first out when the 101st entry is added.

Tab Name	Purpose
Quality Assurance	• When this tab is selected, the display provides access to various quality assurance techniques via the tabs at the bottom of the window.
Controls	• Displays recent runs (up to five) for the selected file and designated shift unless zero (24 hour shift) is selected. If zero is designated for the shift, the most recent runs (up to five) for the selected file are displayed. Most recent run is displayed at the bottom. • Shift times are configured by the individual laboratory during set up. • Control source may be commercial or patient. • Control types may be CBC or CBC/DIFF. • Control levels may be Low, Normal, or High. • Up to three thumbnail graphs are displayed at a time. ▸ Display is determined via selection of the mini-tab. ▸ Up to the last 10 data points are displayed. ▸ Runs are plotted right to left with the last included entry on the right. ▸ Data points are not displayed for any control run excluded from the control file. • Additional control information is located in Topic 4 (Quality Assurance).
Reproducibility	• Tab used to access the Reproducibility display. • Study may be done using the CBC or CBC/DIFF panel. • Reproducibility studies are done to check the instrument precision. • Precision is the ability to obtain the same results, time after time, on the same sample. • Reproducibility, precision, and repeatability are synonymous terms. • Additional reproducibility information is located in Topic 4 (Quality Assurance) and Topic 8 (Basic Troubleshooting).
Calibration	• Tab used to access the Calibration display. • Calibration is needed to establish the accuracy of the instrument. • Accuracy is how close a test result is to the true value. • Calibration is performed using a special reference material such as COULTER® A^{C}•T™ 5diff Cal calibrator. • Additional calibration information is located in Topic 5 (Calibration).
IQAP	• Tab is used to access the IQAP download screen • Refer to the Help System or Instructions For Use manual for the IQAP download procedure

Tab Name	Purpose
XM	• Tab used to access the XM display. • XM tab is only visible if the XM option is **On**. Default setting is **Off**. (**Setup** ▸▸ **Quality Assurance** ▸▸ Enter Password ▸▸ **General** tab ▸▸ **XM Options**) • XM analysis is: ▸ An on going method of monitoring your automated hematology analyzer. ▸Similar to $\overline{X}_B$ analysis in that it uses a weighted moving average of patient sample results. ▸ An optional form of quality control that your laboratory may or may not choose to use. • If your laboratory decides to use the XM option, you may choose to monitor either three parameters (MCV, MCH, and MCHC) **or** nine parameters (WBC, RBC, Hgb, Hct, MCV, MCH, MCHC, RDW, and Plt). • Laboratory must establish their own mean values. • Additional information about the XM option is located in Topic 4 (Quality Assurance).
Results	• When this tab is selected, the Result List appears. • Result List is a sortable listing of processed samples in the current active archive. • To view the Detailed Results, first highlight the desired patient then click  , the **Results/List** icon. • If the Detailed Results view is currently displayed and the operator clicks the **Results/List** icon, the Result List reappears. • There are two tabs at the bottom of the window. Results and Search Results. • Detailed information concerning how parameter results are generated and reviewed is located in Topic 7 (Data Review).
Results	• Select this tab for routine Result List activities.
Search Results	• Select this tab to search for results based on your choice of criteria: ▸ Sample ID ▸ Patient ID ▸ Patient Name • Can search either current or closed archives
Run	• When this tab is selected, the Run window appears. • Displays the results for the last sample analyzed (blank, calibration, reproducibility, control, or patient sample). • Displays the next sample ID to be processed if samples are preassigned in the Worklist or autonumbering is ON. • A Sample ID must be entered prior to processing a sample. • Detailed information concerning the processing of patient samples is located in Topic 6 (Sample Analysis).
Worklist	• When this tab is selected, the Worklist appears. A worklist is a list of work to be done. This worklist identifies the patient samples that need to be processed. • Entries may be received (via download) from a host computer or individually added by the operator. • Detailed information concerning the Worklist and its usage is located in Topic 6 (Sample Analysis).

Other Navigation Tools

- Menu bar allows access to pull-down menus

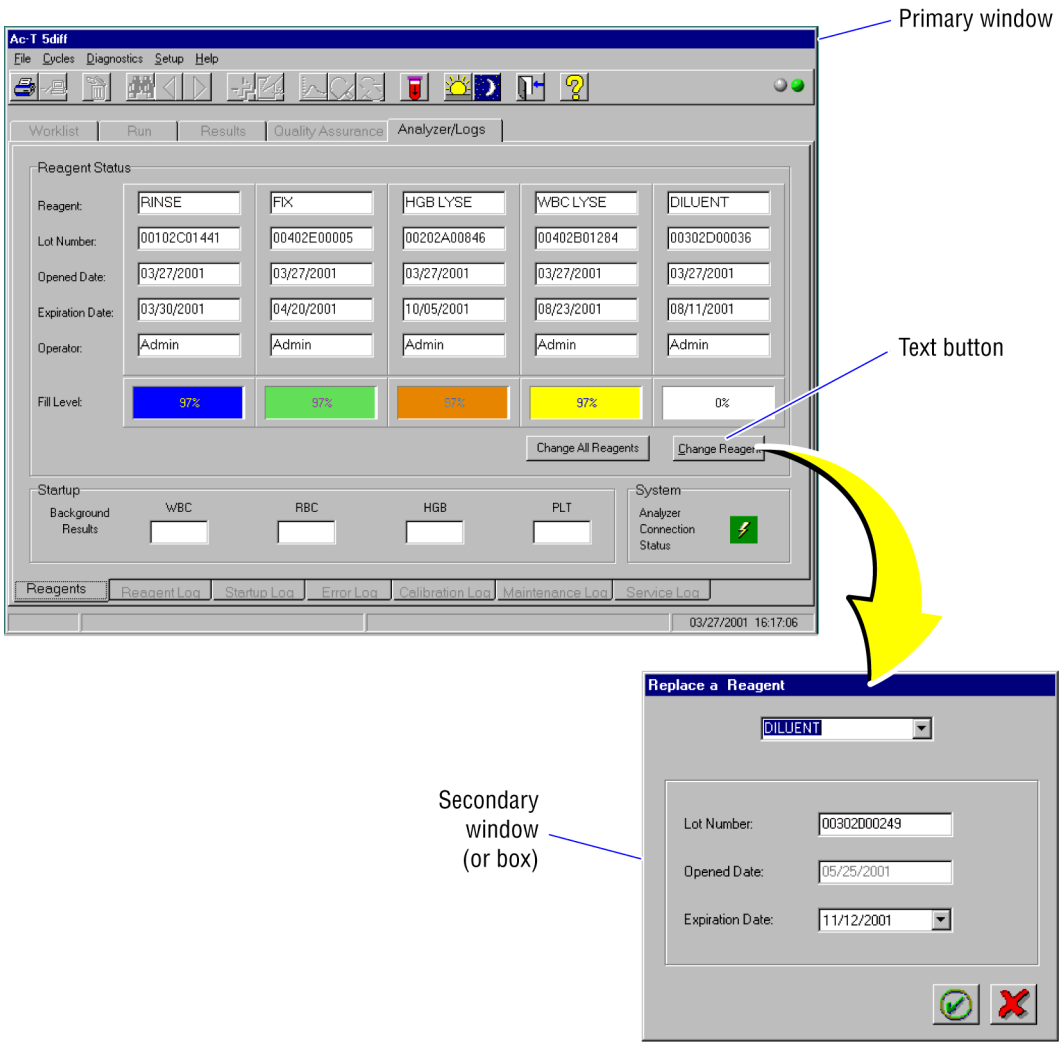


- Tool bar (may also be referred to as Icon bar)

- Text buttons access secondary windows

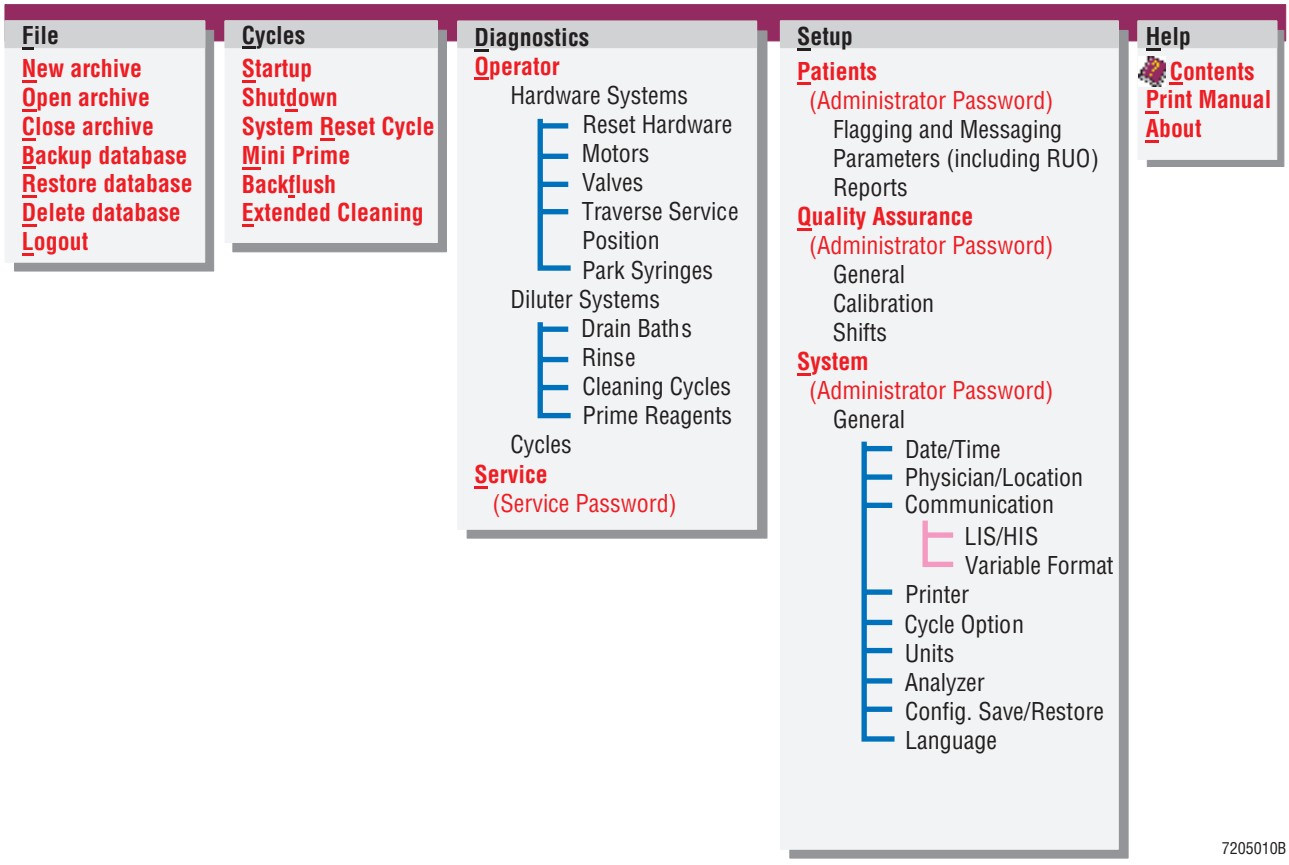
Primary Windows versus Secondary Windows

- Primary windows are those accessed by clicking on a tab; any other window (usually accessed by clicking on a text button) is a secondary window (or box)



- Menu bar and tool bar (icon bar) are:
  - Active while primary window displayed
    - Options can be selected
    - Icons that appear in color are active
  - Not active while a secondary window or box is displayed
    - Options cannot be selected
    - Icons that appear in color are not active
    - Must click either  (to save and exit) or  (to cancel and exit)

SOFTWARE MENU TREE



Software Icons

- Icons depict executable functions
- Icon activity correlates with window function:
  - If icon active, it appears in color
  - If icon inactive, coloring is removed (greyed-out)


Print


Transmit


Delete


Find


Previous


Next


Add


Edit


Results / List


Validate


Rerun Sample


Load Sample


Startup


Shutdown


Logout or Exit

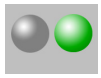

Help


Cancel


OK


Save


Restore Defaults


System State
(Double-click to quickly access the Error Log)

ONLINE HELP

Using the Online Help System on Your A^{C}•T 5diff CP Hematology Analyzer

- Help system is an electronic version of the Instructions For Use manual that includes:
  - Table of contents
  - Index for finding information quickly
  - Glossary of definitions
- Access online Help one of two ways:
  - Click  on the icon bar

    or
  - From Menu bar, select **Help ▸▸ Contents**

  Note: In the event that you cannot access the A^{C}•T 5diff Cap Pierce Online Help System from your instrument, contact your Beckman Coulter Representative.
- Three options available from the Help menu:
  - **Contents**

    Allows operator to search for information on specific system-related topics
  - **Print Manual**

    Allows operator to print the Instructions For Use manual in whole or in part (prints with the same page breaks and document structure as printed document available by order)
  - **About**

    Shows the software version currently installed on Workstation

Using Your A^{C}•T 5diff CP Hematology Analyzer Operator Manuals CD-ROM

- An Operator Manuals CD-ROM was shipped with your instrument
- CD-ROM contains:
  - COULTER® A^{C}•T™ 5diff CP Hematology Analyzer Instructions For Use manual (.PDF and .HTML files)
  - COULTER® A^{C}•T™ 5diff CP Hematology Analyzer Host Transmission Specification, English (.PDF file)
  - Adobe® Acrobat® Reader 4.0 (or higher) for reading the .PDF files
- Allows you to access the same information you can access through the Help menu on the A^{C}•T 5diff CP Workstation from any compatible personal or office desktop computer that meets the requirements listed in the Minimum System Requirements for Using the CD-ROM

ATTENTION: The Workstation PC that is part of your laboratory's A^{C}•T 5diff CP system is configured to work only with the A^{C}•T 5diff CP system and is not intended for other PC operations, such as running other software applications, playing computer games, or accessing the Internet. **This means that you cannot read the contents of this CD-ROM on the A^{C}•T 5diff CP Workstation.** You must use a different personal or office desktop computer.

Minimum System Requirements for Using the CD-ROM

- **Do not use the CD-ROM on your A^{C}•T 5diff CP Workstation**
- You can use this CD-ROM on any PC that meets these *minimum* requirements:
  - Microsoft® Internet Explorer 4.0 (or higher) for displaying the .HTML files
  - An IBM® PC or compatible with a CD-ROM drive
  - A Microsoft® operating system: Windows® 95, Windows 98, Windows 2000, Windows ME, or Windows NT® 4.0 (or higher)
  - 32 MB RAM
  - Display settings: 800 x 600 and 256 colors

Launching the Manual from the CD-ROM

- **CD-ROM contents do not automatically install on your PC**
- Can view or print direct from the CD-ROM
  - View option is an .HTML file that looks and responds the same as your A^{C}•T 5diff CP Workstation online Help system
  - Print option is a .PDF file that prints the Instructions For Use manual in whole or in part (prints with the same page breaks and document structure as the printed document that is available by order)
- Contact your Beckman Coulter Representative if you cannot access the Instructions For Use manual on the CD-ROM

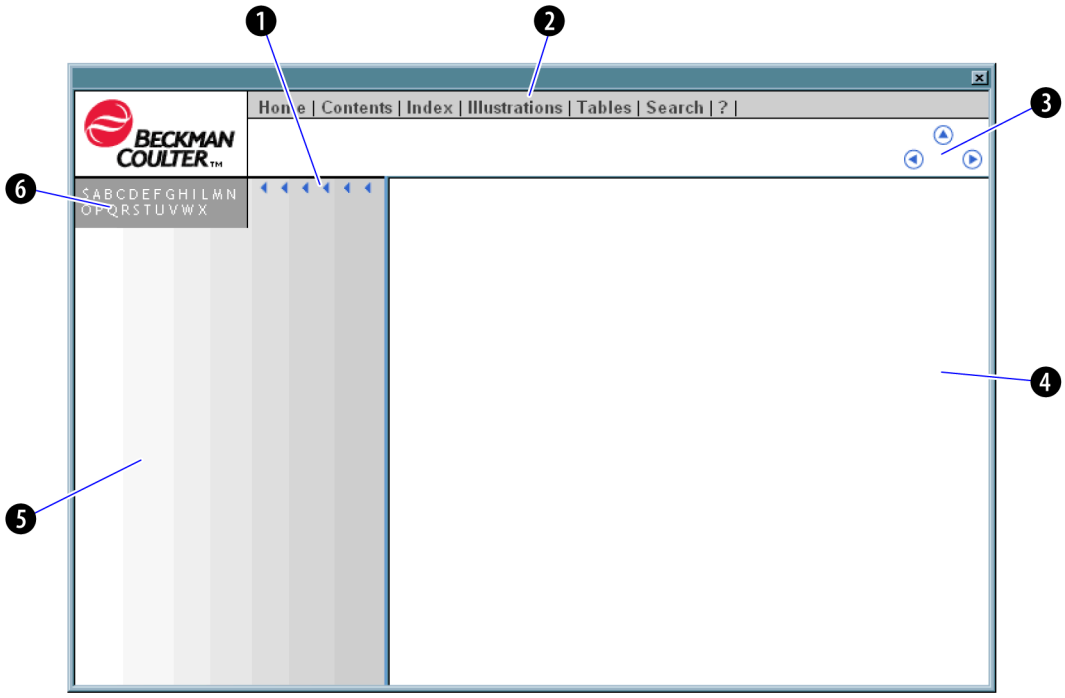
Viewing the Instructions For Use Manual

- Viewing the Instructions For Use manual using your A^{C}•T 5diff CP online Help system or the View file on the CD-ROM is the same
- Both are .HTML files
- When the Help window is accessed, operator may:
  - Move or resize the window
  - Open or close topic panes
- Screen that appears when an operator
  - From the Menu bar, selects **Help ▸▸ Contents**

    or
  - From the icon bar, clicks 

Help Screen

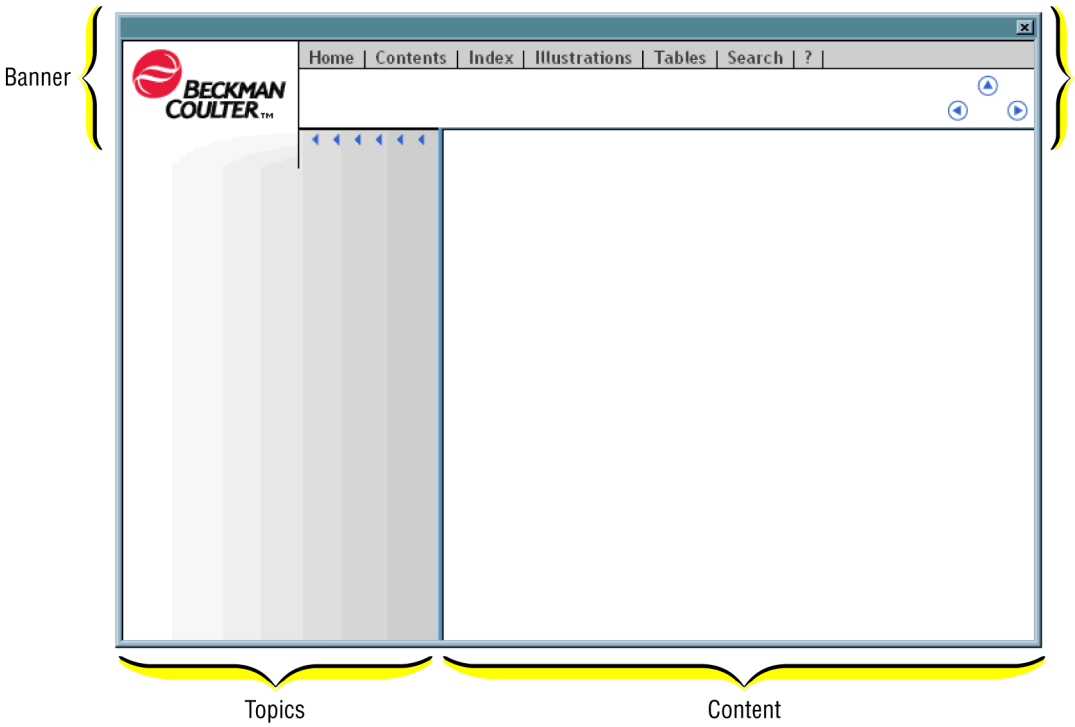
- Screen that appears when **Contents** is selected from **Help** menu:



1. Closes the topics pane when selected
2. Displays the menu options for the online Help
3. Allows an operator to navigate through the Help
4. Displays the contents of the selected Help topic
5. Displays the topics (contents, illustrations, or tables)
6. Displays the Index letters (only when Index is selected)

General Instructions for Using Help

- Help window is divided into three panes: **Banner**, **Topics**, and **Content**



- **Banner** pane contains the Help menu and navigational arrows
- **Topics** pane displays different lists based on the Help menu options selected
- **Content** pane displays information on the selected topic
- Easiest way to find specific information is to use the contents or index lists

Help Menu Options

Home
- Displays title screen

Contents
- Displays list of top-level subject headings
- Select a top-level heading to expand and view its sub-topic headings
- Select a sub-heading to display information about that topic in the Content pane

Index
- Displays list of index entries sorted alphabetically
- From the box that appears, select a letter or symbol to display an alphabetically sorted list of index entries that start with that letter / symbol
- Scroll through the topics and select the number after an index entry to view that topic

Illustrations
- Displays list of illustrations from all topics
- Scroll through the list and select the illustration you want to view

Tables
- Displays list of tables from all topics
- Scroll through the list and select the table you want to view

Search
- Used to locate specific words
- Click inside the **Search for:** text box and type one or more words separated by spaces; do not use punctuation marks
- Select **OK** button to begin search through the Help topics
- When search complete, lists all the topics that contain those specific words
- Scroll through the list and select the topic you want to view

?
- Allows operator to view Help file for using Help

Navigational Aids

- Click  to display the previous topic in the sequence
- Click  to display the next topic in the sequence
- Click  to display the previous link
- Click  to slide the Topics pane off the screen
  - Selecting an option from the Help menu slides Topics pane back into the screen
  - Icon may not be visible in some browsers
  - Alternative method to reduce/enlarge the width of the Topics pane is to use slider bar located between the panes

➤ ONLINE HELP SYSTEM QUICK REFERENCE

Accessing Online Help

ATTENTION: In the event that you cannot access the A^{C}•T 5diff Cap Pierce Online Help System, contact your Beckman Coulter Representative.

Using the COULTER® A^{C}•T™ 5diff CP Workstation

- Click  to display the Help screen

Using Your COULTER A^{C}•T 5diff CP Hematology Analyzer Operator Manuals CD-ROM

ATTENTION: Do not use this CD-ROM on your A^{C}•T 5diff CP Workstation.

1. Turn on your personal or office desktop PC and allow it to boot up. Refer to the PC manual for instructions, as needed.
2. Insert the CD-ROM into the CD-ROM drive.
   - If Autorun is enabled on your PC, the PC automatically launches the program.
   - If Autorun is disabled on your PC, do steps 3 through 4.
3. Locate the **Start.HTM** file (*CD-ROM drive letter*:\Start.HTM):
   - Click the Windows **Start** button.
   - Click **Run**.
   - Click **Browse** and locate the CD-ROM drive.
   - Double-click the CD-ROM drive letter (usually **D** or **E**).
4. Double-click **Start.HTM** to launch the program.
5. Click **For Viewing** (or **For Printing**) as desired.

Moving or Resizing the Help Screen

1. Access online Help.
2. To move the Help window:
   - Click the title bar and hold down the left mouse button.
   - Move the mouse to drag the window to a new location.
   - Release the mouse button when the window is located as desired.
3. To resize the Help window:
   - Move the cursor over the window border until the arrows appear.
   - Click the left mouse button and drag the border to resize the window.
   - Release the mouse button when the window is the desired size.

Opening / Closing the Topics Pane

1. Access online Help.
2. Click the arrows to close the topics pane.
3. Click **Contents**, **Index**, **Illustrations**, **Tables**, or **Search** to re-open the topics pane.

Viewing Help Topics

1. Access online Help.
2. Navigate through Help as needed:
   - Click **Contents** to browse through topics by heading.
   - Click **Index** to see a list of index entries.
   - Click **Tables** to see a list of tables.
   - Click **Illustrations** to see a list of illustrations.
   - Click **Search** to search for words or phrases.
   - Click **Home** to return to main Help screen.
3. Move through topics as needed:
   - Click  to display the previous topic in the sequence.
   - Click  to display the next topic in the sequence.
   - Click  to display the previous link.
4. View referenced or related topics:
   - Click the highlighted words in a topic body to link directly to the referenced topic.
   - Click a word in the highlighted hierarchy – at the top of the topic – to change topic levels. The current topic's position also appears in the hierarchy.
5. To view information not visible in the Help window, scroll through the window by using the scroll bar.

Using the Contents Option

1. Access online Help.
2. Click **Contents** to browse through topics by heading.
3. Select a top-level heading to expand and view its sub-topic headings.
4. Select a sub-heading to display information about that topic in the Content pane.
5. Use the scroll bar to scroll through the information.

Using the Index Option

1. Access online Help.
2. Click **Index**.
3. Click the letter corresponding with the list of index entries you want to see.
4. Scroll through the topics as needed.
5. Click a number after the index entry to display information about that topic.

Using the Tables Option

1. Access online Help.
2. Click **Tables** to display the list of tables.
3. Scroll through the tables to locate the table you want to view.
4. Click the table to display it on the screen.

Using the Illustrations Option

1. Access online Help.
2. Click **Illustrations** to display the list of illustrations.
3. Scroll through the illustrations to locate the table you want to view.
4. Click the illustration to display it on the screen.

Using the Search Option

1. Access online Help.
2. Click **Search** to display the search field.
3. Type the word you want to locate.
4. Click **Search**. If the word you typed appears in the document, each instance where it appears will be shown.

Printing Operator Manuals

To Print the Instructions For Use Manual from the Workstation

Note: Prints with the same page breaks and document structure as the printed document that is available by order.

1. At the Menu bar, click **Help ▸▸ Contents ▸▸ Print Manual**.
2. Click **Print**.
3. Define what pages you want to print. You can print all or part of the document.
4. Follow the instructions on the screen.

To Print Operator Manuals from the CD-ROM

Note: Prints with the same page breaks and document structure as the printed document that is available by order.

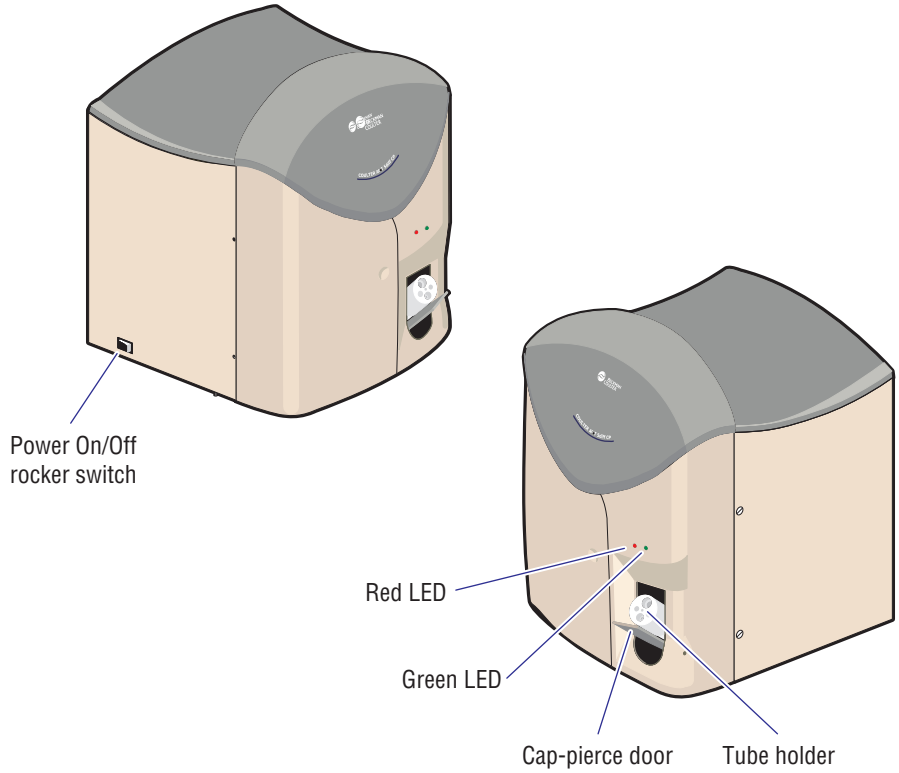
ATTENTION: Do not use this CD-ROM on your A^{C}•T 5diff CP Workstation.

1. Turn on your personal or office desktop PC and allow it to boot up.
   Refer to the PC manual for instructions, as needed.
2. Insert the CD-ROM into the CD-ROM drive.
   - If Autorun is enabled on your PC, the PC automatically launches the program. (The contents of this CD-ROM are not installed on your PC.)
   - If Autorun is disabled on your PC, do steps 3 through 4.
3. Locate the **Start.HTM** file (*CD-ROM drive letter*:\Start.HTM):
   - Click the Windows **Start** button.
   - Click **Run**.
   - Click **Browse** and locate the CD-ROM drive.
   - Double-click the CD-ROM drive letter (usually **D** or **E**).
4. Double-click **Start.HTM** to launch the Home page.
5. Select a language.
6. Under the name of the manual you wish to print, click **For Printing**.
7. Define what pages you want to print. You can print all or part of the document.
8. Follow the instructions on the screen.

➤ ➤

ANALYZER

Controls and Indicators



WARNING Risk of operator injury when covers and doors are not closed and secured in place before you operate the instrument. Ensure all covers and doors are closed and secured before operating the instrument.

POWER On/Off Rocker Switch

- Main POWER On/Off switch located on the left side of the Analyzer
- Recommend the Main Power be left on at all times to increase the stability of electronic components

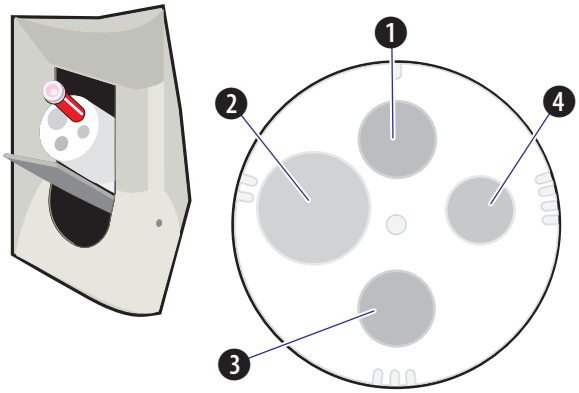
Red and Green LEDs (Light Emitting Diodes)

- Used to quickly identify instrument status
- If either light is glowing, Main Power is ON; if neither light is glowing, analyzer power is OFF
- When green LED is glowing, instrument is ready for operation
- When red LED is glowing, instrument is busy or in a standby state

Tube Holders

- Two tube holders are available for accommodating various size specimen tubes, microcollection devices, and control vials
- Identifying characteristics
  - Tube holder #1 has one dot in its center; tube holder #2 has two dots
  - Sample positions within the tube holder are notched to correlate with the number assigned to that position; for example, sample position 3 has three notches next to its tube slot
  - Slot numbering sequence in tube holder #1 is counterclockwise; slot numbering sequence in tube holder #2 is clockwise

Tube Holder #1

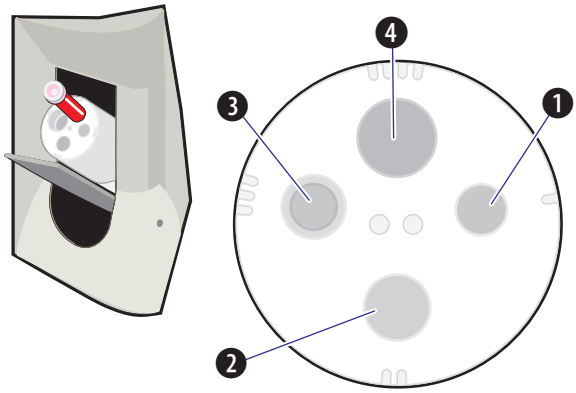


Slot Designations for Tube Holder #1

1. Position 1
2. Position 2
3. Position 3
4. Position 4

Note: Tube Holder #1 has one dot in the center.

Tube Holder #2



Slot Designations for Tube Holder #2

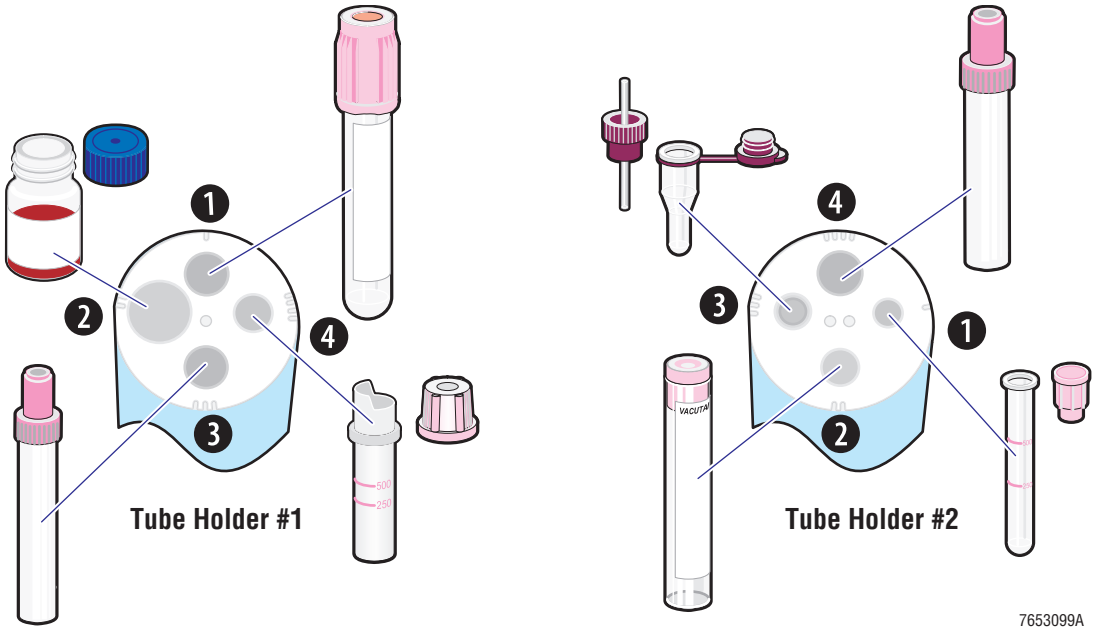
1. Position 1
2. Position 2
3. Position 3
4. Position 4

Note: Tube Holder #2 has two dots in the center.

- Tube holders are interchangeable
  - To remove, rotate and pull the holder off the metal shaft
  - To exchange, slide the desired holder over the metal shaft then rotate the tube holder until it is properly seated

Approved Collection Devices and Control Vials

- Each specimen tube, microcollection device, and control vial approved for use with these tube holders have an assigned slot in the tube holder



- Detailed list of whole-blood collection devices and control vials approved for use with these holders is available in the Instructions For Use manual, Appendix D
  - Lists whole-blood collection devices and control vials compatible with the cap-pierce mechanism used on the A^{C}•T 5diff CP hematology analyzer
  - List is not a recommendation for using one tube in preference to another nor does it guarantee the acceptability of the specimen tube to produce quality results
- Summarized tube list is available in the Sample Analysis section of this training guide
- For those evacuated collection tubes, microcollection devices, and calibrator vials without pierceable stoppers or caps:
  - **Stopper or cap must be removed prior to sampling** (open-vial sampling)
  - **Open-vial sampling increases the operator's exposure to the whole-blood specimen which is considered a biohazardous material**

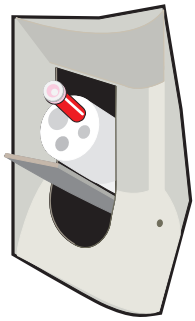
Operation

Single Point Aspiration System

- Means there is **only one pierce position**
- Specimen tube vial must be in the pierce position for the sample to be aspirated
- If the specimen tube is in any other slot:
  - Cap-pierce door may not close
  - If cap-pierce door does close (which initiates the cycle), analyzer aspirates air and results reflect analysis of reagents only (background results)

Pierce Position

- Single-point of aspiration is the **12 o'clock** position



- Holder can be manually rotated (clockwise or counterclockwise as needed) to position the desired tube slot in the pierce position
- An open or closed collection device or vial may be placed in the tube slot
- Sampling probe pierces a closed-vial specimen tube before aspirating the sample
- May run specimen tubes with or without bar-code labels
- If a bar-code label is used, barcode must be scanned before placing the specimen tube in the tube holder

Cap Pierce Door

- Closing the door initiates a cycle
- When the door is pushed into its closed position:
  - Red and green LEDs alternately flash approximately six seconds then the red LED glows steady the remainder of the cycle
  - Four switches inside the cap-pierce mechanism sense the indentations on the side of the tube holder to determine which tube holder is inserted and the size of the tube slot located in the pierce (aspirate) position (Analyzer's computer then knows the downward movement needed to completely submerge the sampling probe's tip in the specimen)
  - Door opens automatically when the aspiration is complete
- If the door is closed and the Analyzer is not cycling, click  to open the door

Panels

- Samples are processed as either a CBC/DIFF or CBC panel

CBC/DIFF Panel

- Default panel
- Aspirates 53 µL of sample from the specimen tube (or vial) via the sampling probe
- Reports 20 parameters
  - CBC parameters: WBC, RBC, Hgb, Hct, MCV, MCH, MCHC, RDW, Plt,
  - and MPV
  - DIFF parameters: NE%, NE#, LY%, LY#, MO%, MO#, EO%, EO#, BA%, and BA#
- If RUO (Research Use Only) is enabled, six additional parameters are reported
  - Qualitative parameters that must not be used for the purpose of diagnosis
  - RUO parameters include Pct, PDW, IMM%, IMM#, ATL%, and ATL#

CBC Panel

- Must be selected before the specimen is processed
- Aspirates 30 µL of sample from the specimen tube (or vial) via the sampling probe
- Reports 10 CBC parameters only
  - WBC, RBC, Hgb, Hct, MCV, MCH, MCHC, RDW, Plt, and MPV
- If RUO (Research Use Only) is enabled, two additional parameters are reported
  - Qualitative parameters that must not be used for the purpose of diagnosis
  - RUO parameters include Pct and PDW
- Final results are calculated automatically

NOTES

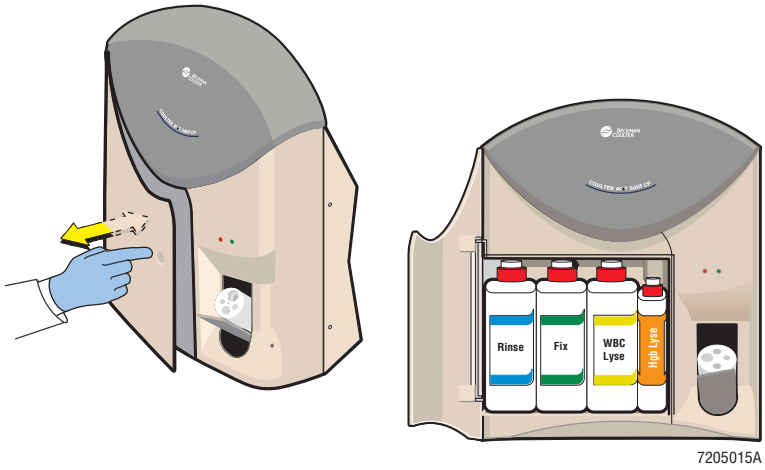
REAGENTS

To obtain the optimum performance characteristics stated in the Instructions For Use manual, use the following reagents on your A^{C}•T 5diff CP hematology analyzer.

Reagent	Label Color	Function	CBC	DIFF
A^{C}•T 5diff Diluent	Purple	• Isotonic solution that dilutes the sample • Stabilizes cell membranes • Conducts aperture current • Provides the sheath medium for focused flow through the flow cell • Rinses analyzer components after analysis • Contains less than 0.1% sodium azide which may cause an explosion if not properly flushed down the drain with large volumes of water	✓	✓
A^{C}•T 5diff Hgb Lyse	Orange	• Disrupts erythrocytes (lysis) • Frees hemoglobin • Reduces cellular debris • Converts hemoglobin to a stable cyanide-containing pigment • Contains potassium cyanide, a quarternary ammonium salt; special instructions are provided for proper disposal	✓	
A^{C}•T 5diff WBC Lyse	Yellow	• Dilutes the sample for the total WBC count • Specifically differentiates between the basophils and other white blood cells for the basophil percentage analysis • Lyses red blood cells • Reduces cellular debris	✓	✓
A^{C}•T 5diff Fix	Green	• Lyses red blood cells • Preserves the white blood cells in their native state • Stains the granules of eosinophils, neutrophils, and monocytes • Provides the dilution used to differentiate the white blood cell subpopulations using the absorbance cytochemistry technology		✓
A^{C}•T 5diff Rinse	Blue	• Cleans and rinses the diluter parts • Prevents protein buildup • Eliminates routine aperture bleaching	✓	✓

Reagent Container Locations

Accessing the Reagent Compartment



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- Pushing on the reagent compartment door allows access to on-board reagent bottles

  From left to right:

  - A^{C}•T 5diff Rinse reagent bottle
  - A^{C}•T 5diff Fix reagent bottle
  - A^{C}•T 5diff WBC Lyse reagent bottle
  - A^{C}•T 5diff Hgb Lyse reagent bottle
- 20 liter A^{C}•T 5diff Diluent reagent container generally sits on floor below the instrument
- If a waste container is being used, this container tends to be placed near the diluent container

Storage and Handling

- Always handle the reagents as instructed in the Instructions For Use manual
- All reagents should be stored and used at an ambient temperature of 18°C to 25°C
- Discard any reagent that has been or that you suspect has been frozen.

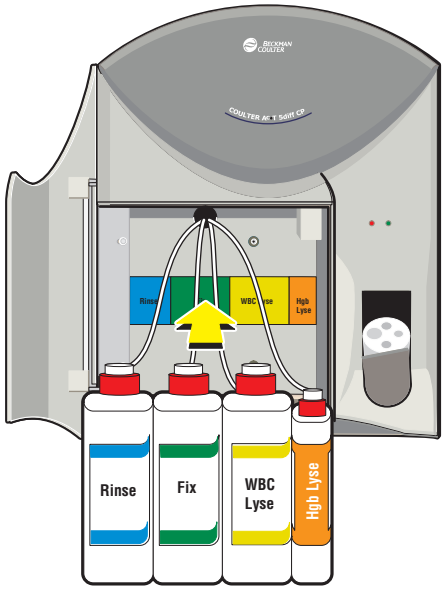
Before Using a Reagent

- Gently mix by inversion
- Allow sufficient time for any microbubbles to dissipate

  Note: Microbubbles may increase the background count and affect parameter results.

When Changing a Reagent

- Do not pool reagents; pouring reagents from one container to another may contaminate the new reagent container which will cause unacceptable background results, especially for platelets
- Always make sure the label on the stopper assembly tubing correlates with the reagent name on the new container
- When transferring the stopper assembly from the old container to the new container, do not touch the pickup tubes or lay the assembly on an uncovered tabletop or the floor
  - If the pickup tube portion of a stopper assembly is contaminated during transfer, bacterial and/or fungal growth may occur inside the reagent container
  - Contamination may cause unacceptable background results especially for platelets
  - If a pickup tube touches you or anything outside the container, flood the pickup tube with distilled water then wipe it with a lint-free tissue before inserting it in the new reagent container
- Always correlate the position of the color-coded reagent bottle with the color-coded label attached to the rear of the reagent compartment



IMPORTANT Risk of misleading results. If a reagent is connected in the wrong position, contamination of the other reagents occurs which means all reagents must then be replaced. When replacing on-board reagents, always correlate the position of the color-coded reagent bottle with the color-coded label attached to the rear of the reagent compartment and with the reagent-name label on the reagent pickup tubing.

Reagent Lot Numbers

- Reagent lot numbers contain 11 alphanumeric characters consisting of five numeric digits, an alphabet letter, and five more numeric digits

  For example, lot number for A^{C}•T 5diff Diluent might be 00102D00002
- If an operator tries to enter an invalid lot number, a message appears

Reagent Log

- Click on **Analyzer / Logs** tab ▸▸ **Reagent Log** tab to view the log
- Maintains most recent 50 entries displaying the most recent log entry at the top of the log and the oldest entry at the bottom of the log
- Rollover is first in, first out when the 51st entry is added
- Log contains the following information fields

  Note: It is necessary to scroll right to see some of these fields.

Information Fields

Field	Function
Date/Time	Displays when the reagent change was made
Reagent	Displays operator entry which identifies the reagent that was changed
Lot Number	Displays operator entry that identifies the specific reagent container connected to the Analyzer
Opened Date	Displays the date the reagent in that container began to be used for sample analysis
Opr	Displays the User name logon information in the Workstation when the reagent was changed
Expiration Date	• Displays the new reagent container's expiration date as manually selected by the operator from a drop-down calendar • If operator does not select a date, current date is displayed
Comment	Displays any comments entered by the operator changing the reagent: • Optional entry that provides an opportunity for the operator to type a personal observation or appraisal • Operator may use the **Add Comments** button to access the Add Comments box, as desired • If the Prompt User For Comments check box is checked (✓), the Add Comments box pops up automatically during the entry process • If the **Prompt User For Comments** check box is checked (✓), the Add Comments box pops up automatically to prompt the operator to enter a comment **Note:** When Add Comments box is displayed, operator cannot access any other window until they either: ▸ Enter a comment then click  to save and exit the box or ▸ Click  to exit the box without entering a comment

Before Disposing of a Reagent or Its Container

- Always dispose of reagents and containers according to your laboratory's guidelines
- If disposing expired A^{C}•T 5diff Hgb Lyse reagent, always follow the instructions for eliminating cyanides before discarding the expired reagent according to your laboratory's guidelines

  Note: See the Instructions For Use manual for specific instructions.

Before Disposing of a Waste Container

- If waste is being collected in a container, it must always be considered a biohazard and therefore, must be treated as a biohazard
- To disinfect 20 liters of waste liquid:
  - 250 mL of sodium hypochlorite solution (12% available chlorine) should either be placed in the empty container before it is connected to the instrument or before moving a full waste container to another location
  - Sodium hypochlorite solution with 12% available chlorine may be ordered from a chemical company or swimming pool chlorine is an acceptable substitute
  - 500 mL of a good quality household bleach may also be used

    Note: Since household bleach only has about 6% available chlorine, the volume must be doubled.
- If the waste container is to be capped:
  - Always follow the instructions for neutralizing the waste before capping the container for disposal according to your laboratory's guidelines
  - Non-neutralized waste may produce gas which can build up pressure in a capped container
  - Neutralize waste after removing the waste container but before capping the container for disposal

  Note: See the replacement procedure in Chapter 11 of the Instructions For Use manual or the Replacing a Waste Container Summary for specific instructions.

➤ CHANGING REAGENT SUMMARY

Note: If **all** reagents are being changed, see Chapter 11 of the Instructions For Use manual.

1 Verify a New Reagent Container is Needed

- Display the Reagents window by choosing **Analyzer/Logs** tab ▸▸ **Reagents** tab.
- Check reagent levels to determine which reagent triggered the *REAGENT LOW LEVEL* message.
  - Check all levels because more than one may be low.
  - Obtain a new unopened container of each reagent that according to your laboratory's protocol requires the reagent to be changed.

2 Replace the Reagent Container

- Open the new reagent container.
- Remove the stopper assembly from the old container then transfer it directly to the new container and tighten to ensure an adequate seal.

  Note: If the lower part of the assembly touches you or anything outside the container, flood that lower part of the assembly with distilled water then wipe it with a lint-free tissue.
- Put the cap from the new container onto the empty container.
- Properly dispose of the empty bottle according to your laboratory's protocol.

3 Record the New Reagent Information

- At the Reagents window, click **Change Reagent**.
- Choose the reagent that was changed from the drop-down menu.
- Type the new lot number.

  Note: Today's date automatically appears in the Opened Date box.
- Choose the Expiration Date (or open container stability) from the drop-down calendar.
  - For all reagents except A^{C}•T™ 5diff Hgb Lyse reagent, select the date printed on the reagent container.
  - For the A^{C}•T 5diff Hgb Lyse reagent which has a 90 day open container stability, select the date that is 90 days from today.
- Click  to save the information and return to the Reagents window.

4 Wait for the Reagent Lines to Prime

- System automatically primes the reagent and updates the level indicator.

  Note: Reagent level may not be displayed at 100% due to this priming.
- Once priming is complete, continue normal operation.

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➤ REPLACING A WASTE CONTAINER SUMMARY

Note: If the waste container is not full and the waste alarm is chirping (beeping) at regular intervals, replace the 9 Volt alkaline battery in the waste sensor alarm.

1 Power Off the System

At the Workstation

- Click  .
- Select **Quit Application**.
- Press Enter or click  .
- Wait while the Workstation closes its program.
- When the Begin Logon box appears, press Ctrl + Alt + Delete simultaneously.
- Click **Shut Down** at the Logon Information box.
- Select **Shutdown** then click **OK** at the Shutdown Computer box.
- When the *It is now safe to turn off your computer* message appears, power the Workstation computer off.

At the Analyzer

- Toggle the Power On/Off rocker switch OFF (position O). This rocker switch is located at the base of the left side panel.

2 Replace the Waste Container

- Clearly mark or apply a waste label to an empty 20 L container.
- Using biohazardous precautions:
  - Carefully remove the cap (with waste sensor attached) from the full waste container.
  - Transfer the waste assembly directly to the empty waste container and tighten.

3 Neutralize and Disinfect the Waste Before Capping the Container

- For 20 liters of waste liquid, add the following to the waste container:
  - 250 mL of sodium hypochlorite solution (if 12% available chlorine) **or** 500 mL of sodium hypochlorite solution (if 6% available chlorine) to disinfect waste.
  - 50 mL of 200 g/L sodium hydroxide solution to prevent gas from forming if the container is being capped.
- Dispose of the biohazardous waste according to your laboratory's protocol.

4 Power Up the System

At the Analyzer

- Toggle the Power ON/OFF rocker switch ON (position –). This rocker switch is located at the base of the left side panel.
- Verify the red LED is glowing steady.

At the Workstation

- Power ON the Workstation computer.
- Wait while the computer performs its internal checks.
- When the Begin Logon box appears, press Ctrl + Alt + Delete simultaneously.
- Type the User name and Password then press Enter or click **OK** to log on.
- Type your 3-character (alphanumeric) Operator ID.
- Press Enter or click  .
- Wait for the Reagents window to appear.
- Verify the background behind the "lightening bolt" is green.

  Note: Analyzer and Workstation should begin communicating within 30 seconds. In the upper right corner, the right circle should also be green.

5 Prepare the System for Processing Samples

- If the Automatic Startup function is enabled:
  - Startup routine was automatically activated when the power was toggled back ON.
  - *Cycle in progress : Startup* status bar provides a visual display of how close the routine is to completion.
  - When complete, resume normal operation.
- If the Automatic Startup function is not enabled:
  - At the Menu bar, select **Cycles ▸▸ Mini Prime**.
  - *Cycle in progress : Mini Prime* status bar provides a visual display of how close the routine is to completion.
  - When complete, resume normal operation.

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OBJECTIVES

When the subject is complete, you will be able to . . .

Startup

- Initiate a Startup.
- Recognize an out-of-limit result on the Reagents window.
- Identify the Reagent Status, Startup, and System areas of the Reagent window.
- Cite the two ways to monitor Analyzer connection status.
- Access the Startup Log.
- Add a comment to the Startup Log.
- Explain how to handle an unacceptable background result.

Mini Prime

- Initiate a Mini Prime.
- State the difference between a Startup and a Mini Prime.
- Identify when to do a Startup versus when to do a Mini Prime.

Autoclean Function

- Explain the purpose for the Autoclean function.
- Set up the Autoclean function based on your laboratory's desired protocol.

Shutdown

- Initiate a Shutdown.
- State how often a Shutdown must be performed.
- State the minimum time the A^{C}•T 5diff Rinse reagent should stay in the instrument while in Shutdown.

NOTES

STARTUP

- Startup must be completed before running patient specimens or controls anytime A^{C}•T 5diff Rinse reagent has replaced diluent in the aperture baths
- Startup routine is a sequence of automated Analyzer cycles designed to exchange the A^{C}•T 5diff Rinse reagent left in the diluter after the Shutdown routine with A^{C}•T 5diff Diluent reagent
- Also primes the A^{C}•T 5diff Hgb Lyse, A^{C}•T 5diff WBC Lyse, and A^{C}•T 5diff Fix reagent lines to prepare the Analyzer for sample analysis
- Checks for sufficient reagent volume based on the **Daily Workload** statistics entered under **Setup ▸▸ System ▸▸ Cycle Option** tab
  - Can enter the estimated number of CBC/DIFF and CBC cycles run per day
  - Default value for CBC/DIFF Runs Per Day is 40; for CBC Runs Per Day is 10
  - Cycle estimates are used to determine if sufficient reagent is available to complete the daily workload; check is done during Startup
  - If reagent volume is insufficient to complete the estimated daily workload, the message *Reagent(s) Low. Insufficient Reagents To Complete Daily Workload.* appears
- Performs a background count and check at the end of the Startup routine and displays the results as either Passed or Failed across the bottom of the Reagents window

  Note: Background count is an analysis cycle on reagents without the blood sample.
- *Cycle in progress* status bar at the base of the Reagents window provides a visual display of how close the routine is to completion
- If Autoprint is enabled, results automatically print when the routine is complete
- Perform quality control checks before running patient specimens according to your laboratory protocol

Two Ways to Initiate a Startup

Automatic

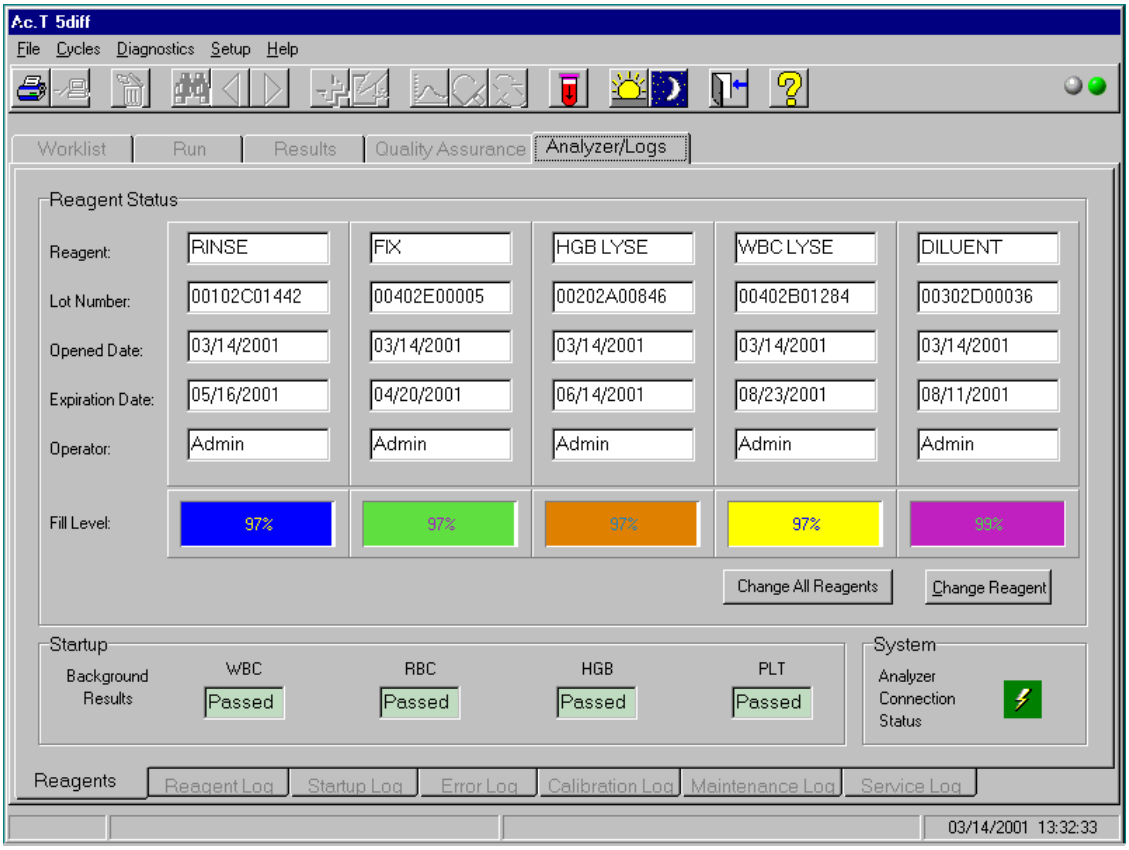
- If automatic startup is selected under System Setup and the system is powered off, when system (Analyzer then Workstation) is powered back on, software automatically performs the Startup routine
- Recommended setting
- Default setting when the instrument is installed
- To set up this automatic feature (if it is disabled):
  - At the Menu bar, click **Setup ▸▸ System**
  - Type Administrator password then press Enter or click 
  - Click **Cycle Option** tab
  - At the Startup box, click the empty box next to Automatic (✓ appears in box)
  - Click  to save the selection and exit the window

On Demand

- At the icon bar, click  to initiate a Startup routine

STARTUP SCREENS

Reagents Window



- Also referred to as the Analyzer / Logs window
- Displays system status in three boxes:
  - Reagent Status box provides an overview of the status of each reagent
  - Startup box shows the background status of the last completed Startup
  - System box provides a visual indication as to whether or not the Analyzer and Workstation are communicating

Reagent Status Box

Field	Information
Reagent	Displays the reagent name
Lot Number	Displays operator entry that identifies the specific reagent container connected to the Analyzer
Opened Date	Displays the date the reagent in that container began to be used for sample analysis
Expiration Date	Displays the new reagent container's expiration date as manually selected by the operator from a drop-down calendar; if operator does not select a date, current date is displayed
Operator	Displays the User name logon information in the Workstation when the reagent was changed
Fill Level	Displays the percentage of reagent still available in the container

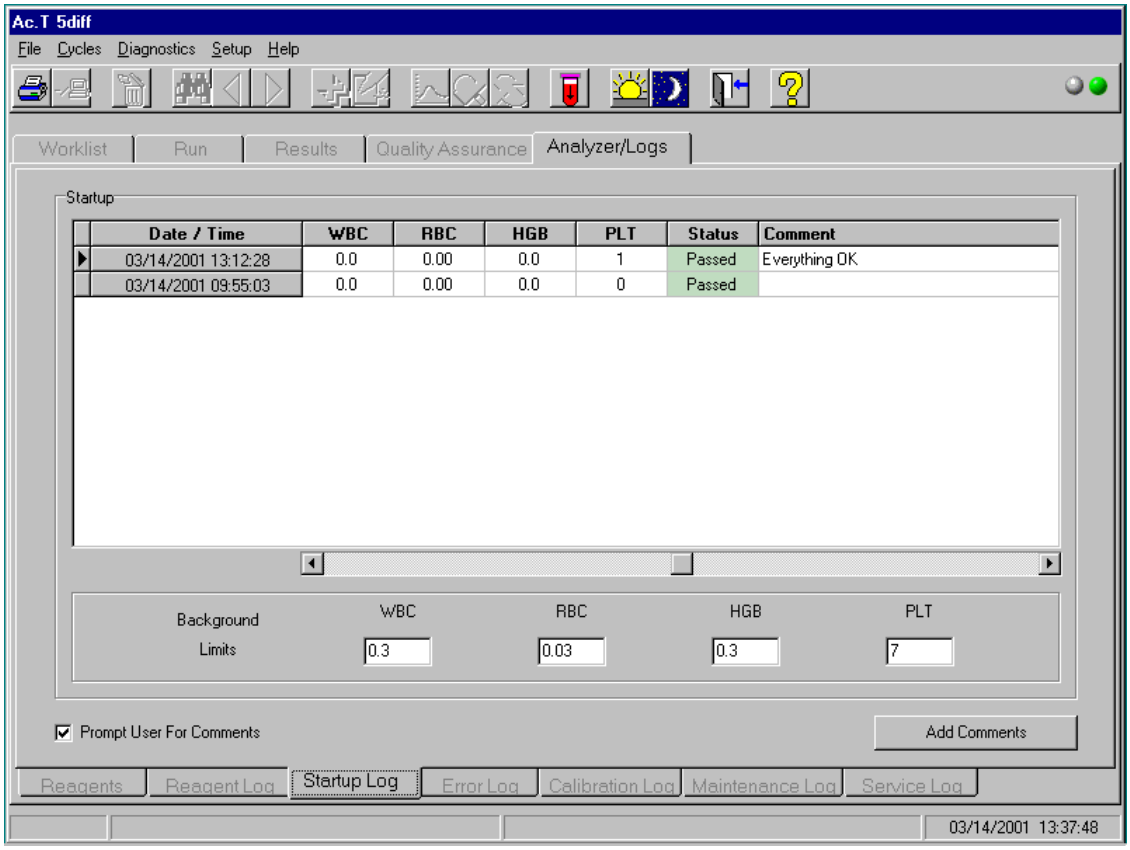
System Box

- Provides a visual of the communication connection status between the Analyzer and the Workstation:
  - If Analyzer and Workstation are communicating, background behind the "lightening bolt" is green
  - If Analyzer and Workstation are not communicating, background behind the "lightening bolt" is red
- Connection status indicated in the System box must correlate with the connection status indicated by the System State indicators (two circles) in the top right corner:
  - If background behind the lightening bolt is green, the right circle must also be green
  - If background behind the lightening bolt is red, the left circle must also be red
- Two tools for monitoring system connection status:
  - Lightening bolt only appears on the Reagents window
  - System State indicators (two circles) provide continuous system connection status information, regardless of the window being displayed

Startup Box

- Displays background status of the last completed Startup
- Shows status on a Passed/Failed basis:
  - "Passed" is displayed on a green background when a parameter is within the internal background limit
  - "Failed" is displayed on a red background when a parameter is outside the acceptable internal background limit
- If background results are not within acceptable limits after the first background check:
  - Analyzer automatically performs the background count up to two more times
  - Only the status of the final background check is recorded
- Numeric background results may be reviewed by accessing the Startup Log
  - Click on **Analyzer / Logs** tab ▸▸ **Startup Log** tab

Startup Log



- Maintains the most recent 50 entries displaying the most recent log entry at the top of the log and the oldest entry at the bottom of the log
- Rollover is first in, first out when the 51st entry is added
- Allows an operator to review numeric background results
- Preset background limits, listed at the bottom of the window, aid in the evaluation of results that fall outside these acceptable background limits

Information Fields

Field	Function
Date/Time	Displays when the Startup was completed
WBC	Displays the numeric white blood cell background result determined during that Startup
RBC	Displays the numeric red blood cell background result determined during that Startup
HGB	Displays the numeric hemoglobin background result determined during that Startup
PLT	Displays the numeric platelet background result determined during that Startup
Status	Startup background status indication: • Field contains the word "Passed" backlighted in green when all background results are within the designated Background Limits • Field contains the word "Failed" backlighted in red when one or more background results exceed the preset Background Limits
Comment	Displays any comments entered by operator performing the Startup: • Optional entry that provides an opportunity for the operator to type a personal observation or appraisal • Operator may use the **Add Comments** button to access the Add Comments box, as desired; highlight the specific log entry before clicking the **Add Comments** button • If the **Prompt User For Comments** check box is checked (✓), the Add Comments box pops up automatically at the end of Startup, after the background check is complete **Note:** When Add Comments box is displayed, operator cannot access any other window until they either: ▸ Enter a comment then click  to save and exit the box or ▸ Click  to exit the box without entering a comment

Indications of a Failed Background

- On the Reagents window:
  - Message appears in the Startup box
  - When a parameter is outside the acceptable background limit, the word “Failed” is displayed on a red background inside the parameter box
  - For example, if the WBC background exceeds the preset WBC background limit, the word “Failed” (on a red background) appears inside the WBC box
- On the Startup Log:
  - Message appears in the Status field
  - “Failed” (on a red background) appears in the Status field when one or more background results exceed the internal background limits

    Note: If “Failed” displayed on a red background appears but all the background results are acceptable, the instrument most likely made an entry on the Error Log.
  - Numeric results must be compared with the displayed background limits to determine which parameter or parameters exceeded the acceptable preset limits

How to Handle a Failed Background

If One or More Background Results Failed

- Click  to redo Startup routine
- If Startup continues to fail, contact your Beckman Coulter Representative

If All Background Results Passed but the Startup Log Status Displays Failed

Note: When “Failed” is displayed on a red background in the Startup Log Status column but all the background results are acceptable, the instrument probably made an entry on the Error Log.

- Click the **Startup Log** tab and evaluate numeric results to verify all background results are within acceptable background limits

  Note: May also click the **Run** tab to view Startup results.
- If “Failed” displayed on a red background appears in Status column but all the background results are acceptable:
  - Double-click either System State indicator to quickly access Error Log
  - Check last entry on the Error Log
  - Verify the date and time correlate with this incident
  - Locate message in the Error Messages table in the Operator’s Guide, Chapter 11
  - Complete suggested action
  - For more assistance, contact your Beckman Coulter Representative

➤ STARTUP SUMMARY

Before starting this summary, verify both Workstation and Analyzer power are ON. If the system is powered OFF, do not use this summary. Use the System Power Up Summary instead.

1 Preliminary Checks

- If the Workstation screen is blank, power on the monitor.
- Verify the green circle is displayed in the upper right corner.
- Check the waste container level.
  - If the container needs to be replaced, change the container using the Replacing a Waste Container Summary as a guide.
- Make sure the Printer is ready to print.
  - If the paper supply is insufficient, add paper.
  - Make sure the power is ON and ready to print.

2 Perform the Startup

- Click  to initiate a Startup routine.
- Monitor the *Cycle in progress : Startup* status bar, as desired.
- Hard copy of the results prints automatically if the Auto-Print function is enabled.

3 Review Results

- Once the Startup routine is complete, verify *Passed* is displayed for each parameter.
  - If *Failed* appears for any parameter, click **Startup Log** tab to evaluate numeric results.

    Note: If the **Add Comments** box automatically appears, click  to exit.
  - Click  to initiate another Startup routine.
  - If the Startup continues to fail, contact your Beckman Coulter Representative.
- Save individual or log printout if required by your accreditation agency.
- Add comments as desired.
  - If the **Add Comments** box appeared automatically, type in your personal observation or appraisal.
  - To access the **Add Comments** box, click **Startup Log** tab ▸▸ **Add Comments** button then type your personal observation or appraisal inside the box.
  - Click  to save your comments.

4 Perform Quality Control Checks

- Perform quality control checks before running patient specimens according to your laboratory's protocol.

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MINI PRIME

- Primes reagent lines including A^{C}•T 5diff Diluent, A^{C}•T 5diff Hgb Lyse, A^{C}•T 5diff WBC Lyse, and A^{C}•T 5diff Fix
- Does not preform a background count
- To initiate a Mini Prime:
  - At the Menu bar, select **Cycles ▸▸ Mini Prime**
  - *Cycle in progress : Mini Prime* status bar provides a visual display of how close the routine is to completion

STARTUP VERSUS MINI PRIME

When to do a Startup versus When to do a Mini Prime

- Startup must be done before running patient samples or controls:
  - After a Shutdown
  - Anytime the Analyzer has set idle (uncycled) more than four hours
- Mini Prime must be done before running patient samples or controls:
  - Anytime the Analyzer has set idle (uncycled) more than two hours but less than four hours
  - Anytime the Analyzer is powered off then back on again

    Note: If your laboratory selected Startup be done automatically when the Analyzer is powered up, a Mini Prime is not necessary.

AUTOCLEAN FUNCTION

- Analyzer automatically performs an Autoclean cycle after a specified number of analyses
- Default number of analyses is 75
- May select from 1 to 75 analyses based on your laboratory's desired protocol
- When the designated number of analyses is reached:
  - A^{C}•T 5diff Rinse reagent is delivered to the baths assembly
  - Cycle takes approximately 2 minutes
- To change the frequency:
  - At the Menu bar, click **Setup ▸▸ System**
  - Type Administrator password then press Enter or click 
  - Click **Cycle Option** tab
  - At the Autoclean Frequency box, highlight the current number then type the desired number of analyses between Autoclean routines
  - Click  to save the selection and exit the window

SHUTDOWN

- Shutdown is a sequence of automated Analyzer cycles designed to exchange the A^{C}•T 5diff reagents inside the diluter with A^{C}•T 5diff Rinse reagent
- Allowing A^{C}•T 5diff Rinse reagent to remain in the instrument a minimum of 30 minutes minimizes protein buildup in the instrument
- Perform a Shutdown once every 24 hours that the instrument is in use
- Startup must be completed prior to running patient specimens or controls

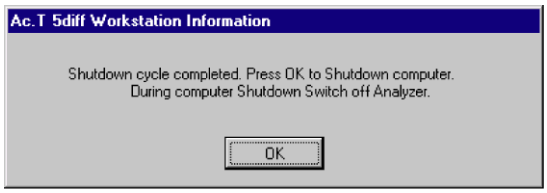
➤ SHUTDOWN SUMMARY

1 Perform Instrument Shutdown

- Click  to initiate a Shutdown routine.
- Monitor the *Cycle in progress : Shutdown* status bar, as desired.

2 Shut Down Computer then Power Off Analyzer and PC

When Shutdown is complete, a message box appears:



- Click **OK** to shut down the computer.
- Turn off the Analyzer power switch.
- Wait for the *"It is now safe to turn off computer"* message to appear.
- Turn off the PC power switch.

Notes:

- Allow A^{C}•T 5diff Rinse reagent to remain in the instrument a minimum of 30 minutes.
- Wait at least 30 seconds before performing the Power Up and Log On procedures.
- After doing Shutdown, you must do a Power Up and Startup before running patient specimens or controls.

➤➤

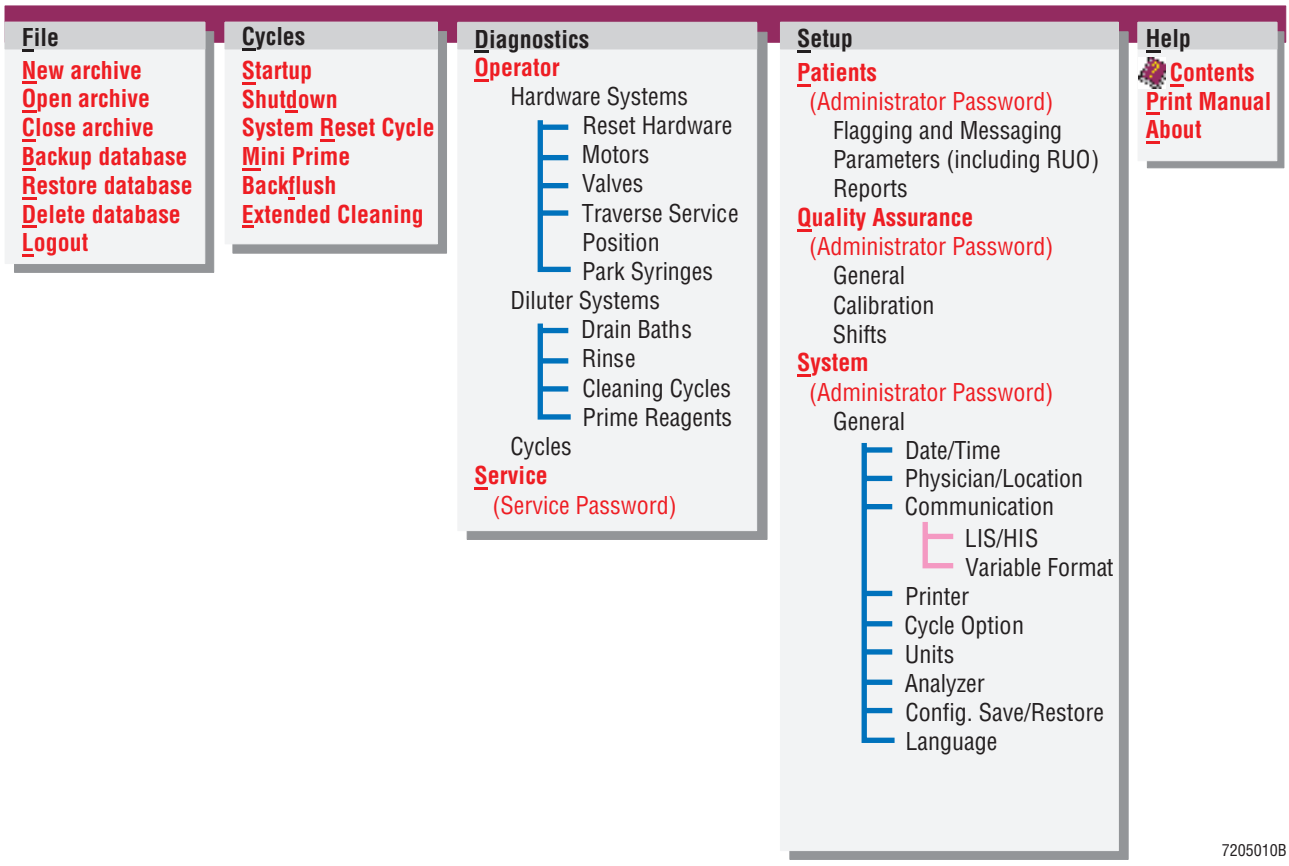
OBJECTIVES

When the subject is complete, you will be able to . . .

- Enter data under **System** including the communications definition if a host computer is in use.
- Enter data under **Quality Assurance** to reflect your laboratory's decisions regarding XM analysis, automatic printing, and shift designations.
- Enter data under **Patients** to include flagging limits, display formats, and print options.
- Set up the initial control files under **Quality Assurance ▸▸ Control** tab.

MENU TREE

This is a menu tree of the Workstation software. Use this menu tree as a reference, as needed.



SETUP MENU OPTIONS

- At the Menu bar, select **Setup**
- **Setup** drop-down menu consists of three items
  - **Patients**
  - **Quality Assurance**
  - **System**
- All Setup options require the Administrator Password 123 for access
- To save changes:
  - Click  to save the entry and remain on the window

    or
  - Click  to save the entry and exit the window
- A red prompt is displayed on the window when it's necessary to restart the application after making a change; user is automatically logged out of the application
- All the options available in the Setup area of the Workstation software are listed here
  - Refer to the menu tree on the opposite page as needed
  - Each menu item is listed with a short description
  - For details, see Appendix A of the Instructions For Use manual.

SYSTEM SETUP

- To access these options, at the Menu bar:
  - Select **Setup ▸▸ System**
  - Type **Administrator Password** then press Enter or click 
  - Click **General** tab and a group of sub-option tabs appear
- To save changes:
  - Click  to save the entry and remain on the window

    or
  - Click  to save the entry and exit the window
- For additional details, see Appendix A of the Instructions For Use manual.

Date/Time Tab

- Allows the operator to adjust the current date and/or time (for example, daylight savings time)
- Allows the operator to select a date and/or time format for display
  - Default date setting is MM/dd/yyyy (month/day/year)
  - Default time setting is hh:mm:ss ampm (hours:minutes:seconds AM/PM)

Physician/Location Tab

- This is where you can delete Physician and/or Location names that are no longer needed.
- Once deleted, they will no longer appear in the drop-down lists of the Add/Edit Worklist screen but they will remain with stored patient results.

Communication Tab

LIS /HIS Tab

- Host computer definition
- Configures the communication format for transfer of sample information to a host computer
- Allows you to enable or disable the automatic transmission of control results to a host computer

Variable Format

- Additional host computer definition used only for the variable format

Printer Tab

- Can access the Windows-NT Printer Properties window
- Can add a printer
- Only certified printers recommended by Beckman Coulter ensure proper printing of designated codes and flags

Cycle Option Tab

- Daily Workload:
  - Can enter the estimated number of CBC/DIFF and CBC cycles run per day
  - Default value for CBC/DIFF Runs Per Day is 40; for CBC Runs Per Day, is 10
  - Cycle estimates are used to determine if sufficient reagent is available to complete the daily workload; check is done during Startup
  - If reagent volume is insufficient to complete the estimated daily workload, the message *Reagent(s) Low. Insufficient Reagents To Complete Daily Workload* appears
- Autoclean Frequency:
  - Enter number of cycles 1 to 75 that will run before an automatic Autoclean is run by the system
  - Default value is 75
- Startup:
  - Can select whether or not the system will automatically run a Startup whenever the system power is turned off then back on
  - Automatic Startup is the default setting

Units Tab

- Select the appropriate reporting unit format for the laboratory
- Format used to report numeric results
- Default setting is US
- Changing the units requires restarting the application; user is automatically logged out of the application

Analyzer Tab

- Accepts entry of the Analyzer's serial number

Config Save / Restore Tab

- Selecting **Save** allows laboratory to save the Analyzer setup and/or the Workstation setup to a floppy disk
  - Recommended after installation and after making any changes
- Selecting **Restore** allows laboratory to select a directory to restore a previous setup file (from the Analyzer setup and/or the Workstation setup) using the information previously saved on the floppy disk
- Selecting **Print** provides a hard copy of the current Analyzer or Workstation setup

Language Tab

- Used to select the language displayed by the Workstation's software
- Allows selection of English, French, German, Italian, or Spanish
- Default setting is English

QUALITY ASSURANCE SETUP

- At the Menu bar:
  - Select **Setup ▸▸ Quality Assurance**
  - Type **Administrator Password** then press Enter or click 
- To save changes:
  - Click  to save the entry and remain on the window

    or
  - Click  to save the entry and exit the window
- For additional details, see Appendix A of the Instructions For Use manual.

General Tab

- Window contains five areas:
  - Reserved Lot Number
  - CV Limits
  - XM Options
  - IQAP ID
  - Auto-Print

Reserved Lot Numbers

- Can list CBC controls files 1 through 12
- Can list CBC/DIFF control files 13 through 24
- Reserved lot numbers allow a control to be processed using the Run window rather than the Control window

CV Limits

- Only the QC CV limits can be changed
- Reproducibility and calibration CV limits are set by the manufacturer

XM Options

- Allows selection of either 3 or 9 parameter mode for XM analysis
- Default setting is Off

IQAP ID

- This is where you enter your instrument's IQAP ID number
- Required in order to download control file data to diskette for submission to IQAP

Auto-Print

- Control box allows selection or deselection of Auto-Print of the control report
- Repro box allows selection or deselection of Auto-Print of the reproducibility report
- Calibration box allows selection or deselection of Auto-Print of the calibration report
- Default setting is Off for all three options

Calibration Tab

- Displays the current calibration factors, the date the calibration factors were modified, and the operator who authorized the changes
- At the end of the calibration process, these values are replaced with new calibration values

Shifts Tab

- Used to separate the control files into different shifts
- May select 24 hour shift or multiple shifts
- If **24 Hour Shift** is selected, shift 1 through 3 fields are disabled and blank
- If **Multiple Shifts** is selected, the From column is enabled and the To column is disabled
  - Gaps in time between shifts is not permitted
  - When a change is made in the From column, the preceding To column automatically increments or decrement to prevent any time gap between shifts

PATIENTS SETUP

- At the Menu bar:
  - Select **Setup ▸▸ Patients**
  - Type **Administrator Password** then press Enter or click 
- To save changes:
  - Click  to save the entry and remain on the window

    or
  - Click  to save the entry and exit the window
- For additional details, see Appendix A of the Instructions For Use manual.

Flagging and Messaging Tab

- Possible to have 20 different flagging sets
  - Flagging sets 1 through 6 contains default values
  - Flagging set 1 is named Standard Range and cannot be edited
  - Flagging sets 2 through 6 action limits and patient limits may be edited as desired, except age may not be edited
  - Additional flagging sets may be set up in the 7 through 20 rows
- Must select one flagging set as a default set
  - Default setting is the Standard Range flagging set
  - May change this default setting as desired
- To edit flagging sets 2 through 6 or add new flagging sets, must click the **Setup** button to access the Flags and Messages window

Flags and Messages Window

Note: The **Flags Sensitivity and Thresholds** tab is for Service use only.

- May create a flagging set by copying an existing flagging set then editing new flagging set as desired (except the age range)
- Two factors necessary to set up a flagging set are:
  - Flagging Set Name
  - Limits (Patient and Action)

- If patient results are outside the **Patient Limits** in the selected flagging set, result is flagged
  - *H* for a result above the upper patient limit (limits in the Patient H column)
  - *L* for a result below the lower patient limit (limits in the Patient L column)
  - Numeric result and flag are displayed with a yellow background
- If patient results are outside the **Action Limits** in the selected flagging set, result is flagged
  - *HH* for a result above the upper action limit (limits in the Action H column)
  - *LL* for a result below the lower action limit (limits in the Action L column)
  - Numeric result and flag are displayed with a red background
  - Generates an interpretive message based on specific parameter results

    Note: Interpretive messages are detailed in the Instructions For Use manual, Chapter 9

Parameters Tab

- Displays the parameters the A^{C}•T 5diff CP hematology analyzer is able of directly measure, derive from a histogram, derive from the DiffPlot, or compute

Enable / Disable RUO Parameters

- RUO (Research Use Only) parameters
- Parameter defined as "For Research Use Only. Not for use in diagnostic procedures."
- Six RUO parameters on this instrument
  - Two platelet parameters: Pct (Plateletcrit) and PDW (Platelet Distribution Width)
  - Four white blood cell parameters: ATL % and # (percentage and absolute number of atypical lymphocytes) and IMM % and # (percentage and absolute number of immature cells)
  - In the United States, when an RUO parameter label is displayed, printed, and/or transmitted, the message *For Research Use Only. Not for use in diagnostic procedures.* is displayed, printed, and/or transmitted
- Default setting is Disabled
  - Button reflects the opposite state of the current RUO parameter selection
  - When button displays **Enable RUO Parameters**, RUO parameters are disabled
- Changing the state from **Disabled** to **Enabled** (or vice versa) requires restarting the application; user is automatically logged out of the application
- If enabled for use in the United States, system prints an RUO Certificate that **must** be completed and returned to Beckman Coulter as instructed

Reports Tab

- Window contains six areas:
  - Reports Headers (six different header positions are available)
  - Auto-Print
  - # of Copy
  - Report Format
  - Printed Parameters
  - Enable

Reports Headers

- Sets up the banner for the top of each printout
- Provides space for six different header entries
- May enter up to 25 alphanumeric characters in each header

Auto-Print

- Allows selection of the Auto-Print option
- Default setting is Off
- If automatic printing is desired, allows selection of **All**, **Normals**, or **Selected Abnormals**
- Selected Abnormal categories include:
  - If **No Parameter Value** selected, patient reports without at least one numeric value automatically print
  - If **With Parameter Flags** selected, patient reports with at least one parameter flag automatically print
  - If **Outside Patient Limits** selected, patient reports containing at least one parameter result falling below the low or above the high patient limits of the selected flagging set automatically print
  - If **Outside Action Limits** selected, patient reports containing at least one parameter result falling below the low or above the high action limits of the selected flagging set automatically print

of Copy

- Default setting is 1

Report Format

- Three report format options are available
- Default setting is Option 1 (full report)

Printed Parameters

- Select the parameters you want to appear on the printed report

Enable

- Six options are available under this category.

  Note: With the exception of the **Raw Values** option, each default setting is enabled ☑.

  - **Range** - patient limit ranges are printed on the report when this option is enabled
  - **Messages** - messages are printed on the report when this option is enabled
  - **Detailed Flags** - detailed flagging prints on the report when this option is enabled; if this option is disabled ☐ (no ✓), simplified flagging prints

    Note: On the Run window, the **Flags and Messages** label changes to **Suspect and Messages** when this option is disabled (simplified flagging prints).
  - **Diffplot Thresholds** - thresholds are printed with Diffplot when this option is enabled
  - **Histogram Thresholds** - thresholds are printed with each histogram when this option is enabled

- Default setting for **Raw Values** is disabled ☐ (no ✓)
  - When enabled, raw values print on the report (which may be useful to service)
  - **Do not enable ☑ this option unless your Beckman Coulter Representative instructs you to enable this option!**

INITIAL SET UP OF CONTROL FILES

- Control files are set up through the Control window (not using the **Setup** option on the Menu bar as the other set ups were in this topic)
- To display the Control window, click on the **Quality Assurance** tab
- 12 CBC control files and 12 CBC/DIFF control files are available for set up
  - CBC control files (1 through 12) contain fields for entering only CBC parameter values
  - CBC/DIFF control files (13 through 24) contain fields for entering values for CBC and DIFF parameters
  - CBC and CBC/DIFF files are type specific and should be set up with a control that correlates:
    - Control that monitors only CBC parameters should be set up in a CBC control file, not a CBC/DIFF file
    - Control that monitors both CBC and DIFF parameters should be set up in a CBC/DIFF control file, not a CBC file
- There are two ways to set up a control file:
  - Control disk download (transfer data from control disk to Workstation)
  - Manual entry of information from control package insert (type everything)
- A^{C}•T 5diff Control Plus is the recommend cell control
  - Quality control material for monitoring CBC and DIFF parameters
  - Three levels of control: Low, Normal, High
- Controls may be processed with the Control window displayed or on the Run window if the lot number for that control is reserved
  - Later when controls are covered in more detail, you will run controls into the files you are setting up
  - If you are interested in running controls on the Run window, after a control file is set up, reserve the lot number by accessing the Reserved Lot Number box as follows:

    At the Menu bar, click **Setup** ▸▸ **Quality Assurance** ▸▸ type Administrator Password `123` then press Enter or click  ▸▸ click **General** tab, if necessary

Set Up

- Since initial control file set up is less involved than management of a control file containing results, initial set up is being included with these other set up options.
- Get the control disk for the controls you need set up then use the instructions under the Initial Set Up of Control Files Summary heading to set up these files

  Note: The Instructions For Use manual has the procedure for setting up control files manually should you ever need it.
- When it is time to record stored control results and set up other control files, use the Control File Management Summary

➤ INITIAL SET UP OF CONTROL FILES SUMMARY

Setting Up a Control File

Note: Before starting this set up, obtain the control disk for the controls you plan to set up.

1 Select the File

- Click the **Quality Assurance** tab to display the Control window.

  Note: If the Control window is not displayed, click the **Controls** tab at the bottom.
- Choose a CBC/DIFF file from the Select Control drop-down menu.

2 Access the Setup Control Window

- Click the **Setup Control** button.

3 Download the Control Data from the Disk

- Insert the control disk into the floppy drive.
- Select the **Download Values** button.
- Select the control level then click  .
- Select Commercial as the source of control material (if it is not already displayed).
- Print if desired then click  .

4 Reserve the Control Lot Number

- Click **Setup ▸▸ Quality Assurance**.
- Type the password then click  .
- Click the **General** tab, if necessary.
- Click the box in the Reserved column that corresponds to the desired control lot.
- Click  .

5 Set Up Another Level of Control, if applicable

IMPORTANT Risk of misleading control results. Old control runs remaining in a newly set up control file are included in the statistical data for the new file. Delete all controls runs before starting to run the new control.

- Review, print, and delete other files, as applicable.
- Choose another empty, inactive file from the Select Control drop-down menu.
- Repeat steps 2, 3, and 4.

➤➤

Note: This summary is only for initial set up of control files; therefore, it is not included in the Summary Pages. The next time you set up a control file, use the Control File Management Summary.

OBJECTIVES

When the subject is complete, you will be able to . . .

Controls

- Explain the storage and handling requirements.
- State the open-vial stability.
- Define an event.
- State indications of instability or deterioration.
- Prepare and run A^{C}•T 5diff Control Plus.
- Recognize an out-of-limit result on the control window.
- Explain what to do if a cell control result is out-of-limits.
- Explain the statistics associated with the control files and graphs.

Interlaboratory Quality Assurance Program (IQAP)

- Explain the purpose of the Interlaboratory Quality Assurance Program (IQAP).

Control File Management

- Review numeric and graphic control data for errors and make appropriate corrections.
- Print control file data.
- Exclude individual control runs.
- Delete a control file.
- Properly set up control files for running A^{C}•T 5diff Control Plus.
- Establish running means and expected ranges for BASO % and #.
- Explain how to make changes to an existing control file.

Optional Checks

- Explain the purpose of a reproducibility check.
- Explain the purpose of a carryover check.

XM Analysis

- Explain the purpose XM analysis.
- Explain why the use of RBC indices is so effective in the XM analysis.
- Define the terms batch, mean values, and limits as they apply to the XM analysis.
- State the minimum number of samples collected for an XM batch.
- Describe the three areas of the XM graph.
- Identify the problem parameter (or parameters) causing an XM batch to be out-of-control.

NOTES

COULTER® AC•T™ 5diff CONTROL PLUS

- Recommended cell control
- Quality control material for monitoring CBC and DIFF parameters
- Three levels of control: Low, Normal, High

Storage

- Control must be stored at 2 to 8°C (35 to 46°F)
- Storage of the control product in the cap down (inverted) position is not recommended
  - Might require additional mixing for complete resuspension of cellular components

Proper Handling

Prepare Cell Controls according to the Package Insert Instructions

- Warm control vials at ambient temperature for at least 15 minutes before mixing
- Mix gently by hand using the 8 x 8 x 8 method twice
  - After the second mixing, check the sides and bottom of the tube to verify the control is mixed completely
  - Overmixing causes hemolysis which indicates product deterioration
- Do not use mechanical mixers or rotators
- Return to the refrigerator within 30 minutes to ensure stated open-vial stability

Stability

Open-Vial Stability

- Open-vial stability stated on the package insert is listed according to days or events (whichever comes first)
- An event occurs anytime an operator completes the following sequence:
  - Removes a control vial from the refrigerator
  - Allows the control vial to stand at room temperature 15 minutes
  - Mixes the control vial gently by hand using the 8 x 8 x 8 method two times
  - Aspirates sample
  - Returns the control vial to the refrigerator within 30 minutes
- MCV and/or RDW parameters may show trending through the product's shelf life; however, this is inherent to the product and should not be considered an indicator of product instability

Indications of Instability

- A slight pink color to the supernatant is normal
- Inability to obtain expected values in the absence of known instrument problems or gross hemolysis (darkly colored supernatant) indicates deterioration of the control

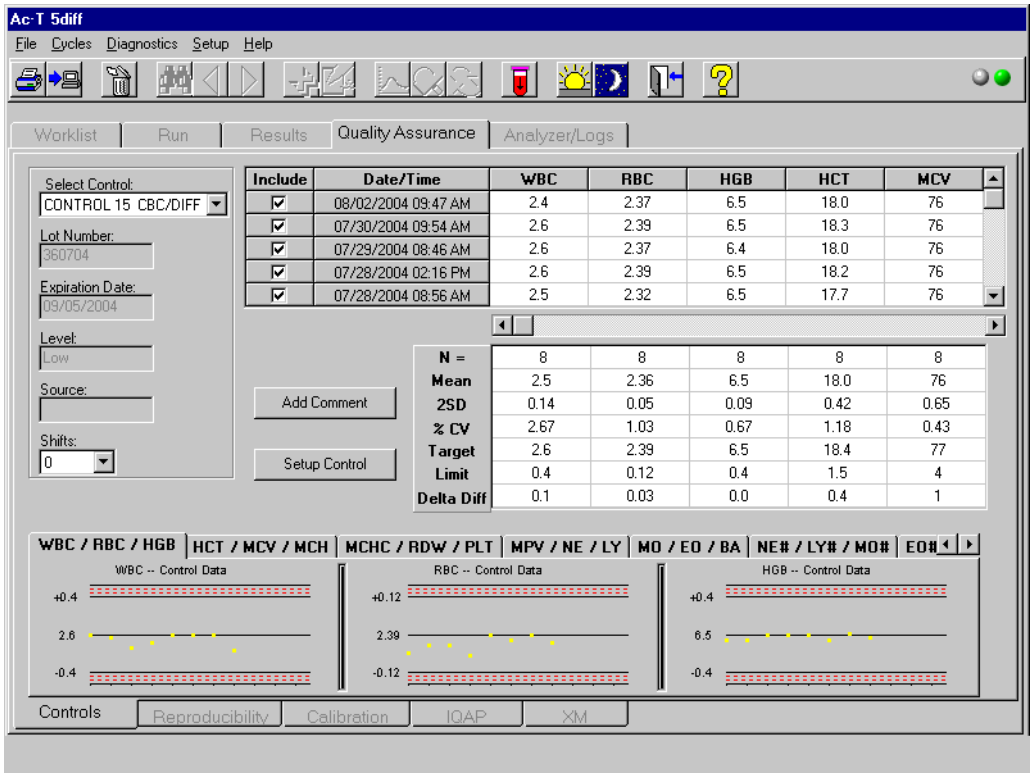
Running A^C•T 5diff Control Plus

- Control is designed with a pierceable cap
- Use the instrument's piercing capability to prevent spillage and reduce biohazard exposure
- Make sure the cap is secure before placing the well-mixed, closed control vial in sample position 1 of tube holder #1 or sample position 4 of tube holder #2
- Make sure tube holder sample position is rotated to the pierce position before closing the cap-pierce door (pierce position is the 12 o'clock position)
- Although control levels may be run in any order, Low, Normal, then High provides the best statistical results
- Controls may be processed with the Control window displayed or on the Run window if the lot number for that control is reserved
- Do not preassign controls into the Worklist
- Hard copy of the results (in a line list format) prints automatically if the Auto-Print function is enabled
- BASO % and # mean values and expected ranges
  - Establish your running mean and expected range for BASO % and # according to your laboratory protocol

Using the Run Window to Run Controls

- Identifier entered in the Sample ID field is compared to the list of Reserved lot numbers
- If the identifier in the Sample ID field matches a Reserved lot number:
  - Corresponding entry fields are automatically completed
  - Workstation places the results in the appropriate control file based on the information entered in the Sample ID field
  - Control results are displayed in the patient sample format but are flagged as a control
    - If a control result is lower than the expected range low limit, the parameter field backlights in yellow and out-of-limit result is flagged with an *L*
    - If a control result is higher than the expected range high limit, the parameter field backlights in yellow and the out-of-limit result is flagged with an *H*
  - Cannot see how the results of this control relates to other stored control results until you access the Control window
- If the control lot number is not reserved:
  - Software searches for a duplicate sample ID number throughout the current active archive then among the sample ID numbers waiting to be processed, the number is accepted only if it is unique
  - Control results are stored as a patient not a control
  - Control results are flagged as a patient
  - Any control with an unreserved lot number must be processed with the Control window displayed (showing the control file for that specific lot number of control)

Reviewing Control Results



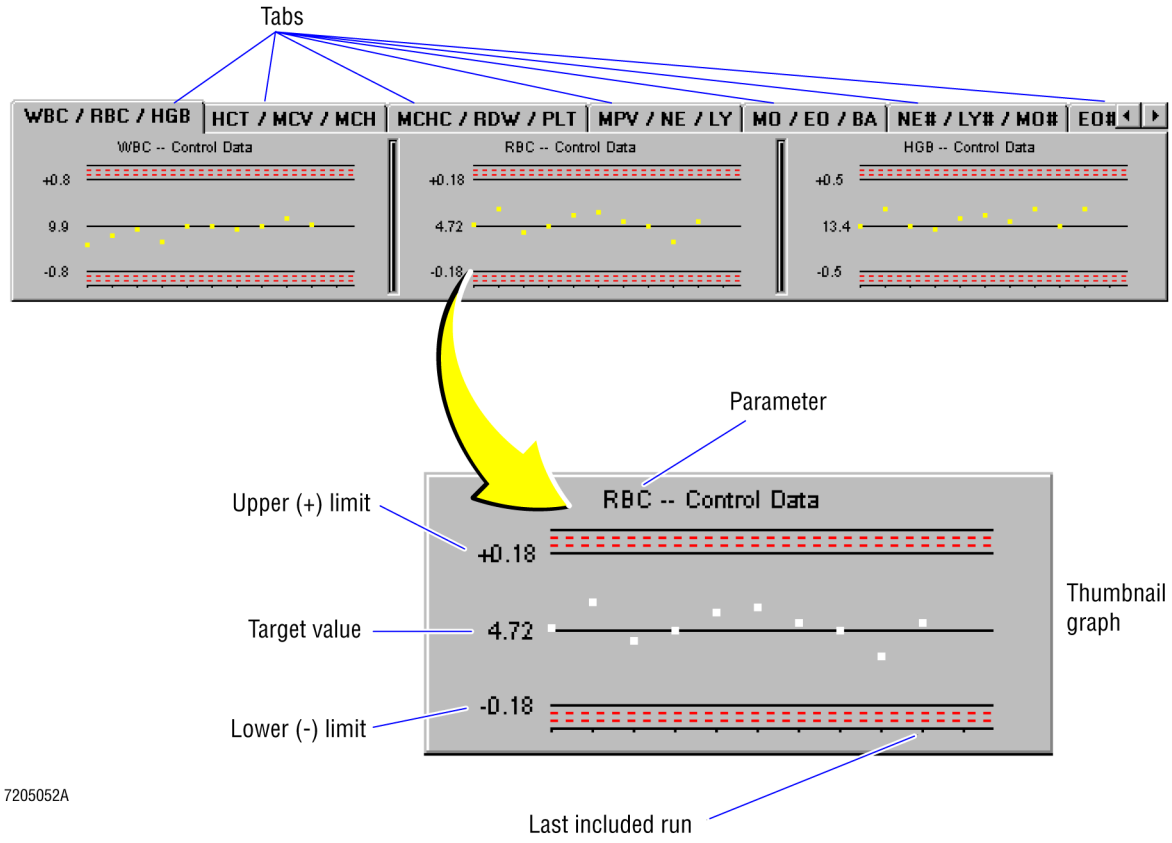
Tabular Results

- The most recent control results are displayed and printed first (at the top)
- Operator can add a comment to a selected set of results using the comment button
- Control results are compared to expected ranges:
  - When a result exceeds the low limit, an *L* is displayed next to the result
  - When a result exceeds the upper limit, an *H* is displayed next to the result
- Each control file can store more than 100 runs:
  - Must scroll to see older data
  - History of previous control results may be helpful during troubleshooting process
  - After 400th control is run into a file, message *Result Could Not Be Saved (File Full)* appears; no further runs can be stored in this control file
- When a % CV exceeds the QC CV limit for that parameter, the parameter field containing the out-of-limit % CV value backlights red

  To access QC CV limits:
  At the Menu bar, click on **Setup** ▸▸ **Quality Assurance** ▸▸ type **Password** then click  ▸▸ click **General** tab, if necessary

- Operator may decide to exclude an analysis:
  - Control results should only be excluded immediately if you can trace an operator error to the unacceptable results
  - Excluding too many runs can cover up an instrument problem and bias results

Graphs



Tabs

- Each of seven tabs allows access to three thumbnail graphs at a time

  Note: All seven tabs appear even when a CBC file is selected.
- From left to right, clicking the graph tab provides access to the following parameters:
  - If **WBC / RBC / HGB** tab selected, thumbnail graphs for the WBC count, RBC count, and hemoglobin parameters are displayed
  - If **HCT / MCV / MCH** tab selected, thumbnail graphs for the hematocrit, Mean Cell Volume, and Mean Cell Hemoglobin parameters are displayed
  - If **MCHC / RDW / PLT** tab selected, thumbnail graphs for the Mean Cell Hemoglobin Concentration, Red cell Distribution Width, and platelet parameters are displayed
  - If **MPV / NE / LY** tab selected, thumbnail graphs for the Mean Platelet Volume, neutrophil %, and lymphocyte % parameters are displayed

- If **MO / EO / BA** tab selected, thumbnail graphs for the monocyte %, eosinophil %, and basophil % are displayed
- If **NE# / LY# / MO#** tab selected, thumbnail graphs for the neutrophil absolute number, lymphocyte absolute number, and monocyte absolute number are displayed
- If **EO# / BA#** tab selected, thumbnail graphs for the eosinophil absolute number and basophil absolute number are displayed (only set with two graphs instead of three)

Thumbnail Graphs

- Each thumbnail graph contains three areas
  - A central line representing the actual mean value for that parameter
  - A line above mean value (central line) represents the upper limit and a line below the mean value represents the lower limit boundaries
  - Area showing dashes is the out-of-limit area
- Each thumbnail graph displays the last 10 data points that are included in the statistics
  - Last data point included in the graph is located on the far right
  - Data points are not displayed for control runs that are excluded from the control file

Expanded Graphs

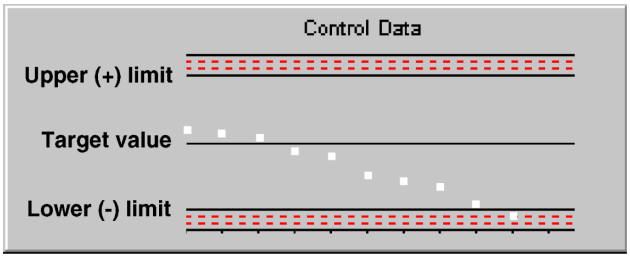
- When a thumbnail graph is double-clicked, graph expands to display up to 100 data points
- Only data points for control runs included in the statistics are displayed
- After the first 100 controls are processed, this option always displays the plot points for the most recent 100 controls
- Provides a history of previous control results which may serve as an aid during the troubleshooting process
- After the 100th control is run, the graph begins to rollover where the oldest control run is deleted to make space for the newest control run
- Most recent data point included in the graph is located on the far right
- Data points are not displayed for control runs that are excluded from the control file

Graphing Control File Data Facilitates Monitoring

- Graphing provides a convenient mechanism that allows daily inspection for trends, shifts, outliers, and/or other violations of the laboratory's quality assurance guidelines
- Data plotted on a graph:
  - Allows easy visual detection of "in control" and "out of control" results
  - Allows faster detection of subtle developing problems
- Different kinds of changes suggest different sources for error
- Graphing is ideal for spotting trends and shifts

Trend

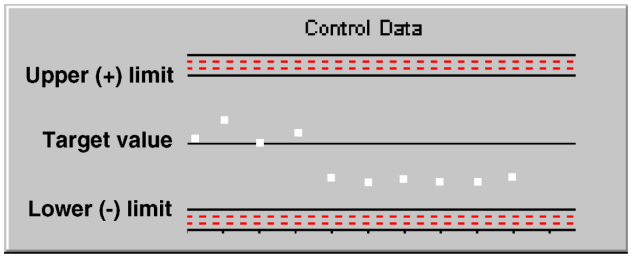
- Gradual change in 5 or more consecutive results, all going upward or all going downward



- Systematic drift or trend in control values suggests that a problem is progressively developing
- Generally due to the deterioration of a reagent or control material

Shift

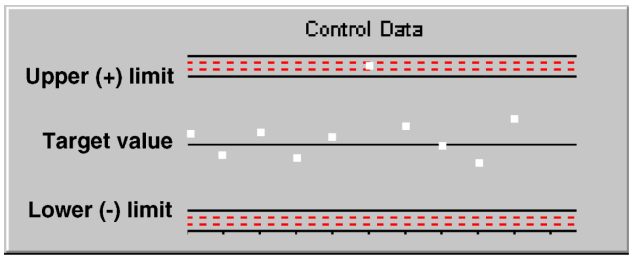
- Sudden or abrupt change in 5 or more consecutive results all above or all below the mean



- This type of systematic error is usually associated with the malfunction of the instrument or an error in technique

Outlier

- Single result that falls outside the upper or lower limit
- Chance probability for a value to be outside the limit is about one time out of every 20 times control is run
- In the example below, run #6 (from the right) illustrates an outlier; notice plot point is located inside the upper double line



- With computerized graphing, small double-lined area typically serves as a limit for the plot point
- Since plot point for an outlier cannot extend beyond this set boundary, operator must look at the numeric results to determine if the result in question is slightly outside the limit or greatly exceeds the limit according to the laboratory's protocol
- If the numeric result for the outlier is only slightly beyond the limit, it may be a statistical outlier (control value is out by chance)
- To confirm a statistical outlier, operator must run the control one more time
  - If the result of the rerun is now in-control, the analytic run is accepted and both runs (the statistical outlier and the acceptable rerun) are to be included in control data
  - If rerun of control reflects the same parameter(s) as still being out-of-control, there may be a control or system problem
- When considering a control problem:
  - Any change in the control material may cause control to be out
  - Control must be handled carefully as instructed on the package insert
  - If control material is treated roughly, stored improperly, or used beyond the expiration date, results may be outside the limits
  - Generally only one level of control or one vial shows the problem
- When considering a system problem:
  - Any change to the instrument regarding the test system may cause controls to be out
  - Usually affects more than one level of control
- Important considerations when a statistical outlier occurs:
  - Frequency of outlier occurrences should be carefully monitored; high outlier frequency may indicate an underlying problem with the instrument, control material, or operator technique
  - When a single control result is analyzed and the run is repeated, the original control result should remain in the control file and used in the statistical calculations

Guidelines for Acceptable Operation

- Performance specifications for your instrument are located in Operator's Guide, Chapter 3
- Laboratory may choose to use Beckman Coulter assigned values or may choose to establish their own laboratory means
- Instrument is considered well maintained and operating correctly if before current cell control lot(s) expire, on the new lot(s) of cell control, the laboratory:
  - Confirms recovered mean values are within the TABLE OF EXPECTED RESULTS

    or
  - Establishes its own mean values and acceptable ranges and periodically reevaluates those means

For More Basic Concepts

- Consider ordering **Basic Concepts of Quality Control**, PN 4235526
  - Designed for someone with little or no quality assurance experience
  - If desired, contact your Beckman Coulter Representative to order a printed copy
- Information covered in this booklet includes:

  Note: This list is not inclusive.
  - Fundamental components of a quality assurance program
  - Definitions of common terminology
  - How to read a package insert
  - Difference between a control and a calibrator
  - Why calibration is necessary
  - How to determine if a control is "in" or "out"
  - What to do if a control is "in"
  - What to do if a control is "out"
  - Outliers
  - Trends
  - Shifts
  - Interlaboratory Quality Assurance Program (IQAP)
- Booklet also contains a glossary and a self-evaluation

➤ RUNNING COULTER AC•T 5diff CONTROL PLUS SUMMARY

1 Get the Workstation Ready

To run controls with the Control window open

- Click the **Quality Assurance** tab.
- From the Select Control drop-down menu, locate the control file that correlates with the lot number of control you wish to run.

To run controls with the Run window open

- Click the **Run** tab.

  Note: To use this option, the control lot number must be reserved. If you are not familiar with this protocol, use the Control window to run the controls.
- Enter the control lot number as the next Sample ID.

2 Prepare the Control according to the Package Insert

- Warm controls at room temperature for 15 minutes.
- Control levels may be processed in any order.
- Mix the control vial using the 8 x 8 x 8 method.
  - Do not use a mechanical mixer!
  - Inspect the control vial to ensure all cells are uniformly distributed, if not, repeat the 8 x 8 x 8 method of mixing again.

3 Run the Control

- At the tube holder, make sure sample position is in the pierce position (12 o'clock). (In tube holder #1, use sample position 1; in tube holder #2, use sample position 4.)
- Place the well-mixed, closed-vial of control in sample position 1.
- Close the cap-pierce door.

4 Review Results

Note: A hard copy of the results prints automatically if the Auto-Print function is enabled.

- Verify control results are within the acceptable ranges.
- If a result is out-of-limits:
  - Rerun the control after gently remixing the control vial.

    or
  - Refer to When CBC/DIFF Control is Outside Its Expected Ranges that follows.
- Compare the results of this run with previous runs, as desired
  - To evaluate stored numerical data or evaluate the graphs for the presence of shifts or trends, the Control window must be displayed.
  - Click **Quality Assurance** tab to display the Control window, if needed.

When CBC/DIFF Control is Outside Its Expected Ranges

1. Check for a control problem.
   - Ensure the control material was properly mixed. If not, mix it according to the package insert.
   - If the control was processed in the Control window, make sure the control file with the correct lot number was selected.
   - If the control was processed in the Run window, make sure the lot number was entered correctly and verify the lot number is reserved.
   - Unless you have established your own running mean and/or expected ranges, make sure the setup information (assigned values and expected ranges) matches the control package insert. If they do not, change the control setup information to match the package insert.
   - If any of the above problems exist, rerun the control; otherwise, proceed to the next check.
2. Rerun the control to determine if the out-of-limit result is a statistical outlier. If the rerun reflects the same parameter(s) as still being out-of-control, proceed to the next check.
3. Ensure the control material was not contaminated by running another vial or level of control. Follow the instructions on the package insert for proper handling. If the results are still out, proceed to the next step.
4. Perform the Extended Cleaning procedure in the Instructions For Use, Chapter 11.

   Use the Online Help to locate this procedure:

   a. Click  .

   b. Click on **Contents**. The table of contents appears in the left frame.

   c. Inside the left frame, click on **11 Diagnostics** to list the headings in that chapter.

   d. Scroll through the headings and click on **Extended Cleaning**. The cleaning procedure appears inside the right frame.

   e. Complete the procedure.
      - Follow the instructions on the Workstation screen.

        or
      - Print a copy of the instructions.
        - Right click on the procedure (anywhere inside the right frame).
        - Click **Print** on the pop-up menu.
        - Click **OK** to initiate printing.

        Note: All the information under the primary heading is printed. Locate the Extended Cleaning instructions and complete the procedure as written.

   f. Rerun the control.
      - If the results are acceptable, run other control levels that need to be processed then resume normal operation.
      - If the results are still out, call your Beckman Coulter Representative to help you troubleshoot.

➤➤

➤ MAKING CHANGES TO A CONTROL FILE CURRENTLY IN USE SUMMARY

Use this summary only if you need to make a change in a control file that is currently in use and only if you are changing an individual item:

- To correct a typographical error.

  or

- To edit an assigned value (obtained from the Table of Expected Results) to a running mean (established by the laboratory).

If you need to make a set up change (such as changing the lot number), use the Control File Management Summary.

1 Select the File

- Click the **Quality Assurance** tab ▸▸ **Control** tab to display the Control window.
- Choose the existing file you want to change from the Select Control drop-down menu.

2 Access the Setup Control Window

- Click the **Setup Control** button.

3 Make the Changes

- Enter the new information.

4 Verify All Changes

- Check all entries are correct.
- Click  .
  - ▸ If control runs are stored in this file:
    - — When the *Do You Want The Existing Results To Be Saved?* message appears, click on the desired action.
    - — Selected action makes the requested changes then exits the window.
  - ▸ If no control runs are stored in this file, requested changes are saved before exiting the window.
- Control file statistics are recalculated based on the changes.

➤ ➤

IQAP (Interlaboratory Quality Assurance Program)

- Coulter Interlaboratory Quality Assurance Program is a service offered to all regular users of A^{C}•T 5diff Control Plus cell controls
- It complements a laboratory's in-house quality control program by providing peer group comparisons for each lot number of a given control product run on the same instrument type
- The assessment is completely independent of the assay value issued when the product was supplied
- A separate IQAP manual (PN 4206266) is available with information on enrollment, data submission, understanding the IQAP report, and troubleshooting IQAP problems

  Note: In software version 2.00 and above you can download your control file data to diskette for submission to IQAP. See the Instructions For Use Manual or Help System for more details.

CONTROL FILE MANAGEMENT

- Once a control file is initially set up, the file is considered an existing control file
- Two types of existing control files:
  - Control file that contains control run data (may be either current or old data)
  - Control file with no stored control run data because the file is either newly set up or has had all its control runs deleted from it
- 12 CBC control files and 12 CBC/DIFF control files are available for set up:
  - CBC control files (1 through 12) contain fields for entering CBC parameter values only
  - CBC/DIFF control files (13 through 24) contain fields for entering CBC and DIFF parameter values
  - CBC and CBC/DIFF files are type specific and should correlate with the control
- When setting up a control file:
  - You can enter all of the information manually or you can download data from the control disk included in your control kit (software version 2.00 and above)
  - Most information is selected from a drop-down menu
  - If manually typing parameter values and limits, use Tab to move from field to field. Refer to the Instructions For Use manual for the complete procedure.
  - If your laboratory designates a specific file for running each low, normal, or high control, it will not be necessary to enter the expected range during setup since the expected ranges are not deleted when the results are deleted
- Initial set up of the control files is located under Topic 3, Setup Options

➤ CONTROL FILE MANAGEMENT SUMMARY

Review the Numeric Control Data and Graphs, if applicable

1 Locate the Control File

- Click the **Quality Assurance** tab to display the Control window.
- Choose the specific file you want to review from the Select Control drop-down menu.

2 Review the Graphs

- Double-click on any thumbnail graph you wish to review.
- Look for any skewed or out-of-limit plot points.
- Check for a shift or a trend.

3 Review Numeric Data

- Review % CV of all parameters (press → to see additional parameters).
- If a CV is high, scroll through the rows and examine each run for results mistakenly run into the file (the graph review should correlate).
- May review and manage the data by shifts, if desired.
- Exclude any mistakes from the control calculations.
  - Locate the control that needs to be excluded from the calculations.
  - Click the ☑ to the left of that control (inside the Include column).
  - ☐ (an empty check box) indicates those control results are no longer being included in the control calculations. N = is decreased by 1.
  - Repeat as needed.

Print the Control File Data

- Click  .
- **Print All Rows** is preselected but if a hard copy of the graphs is also desired select the **Print All Rows and L J Graphs** option.
- If you are submitting data for IQAP evaluation, print a second copy.

Delete the Control File Data

- Click  .
- Choose **Erase All rows** from the drop-down menu.

  Note: All the control results are deleted. Target values and +/- limits remain in the control file and may not need to be entered if the same type and level of control is set up again in that same file.

Set Up the New Lot Number of Commercial Control

IMPORTANT Risk of misleading control results. Old control runs remaining in a newly set up control file are included in the statistical data for the new file. Delete all controls runs before starting to run the new control.

Note: Before starting this set up, obtain the control disk for the controls you plan to set up.

1 Select the File

- Click the **Quality Assurance** tab to display the Control window.

  Note: If the Control window is not displayed, click the **Controls** tab at the bottom.
- Choose a CBC/DIFF file from the Select Control drop-down menu.

2 Access the Setup Control Window

- Click the **Setup Control** button.

3 Download the Control Data from the Disk

- Insert the control disk into the floppy drive.
- Select the **Download Values** button.
- Select the control level then click  .
- Select Commercial as the source of control material (if it is not already displayed).
- Print if desired then click  .

4 Reserve the Control Lot Number

- Click **Setup ▸▸ Quality Assurance**.
- Type the password then click  .
- Click the **General** tab, if necessary.
- Click the box in the Reserved column that corresponds to the desired control lot.
- Click  .

5 Set Up Another Level of Control, if applicable

IMPORTANT Risk of misleading control results. Old control runs remaining in a newly set up control file are included in the statistical data for the new file. Delete all controls runs before starting to run the new control.

- Review, print, and delete other files, as applicable.
- Choose another empty, inactive file from the Select Control drop-down menu.
- Repeat steps 2, 3, and 4.

➤ ➤

OPTIONAL CHECKS

Reproducibility Check

- Check measures how close several results (from the same specimen) are to each other; in other words, this check measures repeatability or precision
- Check may be done using CBC or CBC/DIFF panel
- For best results, a fresh, normal whole-blood specimen must be used
- One normal, fresh whole-blood specimen is cycled at least 5, but no more than 11 times
- CV or Coefficient of Variation is a percentage of deviation from the mean
- Results that exceed the limits appear against a red background
- Reproducibility does not measure accuracy, but true accuracy is not possible unless the instrument is precise
- Performing a reproducibility check before performing a calibration may catch imprecision problems that may affect the calibration process
- Procedure is located in the Operator's Guide, Chapter 11

Carryover Check

- Carryover is the interaction of the previous sample with the current sample
- High to low carryover check verifies the high results of one sample do not affect the low results of the next sample
- Check is done by analyzing a whole-blood specimen with high values followed by a whole-blood specimen with low values; each specimen is run consecutively in triplicate
- Carryover is calculated as follows:

$$\%\ \text{Carryover} = \frac{\text{1st low sample} - \text{3rd low sample}}{\text{1st high sample} - \text{3rd low sample}} \times 100$$

NOTES

XM ANALYSIS

Quality Assurance Tool

- XM analysis is a quality assurance method that allows a laboratory to continuously monitor the performance of their automated hematology system and thus control the quality of their reported results by monitoring their patient results
- Since this quality assurance tool is based on patient results:
  - The more patient samples processed per day, the more frequent the system monitoring
  - Proper monitoring is best accomplished if the laboratory runs more than 100 patient samples per day
  - Running less than 100 patient samples per day means the system is not checked frequently enough to be considered a viable form of quality assurance
  - If your laboratory processes less than 100 samples per day but you still like to consider using this form of control, checking with your regulatory agencies may help you make a final decision
- Any method used for quality control must use a material that meets two requirements:
  - Material must be stable
  - Material must be similar in content to the patient samples that will be analyzed
- XM analysis meets these requirements:
  - Uses the patient samples themselves as a material so the similarity requirement is met
  - Uses the red blood cell indices (MCV, MCH, and MCHC) so the stability requirement is met
- No additional cost to the laboratory because the method uses patient sample results
- Concept of this analysis is based on $\overline{X}_B$ Analysis developed by Dr. Brian Bull but is modified to include the selection of additional parameters:
  - May select the more traditional three parameter analysis that includes MCV, MCH, and MCHC

    or
  - May select a nine parameter analysis that includes WBC, RBC, HGB, HCT, RDW, PLT as well as MCV, MCH, and MCHC
- Traditional three parameter evaluation focuses on the more stable RBC indices

RBC Indices

- MCV, MCH, and MCHC are relatively stable parameters:
  - RBC indices that comprise the 3-parameter XM analysis are very tightly controlled by the body because the red cells function best within a very narrow range of size and hemoglobin content
  - Other parameters such as the white blood cells and platelets that are included in the 9-parameter XM analysis tend to have a wider physiological range and are, therefore, not as predictable as the red cell indices
- Are typically stable for an individual patient from day to day
- Are stable for a patient population over time
  - Many hospitalized patients have been investigated and it has been found that there is no significant day-to-day or week-to-week variability in the mean of their indices

Establishing Mean Values

For the 3 Parameter XM Analysis

- A constant for each RBC index (MCV, MCH, or MCHC)
- XM mean values should reflect the entire patient population of the laboratory:
  - Patient population of a general hospital usually includes samples from all patient age groups, disease states and many hematologically "normal" samples
  - A patient population that includes only one age group or only one disease state will yield different target values than a patient population that includes all groups
- As long as the patient population remains constant, the XM mean values of each index also remain constant
- If the patient population changes, the mean of an index may also change and needs to be reevaluated
- May want to start with the target values suggested by Dr. Brian Bull for his $\overline{X}_B$ Analysis:
  - MCV = 89.5
  - MCH = 30.5
  - MCHC = 34.0
  - If you begin with these values, your laboratory must then evaluate and adjust them for their own patient population
  - Results from at least 250, but ideally 1000 blood samples should be collected to find your laboratory's XM mean values
  - Results should include all types of patients (oncology, presurgical, OB, dialysis, out patients, and so forth)

For the 9 Parameter XM Analysis

- MCV, MCH, and MCHC are part of these nine parameters so the information under the Heading For the 3 Parameter XM Analysis still applies
- For the other six parameters, your laboratory must evaluate the results of your population and decide what represents an acceptable mean value for each parameter to begin establishing the XM mean values for your patient population

Getting Started

Turning the XM Analysis ON

- Default setting for XM analysis is **Off**
  - **XM** tab is invisible when **Quality Assurance** tab is selected
- To activate XM analysis:
  - At the Menu bar, select **Setup** ▸▸ **Quality Assurance**
  - Type the **Administrator Password** then press Enter or click 
  - At the XM Options box, select **3 Param** (parameters) or **9 Param** (parameters)
- Once activated, laboratory must establish their own mean values (XM limits)

Enter XM Mean Values and Limits

- XM mean values and limits must be entered in the Workstation

  Click **Quality Assurance** tab ▸▸ **XM** tab ▸▸ **Setup XM Limits** button
- In the XM Limits box:
  - Type the mean values established by your laboratory (or for MCV, MCH, and MCHC the values suggested by Dr. Bull)
  - Multiply each mean value by 5% and type this number in the +/- box (5% is only a suggested percentage. While establishing the means that reflect your patient population, you may wish to use a higher percentage.)

    For example, if for MCV you use Dr. Bull's suggested mean of 89.5 and a 5% limit.

    XM mean value x percentage = +/- value (89.5 x 0.05 = 4.5)

    You would enter the 89.5 as the mean value and 4.5 as the limit. The Workstation will calculate the upper and lower limits and display them as a whole number. In this case, the upper limit would be 94 and the lower limit, 85. The upper and lower limits are displayed in whole numbers.
- Once your laboratory has established the XM mean values for your population:
  - Enter new mean values and limits.
  - For MCV, MCH, and MCHC, adjust the 5% limits according to your laboratory protocol.

    For example:

    MCV XM mean value x percent limit = +/- value for MCV

    MCH XM mean value x percent limit = +/- value for MCH

    MCHC XM mean value x percent limit = +/- value for MCHC
  - Due to the wide physiological range for the other six parameter, you may wish to use a higher percent limit than you use for the MCV, MCH, and MCHC parameters

Using the XM Analysis

- When XM option is turned on, the:
  - Workstation stores results of all patient samples (including any repeat samples) as they are cycled
  - XM analysis is performed on sets of 20 patient results at regular intervals (each patient set is referred to as a batch)
- Stored results being displayed on the current batch table may be accessed by clicking the **Batch Details** button:
  - If the 3 parameter XM option is selected, table shows the MCV, MCH, and MCHC parameter results collected towards this batch of 20
  - If the 9 parameter XM option is selected, table shows the WBC, RBC, HGB, HCT, MCV, MCH, MCHC, PLT, and RDW parameter results collected towards this batch of 20

    Note: It is necessary to use the scroll bar to view the MCHC, PLT, and RDW parameter results.
- Results stored on the current batch table cannot be deleted from the table
- Samples that will not be stored on the current batch table and, therefore, not used in the XM batch analysis calculation include:
  - Samples processed in the reproducibility mode of operation
  - Samples processed in the calibration mode of operation
  - Controls processed with the Control window displayed
  - Controls (with reserved lot numbers) processed with the Run window displayed
  - Blank samples
  - Patient samples with at least one non-numeric result
- When the batch of 20 patient samples is completed, the Workstation performs the XM analysis and determines the batch mean of each parameter:
  - Calculation done automatically by the Workstation
  - This batch mean is not a simple average value of the patients' parameter results, but a type of "weighted moving average" that statistically allows the small batch of 20 patient samples to appear as if it were a larger set of approximately 100 samples
- Workstation then compares each batch mean to its expected XM mean value
  - Results are displayed on the XM Batch Means table on the XM window
  - If the batch mean is inside the expected XM limits, the batch is said to be IN and the instrument is functioning properly
  - If the batch mean is outside the expected XM limits, the batch is said to be OUT:
    - One or more parameters are not close enough to the expected XM mean values
    - An *H* or *L* flag appears beside the out-of-limit results
    - Red **XM** icon appears to the left of the System State indicators (connection status circles in the upper right corner)

Reviewing XM Data

XM Batch Table

- Table appearing on the XM window that collects completed batch information
- Used to view the calculated batch means for the last 60 batches of 20 samples
- Most recent batch is located at the bottom of the table
- May be used to identify the affected parameters and to determine the direction and amount of the change
- After the first 60 batches are completed, this option always displays the last 60 completed batches
  - Provides a history of recent batch means which may serve as an aid during the troubleshooting process
  - After the 60th batch is completed, the table begins to rollover where the oldest batch results are deleted to make space for the last completed batch results
  - Individual batches cannot be deleted
- Details of a selected batch may be viewed by clicking the **Batch Details** button
  - Table that collects the individual sample results
  - Individual samples cannot be deleted from the batch of 20 samples

XM Graphs

- Three graphs are displayed at a time
  - If 3 parameter XM option is selected, MCV, MCH, and MCHC graphs are all displayed at one time
  - If 9 parameter XM option is selected, three tabs are available for selecting the three graph grouping for display:
    - If the left tab is selected, the WBC, RBC, and HGB graphs are displayed
    - If the middle tab is selected, the HCT, MCV, and MCH graphs are displayed
    - If the right tab is selected, the MCHC, RDW, and PLT graphs are displayed
- Used to view the batch means in relationship to the XM mean value and XM limit for the corresponding index
- Graphs are good for identifying trends and shifts
- Each graph contains three areas
  - A central line representing the actual XM mean value for that parameter
  - A line above XM mean value (central line) represents the upper limit and a line below the XM mean value represents the lower limit boundaries
  - Area showing dashes is the out-of-limit area
- Each graph displays the last 10 data points
- When a graph is double-clicked, the graph expands to include the last 60 data points

- After the first 60 batches are completed, this option always displays the plot points for the last 60 completed batches
  - Provides a history of recent batch means which may serve as an aid during the troubleshooting process
  - After the 60th batch is completed, the graph begins to rollover where the oldest batch results are deleted to make space for the last completed batch results

What Causes a Batch to go "Out-of-Limits"?

- Change in the patient population
- An instrument problem may exist
- A calibration problem may exist
- A reagent problem may exist

Troubleshooting when an XM Batch is "Out"

Consider a Change in the Patient Population

- Is there a change in the overall patient population?
  - One or more new patient types were added to the population
  - One or more patient types are no longer part of the population
  - Possible seasonal change of the patient population (hospitals or clinics in resort areas)
- If a change in the overall patient population is probable,
  - Future XM batch results will also be outside the limits
  - New XM mean values based on the current population must be established and entered in the computer
- If a change in the overall patient population is not probable, it is possible that the patient population has not really changed but just appears to have changed because the batch of 20 patients does not truly represent the total population

Check for Non-Reportable Results and/or Non-Random Sampling

- An XM batch may go outside the XM limits because the batch is biased by physiologically impossible results caused by a reagent or instrument problem, or by physiologically abnormal specimen
- Use the **Batch Details** window to check for non-reportable results and/or non-random sampling
  - Non-reportable results where a batch of patients is biased by short samples, physiologically impossible results and/or erroneous data accumulated before a reagent or instrument problem was identified and solved
  - Non-random patient sampling where a batch of patients is biased by several abnormal patients of one particular type (oncology, neonatal and so forth)

Consider an Instrument Problem Exists

- Consider an instrument problem only if:
  - A change in the patient population is doubtful
  - All the results in the current XM batch are reportable (physiologically possible)
  - Random sampling is verified
- Instrument problems cause XM batch results to go "out" and stay out
  - Can be a gradual change:
    - If caused by a calibration drift, should go back within limits after calibration
    - If caused by a component going bad over time, should go back within limits after the component is replaced
  - Can be a sudden change:
    - If caused by a component failure, should go back within limits after the component is replaced
    - If caused by any number of instrument problems, should go back within limits after the problem is fixed

Assessing the Situation

- Review the XM graphs and XM batch means to identify the out-of-limit parameter and note the direction of the change (increased or decreased)
- The physiological range of some parameters, like WBCs and Plts, makes it difficult to assess if a problem really exists or the diversity of the values has cause the problem
- Generally more than one parameter may show a change
- It is also important to note any parameter that may still be inside the limit but shows a significant change
- Call your Beckman Coulter Representative if you need assistance.

OBJECTIVES

When the subject is complete, you will be able to . . .

- Explain the purpose of calibration.
- Tell when one needs to verify calibration.
- Perform the preliminary procedures.
- Perform the calibration procedure in Chapter 10 of the Operator's Guide.
- Demonstrate how to access the Calibration Log.

NOTES

CALIBRATION

- A procedure used to standardize the instrument by determining its deviation from calibration references and applying any necessary correction factors
- Never use calibration to adjust for an instrument problem
- Verify the ambient room temperature is typical for the laboratory and within the instrument's operating range (16 to 34°C; 61 to 93°F)
- In the normal process of tracking data for an extended period of time, your laboratory can make a specific decision to recalibrate a given parameter
- **Never** adjust to a specific value based on an individual sample result

When to Calibrate

- At installation, before you begin analyzing samples
- After Service has replaced a major analytical component such as the sampling syringe or an aperture
- If your Beckman Coulter Representative suggests you calibrate

When to Verify Calibration

- As dictated by your laboratory procedures, local, or national regulations
- When controls show evidence of unusual trends
- When controls exceed the manufacturer's defined acceptable limits

PRELIMINARY CALIBRATION PROCEDURES

- Procedures designed to ensure the instrument is operating optimally
- Recommend a thorough review of data stored in current control and, if applicable, XM files to note any possible bias that might require a change in a calibration factor
- Thorough review of IQAP data is also recommended

Pre-Calibration Checks

Check Reagent and Waste Containers

- Check reagent volumes to ensure sufficient volume to complete calibration procedures
  - As needed, use the Changing Reagent Summary to replace any container with insufficient reagent with a new reagent container
- Check the waste container level
  - If needed, use the Replacing a Waste Container Summary as a guide

Ensure the Analyzer is Clean

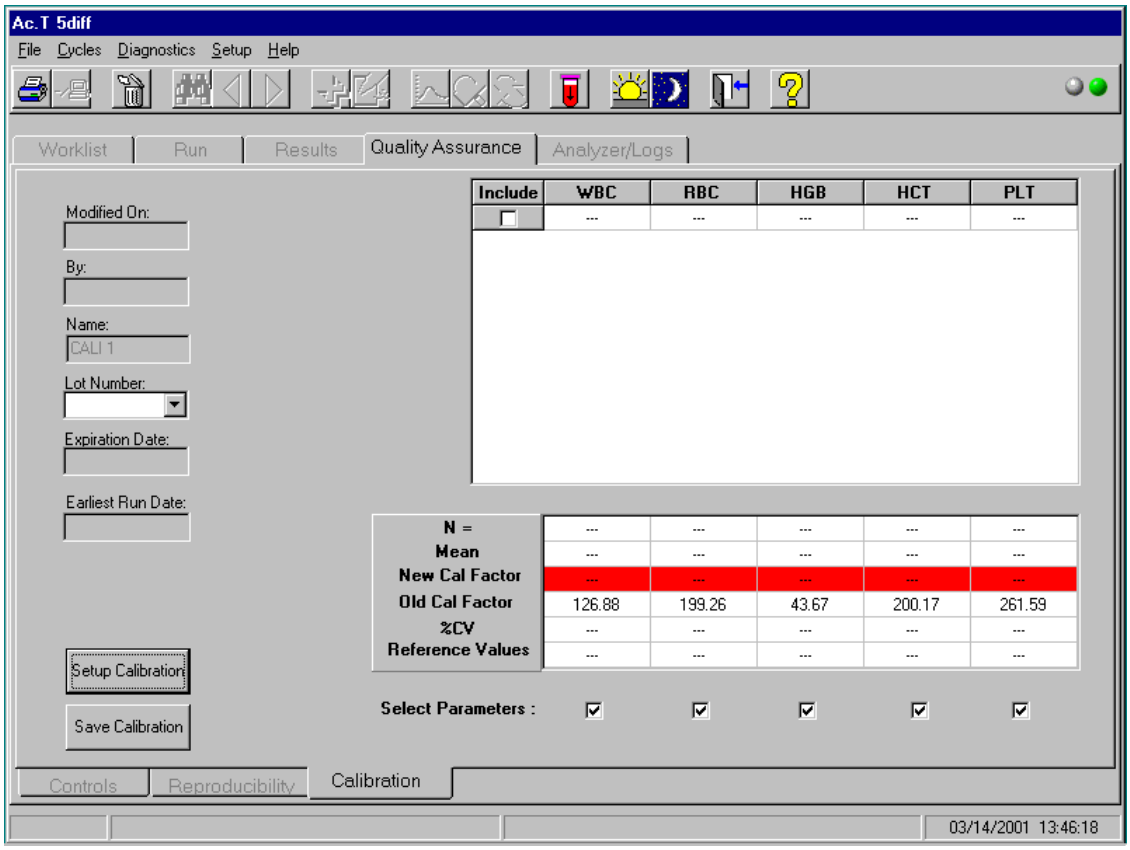
- If the instrument is routinely shut down with A^{C}•T 5diff Rinse reagent for at least 30 minutes every 24 hours it is in use:
  - Do a Startup
  - Run commercial cell controls
- If the instrument is **not** routinely shut down with A^{C}•T 5diff Rinse reagent for at least 30 minutes every 24 hours the instrument is in use:
  - Do the Extended Cleaning procedure in Chapter 11 of the Instructions For Use manual
  - Do a Startup
  - Run commercial cell controls

CALIBRATION PROCEDURE

- Beckman Coulter recommends using COULTER® A^{C}•T™ 5diff Cal Calibrator
- May calibrate one or more CBC parameters including WBC, RBC, Hgb, Hct, and Plt
  - Operator selects those parameters to be calibrated via the Select Parameters boxes
- Carefully follow the instructions on the COULTER® A^{C}•T™ 5diff Cal Calibrator package insert and the Auto-Calibration instructions in Chapter 10 of the Operator's Guide anytime calibration is performed
  - Before accumulating calibration data, prime the instrument using two normal whole-blood specimens and the calibrator material
  - 11 is the maximum number of runs that can be displayed in this window
  - Never accept a calibration factor based on less than five calibrator runs
- Perform each analysis using well-mixed calibrator
  - Before the first sample is aspirated, make sure the calibrator is mixed according to the instructions on the package insert
  - **Insert the tube correctly into:**
    - slot #1 of Tube Holder #1, or
    - slot #4 of Tube Holder #2
  - Close the tube holder door to begin analysis
  - Each time the tube holder door opens
    - remove the calibrator tube
    - mix thoroughly
    - replace the calibrator tube
    - close the tube holder door
  - Analyze the calibrator at least 5 but no more than 11 times

- Operator may decide to exclude an analysis
  - Sample results should only be excluded if you can trace an operator error to the unacceptable results
  - Excluding too many runs can cover up an instrument problem and bias calibration
  - Although the instrument can calculate the calibration statistics on three runs, Beckman Coulter recommends calibration factors based on a minimum of five acceptable results
- From the Menu bar, selecting **Quality Assurance** tab ▸▸ **Calibration** tab displays the Calibration window

Calibration Window



Terms and Formulas used in the Calibration Procedure

N=

- Number of A^{C}•T 5diff Cal calibrator runs included in the statistics for the parameter
- ☑ next to a set of results in the accumulated results table indicates that set of results is included in the statistics
- ☐ (no ✓) indicates that set of results is not included in the statistics; therefore, N may be less than the number of results displayed on the table

Mean

- Average of the A^C•T 5diff Cal calibrator runs
- Calculation can be based on at least 5, but no more than 10 runs

New Cal Factor

- Calibration Factor needed to recover the A^C•T 5diff Cal Calibrator Reference Value
- It is calculated and displayed regardless of whether or not you need to change it

$$New\ Cal\ Factor\ = \frac{Reference\ Value \times Old\ Cal\ Factor}{Calibrator\ Mean\ Value}$$

Old Cal Factor

- The current Calibration Factor in use by the analyzer
- Remains the current calibration factor until the save calibration function is performed

% CV

- Indicates the reproducibility of the A^C•T 5diff Cal calibrator run
- Checked to ensure valid data is used as you are deciding whether or not to recalibrate
- Compared to the preset Cal CV limits displayed inside the CV Limits box

  Setup ▸▸ Quality Assurance ▸▸ Administrator Password required ▸▸ **General** tab

Reference Values

- The assigned value for each parameter
- Assigned value obtained from the A^C•T 5diff Cal calibrator package insert

Calibration Log

- Click on **Analyzer / Logs** tab ▸▸ **Calibration Log** tab to view the log
- Maintains the most recent 50 calibration entries displaying the most recent log entry at the top of the log and the oldest entry at the bottom of the log
- Rollover is first in, first out when the 51st entry is added
- Log contains the following information fields

  Note: It is necessary to scroll right to see some of these fields.

Information Fields

Field	Function
Date/Time	Displays when the calibration was completed
Lot Number	Displays the lot number of calibrator used to perform the calibration; the word "Manual" appears in this field if the operator manually enters the calibration factors
Opr	Displays the User name logon information in the Workstation when the calibration was completed
Expiration Date	Displays operator entry identifying the expiration date for the calibrator material used to perform the calibration; field is blank if calibration factors are entered manually
Forced	If checked, indicates the accepted calibration factors are not based on ideal results (at least one CV was too high or at least one calibration factor was not within specified limits) or the calibration factors were entered manually **Never force a calibration without first consulting with a Beckman Coulter Representative!**
WBC Cal Factor	Calibration factor used in determining the reported WBC count result
RBC Cal Factor	Calibration factor used in determining the reported RBC count result
HGB Cal Factor	Calibration factor used in determining the reported HGB result
HCT Cal Factor	Calibration factor used in determining the reported HCT result
PLT Cal Factor	Calibration factor used in determining the reported PLT count result
RDW Cal Factor	Calibration factor used in determining the reported RDW result: • Is not determined during Auto-Calibration • Must be determined using the Manual Calibration procedure in Appendix C of the Operator's Guide • Only appears if the calibration factor is manually entered
MPV Cal Factor	Calibration factor used in determining the reported MPV result: • Is not determined during Auto-Calibration • Must be determined using the Manual Calibration procedure in Appendix C of the Operator's Guide • Only appears if the calibration factor is manually entered

Comment	Displays any comments entered by operator performing calibration: • Optional entry that provides an opportunity for the operator to type a personal observation or appraisal • Operator may use the **Add Comments** button to access the Add Comments box, as desired • If **Prompt User For Comments** check box is checked (✓), the Add Comments box pops up automatically at the end of calibration **Note:** When Add Comments box is displayed, operator cannot access any other window until they either: ▸ Enter a comment then click  to save and exit the box or ▸ Click  to exit the box without entering a comment

If A^{C}•T 5diff CP Hematology Analyzer is a Backup Instrument

- If your A^{C}•T 5diff CP system is being used as a backup to another COULTER hematology analyzer, you may need to perform a manual calibration to determine the RDW calibration factor
- See the instructions in Appendix C of the A^{C}•T 5diff CP Operator's Guide, as needed
- Operator should contact their Beckman Coulter Representative if there are questions

OBJECTIVES

When the subject is complete, you will be able to . . .

Specimen Handling

- State the proper anticoagulant to use for specimens processed on an A^{C}•T 5diff CP hematology analyzer.
- State the minimum volume of blood needed for processing a specimen.
- State the maximum storage time prior to running a specimen for CBC versus CBC/DIFF analysis.
- State the proper storage temperature.
- Properly insert a tube into the tube holder and close the cap-pierce door.

Specimen Identification

- Cite the window used to run patient specimens
- State the identifier that must be entered prior to running a patient specimen.
- Cite three options for entering a sample ID.
- Explain the autonumbering feature.
- Demonstrate how to start autonumbering then how to cancel its usage.
- Cite two situations that allow the sample ID to be duplicated.

Flagging Sets

- Explain the purpose of a flagging set.
- Demonstrate how to set up a flagging set.
- Contrast patient ranges versus action ranges.
- When given different scenarios by your instructor, cite the flagging set that would be applied to the patient results.

Workflow Options

- Define Worklist.
- Explain how the database, archive, and worklist are related.
- Demonstrate how to create an archive.
- Explain how to identify when a previously closed archive is open.
- Identify the two reasons why your laboratory may want to use the Worklist.
- Run patient samples using the Worklist and without using the Worklist.

Detailed Run Results

- Name and define each area on the Run window.

NOTES

SPECIMEN HANDLING

Specimen Collection

Approved Collection Devices and Control Vials

- Wide variety of specimen tubes, microcollection devices, and control vials are approved for use on the A^{C}•T 5diff CP hematology analyzer
- Closed-vial sampling is recommended for those evacuated collection tubes and control vials with pierceable stoppers or caps
- For those evacuated collection tubes, microcollection devices, and calibrator vials without pierceable stoppers or caps:
  - Stopper or cap must be removed prior to sampling (open-vial sampling)
  - Open-vial sampling increases the operator's exposure to the whole-blood specimen which is considered a biohazardous material
- List of approved specimen tubes, microcollection devices, and control vials is located in Appendix D of the Operator's Guide

Using Evacuated Collection Tubes

- Collect venous blood specimens in K_3EDTA or K_2EDTA
  - A proper proportion of whole blood to anticoagulant is critical
  - Collect venous blood according to the tube manufacturer's requirements
  - K_3EDTA is the preferred anticoagulant
- At collection and before analysis, mix each blood specimen gently and thoroughly according to the tube manufacturer's recommendations and your laboratory's protocol

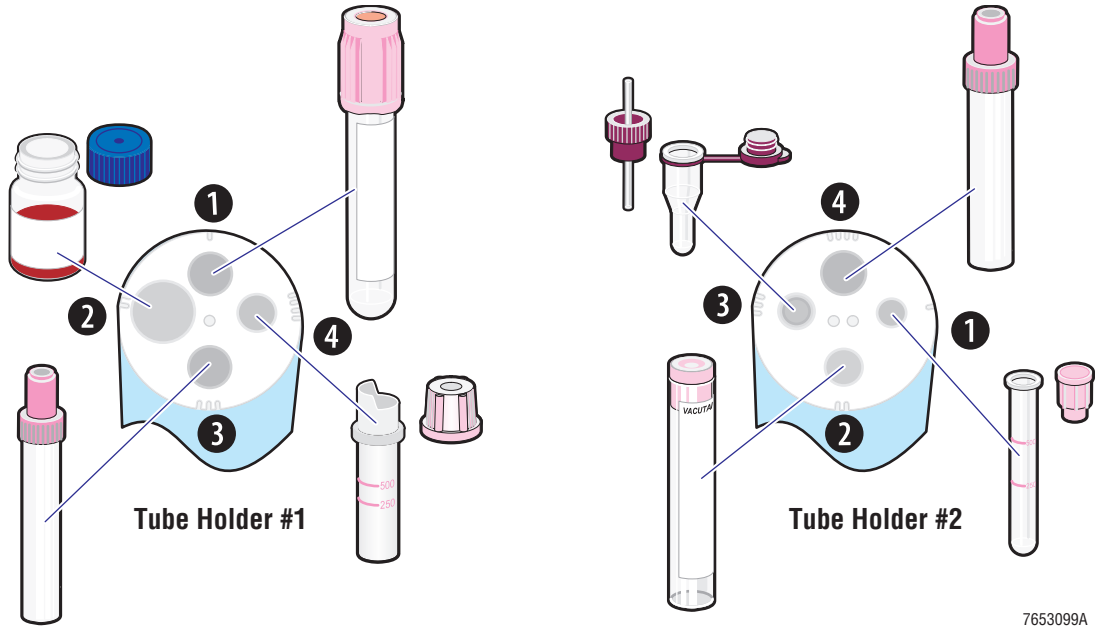
Using Microcollection Devices

- Anticoagulated capillary specimens collected in a microcollection device may be aspirated as an open-vial specimen
- Proper collection is critical to avoid micro-clots and/or debris
- Review and follow the collection and mixing instructions in the package insert provided by the manufacturer
- Make sure the manufacturer's minimum fill volume is achieved

Storage

- Run a CBC/Diff specimen within 24 hours of collection
- Run a CBC specimen within 48 hours of collection

Overview of Tube Holder Sample Positions



Note: This list below is not comprehensive. It is simply provides an overview of the tube holders and the approved collection devices and control vials they accommodate. For a detailed list, see Appendix D in the Operator's Guide.

Tube Holder #1	Type of Collection Device or Control Vial
Sample Position ❶	✓ Most 13 mm x 75 mm evacuated specimen tubes containing either K_3EDTA or K_2EDTA for collecting whole-blood volumes of 2 to 5 mL ✓ COULTER® A^C•T™ 5diff Control Plus control tubes
Sample Position ❷	✓ COULTER® A^C•T™ 5diff Cal Calibrator vial
Sample Position ❸	✓ Sarstedt Monovette® 11.5 mm x 66 mm specimen tube collecting 2.7 mL of whole-blood
Sample Position ❹	✓ Becton-Dickinson Microtainer for collection of 0.25 to 0.50 mL of whole-blood
Tube Holder #2	
Sample Position ❶	✓ Becton-Dickinson Microtainer for collection of 0.25 to 0.50 mL of whole-blood
Sample Position ❷	✓ Becton-Dickinson 10.25 mm x 64 mm Vacutainer® for collecting 3 mL of whole-blood
Sample Position ❸	✓ RAM Scientific microcollection device for collecting 125 µL of whole-blood
Sample Position ❹	✓ 13 mm x 75 mm specimen tube with multiple labels ✓ COULTER® A^C•T™ 5diff Control Plus control tubes

SAMPLE IDENTIFICATION

- Patient samples must be processed with the Run window open (click **Run** tab)
- Sample identifiers may be entered on the Run window or on the Worklist

  Note: A worklist is a list of samples with preassigned identification and demographic information that are pending analysis. Details about using the Worklist are covered later.

Sample ID

- Sample ID is required to process a patient sample on this instrument
- Sample identifier may be entered in the Next Sample ID field on the Run window or in the Sample ID field on the Worklist
- Up to 16 alphanumeric characters can be entered in either field
- Must be entered before the specimen can be processed

Options for Entering Sample ID

Manual Entry

- On the Run window inside the sample identification box:
  - Sample ID for the next specimen can be manually typed into the Next Sample ID field
  - Two other entries can also be made inside this box
    - Panel selection for the next specimen - default is CBC/DIFF
    - Patient ID for the next specimen - optional entry of up to 25 alphanumeric characters
- On the Worklist:
  - Sample ID for the next specimen can be manually typed into the Sample ID field
  - Panel selection and other demographic information can also be entered for this patient

Optional Bar-Code Reader

- Hand-held scanner for reading the bar-code label on a specimen tube
- Sample ID may be scanned into the Sample ID Next field on the Run window or into the Sample ID field on the Worklist
- Operator must press the button on the bar-code reader before scanning a label
- Risk of sample misidentification if the entire barcode is not captured with the bar-code reader, especially with the interleaved 2-of-5 bar-code format
- Position the reader over the entire barcode to capture the entire sample ID
- An audible beep indicates the barcode was read successfully
- Verify the correctness of each bar-code reading to ensure proper sample identification
- Barcode specifications are located in Appendix B of the Operator's Guide
- Set up is performed by your Beckman Coulter Representative

Autonumbering Feature

- Instrument automatically assigns a sample ID (from 1 to 999999)
- Sample ID number automatically increments by 1 from the previously assigned number before each analysis
- Autonumbering is turned on via the Add/Edit Worklist box; however, it is not necessary to use the Worklist to use Autonumbering
  - Click **Worklist** tab ▸▸  to display the Add/Edit Worklist box
  - Inside the Autonumbering box, click ☐ to place a ✓ inside the box (☑); this enables the option
  - Enter the starting number (1 to 999999) for the first specimen
  - Click **Apply** to save the change
  - Click  to exit the box
  - Click **Run** tab to verify the designated starting number is in the Sample ID Next field
  - When the first specimen is processed:
    - Designated starting number moves to the Sample ID In Progress field
    - Number in the Sample ID Next field is one digit higher
- When autonumbering is enabled, the sample ID number increments inside both the Sample ID Next field on the Run window and the Sample ID field on the Worklist
- Operator may override the current autonumber as desired; autonumbering automatically begins again with the autonumber that was overwritten

Duplicate Sample ID Check

- Workstation database maintains a record of all sample IDs processed
- Identifier entered in the Sample ID field is compared to the list of Reserved lot numbers
  - If the number matches a Reserved lot number, corresponding entry fields are automatically completed
  - If the number does not match a Reserved lot number, the software searches for a duplicate sample ID number throughout the current active archive then among the sample ID numbers waiting to be processed, the number is accepted only if it is unique
- If an operator enters a sample ID that has already been used,
  - Message *Patient Results With Matching Sample ID Already Exist. Existing Sample ID Cannot Be Reused. Enter Another Sample ID or Use Rerun* appears
  - When operator clicks **OK**, the message *Halted Sample Processing. If Analyzing Samples Please Verify The Next Sample ID Before Continuing.* appears

  Note: This duplicate check is not performed if  (the Rerun feature) is being used.
- If your laboratory sample ID sequence repeats **or** if your laboratory is using the autonumbering feature and the number sequence is being restarted on a daily, weekly, or monthly cycle, a storage process called archiving must be done on a routine basis to clear the current memory of used sample ID numbers

- Archiving process
  - Resets the duplicate sample ID process so that the sample IDs can be reused
  - How often your laboratory archives depends on the frequency your laboratory repeats sample IDs - daily?, weekly?, monthly?, annually?

    Note: If you repeat sample IDs on an annual basis, it is recommended that you archive monthly. Small archives are easier to navigate through and easier to manage than large ones. Large archives may cause the system response time to decrease.

Rerunning a Patient Sample

- When you want to repeat a sample, must use  (system's Rerun feature)
- Due to the duplicate ID feature, if sample ID to be repeated was manually re-entered into the instrument, a *Duplicate Sample ID* message appears and specimen cannot be processed again
- Can indicate you want to rerun any specimen from the Results window or the Detailed Results window
- From Run window, can indicate you want to rerun the last specimen
- When in the Detailed Results view and the operator clicks  :
  - Sample Results Status box turns red to indicate a rerun is requested on this sample
  - Sample Results Status box appears in the top right of the screen, just to the right of the Panel field
  - When specimen is rerun and the rerun results are displayed, Sample Results Status box associated with the rerun results is white which indicates the most recent set of results
    - Run can be validated (Sample Results Status box turns green)
    - Once validated (confirmed acceptable to report), specimen with that sample ID number cannot be rerun
  - Sample Results Status box associated with the original results remains red to indicate the results are the old (or previous) results; this run cannot be validated
  - Either set of results may be printed and/or transmitted
- In the Results List view, the first analysis and the rerun appear in the order processed

FLAGGING SETS

- A flagging set contains patient ranges and action ranges
- If a parameter result is outside the designated range, the result is flagged
- A^{C}•T 5diff CP hematology analyzer provides the potential for 20 different flagging sets
- At the Menu bar, select **Setup ▸▸ Patients ▸▸ Administrator Password ▸▸ Flagging and Messaging** tab to locate the available flagging sets

Pre-Defined Flagging Sets

- When installed, the system has six pre-defined flagging sets

Flagging Set Number	Flagging Set Name	Age Range
1	Standard Range	No defined range
2	Man	>12 years
3	Woman	>12 years
4	Newborn	0 to <= 30 days
5	Infant	1 month to < 6 years
6	Child	>= 6 years to <= 12 years

- Standard Range is the default flagging set, but can change the default selection to any of the 20 flagging sets to meet your laboratory's needs
- With the exception of the Standard Range flagging set, both action and patient ranges may be edited to best fit your laboratory's needs

Additional Flagging Sets

- Can add up to 14 additional flagging sets (numbers 7 through 20)
- When an operator clicks the **Setup** button to set up another flagging set, the Standard Range patient and action values are automatically copied into the table for the new flagging set; values may be edited as desired
- Action and/or Patient Limits may be copied from one flagging set to another

Flagging

Patient Ranges

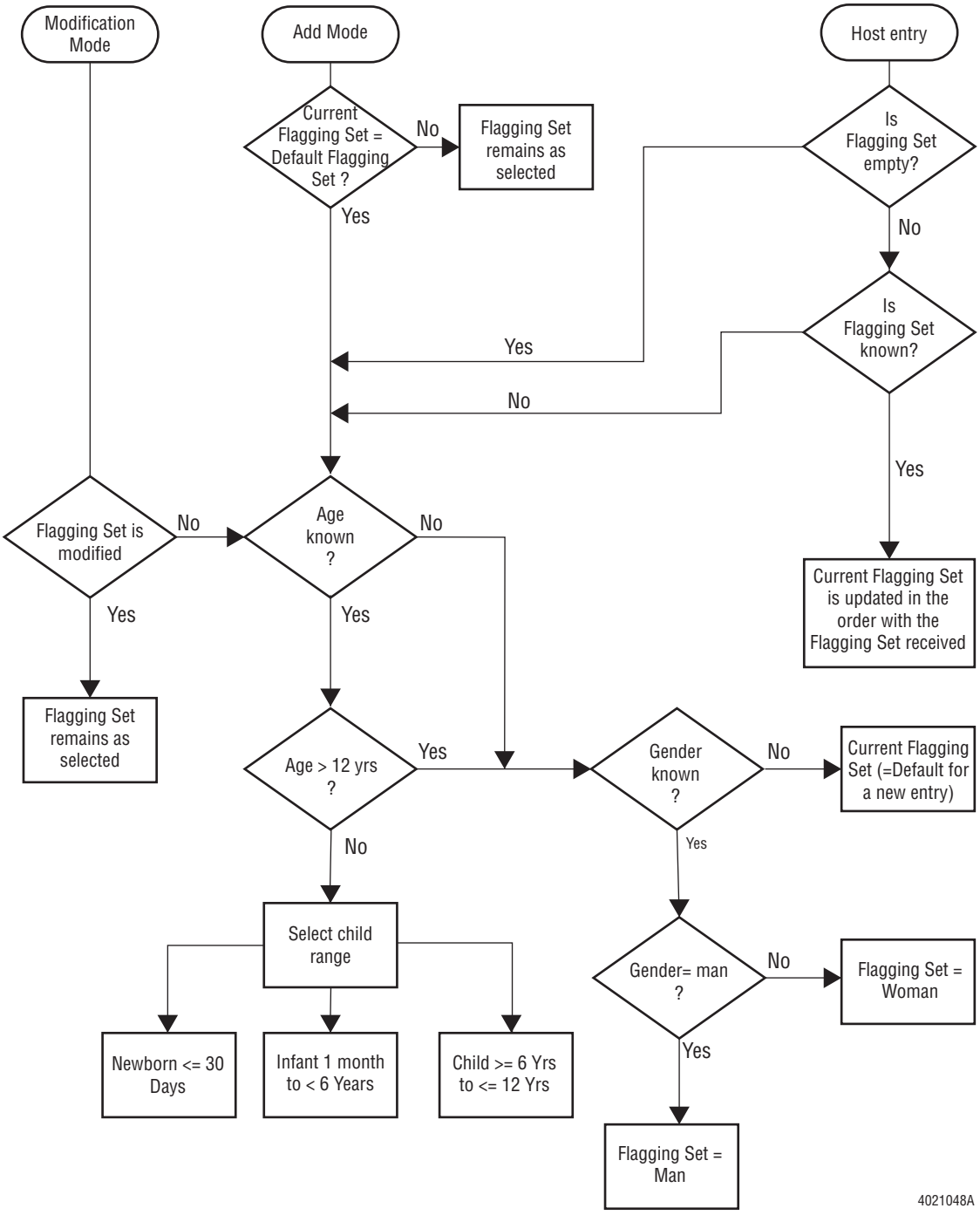
- *L* flag appears to the right of the parameter result that is below the lower limit in the Patient L column
- *H* flag appears to the right of the parameter result that is above the upper limit in the Patient H column
- Yellow background accompanies the flag appearing inside the parameter result field

Action Ranges

- *LL* flag appears to the right of the parameter result that is below the lower limit in the Action L column
- *HH* flag appears to the right of the parameter result that is above the upper limit in the Action H column
- Red background accompanies the flag appearing inside the parameter result field
- May also generate an interpretive message

Flagging Set Hierarchy

- Worklist may be used to assign a flagging set
- Flagging set is automatically selected based on the gender and age (or date of birth) entered on the Worklist unless a flagging set is manually selected by the operator
- If both gender and age (or date of birth) are not entered and no flagging set is specified, the following hierarchy is used to decide which flagging set will be applied to the results for that particular sample ID



WORKFLOW OPTIONS

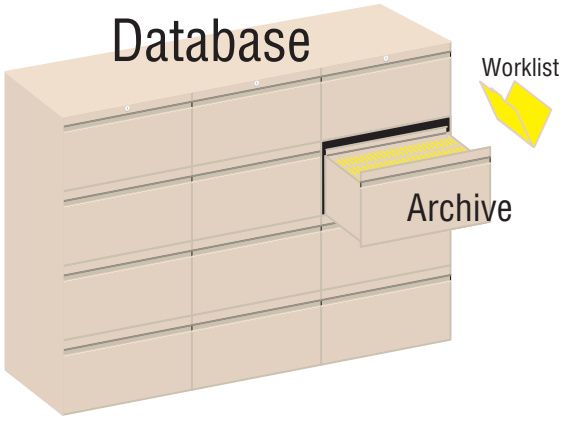
- Workflow patterns on the A^{C}•T 5diff CP hematology analyzer evolve around whether or not your laboratory will use a Worklist
- Worklist provides several functions that may be important to your laboratory
  - Provides a way to enter patient demographics such as patient name, gender, date of birth, age, physician's name, clinic location, and comments so that this information can be linked with the final results for storage
  - Allows a choice of flagging sets

Worklist

- A worklist is a list of work to be done
- Worklists are used to preassign identifiers and/or demographics for specimens that are pending analysis
  - Sample ID identifier is a required entry (up to 16 alphanumeric characters)
  - Demographics include optional information such as patient ID, patient name, gender, date of birth, age, physician's name, clinic location, and comments
  - Identifiers and demographic information must be entered before running the patient specimen
  - Sample ID is the only required entry; all other entries are optional
- Host transmission protocol can be established so that demographics are automatically downloaded from the host computer to the Worklist
- After the specimen is processed, all entered information is printed on the final report, stored in the active archive, and transmitted to a host computer (if applicable)
- To decide if using a worklist best fits your laboratory's needs means considering several factors including the type of information you desire archived (stored) in your database

Database, Archive, and Worklist Relationships

- On this system, the database is like a large electronic storage "cabinet" on the Workstation's hard drive
- Within the database, the archive is like a "file cabinet" and the worklist like a "folder" within the file cabinet



- Workstation database maintains a record of all sample IDs processed
- Worklist checks for duplicate sample IDs within the active archive (current archive)
- If your laboratory sample ID sequence repeats **or** if your laboratory is using the autonumbering feature and the number sequence is being restarted on a daily, weekly, or monthly cycle, archiving must be done on a routine basis to reset the duplicate sample ID process so that the sample IDs can be reused
  - How often your laboratory archives depends on the frequency your laboratory repeats sample IDs - daily?, weekly?, monthly?, annually?
  - If sample IDs are repeated annually or not repeated at all, monthly archiving is recommended
  - Small archives are easier to navigate through and manage than large ones
  - Large archives may cause the system response time to decrease

Creating an Archive

- To store patient samples, an archive must be created
- At the Menu bar, select **File ▸▸ New archive**
- Beckman Coulter recommends a new archive be created at frequent intervals to ensure system processing is not affected by a large current archive
- When a new archive is created:
  - Previous archive is automatically closed
  - New archive is now the current or active archive
  - Create a new archive at least once a month
- When a new archive is created, the date and time are recorded and act as the identifier for that archive and remains with the archive for as long as it exists
- It is not possible to close an active archive without creating a new archive!
  - Action of selecting **File ▸▸ Close archive** does not close an active archive
  - Only way an active archive can be closed is to create a new archive

Using the Active Archive

- All analyzed results including reproducibility, blanks, controls, and calibrators are stored in the active archive until another new archive is created
- If your laboratory created an archive it is likely you will want to retrieve those results at some time; therefore, the identifiers your laboratory chooses to use are important
- Sample ID cannot be duplicated within an active archive without using the Rerun feature
- Preassigned identifiers and/or demographics for each specimen and the results obtained during sample analysis are linked together and are considered as a single archived component within the archive
  - Means that when an archive is closed, the Worklist information and parameter result information for all the samples processed while the archive was the active archive are stored at the same time

Opening a Closed Archive

- **Sample analysis is not permitted while a previously closed archive is open!**
- Never attempt to open a previously closed archive while the active archive is still accumulating data
- To open a previously closed archive:
  - At the Menu bar, select **File ▸▸ Open archive**
    - Table listing all closed archives appears
    - Archive date is the date the archive was first created
  - Click on the archive you wish to open
- When a closed archive is opened:
  - All analyzed samples are listed in the worklist format, including reproducibility, blanks, controls, and calibrators
  - Background color changes to "light green" to indicate you are reviewing archived, not current, information
  - At the Menu bar, select **File ▸▸ Close archive** to close the open archive

Summary

- Use the Worklist if choosing a flagging set or entering patient demographics is important (either manually or via an automatic download from your host computer)

Using the Worklist

Adding New Entries to the Worklist

- Click **Worklist** tab ▸▸  to display the Add/Edit Worklist box
- Sample ID is still a required entry (up to 16 alphanumeric characters)
- CBC/DIFF panel is default selection; select CBC panel from the drop-down box if desired
- Flagging Set for a patient older than 12 years
  - Flagging set is automatically selected based on the age and gender entered on the Worklist unless a flagging set is manually selected by the operator
  - If both age and gender are not entered and a flagging set is not selected, the default flagging set is automatically selected when the Worklist entry is saved
  - If the age and/or gender for an existing Worklist entry is modified, the flagging set is automatically modified to match the new age and/or gender
- Flagging Set for a patient 12 years old or younger
  - Flagging set is automatically selected based on the age entered on the Worklist; gender is not considered
  - If the age for an existing Worklist entry is modified, the flagging set is automatically modified to match the new age
- Collect date must follow an eight-digit format; for example, 04 02 2001
- Collect time uses AM/PM format
- Comment field - may enter up to 50 alphanumeric characters

- When Patient ID field is selected for entry:
  - List of existing Patient IDs with their respective patient names are listed
  - If operator selects from the list, the existing patient demographics are automatically entered into the respective fields
  - Operator may choose to make a manual entry of up to 25 alphanumeric characters
- Patient Name field - may enter up to 30 alphanumeric characters
- If no date of birth is available, operator can enter the patient's age
- Gender
  - Unknown is the default
  - Male or female may be selected from a drop-down box
- Physician field
  - Select name from a list of physicians or manually enter
  - May enter up to 30 alphanumeric characters
- Location field
  - Select name from a list of locations or manually enter
  - May enter up to 15 alphanumeric characters
- Click  to make enter another specimen
- When last entry to Worklist is complete, click  to save entries and exit box
- If after the last entry operator clicks  then  , blank entry is inserted on Worklist (because  saves and exits)

Using the Autonumbering Feature with the Worklist

- Instrument automatically assigns a sample ID (from 1 to 999999)
- Sample ID number automatically increments by 1 from the previously assigned number before each analysis
- Autonumbering is turned on via the Add/Edit Worklist box:
  - Click **Worklist** tab ▸▸  to display the Add/Edit Worklist box
  - Inside the Autonumbering box, click ☐ to place a ✓ inside the box (☑); this enables the option
  - Enter the number (1 to 999999) for the first specimen
  - Click **Apply** to save the change
  - Click  to exit the box
  - Create the Worklist
  - Click **Run** tab to verify the designated starting number is in the Sample ID Next field
  - When the first specimen is processed,
    - Designated starting number moves to the Sample ID In Progress field
    - Number in the Sample ID Next field is next entry on the Worklist (which should be one digit higher)
- When autonumbering is enabled, the sample ID number increments inside both the Sample ID Next field on the Run window and the Sample ID field on the Worklist
- Operator may override the current autonumber as desired; autonumbering automatically begins again with the autonumber that was overwritten

Processing Patient Samples Using the Worklist

- Not necessary to process patient specimens in the Worklist order
- If when you check the Sample ID Next field, you want to run a different specimen:
  - Manually type the desired sample ID or scan the bar-code label on its specimen tube into the Sample ID Next field
  - System automatically matches the desired sample ID with the panel and demographic information previously entered on the Worklist
  - Ready to process the specimen

➤ RUNNING PATIENT SAMPLES USING THE WORKLIST SUMMARY

Note: Use this summary only if you want to use the Worklist. If you do not want to run samples using the Worklist, use the Running Patient Samples Without Using the Worklist Summary.

1 Get the Workstation Ready

- Click **Results** tab to verify only the active archive is open (background is white).
  - If the background is green, an old archive is open and must be closed.
  - At the Menu bar, select **File ▸▸ Close archive** to close the old archive.
- If, according to your laboratory protocol, it is time to create a new archive, select **File ▸▸ New archive**.
- Click **Worklist** tab.

If the Worklist is Set Up Manually

Autonumbering OFF

- Click  .

Inside the Add/Edit Worklist box:

- Enter sample ID:
  - Click on the Sample ID field.
  - Manually type the ID number at the keyboard.
    or
    Scan the specimen tube's bar-code label with the bar-code wand.
  - Verify the ID number inside the Sample ID field is correct.
- CBC/DIFF is the default panel selection; or select CBC from the drop-down box.
- Flagging set currently selected as default is entered; select another flagging set from the drop-down box, if desired.
- Enter comment or other patient demographic information as desired. (If enter gender and/or age and a flagging set is not manually selected, the flagging set automatically changes accordingly.)
- Click  to save entries and clear the box for entering the identifiers and demographics for another specimen.
- When the last entry to the Worklist is complete, click  to save and exit.
- Click **Run** tab.

Autonumbering ON

- Click  .

Inside the Add/Edit Worklist box:

- Sample ID entry is automatic.
  To override the autonumber with another sample ID:
  - Highlight the autonumber in the Sample ID field.
  - Manually type the desired sample ID number at the keyboard.
  - Verify the ID number inside the Sample ID field is correct.
    Note: Autonumbering automatically begins again with the autonumber that was overwritten.
- CBC/DIFF is the default panel selection; or select CBC from the drop-down box.
- Flagging set currently selected as default is entered; select another flagging set from the drop-down box, if desired.
- Enter comment or other patient demographic information as desired. (If enter gender and/or age and a flagging set is not manually selected, the flagging set automatically changes accordingly.)
- Click  to save entries and clear the box for entering the identifiers and demographics for another specimen.
- When the last entry to the Worklist is complete, click  to save and exit.
- Click **Run** tab.

If the Worklist is Downloaded from a Host Computer

- Sample ID and patient demographics are automatically downloaded from the host computer. (Information downloaded from the host computer cannot be edited.)
- Click **Run** tab.

2 Process the Sample

- Check the Sample ID Next field for the next specimen to be processed.

  Note: If you want to run another specimen, manually type the sample ID or scan the bar-code label on the specimen tube. The system automatically matches the desired sample ID with the panel and demographic information previously entered on the Worklist.
- Verify the tube holder is appropriate for the specimen tube being analyzed. If not, change the tube holder.
- Mix the specimen gently but thoroughly according to your laboratory's protocol.
- If the specimen tube does not have a pierceable stopper, remove the stopper.
- Place the well-mixed specimen tube in the sample position that best matches the tube. Rotate the specimen tube to the pierce position (12 o'clock position), as needed.
- Close the cap-pierce door to initiate the cycle.
- Remove the specimen tube when the cap-pierce door automatically opens after aspiration is complete. Notice the red LED is still glowing, the Analyzer is busy processing the sample.
- Overlap sample processing, if desired.
  - Repeat step 2 to get ready for the next cycle.
  - When the cycle is complete (green LED glows steady), go to step 3.

3 When the Cycle is Complete

- Verify the sample results appear in the Run window.
- Verify the sample ID entry and results before reporting.
- Click  for a hard copy of the results.

  Note: A hard copy prints automatically if the Auto-Print function is enabled.
- Repeat steps 2 and 3 until all specimens are analyzed.

➤➤

➤ RUNNING PATIENT SAMPLES WITHOUT USING THE WORKLIST SUMMARY

Note: Use this summary only if you do not want to use the Worklist. If you want to run samples using the Worklist, use the Running Patient Samples Using the Worklist Summary.

1 Get the Workstation Ready

- Click **Results** tab to verify only the active archive is open (background is white).
  - If the background is green, an old archive is open and must be closed.
  - At the Menu bar, select **File ▸▸ Close archive** to close the old archive.
- If, according to your laboratory protocol, it is time to create a new archive, select **File ▸▸ New archive**.
- Click **Run** tab.

Autonumbering OFF

- Enter Next Sample ID:
  - Click on the Sample ID Next field.
  - Manually type the ID number at the keyboard.
    or
    Scan the specimen tube's bar-code label with the bar-code wand.
  - Verify the ID number inside the Sample ID Next field is correct.
- CBC/DIFF is the default panel selection; or select CBC from the drop-down box.
- Enter a Patient ID if desired.

Autonumbering ON

- Next Sample ID entry is automatic.

  To override the autonumber with another sample ID:
  - Highlight the autonumber in the Sample ID Next field.
  - Manually type the desired sample ID number at the keyboard.
  - Verify the ID number inside the Sample ID Next field is correct.

    Note: Autonumbering automatically begins again with the autonumber that was overwritten.
- CBC/DIFF is the default panel selection; or select CBC from the drop-down box.
- Enter a Patient ID if desired.

2 Process the Sample

- Verify the tube holder is appropriate for the specimen tube being analyzed. If not, change the tube holder.
- Mix the specimen gently but thoroughly according to your laboratory's protocol.
- If the specimen tube does not have a pierceable stopper, remove the stopper.
- Place the well-mixed specimen tube in the sample position that best matches the tube. Rotate the specimen tube to the pierce position (12 o'clock position), as needed.
- Close the cap-pierce door to initiate the cycle.
- Remove the specimen tube when the cap-pierce door automatically opens after aspiration is complete. Notice the red LED is still glowing, the Analyzer is busy processing the sample.
- Overlap sample processing, if desired.
  - Go to step 3 to get ready for the next cycle.
  - When the current cycle is complete, go to step 4.

3 Prepare for the Next Cycle

- Enter information for the next specimen while the current sample is being analyzed.

Autonumbering OFF

- Enter Next Sample ID:
  - Click on the Sample ID Next field.
  - Manually type the ID number at the keyboard.
    or
    Scan the specimen tube's bar-code label with the bar-code wand.
  - Verify the ID number inside the Sample ID Next field is correct.
- CBC/DIFF is the default panel selection; select CBC from the drop-down box, if desired.
- Enter a Patient ID if desired.
- If you are overlapping, go to step 2.

  If you are not overlapping, when the cycle is complete (green LED glows steady), go to step 4.

Autonumbering ON

- Next Sample ID entry is automatic.

  To override the autonumber with another sample ID:
  - Highlight the autonumber in the Sample ID Next field.
  - Manually type the desired sample ID number at the keyboard.
  - Verify the ID number inside the Sample ID Next field is correct.

    Note: Autonumbering automatically begins again with the autonumber that was overwritten.
- CBC/DIFF is the default panel selection; select CBC from the drop-down box, if desired.
- Enter a Patient ID if desired.
- If you are overlapping, go to step 2.

  If you are not overlapping, when the cycle is complete (green LED glows steady), go to step 4.

4 When the Current Cycle is Complete

- Verify the current sample results appear in the Run window.
- Verify the sample ID entry and results before reporting.
- Click  for a hard copy of the results.

  Note: A hard copy prints automatically if the Auto-Print function is enabled.
- Repeat steps 2, 3, and 4 until all specimens are analyzed.

➤➤

DETAILED RUN RESULTS

Run Window

1. Run tab
2. Sample ID and Patient Demographics
3. DiffPlot
4. Flags and Messages Area
5. DIFF Parameter Results
6. PLT Histogram
7. RBC Histogram
8. Sample Identification Box
9. WBC/BASO Histogram
10. PLT Parameter Results
11. RBC Parameter Results
12. WBC Parameter Result

Detailed Run Window Areas

The information in this section describes the various areas of the Detailed Run Results window. Refer to the Run window example as needed.

Click on the **Run** tab ❶ to display the Run window.

Note: Numbers like ❶ are inserted to help you locate a referenced area on the Run window (located on the previous page).

Sample ID and Patient Demographics Area ❷ ❽

- Seq #
  - Sequence number
  - Automatically incremented by the software
  - Unique chronological number for specimen sampling
- Sample ID
  - Sample identification
  - Alphanumeric character field, maximum of 16 characters
  - Sample ID entry may be made in the Sample ID Next field inside the Sample Identification box on the Run window or in the Sample ID field in the Add/Edit Worklist box prior to analysis
  - Field is compared to the list of Reserved lot numbers

- Patient ID (Optional)
  - Patient identification
  - Alphanumeric character field, maximum of 25 characters
  - Patient ID entry may be made in the Patient ID Next field inside the Sample Identification box on the Run window or in the Patient ID field in the Add/Edit Worklist box
  - Duplicates are allowed
  - Entry must be made prior to analysis
  - May enter both patient ID and patient name in the Add/Edit Worklist box, if desired
- Patient Name (Optional)
  - Name of the patient
  - Alphanumeric character field, maximum of 30 characters
  - Entry can only be made in the Patient Name field in the Add/Edit Worklist box
  - Cannot add a patient name to results with a patient ID
  - If a patient ID is not entered in the Add/Edit Worklist box, patient name can be added while the Results List or the Detailed Results view is displayed
  - Entry must be made prior to analysis
  - May enter both patient name and patient ID in the Add/Edit Worklist box, if desired
- Gender (Optional)
  - Male or Female
  - Unknown displayed when a selection is not made
  - Entry can only be made in the Gender field in the Add/Edit Worklist box prior to analysis
  - Entry required if you want the software to automatically select the most appropriate flagging set for a patient older than 12 years
    - Flagging set is automatically selected based on the age and gender entered on the Worklist unless a flagging set is manually selected by the operator
    - If both age and gender are not entered and a flagging set is not selected, the default flagging set is automatically selected when the Worklist entry is saved
    - Flagging set for a patient 12 years old or younger is automatically selected based on the age entered on the Worklist; gender is not considered
    - If the age and/or gender for an existing Worklist entry is modified, the flagging set is automatically modified to match the new age and/or gender
- Date of Birth (Optional)
  - Patient date of birth
  - Date is displayed in the format selected by the laboratory under **Setup ▸▸ System ▸▸ Date/Time**
  - Entry can only be made in the Date of Birth field in the Add/Edit Worklist box prior to analysis
  - Field for displaying the patient's age does not appear on the Run window

- Date of birth or age entry is required if you want the software to automatically select the most appropriate flagging set
  - Flagging set is automatically selected based on the age and gender entered on the Worklist unless a flagging set is manually selected by the operator
  - If both age and gender are not entered and a flagging set is not selected, the default flagging set is automatically selected when the Worklist entry is saved
  - Flagging set for a patient 12 years old or younger is automatically selected based on the age entered on the Worklist; gender is not considered
  - If the age and/or gender for an existing Worklist entry is modified, the flagging set is automatically modified to match the new age and/or gender

- Panel
  - Displays the panel requested for this analysis
  - CBC/DIFF is the default panel selection
  - If CBC/DIFF panel selected, typically 20 parameters are reported
    - CBC parameters: WBC, RBC, Hgb, Hct, MCV, MCH, MCHC, RDW, Plt, MPV
    - DIFF parameters: NE%, NE#, LY%, LY#, MO%, MO#, EO%, EO#, BA%, BA#
    - If RUO (Research Use Only) parameters are enabled, Pct, PDW, IMM%, IMM#, ATL%, and ATL# are also reported
  - If CBC panel selected, typically 10 parameters are reported
    - WBC, RBC, Hgb, Hct, MCV, MCH, MCHC, RDW, Plt, and MPV
    - If RUO (Research Use Only) parameters are enabled, Pct and PDW are also reported
  - Selection may be made in the Panel field inside the Sample Identification box on the Run window or in the Panel field in the Add/Edit Worklist box prior to analysis

- Sample Results Status Box
  - Provides a visual of the sample's review status
  - Box may be white, red, or green
  - White box indicates the current (latest) set of results
  - Red box indicates the sample is rerun and the results you are viewing are the old (or previous) results; these results can be printed or transmitted
  - Green box indicates a current sample that has been reviewed and validated
    - Once sample results are validated (confirmed acceptable to report), the specimen with that sample ID number cannot be rerun
    - Clicking  while viewing sample results validates those results

- Flagging Set
  - Displays the flagging set being used for the results of this patient sample
  - May be the Default flagging set, a flagging set assigned by the operator, or a flagging set automatically selected by the software based on the patient's gender and/or age (or date of birth)
  - Flagging Set for a patient older than 12 years
    - Flagging set is automatically selected based on the age and gender entered on the Worklist unless a flagging set is manually selected by the operator
    - If both age and gender are not entered and a flagging set is not selected, the default flagging set is automatically selected when the Worklist entry is saved
    - If the age and/or gender for an existing Worklist entry is modified, the flagging set is automatically modified to match the new age and/or gender
  - Flagging Set for a patient 12 years old or younger
    - Flagging set is automatically selected based on the age entered on the Worklist; gender is not considered
    - If the age for an existing Worklist entry is modified, the flagging set is automatically modified to match the new age
  - If the operator wants to assign a flagging set, the selection must be made in the Flagging Set field in the Add/Edit Worklist box prior to analysis
- Run Date / Time
  - Displays the date and time of this run
  - Displayed in the format selected by the laboratory under **Setup ▸▸ System ▸▸ Date/Time**
  - Automatic entry when the specimen is actually run
- Comment (Optional)
  - Displays observations or remarks made by the operator
  - Alphanumeric character field, maximum of 50 characters
  - Can only enter remarks into the Comment field of the Add/Edit Worklist box prior to analysis
- Other patient demographics such as physician and location are not displayed on the Run window, but do appear on the printout if entered via the Add/Edit Worklist box prior to analysis

CBC Parameter Results Areas ❻ ❼ ❾ ❿ ⓫ ⓬

- WBC parameter result ⓬ is displayed with the WBC/BASO histogram ❾
- RBC parameter results ⓫ are displayed with the RBC histogram ❼
- Platelet parameter results ❿ are displayed with the PLT histogram ❻

DIFF Parameter Results Area ❺

- Displays percentage and absolute number for each differential parameter
  - NE and NE# = Neutrophil percentage (NE) and Neutrophil absolute number (NE#)
  - LY and LY# = Lymphocyte percentage (LY) and Lymphocyte absolute number (LY#)
  - MO and MO# = Monocyte percentage (MO) and Monocyte absolute number (MO#)
  - EO and EO# = Eosinophil percentage (EO) and Eosinophil absolute number (EO#)
  - BA and BA# = Basophil percentage (BA) and Basophil absolute number (BA#)

DiffPlot ❸

- Displays the major white blood cell groups
  - Lymphocytes
  - Monocytes
  - Neutrophils
  - Eosinophils
- Basophil percentage is determined from the WBC/BASO histogram. ❾

Flags and Messages Area ❹

- Displays various DiffPlot and histogram flags
- Displays interpretive messages
  - Message is triggered from the flagging limits established by laboratory protocol
  - Message indicates a possible pathological disorder
- Displays analytical alarms
  - Alarm in the form of a message generated by the Analyzer
  - Indicates a condition that may be related to Analyzer operation
  - Also indicates if results will be influenced by this condition
- Area label depends on **Detailed Flags** setting located under Patient Setup
  - At the Menu bar, select **Setup** ▸▸ **Patients** ▸▸ type **Administrator Password** ▸▸ **Reports** tab ▸▸ **Enable** ▸▸ **Detailed Flags**
  - When **Detailed Flags** setting is enabled, Flags and Messages label appears on Run window
  - When **Detailed Flags** setting is disabled, Suspect and Messages label appears on Run window

OBJECTIVES

When the subject is complete, you will be able to . . .

Data Storage and Retrieval

- Explain the purpose of an archive.
- Demonstrate how to create a current, active archive.
- Explain how to know if you are viewing a current, active archive or a previously closed archive that has been opened.
- Describe the process of sorting samples results in an archive.
- Demonstrate how to search for the results of a specific sample.
- Demonstrate how to view the detailed results for a specific sample.
- Print single sample results from an archive in the tabular and detailed format.
- Tag and batch print a group of sample results in the preferred format, tabular or detailed.
- Tag and batch transmit a group of results from an archive to a host computer, if applicable.

Basics of CBC/DIFF Analysis

- Briefly explain the Sequential Dilution System (SDS).
- State the Coulter Principle.
- State the relationship of resistance, cell volume, and pulse height.
- Define threshold, count periods, and voting.
- Explain the concept of channelization.
- Explain how pulses are used to develop a histogram.
- Briefly explain A^{C}V Differential Technology.

WBC Parameters

- List the individual parameters that comprise the WBC Profile.
- Identify which parameters in the WBC Profile are directly measured, derived from the WBC/BASO histogram, derived from the DiffPlot, or computed.
- Identify the flags that occur when the WBC count results do not agree.

RBC Parameters

- List the individual parameters that comprise the RBC Profile.
- Identify which parameters in the RBC Profile are directly measured, derived from the RBC histogram, or computed.
- Compute MCV, MCH, and MCHC results.
- Explain the process of building an RBC histogram.
- Explain the area of the RBC histogram used to determine the RDW parameter.
- Describe a typical RBC histogram.
- Identify the flags that occur when the RBC count results do not agree.

Plt Parameters

- List the individual parameters that comprise the PLT Profile.
- Identify which parameters in the PLT Profile are directly measured, derived from the Plt histogram, or computed.
- Explain the purpose of rinse flow system.
- Explain the process of building a PLT histogram.
- Explain the area of a typical PLT histogram used to determine the MPV parameter result.
- Explain the area of a typical PLT histogram used to determine the PDW parameter result.
- Describe a typical PLT histogram.
- Identify the flag that occurs when the PLT count results don't agree.
- State the condition that generates an SCL flag.
- When the MIC flag is generated, explain how you can determine if the platelet count and associated parameters are reliable.

Flagging

- Demonstrate how to locate details concerning flags, interpretive messages, or analytical alarms in the Instructions For Use manual.

DATA STORAGE

Database

- Database in your A^{C}•T 5diff CP hematology analyzer is capable of storing 10,000 results
- Stored data includes patient sample, blank, reproducibility, control, and calibrator results
- Because the database capacity is large, Beckman Coulter recommends a new archive be created on a daily, weekly, or monthly basis
  - Frequency is dependent on your laboratory workload and sample ID protocol
  - Small archives are easier to navigate through and easier to manage than larger ones

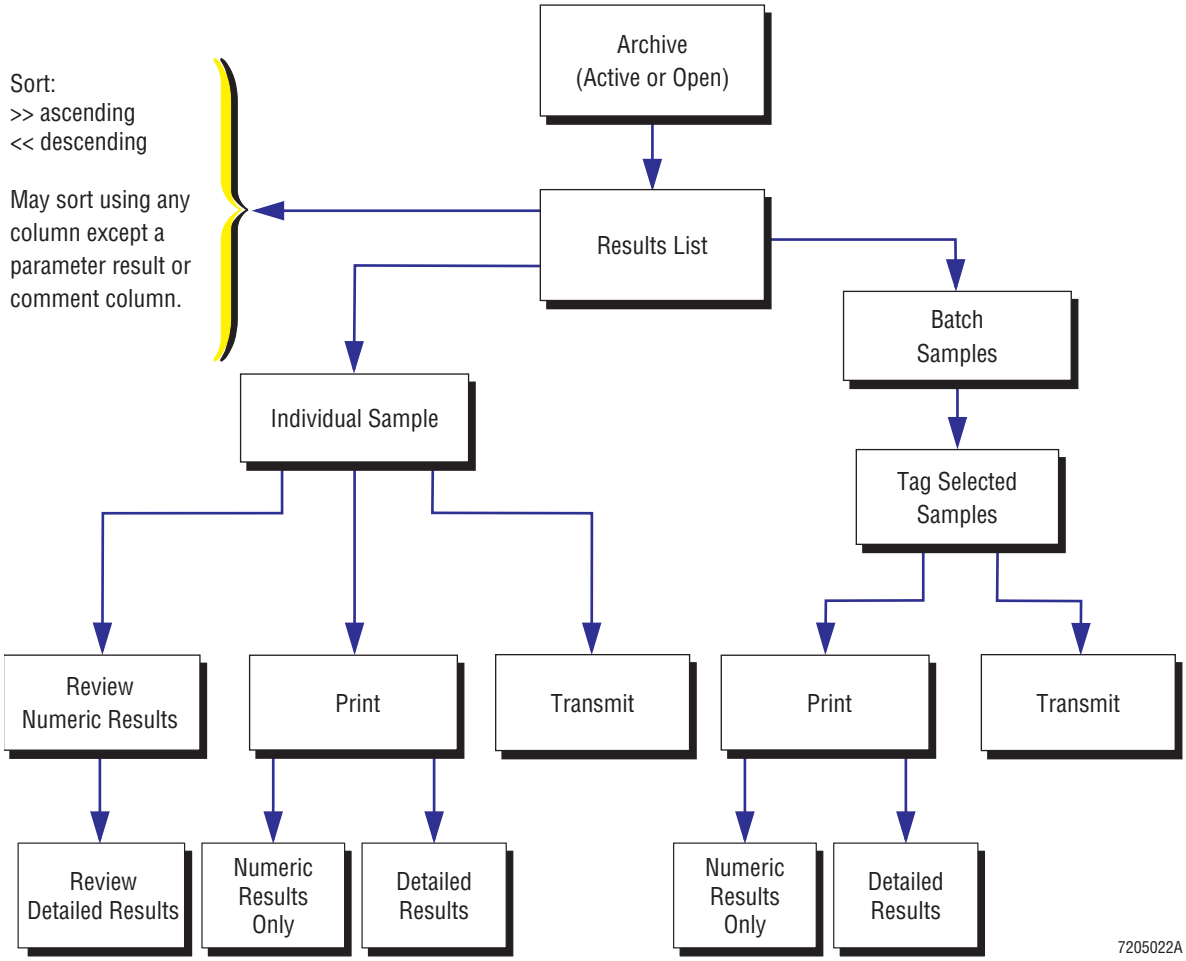
Database Compacting

The database can store 10,000 results. To optimize performance, the system compacts the database each time you log in.

After 10,000 results have been stored, at the next login, the system automatically deletes the oldest results to reduce the database to 9,500 and allow additional results to be saved.

DATA RETRIEVAL

An operator may sort, retrieve, review, print, or transmit data stored in either the current, active archive or a closed archive (that's been opened)



IMPORTANT Risk of compromising system functionality if you batch print and/or batch transmit while receiving a Worklist download from a host and/or while analyzing samples with Auto-Transmit and Auto-Print functions turned on. Always allow sample analysis and/or the host download to complete before batch printing and/or batch transmitting.

➤ ARCHIVED DATA QUICK REFERENCE

Verify the Desired Archive is Being Used

1. Click the **Results** tab to display the list of sample results.
2. Is this the archive you wish to use?
   - If you wish to use the current, active archive:
     - A white background indicates you are viewing the current, active archive.
     - A light green background indicates you are viewing a previously closed archive that has been opened. At the Menu bar, select **File ▸▸ Close archive**.
   - If you wish to use a previously closed archive:

     Note: Do not analyze patient samples when a previously closed archive is open.
     - A white background indicates you are viewing the current, active archive. At the Menu bar, select **File ▸▸ Open archive** then select the date for the archive you wish to use. When the archive opens, a light green background indicates you are viewing a previously closed archive.
     - A light green background indicates you are viewing a previously closed archive that has been opened. If you are not sure that you are viewing the archive you wish to use, at the Menu bar:
       - Select **File ▸▸ Close archive** to close this open archive.
       - Select **File ▸▸ Open archive**, the table of closed archives appears.
       - Select the date for the archive you wish to use.
3. You may choose to sort, view, print, transmit, or delete selected or all results.
4. If you open an archive, make sure you close the archive when your work is completed.

To Sort the Results List

At the Results window, you can sort information in ascending or descending order using any column except a parameter result or comment column. In other words, you can sort by Sample ID but not by an analysis parameter, such as WBC, RBC, Hgb, Plt, and so forth.

1. Click the title of the column you want sorted. For example, if you wish to sort by sample ID, click the Sample ID column title.
   - >> appearing next to the title indicates an ascending (low to high) sort order.
   - << appearing next to the title indicates a descending (high to low) sort order.
   - Repeating sequence for sort functions:
     - Click once, sort ascending with a >> symbol in the column header
     - Click again, sort descending with a << symbol in the column header
     - Click again, defaults back to original order
     - Click again, start again with sort ascending
   - Sample IDs sort alphanumerically by character. For example, 10 will appear before 4 because it is being sorted by the “1”.
   - Numbers appear in a sorted list before letters. For example, Sample ID 482 will appear before Sample ID N482 (unless sorted in descending order).

To Search for a Specific Sample

1 Scroll through the list of results to locate the sample ID.

or

2 Use the search feature:

- Click the **Search Results** tab at the bottom of the Results window.
- Click **Current Archive** or **Closed Archive**.
- Click your choice of search criteria: **Sample ID**, **Patient ID**, or **Patient Name**.
- Type the chosen identifier.
- Click the Search button.

To View Sample Results

1 To view numeric results, scroll right.

- You may choose to print or transmit the numeric sample results.

2 To view detailed results (results and histograms in the Run window format):

- Double-click the desired result.

  or

- Click  to view the selected results.

  Note: Click  to review the previous result or  to review the next result, as desired.

- You may choose to print or transmit the detailed sample results.
- Click  to return to the Results List.

To Output Sample Data: Print and Reprint / Transmit and Retransmit

To Print a Single Sample Data Set

1 At the selected set of sample results, click  to display the print menu.

2 Select the desired print format:

- **Print Summary List For Selected Rows** (prints numeric results only)

  or

- **Print Patient Report For Selected Rows** (prints Run view of results and histograms)

ATTENTION: Beckman Coulter recommends that you do not perform simultaneous batch printing and batch transmission while running the system with the Auto-Print and Auto-Transmit functions enabled. Performing all these activities with a large active archive may affect system processing.

To Batch Print a Group of Samples

1 Display the list of sample results.

2 While pressing the Ctrl key, click on each sample you wish to print. A black dot appears in the far left column and the sample row is highlighted to indicate the sample is selected.

3 Click  to display the print menu.

4 Select the desired print format:

- **Print Summary List For Selected Rows** (prints numeric results only)

  or

- **Print Patient Report For Selected Rows** (prints Run view of results and histograms)

ATTENTION: Beckman Coulter recommends that you do not perform simultaneous batch printing and batch transmission while running the system with the Auto-Print and Auto-Transmit functions enabled. Performing all these activities with a large active archive may affect system processing.

To Batch Transmit a Group of Samples

Note: The transmit option also allows an operator to transmit the last result and all results.

1 Click the **Results** tab if you have not already done so.

2 While pressing the Ctrl key, click on each sample you wish to transmit. A black dot appears in the far left column and the sample row is highlighted to indicate the sample is selected.

3 Click  to display the transmit menu.

4 Select **The selected results** from the transmit menu.

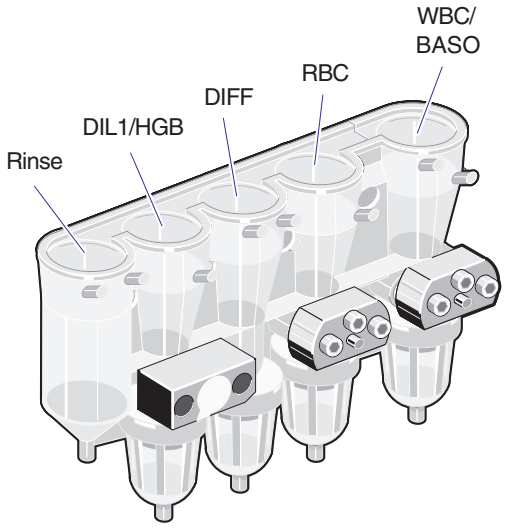
➤ ➤

NOTES

BASICS OF CBC/DIFF ANALYSIS

Sample Dilutions

- During aspiration, whole blood is pulled into sampling probe:
  - 53 µL of whole-blood aspirated when CBC/DIFF panel selected
  - 30 µL of whole-blood aspirated when CBC panel selected
  - Volume of sample aspirated from the specimen tube into sampling probe is sufficient to make all the dilutions needed to develop parameter results for selected panel
- Aspirated sample is partitioned as it is distributed to make a series of dilutions in a series of baths located in the right side compartment; distribution technique referred to as the Sequential Dilution System (SDS) technique



  - Four dilutions are made for a CBC/DIFF panel analysis, including a dilution in the:
    - DIL1/HGB bath,
    - DIFF bath,
    - RBC bath, and
    - WBC/BASO bath.
  - Three dilutions are made for a CBC panel analysis, including a dilution in the:
    - DIL1/HGB bath,
    - RBC bath, and
    - WBC/BASO bath.
  - Steps required to make these dilutions are detailed in Topic 8 of this Training Guide

Dilution Used to Determine RBC and PLT Results

- Dilution made in RBC bath used to analyze red blood cells and platelets
- RBC bath dilution prepared in two stages:
  - Primary dilution: 10 µL whole-blood is diluted with 1.7 mL of diluent in the DIL1/HGB bath to form a 1:170 dilution (commonly referred to as first dilution)
  - Secondary dilution: 42.5 µL of this first dilution is transferred to the RBC bath where it is further diluted with 2.5 mL of diluent to form final 1:10,000 dilution
- Final 1:10,000 dilution inside RBC bath is used to:
  - Determine RBC count
  - Develop RBC histogram which is needed to obtain the Hct, MCV, and RDW parameter results
  - Determine Plt count
  - Develop Plt histogram which is needed to obtain MPV, Pct, and PDW parameter results

Summary of Technical Characteristics for Obtaining RBC and Platelet Counts

Dilution Characteristics	
Primary Dilution for RBC and Plt:	
Initial volume of whole-blood	10 µL
Volume A^{C}•T 5diff Diluent	1700 µL
Primary dilution ratio	1:170
Secondary Dilution for RBC and Plt:	
Volume of primary dilution	42.5 µL
Volume A^{C}•T 5diff Diluent	2500 µL
Secondary dilution ratio	1:58.8
Final dilution for RBC and Plt results	1:170 x 1:58.8 = 1:10,000
Reaction temperature	35°C (95°F)
Measurement Characteristics	
Method of analysis	Coulter Principle
Aperture diameter	50 µm
Count vacuum	200 mb (5.9 in. Hg)
Count period	2 x 5 seconds

Dilution Used to Determine the Hemoglobin Result

- Final dilution made in DIL1/HGB bath is used to determine hemoglobin parameter result
- Final dilution in the DIL1/HGB bath is prepared in three phases:
  - Phase 1: 10 µL whole-blood is diluted with 1.7 mL of diluent in the DIL1/HGB bath to form a 1:170 dilution (commonly referred to as first dilution)
  - Phase 2: 42.5 µL of this first dilution is removed and 0.40 mL of diluent is added during an external probe wash
  - Phase 3: 0.4 mL of Hgb Lyse reagent is added to form final 1:250 dilution
- Hemoglobin released by lysis of the red blood cells combines with potassium cyanide to form a cyanmethemoglobin compound
- Hemoglobin concentration is determined based on transmittance of light through the optical part of the DIL1/HGB bath using a spectrophotometric technique at a wavelength of 550 nm
  - Transmittance of sample dilution is compared to transmittance of a reagent blank
  - Analyzer calculates Hgb using the blank and sample readings

Summary of Technical Characteristics for Measurement of Hemoglobin

Dilution Characteristics	
Volume of whole-blood	10 µL
Volume A^{C}•T 5diff Diluent	1700 µL
Preliminary dilution ratio	1:170
Volume of the 1:170 dilution removed (for making the RBC/Plt dilution)	42.5 µL
Additional volume of A^{C}•T 5diff Diluent	400 µL
Volume of A^{C}•T 5diff Hgb Lyse	400 µL
Final dilution for Hgb determination	1:250
Reaction temperature	35°C (95°F)
Measurement Characteristics	
Method of analysis	Spectrophotometry
Wavelength	550 nm

Dilutions Used to Determine the WBC Count and Differential

- Two dilutions are required to obtain WBC count and differential parameter results:
  - WBC/BASO dilution and
  - DIFF dilution

WBC/BASO Dilution

- Final 1:200 dilution is prepared by simultaneously delivering 10 µL of whole-blood and 2.0 mL of WBC Lyse reagent into WBC/BASO bath
- Final 1:200 dilution inside the WBC/BASO bath is used to:
  - Determine the WBC count, and
  - Develop the WBC/BASO histogram, which is needed to obtain the BASO count
- WBC count and BASO count are determined simultaneously
- WBC count is determined twice using two different methodologies:
  - Count obtained in the WBC/BASO bath is reference WBC count
  - Second WBC count is determined in the flow cell during acquisition of the DiffPlot. The dilution analyzed in the flow cell is prepared in the DIFF bath
  - WBC count results from the two methodologies are compared and if the results exceed predefined limits, the WBC count result is flagged

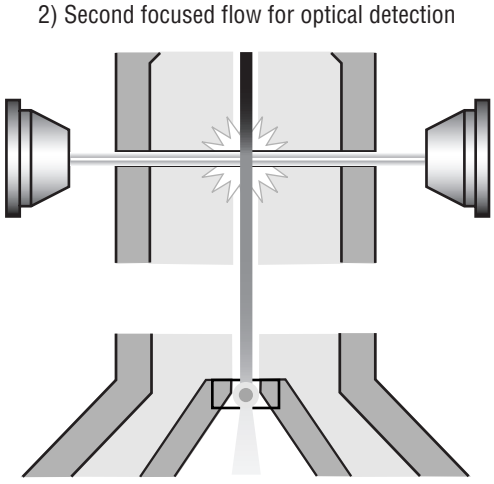
    Note: The comparison between the WBC count from the WBC/BASO bath and the WBC count from the flow cell is not performed when the CBC panel is selected or when this option is disabled in setup.

Summary of Characteristics Required to Obtain WBC and BASO Results

Dilution Characteristics	
Volume of whole-blood	10 µL
Volume A^{C}•T 5diff WBC Lyse	2,000 µL
Dilution ratio	1:200
Reaction temperature	35°C (95°F)
Measurement Characteristics	
Method of analysis	Coulter Principle
Aperture diameter	80 µm
Count vacuum	200 mb (5.9 in. Hg)
Count period	2 x 6 seconds

DIFF Dilution

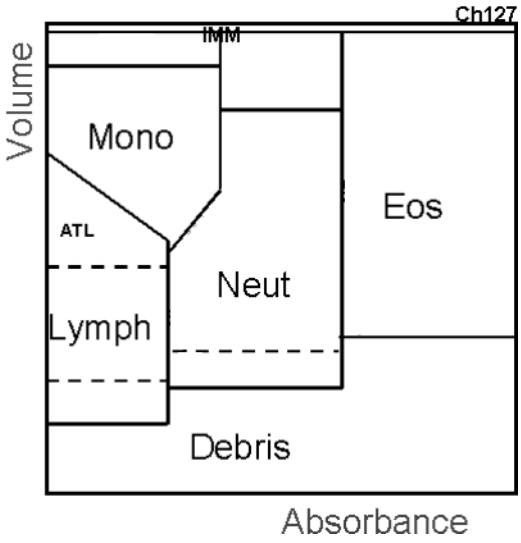
- Final dilution inside the DIFF bath is prepared in three phases:
  - Phase 1: 25 µL of whole-blood is delivered to DIFF bath in a flow of Fix reagent
  - Phase 2: During a 12 second incubation period, Fix reagent lyses the red blood cells, stabilizes the WBC in their native forms, and differentially stains lymphocytes, monocytes, neutrophils, and eosinophils, with the eosinophils staining most intensely
  - Phase 3: 1.0 mL of diluent is added to DIFF bath to stop the cytochemical reaction and form final 1:80 dilution
- Final 1:80 dilution is transferred to the flow cell where each cell is measured for impedance or resistivity (volume) and absorbance (optical detection of cytochemistry changes)



- Final 1:80 dilution prepared in DIFF bath is analyzed in flow cell to:
  - Develop DiffPlot from which four of five leukocyte (white blood cell) populations are determined: lymphocytes, monocytes, neutrophils, and eosinophils

    Note: In a typical whole-blood sample, the basophil population (determined in the WBC/BASO bath) is very small compared to the other four white blood cell populations.
  - Determine a second WBC count that is compared with the reference WBC count determined in WBC/BASO bath; if WBC count results from the two methodologies exceed predefined limits, WBC count result is flagged

    Note: The comparison between the WBC count from the WBC/BASO bath and the WBC count from the flow cell is not performed when the CBC panel is selected or when this option is disabled in setup.



DiffPlot developed with optical transmission (absorbance) on the X-axis and volume on the Y-axis

DiffPlot used to determine four of five leukocyte (white blood cell) populations:

- Lymphocytes (Lymph)
- Monocytes (Mono)
- Neutrophils (Neut)
- Eosinophils (Eos)

Summary of Technical Characteristics for Acquisition of the DiffPlot

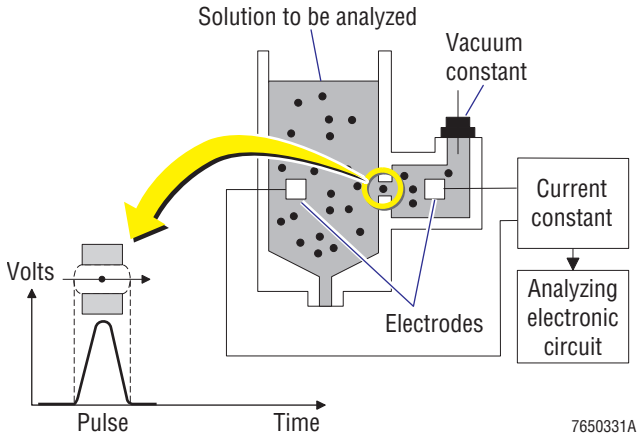
Dilution Characteristics	
Volume of whole-blood	25 µL
Volume A^{C}•T 5diff Fix	1000 µL
Volume A^{C}•T 5diff Diluent	1000 µL
Final dilution ratio	1:80
Reaction temperature	35°C (95°F)
Incubation duration	12 seconds
Measurement Characteristics	
Method of analysis	Impedance with hydrofocus
Aperture diameter	60 µm
Diameter of the flow	42 µm
Volume injected	72 µL
Injection duration	15 seconds
Data accumulation	12 seconds

MEASUREMENT PRINCIPLES

The Coulter Principle

- Electronic method for counting and sizing particles based on the fact that cells, which are poor conductors of a weak current, interrupt the current flow
- Cells, suspended in a conductive diluent, are pulled through the aperture by a low vacuum; this creates a momentary increase in resistance to the electronic flow
- Resistance creates a pulse that is sensed and counted by the instrument as a particle; amount of resistance (the amplitude or height of each pulse) is directly related to the size (volume) of the particle that produced it
- Impedance variation generated by the passage of nonconductive cells through a small, calibrated aperture is used to determine the count (number of particles) and size (volume) of the particles passing through the aperture within a given time period
- Used to analyze the final dilutions in the RBC bath and WBC/BASO bath:
  - RBC bath aperture sensor system determines the cell count and size of red blood cells and platelets
  - WBC/BASO bath aperture sensor system determines the cell count and size of white blood cells. Additionally, this system differentiates between basophils and other white blood cells as related to the WBC Lyse specific lytic action on the white blood cells inside this bath

Aperture Sensor System

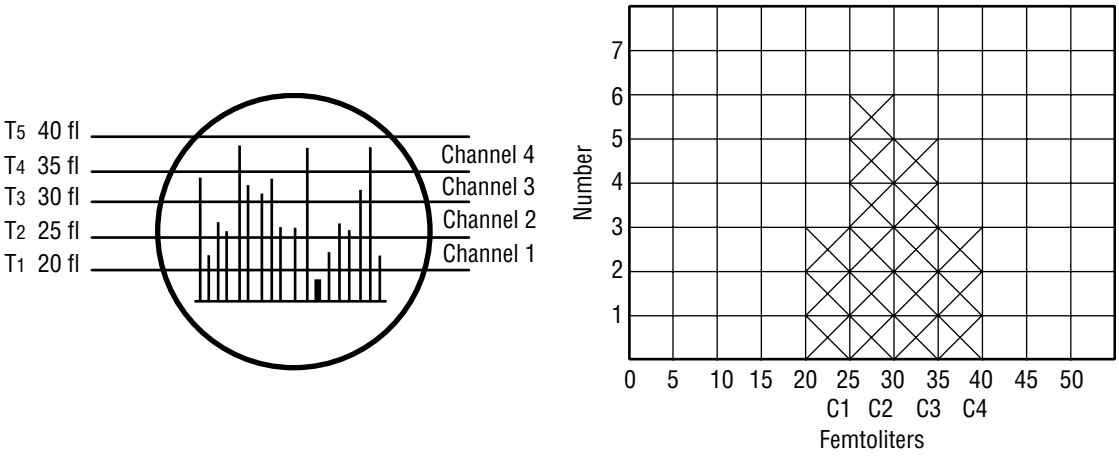


- To sense particles using the Coulter Principle, a current flow is established so changes in that flow can be monitored
- In this sensing system, an electrode is placed on each side of the aperture
  - Most visible electrode (referred to as counting head) is the conductive metallic housing attached to the front of the RBC and WBC/BASO baths
  - Second electrode (referred to as the bath electrode) is not as conspicuous; this electrode is located inside the bath
  - Aperture located between the counting head and the bath electrode

- When count circuit is activated and an electronically conductive reagent is in the RBC or WBC/BASO bath, an electric current continuously passes through the aperture; current moving between the two electrodes establishes the electronic flow through the aperture
- Once a sample is aspirated, an aliquot of that aspirated sample is diluted with reagent (an electrolyte) and is delivered to the RBC or WBC/BASO bath using tangential flow (which ensures proper mixing of the dilution)
- When cells suspended in the conductive reagent are pulled through a calibrated aperture, electrical resistance between the two electrodes increases proportionately with the cell volume
- Resistance creates a pulse that is sensed and counted as a particle by the Analyzer
- Amount of resistance (amplitude of each pulse) is directly related to the size of the particle that produced it
- Generated pulses have a very low voltage, which amplification circuit increases so that the electronic system can better analyze the pulses and eliminate the background noise

Thresholds

- Thresholds are electronically set size limits
- Unwanted particles, such as debris, are excluded from analysis
- Particles equal to or above the threshold are analyzed and particles below the threshold are excluded



Histogram Development

- The lowest threshold (T_1) is sometimes referred to as the Base Threshold
- Succession of thresholds are used to sort particles by size and produce a size distribution curve

- Area between two adjacent thresholds is called a Channel
  - Channel 1 (C1) is between T_1 and T_2
  - Channel 2 (C2) is between T_2 and T_3, and so forth
  - Each threshold represents a size
  - For a particle to be counted in a particular channel, it must be larger than or equal to the lower threshold but smaller than the upper threshold
- To produce a particle size distribution curve or histogram, the number of particles is plotted on the Y-axis as Relative Number of particles and the particle size (or volume) on the X-axis as Femtoliters (fL)
- Histogram information may be used to determine the mean (average) size of the particles, the dispersion of particles around the mean size, and subpopulations within a main population.
- A^C•T 5diff CP instruments develop three histograms: WBC/BASO, RBC, and PLT

Acquisition and Analysis Management

- Analyzer uses three microprocessors
- One microprocessor acts as Master and controls other two microprocessors
- Data acquisition and analysis is accomplished in two phases:

	Acquisition and Analysis Phase 1	**Acquisition and Analysis Phase 2**	**End of Cycle Rinse**
Microprocessor #1	Differential DiffPlot (12 seconds)	Platelet (2 x 5 seconds)	
Microprocessor #2	WBC/BASO (2 x 6 seconds)	RBC (2 x 5 seconds)	
Master Microprocessor		Hgb Sample (3 x 1 second)	Hgb Blank

Phase 1

- Master microprocessor directs actions of two smaller microprocessors
- Lasts 12 seconds
- Microprocessor #1:
  - Collects raw data from the DIFF dilution that's being transferred and sensed inside the flow cell
  - Develops the DiffPlot
  - Determines a second WBC count in the flow cell during acquisition of the DiffPlot
  - Analyzes DiffPlot to determine the percentage of neutrophils, lymphocytes, monocytes, and eosinophils
- Microprocessor #2:
  - Collects raw data as low vacuum pulls the 1:200 dilution through the 80 µm aperture in WBC/BASO bath
  - Raw data for determining WBC count and BASO count is collected over two consecutive time periods of 6-seconds each

    Note: These data collection time frames are commonly referred to as either count periods or duplicate counting.
  - Raw data for developing WBC/BASO histogram is collected through two consecutive time periods of 6-seconds each (for a total of 12 seconds)

Phase 2

- Master microprocessor again directs actions of two smaller microprocessors
- Lasts 10 seconds
- Microprocessor #1:
  - Collects platelet raw data as low vacuum pulls 1:10,000 dilution through the aperture in RBC bath
  - Raw data for determining PLT count is collected over two consecutive time periods of 5-seconds each (duplicate counting)
  - Raw data for developing PLT histogram is collected through two consecutive time periods of 5-seconds each (for a total of 10 seconds)
- Microprocessor #2:
  - Collects RBC raw data as low vacuum pulls 1:10,000 dilution through the aperture in RBC bath
  - Raw data for determining RBC count is collected over two consecutive time periods of 5-seconds each (duplicate counting)
  - Raw data for developing RBC histogram is collected through two consecutive time periods of 5-seconds each (for a total of 10 seconds)
- Master microprocessor:
  - Reads sample hemoglobin concentration in DIL1/HGB bath three different times
  - Later when DIL1/HGB bath drains and is filled with Rinse reagent, reads the concentration again to determine Hgb Blank

Voting

- Instrument generates two counts on WBC, RBC, Hct, and Plt parameters that go through the voting process
- Voting is comparison of the two results with each other to verify results are in close agreement:
  - If they agree, final reported result is the average of the two results
  - If the results differ by more than a predefined limit, WBC, RBC, Hct, or Plt result is flagged with a voteout *V* flag

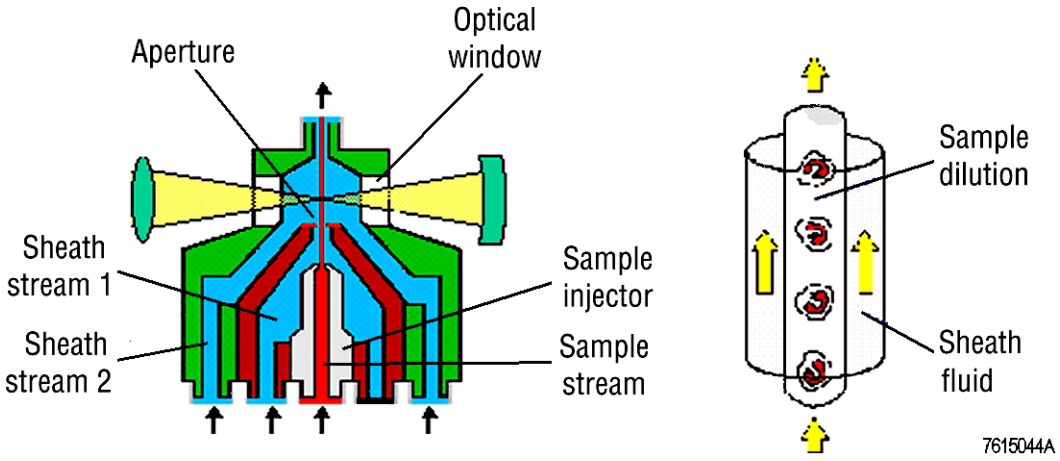
Voteout Flagging

- If WBC result is flagged with a *V*, then the DIFF number results are also flagged with a *V*
- If RBC result is flagged with a *V*, then MCV, MCH, MCHC, and RDW results are replaced by (· · · · ·) code
- If Hct result is flagged with a *V*, then MCV and MCHC results are replaced by (· · · · ·) code
- If Plt count votes out, then Plt result is flagged with a *V*
- For more information about flagging, see Chapter 9 of the Instructions For Use manual.

A^{C}V Differential Technology

- In DIFF (differential) bath, 25 µL of whole blood is mixed with 1,000 µL of A^{C}•T 5diff Fix reagent for 12 seconds, then stabilized with 1,000 µL of A^{C}•T 5diff Diluent for an additional three seconds
- Analyzer maintains the reagents and reaction at a regulated temperature of 35°C (95°F)
- Reaction in the DIFF bath lyses red blood cells, preserves leukocytes at their original size, and differentially stains lymphocytes, monocytes, neutrophils, and eosinophils, with eosinophils staining most intensely
- Each stained cell is individually focused by the Dual Focused Flow (DFF) system and transported through the flow cell using sample pressure and diluent sheath flow

Dual Focused Flow (DFF)



- DFF uses sheath fluid to surround and force cells suspended in diluent to pass one at a time through the center of the flow cell (hydrodynamic focusing process)
  - First sheath flow focuses sample through the impedance aperture
  - Second sheath flow maintains the focused flow of cells as they exit the aperture into the optical flow cell
- Hydrodynamic focusing in the flow cell enables accurate and rapid cell-by-cell measurements on a large number of individual cells
- Sequential analyses for cell volume (impedance) and light absorbance are performed in the flow cell:
  - Total of 72 µL of sample from the DIFF bath is injected through the flow cell for 15 seconds
  - Data for developing the DiffPlot is accumulated 12 seconds
  - Flow cell incorporates a 60 µm aperture for cellular volume analysis and about a 40 µm measurement area for light absorbance
  - Flow cell lamp is a 20 watt incandescent tungsten-halogen lamp

Focused Flow Impedance

- Focused flow impedance technology measures the electrical resistance of a cell as it passes through the aperture in the flow cell
- Change in resistance is directly proportional to volume of the cell

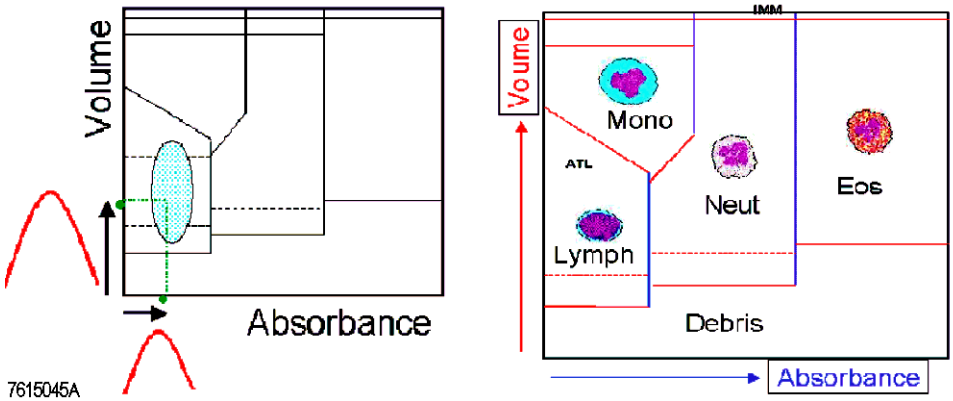
Absorbance Cytochemistry

- As a cell passes through the optical portion of the flow cell, light is scattered in all directions
- Sensor detects only forward scattered light
- Optical measurement is derived as a function of the amount of light lost due to diffraction and absorbance, as compared to full transmission when no cell is present
- Collected signals are converted into voltage pulses and are processed

- Magnitude of the voltage pulses are proportional to the physical and chemical characteristics of the cells being analyzed; light absorbance is related to cellular contents (granularity, nuclear content, and so forth) after cytochemical staining
- Measurements provide the information for lymphocytes, monocytes, neutrophils, and eosinophils, and their precursors

Signal Processing

- Signals from the flow cell aperture and from the optical measurement are correlated by a window of time
- Optical pulse must be detected within 100 to 300 microseconds of the impedance pulse; otherwise, the signal is rejected
- Output signals from the focused flow impedance and the light absorbance measurements are combined to define the WBC differential population clusters



Thresholds

- Most of the population partition thresholds are fixed and give the limits of the morphological normality of leukocytes
- Changes in the morphology of a population are expressed on the DiffPlot by a shifting of the corresponding population; volume and absorbance thresholds are used to detect shifting populations

NOTES

WBC PARAMETERS

- WBC parameter results are generated from two different dilutions:
  - WBC/BASO dilution which is made and analyzed in WBC/BASO bath and
  - DIFF dilution which is made in DIFF bath but analyzed in the flow cell

WBC/BASO Dilution

- WBC and basophil counts are determined from 1:200 dilution made in WBC/BASO bath
- To make this dilution, 10 µL of whole blood is mixed with 2,000 µL of A^C•T 5diff WBC Lyse reagent; reaction that occurs lyses the red blood cells and specifically differentiates between basophils and other leukocytes by volume
- WBC count and BASO count are determined simultaneously from final 1:200 dilution

WBC Count

When the CBC/DIFF Panel is Selected

- Analyzer compares data from the two 6-second count periods then votes and rejects any questionable data; this is reference WBC count
- When the two counts are compared:
  - If two counts agree, reference WBC count reported

    WBC count = Number of cells per volume x calibration factor

    WBC count displayed and printed as: WBC = N x 10^3 cells /µL
  - If two counts differ more than predefined limit, WBC result is flagged with a *V* and DIFF absolute numbers are also flagged with a *V*
- Second WBC count is determined in the flow cell during acquisition of DiffPlot:

  Note: The comparison between the WBC count from the WBC/BASO bath and the WBC count from the flow cell is not performed when the CBC panel is selected or when this option is disabled in setup.
  - If WBC count from flow cell exceeds WBC count from WBC/BASO bath by more than a predefined amount, *DIFF+* is displayed
  - If WBC count from flow cell is less than WBC count from WBC/BASO bath by more than a predefined amount, *DIFF-* is displayed
  - When a *DIFF-* or a *DIFF+* flag occurs, WBC count and all DIFF# parameters are flagged with an *

When the CBC Panel is Selected

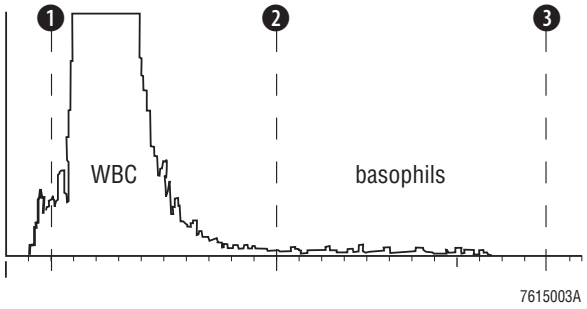
- Analyzer compares data from the two 6-second count periods then votes and rejects any questionable data; this is reference WBC count
- When two counts are compared:
  - If two counts agree, reference WBC count is reported

    WBC count = Number of cells per volume x calibration factor

    WBC count displayed and printed as: WBC = N x 10^3 cells /µL
  - If two counts differ more than a predefined limit, WBC result is flagged with a *V* and DIFF absolute numbers are also flagged with a *V*

BASO Count

- Differentiation between basophils and other leukocytes is obtained by means of the A^C•T 5diff WBC Lyse-specific lytic action
- Raw data for developing WBC/BASO histogram is collected through two consecutive time periods of 6-seconds each (for a total of 12 seconds)



- Referencing the above illustration:
  - Basophils are located in the area between thresholds labeled ❷ and ❸
  - One hundred percent (100%) of the leukocytes is represented by the total number of nucleated particles plus the basophils within area between thresholds labeled ❶ and ❸
  - Basophil percentage is calculated from number of particles existing in the area between thresholds labeled ❷ and ❸
- BASO count = Number of cells per volume x calibration factor in a percentage relative to the number of counted cells (basophils plus other WBC nuclei)

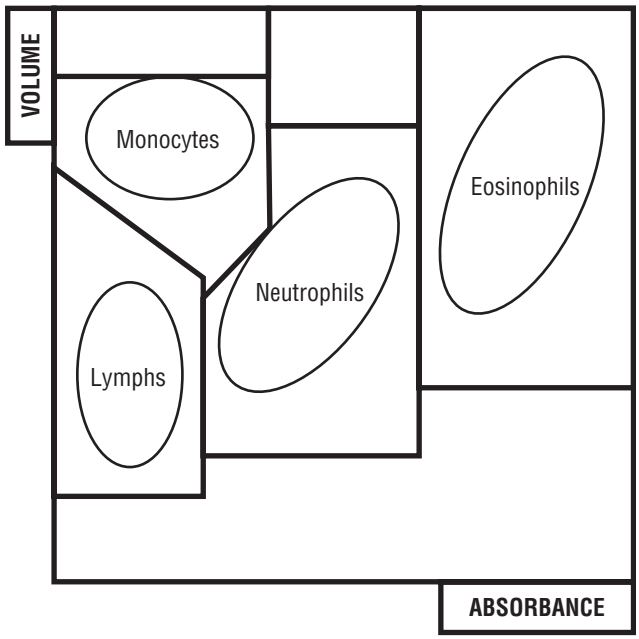
$$\text{BASO count} = \frac{\text{BASO\%}}{\text{WBC\%}} \times \text{WBC count}$$

DIFF Dilution

- DiffPlot developed from analysis of 1:80 DIFF dilution
- From this DiffPlot, neutrophil %, lymphocyte %, monocyte %, and eosinophil % are determined
- To make 1:80 DIFF dilution:
  - 25 µL of the whole-blood sample is mixed with 1,000 µL of A^{C}•T 5diff Fix reagent; Fix reagent lyses the red blood cells, stabilizes the white blood cells, and differentially stains the lymphocytes, monocytes, neutrophils, and eosinophils, with the eosinophils staining most intensely
  - After 12 seconds of incubation, 1,000 µL of A^{C}•T 5diff Diluent reagent added to stop cytochemical reaction
- Dilution is injected through the flow cell 15 seconds; for 12 of these 15 seconds, data for developing the DiffPlot is accumulated

DiffPlot Development

- DiffPlot analysis on A^{C}•T 5diff CP hematology analyzer based on three essential principles:
  - Dual Focused Flow (DFF) fluid dynamics, which is a process by which individual cells or particles are focused in a stream of diluent (hydrodynamic focusing)
  - Volume measurement (Coulter Principle)
  - Measurement of transmitted light with zero degree (0°) angle, which permits a response proportional to the internal structure of each cell and its absorbance
- From these measurements, a DiffPlot is developed with optical transmission (absorbance) on the X-axis and volume on the Y-axis
- DiffPlot regions include:



DiffPlot Regions Defined

- Study of DiffPlot permits clear differentiation of four out of five leukocyte populations
- In typical whole-blood sample, basophil population is very small when compared with the other four white cell populations

Neutrophils

- Neutrophils, with their cytoplasmic granules and segmented nuclei, scatter light according to their morphological complexity
- Hypersegmented neutrophil gives an increased optical response when compared to a young neutrophil population
- Higher the complexity of the cell, the further to the right they appear in the DiffPlot

Lymphs (Lymphocytes)

- Lymphocytes, typically being small with regular shape are smaller in volume and lower in absorbance than the other cells, and are positioned in the lower region of the DiffPlot
- Normal lymphocyte populations typically have a homogeneous volume with a Gaussian (bell-shaped) distribution
- Large lymphocytes, reactive lymphoid forms, stimulated lymphocytes and plasma cells are found in the upper portion of the lymphocyte region
- Lower area of the lymphocyte zone is normally empty; however, when small lymphocytes are present, a population may exist in this area
- Presence of platelet aggregates is indicated by a distribution pattern that moves from the DiffPlot origin into the lymphocyte region
- NRBC cytoplasmic membranes lyse like those of mature erythrocytes; small nuclei that remain appear in the debris and small lymphocyte regions

Monocytes

- Monocytes are typically large cells with a kidney-shaped nucleus and agranular (granule-free) cytoplasm
- Cells neither scatter nor absorb large amounts of light and, therefore, are positioned in the lower end of the absorbance axis
- Due to their size, monocytes are clearly positioned high on the volume axis
- Very large monocytes may be found in the IMM (immature cell) region

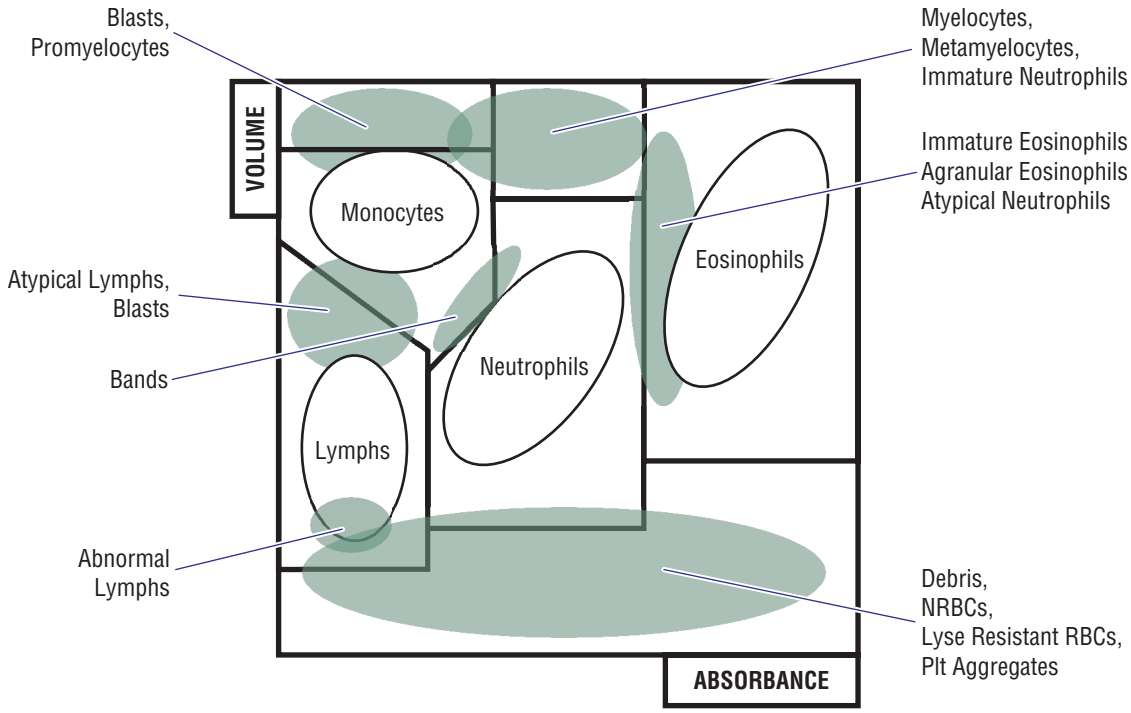
Eosinophils

- With reagent action, eosinophils are the most intensely stained cells for optical separation
- Due to the staining and their size, eosinophils show higher absorbance than neutrophils, but will be of similar volume

Debris

- Platelets and debris from erythrocyte lysis represent the background debris population located in the lower region of the DiffPlot

Immature White Blood Cells



Immature Granulocytes

- Immature granulocytes are detected by their larger volume and by presence of granules that increase the intensity of scattered light
- Due to their increased volume and similar absorbance, promyelocytes, myelocytes, and metamyelocytes are located above the neutrophil population and are typically counted as IMM cells; IMM cells are included in the reported neutrophil value

Band Cells

- Band cells are typically larger or of similar size to the neutrophils; however, due to their low level of cellular complexity, they absorb less light
- Band cells tend to appear in the region between the neutrophils and the monocytes

Blast Cells

- Blast cells are generally larger than monocytes and have similar absorbance
- When blast cells are present, generally located above the monocytes, which means they will be included in the IMM cell count
- Small blasts will be located between the normal lymphocyte and monocyte populations

DiffPlot Thresholds

- Most of the population partition thresholds are fixed and give the limits of the morphological normality of leukocytes

- Changes in the morphology of a population are expressed on the DiffPlot by a shifting of the corresponding population
- Volume and absorbance thresholds are used to detect shifting populations
- When populations in DiffPlot exceed limits set for that region, a review (*R*) flag occurs on the DIFF parameter related to that region, and either DiffPlot and Histogram flags or Analytical Alarms occur to indicate the area within the DiffPlot that is affected
- If *R* flag occurs on a DIFF parameter, further investigate the result
- Twelve different flags may occur related to the position of the populations within the DiffPlot:
  - *CO* (diff reject)
  - *DB* (debris)
  - *SL* (small lymphocytes)
  - *SL1* (small lymphocytes 1)
  - *NL* (neutrophil/lymphocyte)
  - *MN* (monocyte/neutrophil)
  - *UM* (upper monocyte)
  - *LN* (lower neutrophil)
  - *UN* (upper neutrophil)
  - *NE* (neutrophil/eosinophil)
  - *ATL* (atypical lymphocytes)
  - *IMM* (immature cells)
- See the Definition of DIFF Flags table in Chapter 9 of the Instructions For Use manual for a lists of these flags and an illustration of the affected DiffPlot region; table also lists suspected abnormalities

Flags, Interpretive Messages, and Analytical Alarms

- For details concerning flags, interpretive messages, and analytical alarms associated with WBC parameters, see Chapter 9 of the Instructions For Use manual
- To use Online Help to locate flags, interpretive messages, or analytical alarms associated with WBC parameters:

  1) Click 
  2) Click on **Contents**; table of contents appears in left frame
  3) Inside left frame, click on **9 Data Review** to list headings in that chapter
  4) Scroll through headings and click on heading that interests you; information appears inside right frame
  5) Scroll to locate information concerning a specific flag, interpretive message, or analytical alarm

     or

     Print a copy of the information

     - Right click on the messages (anywhere inside right frame)
     - Click **Print** on the pop-up menu
     - Click **OK** to initiate printing

     Note: All the information under the primary heading is printed; locate specific flag, interpretive message, or analytical alarm information you need

WBC Parameters

Category	Parameter	Source of Data
Directly Measured	WBC (White Blood Count)	Differential lysis using the Coulter Principle
	BA % (Basophil Percentage)	Differential lysis using the Coulter Principle
Derived from the DiffPlot	NE % (Neutrophil Percentage)	A^{C}V Technology
	LY % (Lymphocyte Percentage)	A^{C}V Technology
	MO % (Monocyte Percentage)	A^{C}V Technology
	EO % (Eosinophil Percentage)	A^{C}V Technology
Computed	NE # (Neutrophil Absolute Number)	$\frac{Neut\% \times WBC}{100}$
	LY # (Lymphocyte Absolute Number)	$\frac{Lymph\% \times WBC}{100}$
	MO # (Monocyte Absolute Number)	$\frac{Mono\% \times WBC}{100}$
	EO # (Eosinophil Absolute Number)	$\frac{Eos\% \times WBC}{100}$
	BA # (Basophil Absolute Number)	$\frac{Baso\% \times WBC}{100}$

NOTES

RBC PARAMETERS

RBC Dilution

- Final 1:10,000 dilution in RBC bath that is often referred to as RBC/PLT dilution
- RBC dilution contains red blood cells, white blood cells, and platelets:
  - Thresholds are used to separate platelet pulses, which are much smaller, from the red and white blood cell pulses
  - Since white blood cells fall in red blood cell size range, they are counted and sized as RBCs

RBC Count

- RBC count obtained from RBC/PLT dilution in RBC bath
- To obtain an RBC count result, Analyzer compares data from the two 5-second count periods then votes and rejects any questionable data
- When the two counts are compared:
  - If two counts agree, RBC count is reported

    RBC count = Number of cells counted per volume unit x calibration factor

    RBC count displayed and printed as: RBC = N x 10^6 cells /µL
  - If two counts differ more than a predefined limit, RBC result is flagged with a *V* and MCV, MCH, MCHC, and RDW parameters are replaced with (· · · · ·) code
- Since white blood cells fall in the red blood cell size range, they are counted and sized as RBCs when this 1:10,000 dilution is analyzed

  Note: The WBCs are not gated out of the count because any interference is usually insignificant; there are normally very few WBCs (thousands) in comparison to the number of RBCs (millions). Only when the white count is markedly elevated is the red cell count or histogram influenced.

Hematocrit Measurement

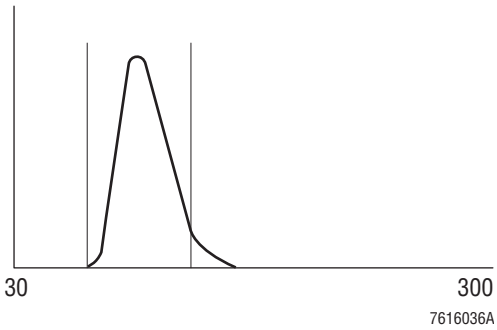
- Height of the pulse generated by the passage of a cell through the aperture is directly proportional to the volume of the analyzed red blood cell
- Hematocrit (Hct) is the sum of all the digitized pulses
- Collects data from the two 5-second count periods then votes and rejects any questionable data
- When two hematocrit results are compared:
  - If two values agree, Hct result is reported

    Hct = Sum of digitized pulses x calibration factor

    Hct displayed and printed as a percentage (%)
  - If two values differ more than a predefined limit, Hct result is flagged with a *V* and MCV and MCHC parameters are replaced with (· · · · ·) code

RBC Histogram

- In addition to being counted, red blood cells are categorized according to size (from 30 fL to 300 fL) by a 256-channel pulse-height analyzer
- Pulse-height analyzer uses a number of thresholds to sort the particles into several size (volume) categories and to develop a size distribution curve of the particles
- Raw data for developing the RBC histogram is collected through two consecutive time periods of 5-seconds each (for a total of 10 seconds)
- RBC distribution curve reflects the native size of the red blood cells and any other particle in red blood cell size range
- Example of an RBC histogram with a normal RBC size distribution is shown below.



Information Obtained Using the RBC Histogram

- RBC histogram provides the information for determining a helpful descriptor of the red cell population, RDW (Red cell Distribution Width)
- RBC histogram is also used to determine if a red blood cell population is typical; if not, descriptive flagging is generated

RDW Determination

- RDW (Red cell Distribution Width) is an index of the variation or spread in the size of the red blood cells
- Study of RBC distribution detects erythrocyte anomalies linked to anisocytosis and enables clinician to follow evolution of the width of the curve relative to the cell number and average volume:
  - Generally, as the red cell variation increases, width of the distribution curve broadens and visible asymmetry of the distribution curve is more likely
  - Width of the distribution curve, however, does not always indicate a true increase in size variation
- RDW index more accurately detects red cell variation than a visual inspection of the histogram (unless two distinct populations of cells are present)

  Note: A primarily microcytic population of red cells (decreased MCV) tends to have a tighter distribution (lower SD) than a normocytic population. A primarily macrocytic population of red cells (increased MCV) tends to have a wider distribution (higher SD) than a normocytic population.

- Displayed and printed as a percentage, RDW is calculated using the standard deviation (SD) of the RBC population and the MCV

$$\frac{K \ SD}{MCV} = RDW\ (\%)$$

where:

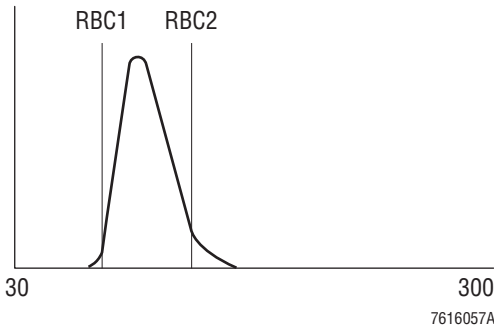
K = System constant

SD = Calculated standard deviation based on the red cell distribution

MCV = Mean Cell Volume of the red cells

RBC Distribution Flags

- Once RBC distribution curve is developed, two positions on the distribution curve are located:



RBC1 and RBC2 Thresholds

- Thresholds RBC1 and RBC2 define the MICRO and MACRO regions and are calculated based on standard deviation (SD) of the RBC population
- RBC1 threshold (monitoring area for microcytes) and RBC2 threshold (monitoring area for macrocytes) identify the points on the curve that are ±2 SD from the mean

Flags

- MICRO flag is generated when the percentage of cells in the microcytic region compared to the total number of RBCs exceeds the preset default limit of 5%
- MACRO flag is generated when the percentage of cells in the macrocytic region compared to the total number of RBCs exceeds the preset default limit of 7.5%
- Laboratory may establish its own limits to replace the preset default values

Note: The MICRO and MACRO flags are independent of the Microcytosis and Macrocytosis flags that are generated from the Low and High patient limits.

Hgb Determination

- Addition of CBC lytic reagent disrupts the red cells to free the hemoglobin which then combines with the potassium cyanide to form a stable cyanmethemoglobin compound
- Converted hemoglobin is directly measured through the optical part of the DIL1/HGB bath using a spectrophotometric technique at a wavelength of 550 nm; the photometric technique is based on Beer's Law
- Transmittance of the sample dilution is compared with the transmittance of a reagent blank; system calculates the Hgb using both the blank and sample readings:

  Hgb result in g/dL represents: absorbance value obtained x calibration factor

  Hgb displayed and printed as: Hgb = N g/dL

Hgb Blank Reading

- Hgb blank value measured during the first patient cycle after a Startup cycle is stored as a reference blank; blank must be greater than 2.5 Vdc
- During each analysis cycle, Analyzer checks the measured Hgb blank against the stored Hgb blank reference value

  Note: If the new Hgb blank reference value is within 3% of the old reference value, the Hgb blank reference value is changed to this new value.

Sample Reading

- Value is based on the sample, diluent, and Hgb Lyse reagent mixture in the DIL1/HGB bath during sample measurement

Hgb Specific Flags

- If the Hgb blank value is less than 2.5 Vdc, reject (*R*) flag occurs on Hgb value
- If difference between the new Hgb blank reference value and the original Hgb blank reference value is greater than 3%, review (*R*) flag is generated
- If three consecutive review (*R*) flags occur on the Hgb blank reference value, (· · · · ·) code replaces the Hgb, MCH, and MCHC results
- For each Hgb sample read value, Analyzer takes three readings; if the difference between these readings exceeds the predefined limits, voteout (*V*) flag is generated
- If the [(Hgb g/dL X 3)/Hct%] is <0.8 or >1.2, the RBC, Hgb, MCV, Hct, MCH, MCHC, Plt, MPV, Pct, and PDW will be flagged with *. The presence of this flag indicates that there may have been an error in the analytical process.

Calculations

MCV Calculation

- MCV (Mean Cell Volume) is calculated using the Hct and the RBC count
- Displayed and printed in femtoliters (fL)
- Calculation for MCV is:

$$\frac{\text{Hct}}{\text{RBC}} \times 10 = \text{MCV (fL)}$$

MCH Calculation

- MCH (Mean Cell Hemoglobin) is calculated from the Hgb value and the RBC count and describes the average weight of hemoglobin in a red cell
- Displayed and printed in picograms (pg)
- Calculation for MCH is:

$$\frac{\text{Hgb}}{\text{RBC}} \times 10 = \text{MCH (pg)}$$

MCHC Calculation

- MCHC (Mean Cell Hemoglobin Concentration) is calculated using the Hgb and Hct values and describes the average concentration of hemoglobin in the red blood cells
- Displayed and printed in grams per deciliter
- Calculation for MCHC is:

$$\frac{\text{Hgb}}{\text{Hct}} \times 100 = \text{MCHC (g/dL)}$$

Flags, Interpretive Messages, and Analytical Alarms

- For details concerning flags, interpretive messages, and analytical alarms associated with RBC parameters, see Chapter 9 of the Instructions For Use manual.
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     Print a copy of the information

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     - Click **OK** to initiate printing

     Note: All the information under the primary heading is printed. Locate the specific flag, interpretive message, or analytical alarm information you need.

RBC Parameters

Category	Parameter	Source of Data
Directly Measured	RBC (Red Blood Count)	Coulter Principle
	Hgb (Hemoglobin Concentration)	Photometric Measurement
	Hct (Hematocrit)	Coulter Principle
Derived from the RBC Histogram	RDW (Red Cell Distribution Width)	RBC Histogram (Developed using the Coulter Principle)
Computed	MCV (Mean Cell Volume)	$\frac{Hct \times 10}{RBC}$
	MCH (Mean Corpuscular Hemoglobin)	$\frac{Hgb \times 10}{RBC}$
	MCHC (Mean Corpuscular Hemoglobin Concentration)	$\frac{Hgb \times 100}{Hct}$

PLATELET PARAMETERS

RBC Dilution

- Platelet parameters are obtained from RBC dilution
- Final 1:10,000 dilution in RBC bath that is often referred to as RBC/PLT dilution
- RBC dilution contains red blood cells, white blood cells, and platelets
- Platelet counting and sizing is done in RBC bath
- Thresholds are used to separate platelet pulses, which are much smaller, from the red and white blood cell pulses

Plt Count

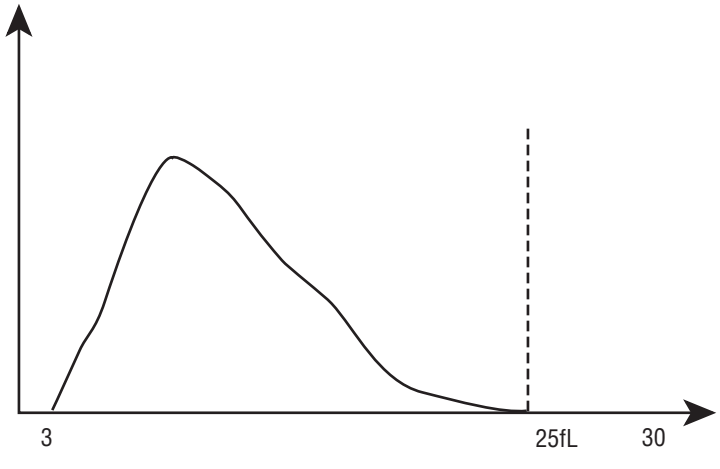
- Plt count is obtained from the RBC/PLT dilution in RBC bath
- To obtain a Plt count result, Analyzer compares the data from the two 5-second count periods then votes and rejects any questionable data
- When two counts are compared:
  - If two counts agree, Plt count reported

    Plt count = Number of cells counted per volume unit x calibration factor

    Plt count displayed and printed as: Plt = N x 10^3 cells /µL
  - If two counts differ more than the predefined limit, Plt result is flagged with a *V*

PLT Histogram

- Platelets are categorized according to size by a 256-channel pulse-height analyzer
- Pulse-height analyzer uses a number of thresholds to sort the particles into several size (volume) categories and to develop a size distribution curve of the particles between 2 fL and 30 fL
- Platelet histogram reflects the native size of the platelets and any other particle in the platelet size range
- Illustration that follows is a PLT histogram with a typical platelet size distribution:



Information Obtained Using the PLT Histogram

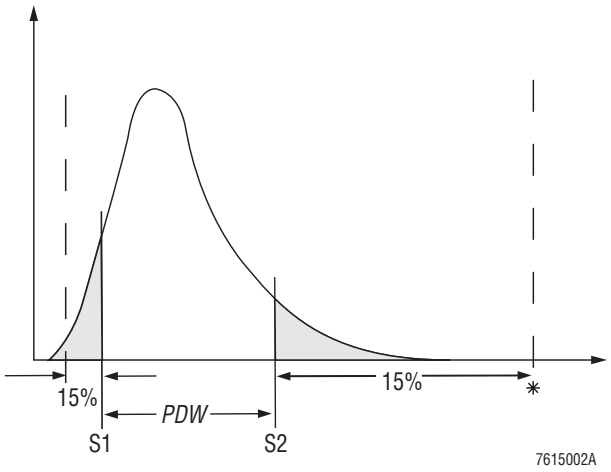
- PLT histogram provides the information for determining a helpful descriptor of the platelet population:
  - MPV (Mean Platelet Volume)
  - PDW (Platelet Distribution Width)
- PLT histogram is also used to determine if a platelet population is typical; if not, descriptive flagging is generated

MPV Measurement

- MPV (Mean Platelet Volume) is measured directly from analysis of the platelet distribution curve
- MPV is displayed and printed in femtoliters (fL)

PDW Determination

- PDW (Platelet Distribution Width) is determined from the PLT histogram as the width of the curve between S1 and S2
- In the illustration that follows, shows the area of the PLT histogram that used to determine the PDW parameter result:



- In this example, S1 and S2 are placed so that:
  - 15% of the platelets occur between 2 fL and S1
  - 15% of the platelets occur between S2 and the variable upper threshold

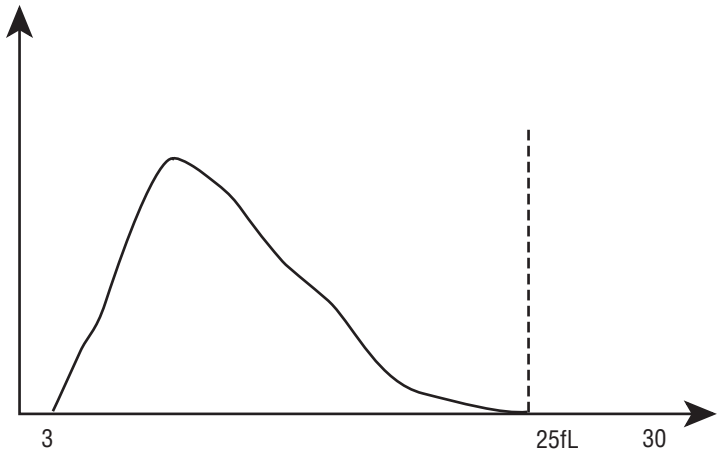
    Note: This threshold is explained under the Detecting Abnormal Platelet Distributions heading that follows.
  - PDW result is determined on the platelets between S1 and S2
- PDW parameter result is displayed and printed as a percentage (%)
- PDW is not an FDA approved parameter; therefore, this parameter label and result are not routinely displayed in the United States, but may be routinely displayed in other countries

Detecting Abnormal Platelet Distributions

- Particles of approximately platelet size can interfere with platelet histogram and count
- Small particles, such as microbubbles or dust, can overlap the low end of the histogram
- Microcytic red cells can intrude at the upper end of the histogram

Identifying a Normal Distribution

- When platelet histogram is being evaluated, a mobile threshold can move from its starting position at 25 fL to 18 fL
- Computer searches for a valley between the platelet and red cell populations
- If no valley is detected between 18 fL and 25 fL, threshold remains at 25 fL and no flag is generated
- In the typical platelet distribution that follows, no valley is detected between 18 fL and 25 fL, the threshold remains at 25 fL and no flag is generated:

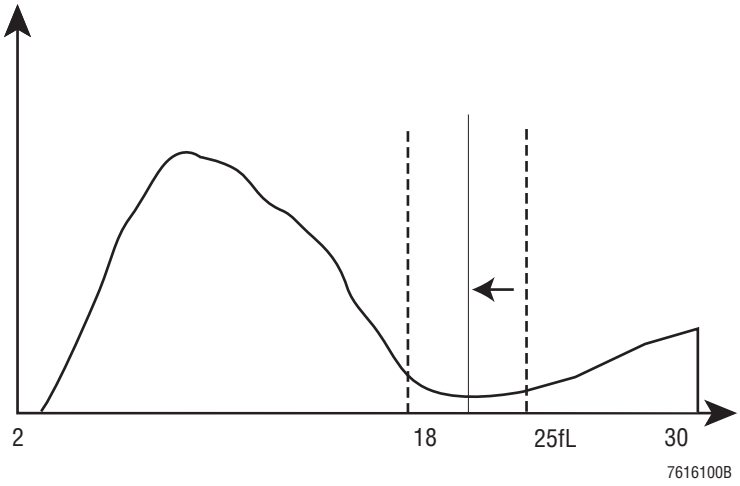


Microcytic Interferences on the Upper End of the Platelet Distribution Curve

- Microcytic red cells can intrude at the upper end of the platelet distribution curve
- If specimen contains microcytes, A^{C}•T 5diff CP hematology analyzer may be able to successfully eliminate the influence of this interference by repositioning the variable threshold (25 fL threshold) and excluding the microcytes

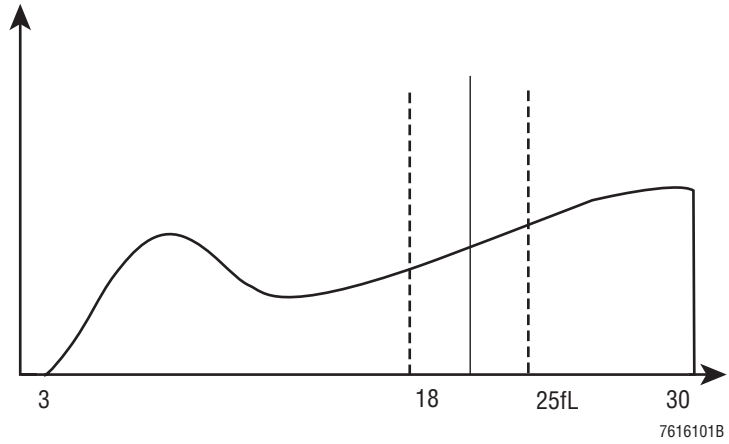
Microcytic Interference with a Distinct Valley between 18 fL and 25 fL

- If intrusion of microcytes creates a valley between the 25 fL and the 18 fL thresholds, the 25 fL threshold is repositioned at the valley to minimize interference to platelet parameter results
- When this occurs, reported platelet result is acceptable; however, MIC (microcytes) flag appears to alert operator that microcytes are present
- Illustration that follows is an example of microcytic interference with a valley between 18 fL and 25 fL:



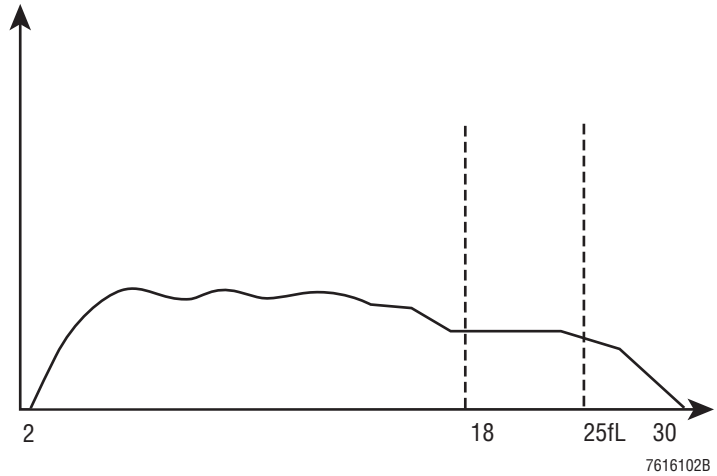
Microcytic Interference with a Valley below 18 fL

- If microcytes are extremely small so that the valley between the platelet population and the microcyte population falls below the 18 fL limit, threshold is placed at the 18 fL limit
- When this occurs, MIC flag appears and the platelet count is flagged to alert the operator that the extremely small microcytes present in this sample could not be eliminated
- Platelet count and associated parameters are not reliable and should be verified by an alternative method; to effectively eliminate the microcytes, the Instructions For Use manual suggests the customer use platelet rich plasma (PRP) or a manual count to verify the results
- Illustration that follows is an example of microcytic interference with a valley below 18 fL:



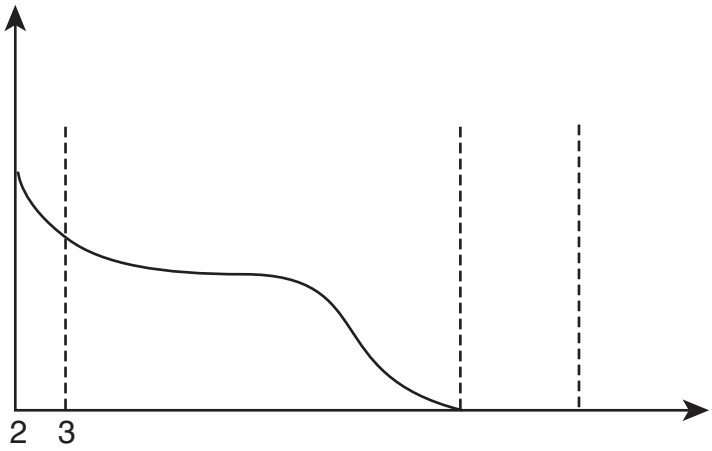
Interference with No Distinct Valley

- Interference present in the upper area of the platelet distribution curve that blends with the platelet population so that there is no clear distinction between the platelets and the interference suggest the presence of schistocytes (fragmented red cells) or platelet aggregates (platelet clumps)
- If threshold cannot be positioned in the 25 fL to 18 fL region, threshold defaults to the 18 fL position
- **SCH** (schistocytes) flag appears and the platelet count is flagged to alert the operator that the interference (which is most likely either schistocytes or platelet clumps) could not be eliminated
- Platelet count and associated parameters are not reliable and must be verified using an alternative method
- Illustration that follows is an example of interference with no distinct valley:



Interference on the Lower End of the Platelet Distribution Curve

- Particles that are approximately platelet size can interfere with platelet histogram and count
- Small particles, such as microbubbles or dust, can interfere at the low end
- If number of pulses in the 2 to 3 fL region is higher than the predefined limits, an SCL flag appears to alert the operator that a significant number of small cells or interference, such as microbubbles, are present
- **SCL** appears under the Analytical Alarms heading, "Small Cells" appears under the Interpretive Messages heading and "..." appears in place of PLT and MPV parameter values
- Rerun the sample and verify the results.
- Illustration that follows is an example of interference on the lower end of the platelet distribution curve:



Pct Calculation

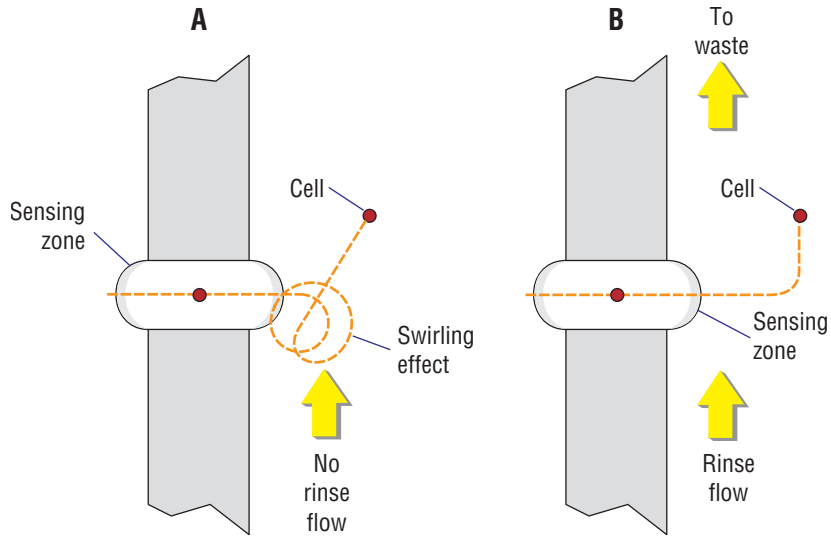
- Pct (plateletcrit or thrombocrit) is calculated according to the formula:

$$\frac{\text{Plt } (10^3/\mu\text{L}) \times \text{MPV (fL)}}{10,\ 000} = \text{Pct\%}$$

- Pct parameter result displayed and printed as a percentage (%)
- No clinical significance has been identified for Pct parameter
- Pct (and PDW) are not FDA approved parameters; therefore, these parameter labels and results are not routinely displayed in the United States, but may be routinely displayed in other countries

Rinse Flow System

- Rinse flow is a steady stream of A^C•T 5diff Diluent reagent that flows behind the RBC aperture during sensing periods
- Without rinse flow (Figure A), there is a characteristic swirling of the dilution at the outlet of the aperture so that red (or white) cells caught up in these eddies behind the aperture may reenter the sensing zone and produce small pulses that could be counted as platelets
- With rinse flow (Figure B), the steady stream of diluent prevents the formation of these eddies



Flags, Interpretive Messages, and Analytical Alarms

- For details concerning flags, interpretive messages, and analytical alarms associated with platelet parameters, see Chapter 9 of the Instructions For Use manual
- To use Online Help to locate flags, interpretive messages, or analytical alarms associated with platelet parameters:

  1) Click 
  2) Click on **Contents**; table of contents appears in left frame
  3) Inside left frame, click on **9 Data Review** to list headings in that chapter
  4) Scroll through headings and click on the heading that interests you; information appears inside right frame
  5) Scroll to locate the information concerning a specific flag, interpretive message, or analytical alarm

     or

     Print a copy of the information

     - Right click on the messages (anywhere inside right frame)
     - Click **Print** on the pop-up menu
     - Click **OK** to initiate printing

     Note: All the information under the primary heading is printed. Locate the specific flag, interpretive message, or analytical alarm information you need.

Platelet Parameters

Category	Parameter	Source of Data
Directly Measured	Plt (Platelet Count)	Coulter Principle
Derived from the PLT Histogram	MPV (Mean Platelet Volume) PDW (Platelet Distribution Width)	Plt Histogram (Developed using the Coulter Principle) Plt Histogram (Developed using the Coulter Principle)
Computed	Pct (Plateletcrit)	$\frac{Plt\ count \times MPV}{10}$

NOTES

OBJECTIVES

When the subject is complete, you will be able to . . .

Accessing Principal Assemblies and Component Identification

- Name each principal assembly.
- Locate and name main components.

Cycle Description

- Identify the start conditions prior to initiating a cycle.
- Identify the pierce position.
- Cite the volume of sample aspirated for a CBC/DIFF panel versus a CBC panel.
- Tell whether the bubble placed between diluent and blood during aspiration is normal or abnormal.
- Explain how the final RBC/Plt dilution is made, including the bath locations and reagents used in the process.
- Explain how the WBC/BASO dilution is made, including the bath location and reagents used in the process.
- Explain how the DIFF dilution is made, including the bath location and reagents used in the process.
- Explain what portions of the cycle are eliminated by selecting the CBC panel.
- Explain how the tangential flow or reagent mixes the sample and reagent.
- Explain the purpose of the mixing bubbles entering the DIL1/HGB bath.
- Explain the purpose of the burn circuit.

Basic Troubleshooting Techniques

- Recognize an instrument problem based on:
  - Abnormal sample results.
  - Abnormal Startup results.
  - Abnormal control results.
  - Error messages.
- Demonstrate how to access the Error Log.
- Demonstrate that you can locate the Error Messages table in the Instructions For Use manual.
- Use what you know about normal cycle flow and the derivation of parameter results to troubleshoot.
- Isolate the components involved, when given a symptom.
- Reproducibility check:
  - Explain the purpose of this check.
  - State at least one situation when performing a reproducibility check would be beneficial.
- Properly power down and power up the system as needed.

Preventive Maintenance

- Perform the proper maintenance procedures based either on a time schedule or on an as needed basis.
- Demonstrate how to access the Maintenance Log.

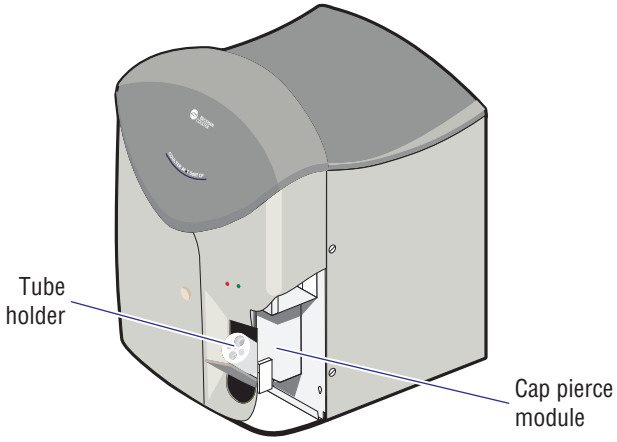
Replacement and Adjustment Procedures

- Locate and perform selected replacement and adjustment procedures such as replacing the flow cell lamp.

ACCESSING PRINCIPAL ASSEMBLIES AND COMPONENT IDENTIFICATION

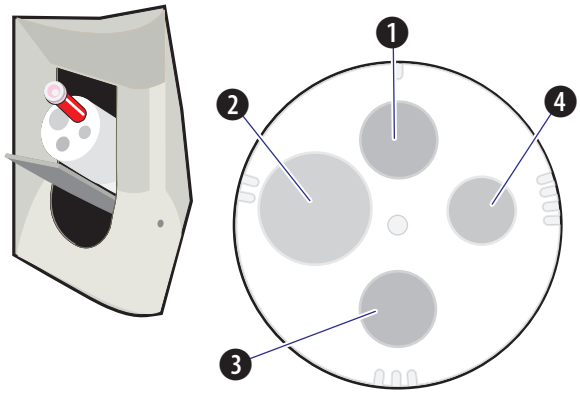
Cap Pierce Module

With the Door Open



Tube Holders

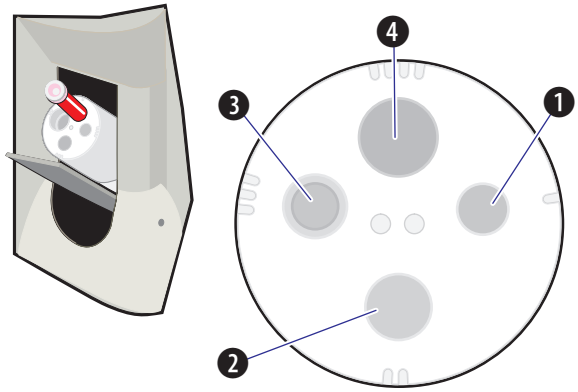
Tube Holder #1



Position Designations for Tube Holder #1

1. Sample position 1
2. Sample position 2
3. Sample position 3
4. Sample position 4

Tube Holder #2



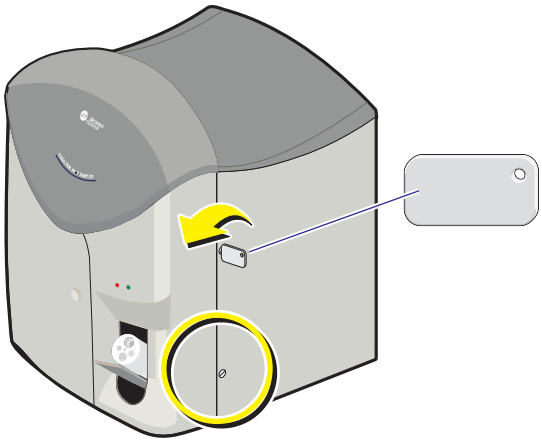
Position Designations for Tube Holder #2

1. Sample position 1
2. Sample position 2
3. Sample position 3
4. Sample position 4

Note: For a detailed list of specimen tubes available to use with these holders, refer to Appendix D in the Instructions For Use manual. A summarized list is also available under Topic 6 in this Training Guide.

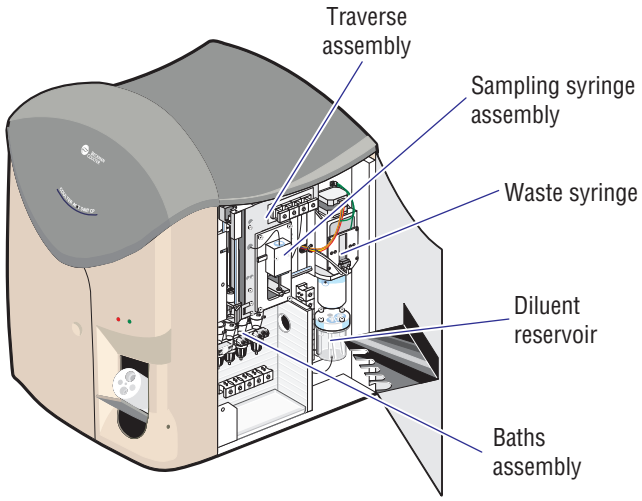
Accessing the Right Side Compartment

Note: Use the special key to loosen the two captive screws securing the right side panel to the instrument frame.



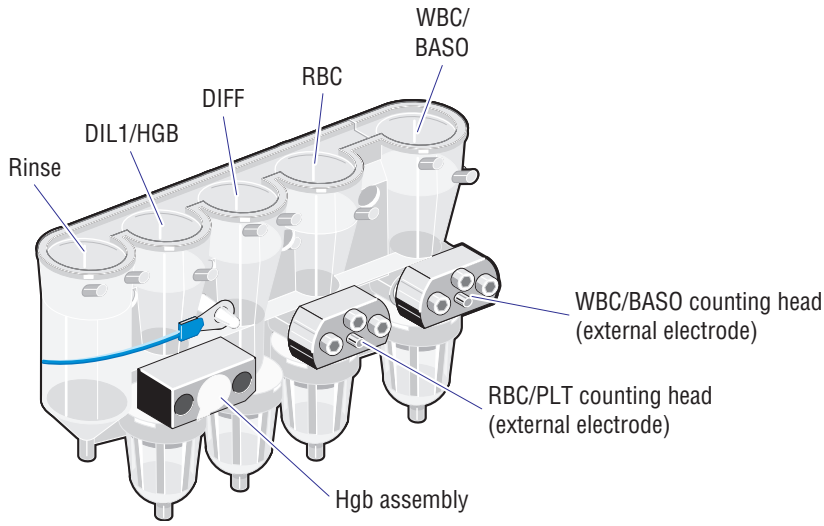
Right Side Compartment Components

Component Locations



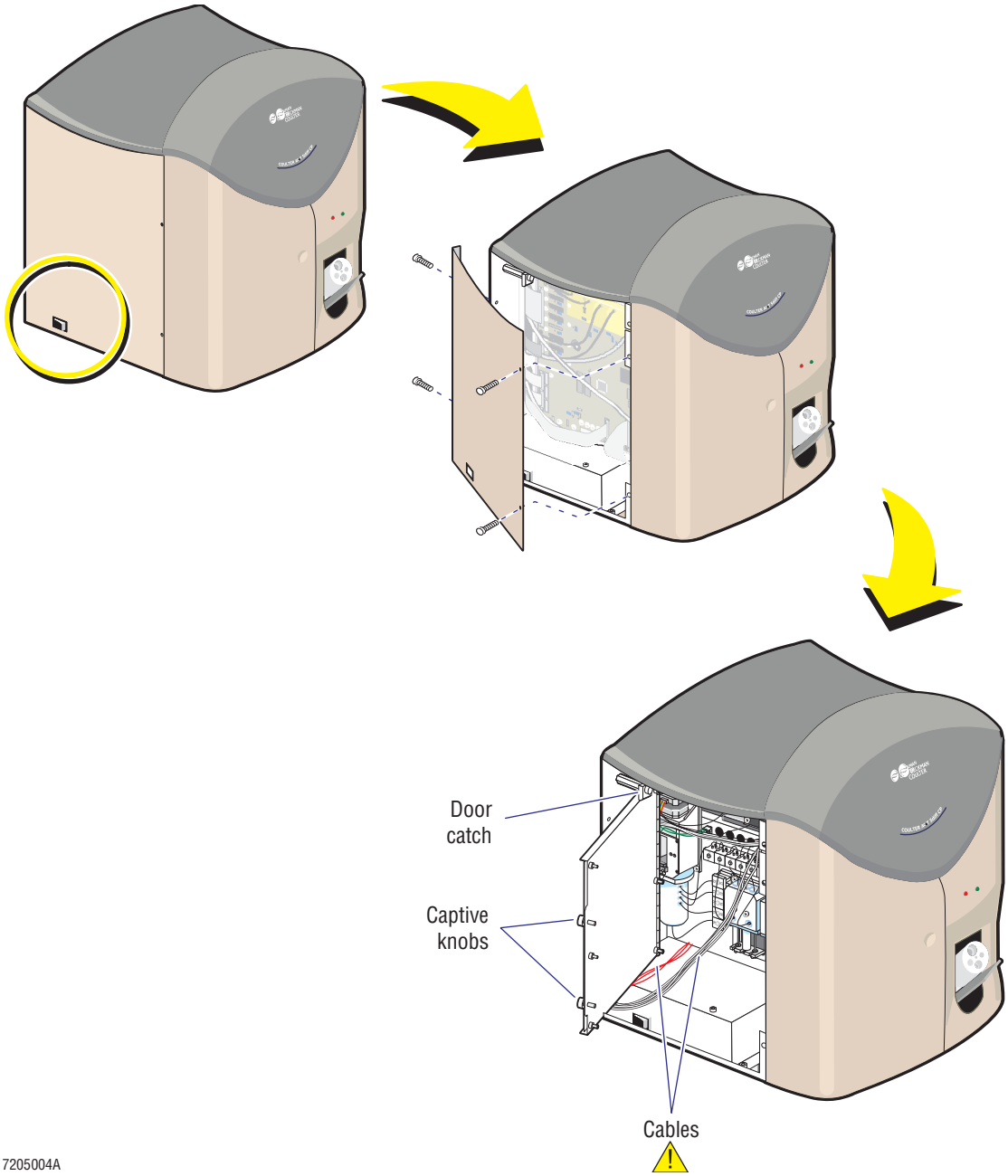
- Allows operator to access hydraulic parts for maintenance operations
- Compartment is heated to ensure cytochemical reaction of the DIFF reagent with the whole-blood sample is consistent
- Mandatory to keep this door locked when running control or patient samples to ensure proper heating of the dilutions

Baths Assembly



Accessing the Left Side Compartment

Note: Use the 3 mm Allen wrench to remove the four hex screws securing the left side panel to the instrument frame. Do not remove this panel unless the instrument is properly powered down using the instructions in the SYSTEM POWER DOWN SUMMARY (summary page from this Training Guide) or the ***Power Down the System*** procedure in Chapter 5 of the Instructions For Use manual.

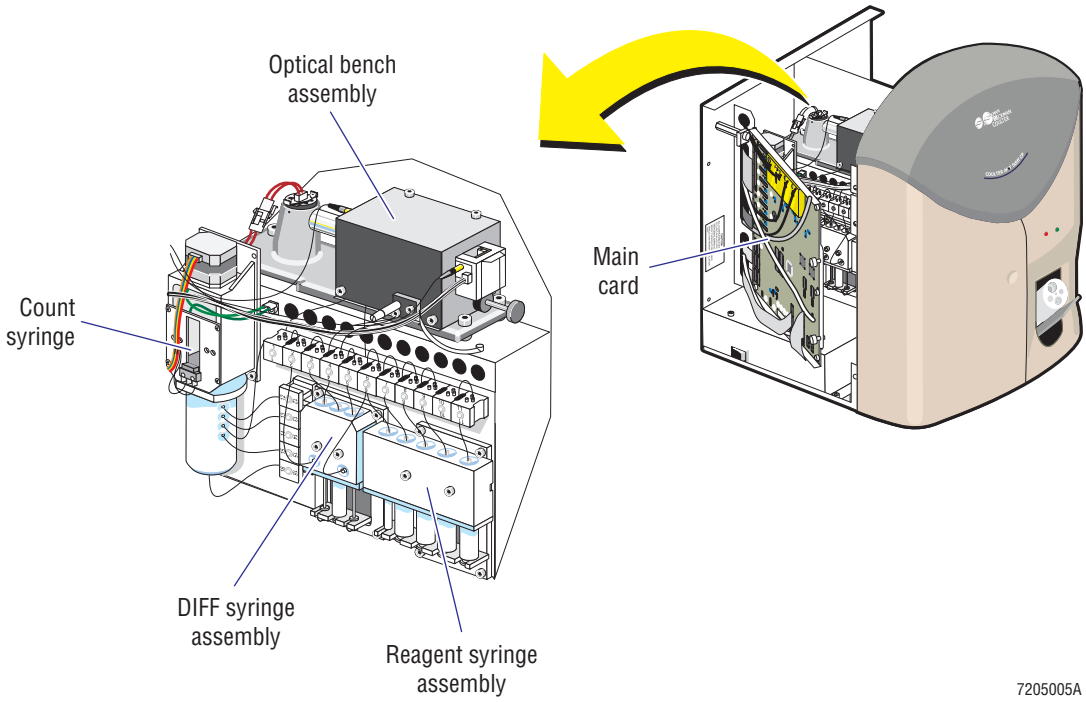


7205004A

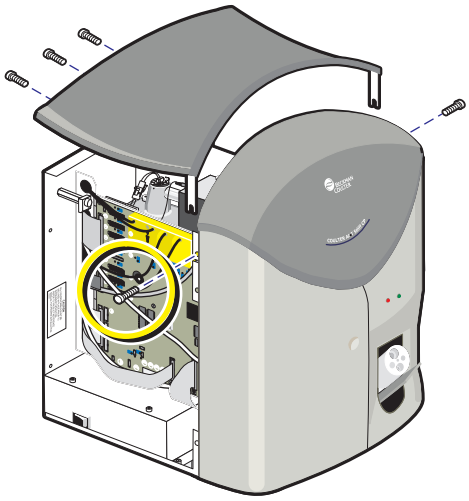
- Allows access to the Main card (circuit card that controls Analyzer operation)
- Allows access to additional hydraulic components maintained by service
- Operator must remove this panel to access the flow cell lamp housing when lamp replacement is necessary

Left Side Compartment Components

Component Locations

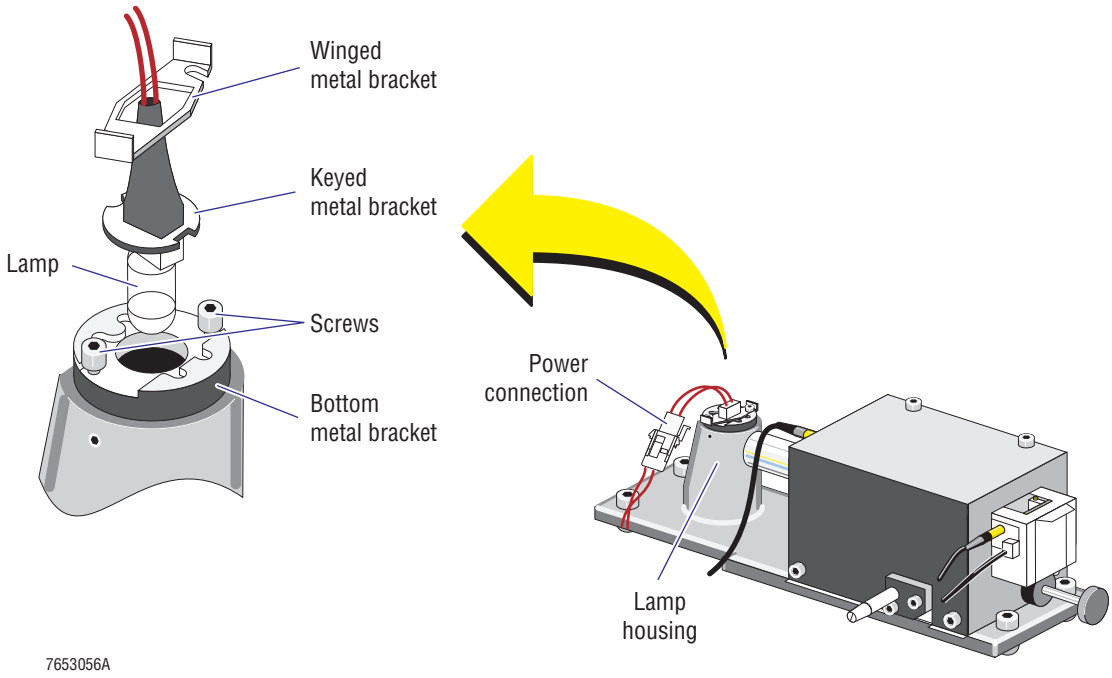


Must Remove the Top Cover to Access the Flow Cell Lamp



- To remove the top cover, operator must locate and remove five screws
  - Left side panel must be removed to remove one screw securing the top cover to the instrument frame; right side door must be opened to access the second screw
  - Three screws secure the top cover to the back panel
- Removal of the left side panel and top cover allows easy access to the flow cell lamp housing located on the optical bench

Flow Cell Lamp Location on the Optical Bench



- Lamp replacement procedure located in the Instructions For Use manual, Chapter 11

  To use Online Help to locate this procedure:

  1) Click 
  2) Click on **Contents**; table of contents appears in left frame
  3) Inside left frame, click on **11 Diagnostics** to list headings in that chapter
  4) Scroll through headings and click on **Replacing the Flow Cell Lamp**; replacement procedure appears inside the right frame
  5) Complete procedure as written:
     a) Follow the instructions on the Workstation screen

        or

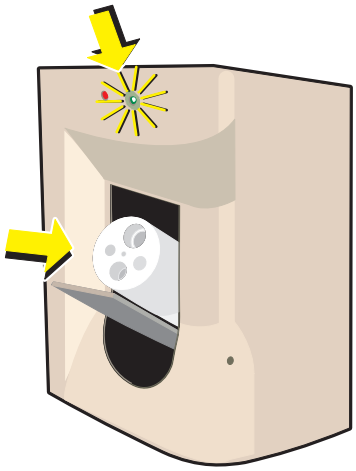
     b) Print a copy of the instructions
        - Right click on procedure (anywhere inside right frame)
        - Click **Print** on the pop-up menu
        - Click **OK** to initiate printing

     Note: All the information under the primary heading is printed. Locate the flow cell lamp replacement instructions and complete the procedure as written.

CYCLE DESCRIPTION

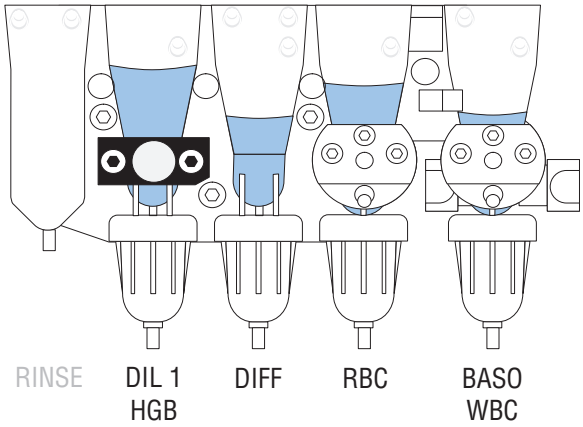
Start Conditions

Front of the Analyzer



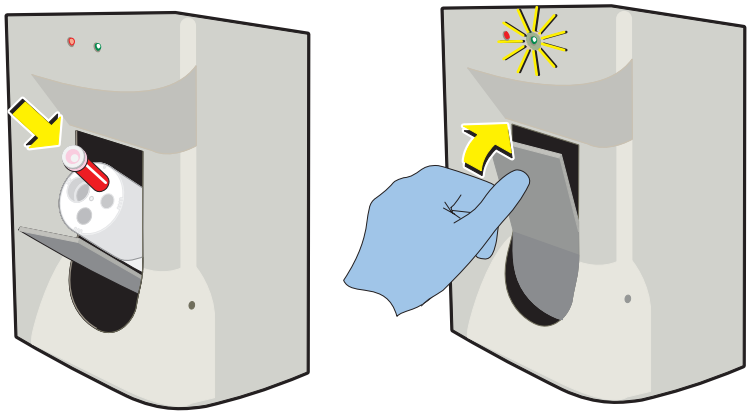
- Cap-pierce door is open
- Green LED is glowing steady indicating the instrument is ready

Baths Assembly (in the Right Side Compartment)



- All baths (except the rinse chamber) are filled with clean diluent

Initiate Cycle

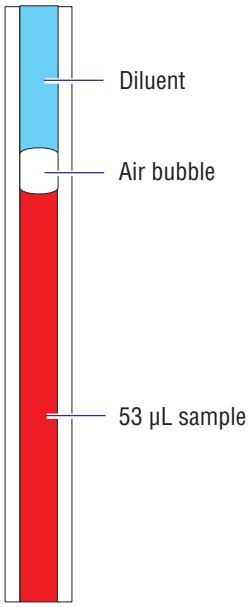


- Select appropriate tube holder
- Insert tube holder on the piercing mechanism and lock it in place
- Place the specimen tube in the tube holder opening that best correlates with size of the specimen tube:
  - Specimen tube may be an open tube or may be sealed with a pierceable stopper
  - For best results, specimen tube must be well mixed before placing the tube in the tube holder
- Verify the specimen tube is in the pierce position (12 o'clock position)
  - Tube holder may be rotated (clockwise or counterclockwise) as needed
- Close door to initiate the cycle
  - Before closing the door, verify the green LED is glowing steady. When the green LED is glowing steady, the instrument is ready to process a sample
  - For best results, time between placing the well-mixed specimen tube inside the tube holder and closing the door should be minimal

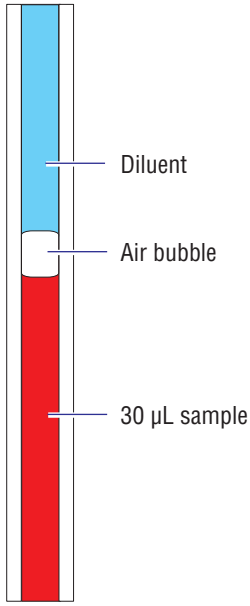
Aspiration

Inside the Sampling Probe

Aspiration in the CBC/DIFF Mode



Aspiration in the CBC Mode

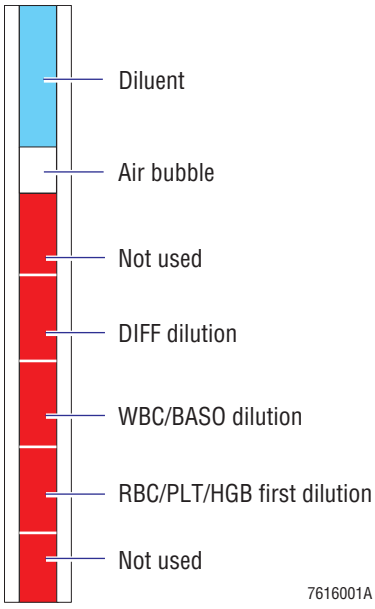


- Sampling probe moves above the pierce position
- Cap pierce mechanism senses the tube holder and determines which tube slot is in the pierce position so the sampling probe can make the appropriate downward movement:
  - If specimen tube has a pierceable stopper, sampling probe pierces the stopper and continues its downward movement until the tip is submerged in the specimen
  - If specimen tube is an open vial, sampling probe makes the same downward movement until the tip is submerged in the specimen
  - If specimen tube is not in the pierce position, cap-pierce mechanism makes these same movements but aspirates air instead of sample
- Volume of whole-blood aspirated into the sampling probe is sufficient to make all the dilutions needed to develop parameter results for the selected panel:
  - 53 µL of whole-blood aspirated when the CBC/DIFF panel selected
  - 30 µL of whole-blood aspirated when the CBC panel selected
- Green and red LEDs alternately flash while the instrument is busy aspirating required sample volume; when aspiration is complete, red LED glows steady as the cycle progresses
- Additionally, all baths drain

Sample Partitioning

- Using the Sequential Dilution System (SDS) technique, aspirated sample is partitioned as it is distributed to make a series of dilutions in a series of baths
- When CBC/DIFF panel selected, three aliquots of the aspirated whole-blood sample are used to make the dilutions
- When CBC panel selected, only two aliquots of the aspirated whole-blood sample are used to make dilutions.; DIFF aliquot is not needed in the CBC mode of operation

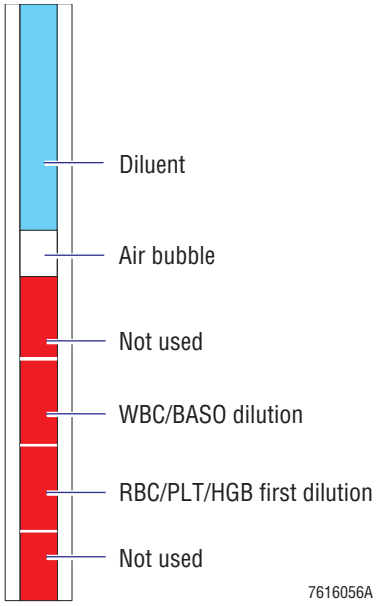
Sample Partitions inside the Sampling Probe when CBC/DIFF Panel is Selected



After aspiration, aliquots of the whole-blood sample are distributed to the various baths as follows:

- 3 µL sample aliquot at the tip of the probe is discarded into the rinse chamber as the exterior of the sampling probe is rinsed, ensuring sample integrity
- 10 µL of sample is delivered to the DIL1/HGB bath for use in preparing the primary RBC/Plt dilution and for measuring Hgb value
- 10 µL of sample is delivered to the WBC/BASO bath for the WBC/BASO count
- 25 µL of sample is delivered to the DIFF bath for development of the DiffPlot
- 5 µL of remaining sample is discarded into the rinse chamber

Sample Partitions inside the Sampling Probe when CBC Panel is Selected

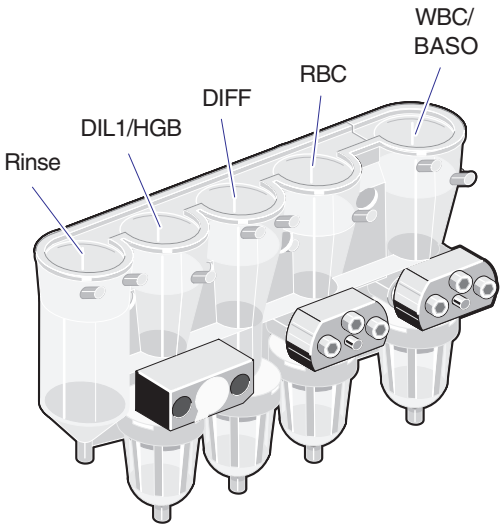


After aspiration, aliquots of the whole-blood sample are distributed to the various baths as follows:

- 3 µL sample aliquot at the tip of the probe is discarded into the rinse chamber as the exterior of the sampling probe is rinsed, ensuring sample integrity
- 10 µL of sample is delivered to the DIL1/HGB bath for use in preparing the primary RBC/Plt dilution and for measuring Hgb value
- 10 µL of sample is delivered to the WBC/BASO bath for the WBC/BASO count
- 7 µL of remaining sample is discarded into the rinse chamber

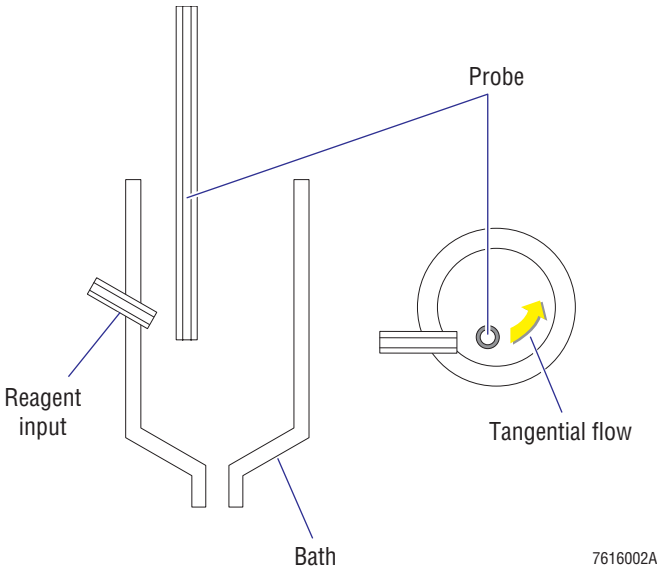
Delivery

Baths Assembly



- Each aliquotted sample is delivered to its appropriate bath
- Sample and reagent are delivered simultaneously using tangential flow

Tangential Flow



- Each aliquotted sample is delivered to its appropriate bath using a tangential flow of reagent which mixes the diluted sample and minimizes viscosity problems
- To create this tangential flow:
  - Delivery port is positioned so that reagent is delivered tangentially to the wall of the bath (In other words, the position of the delivery port allows the reagent to flow along walls of the bath)
  - Sampling probe tip must be properly aligned with the reagent delivery port on the bath
- Simultaneous delivery of sample and reagent at a single point on the curved wall of the bath sets up a flow that not only produces a thoroughly mixed dilution but also sets up a consistent swirling motion that ensures reproducible sample delivery from the probe

Preparing Dilutions

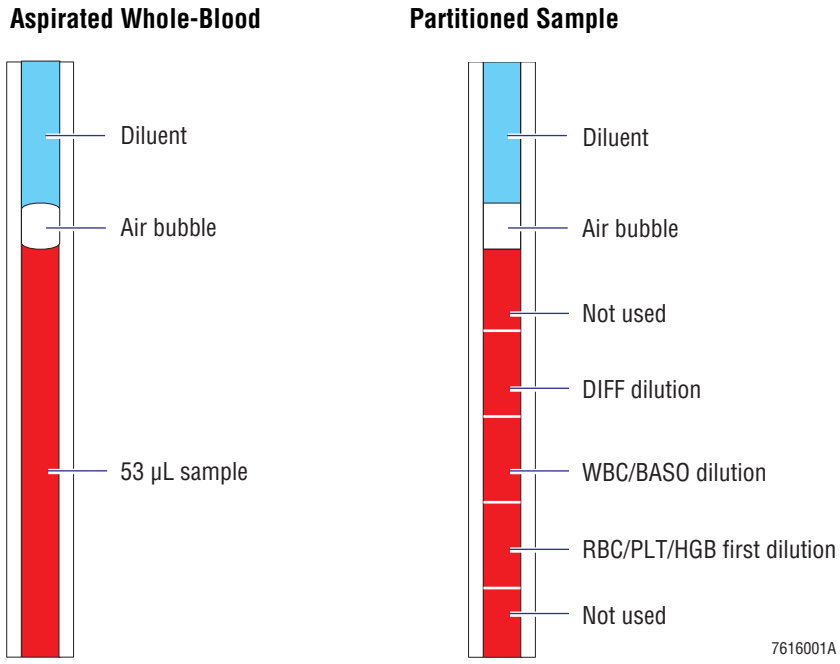
- Using the Sequential Dilution System (SDS) technique, aspirated sample is partitioned as it is distributed to make a series of dilutions in a series of baths
- When a CBC/DIFF panel selected, this series of dilutions occurs in 10 phases

  Note: When a CBC panel is selected, phase 5 (making the DIFF dilution) and phase 7 (stopping the DIFF reaction) are not done because the DIFF was not selected. The elimination of these phases means the volume of reagents used in making the DIFF dilution (1.0 mL of A^{C}•T 5diff Fix and 1.0 mL of A^{C}•T 5diff Diluent) are not dispensed and wasted.

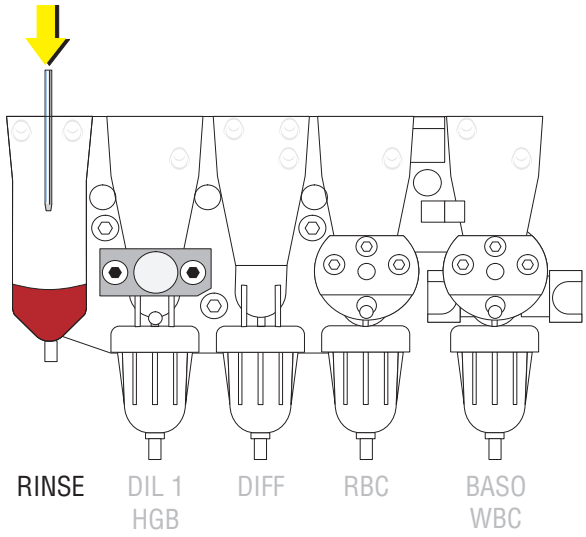
Phase 1 Sample Aspiration

- 53 µL of whole-blood is aspirated into the sampling probe from the specimen tube

  Note: If a CBC panel is selected, 30 µL of whole-blood is aspirated.

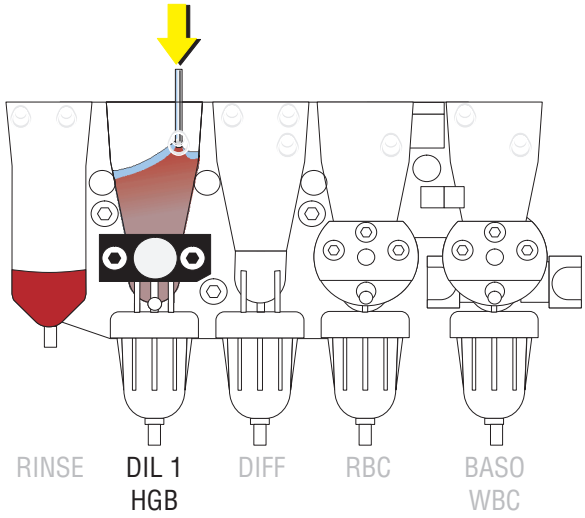


Phase 2 Initial Discard and External Probe Rinse



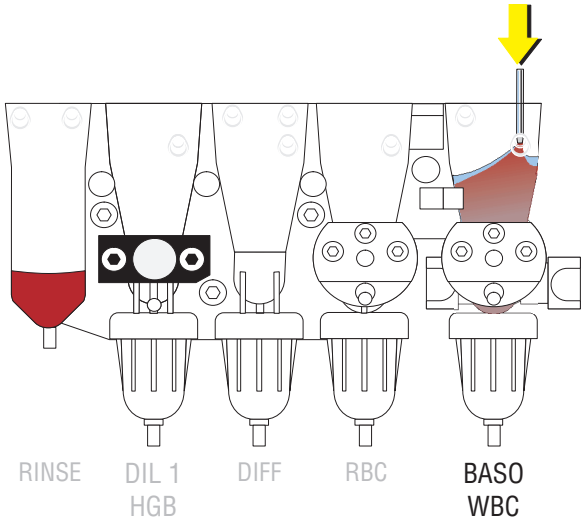
- Horizontal traverse assembly positions sampling probe over rinse chamber
- 3 µL sample aliquot at the tip of the sampling probe is discarded into the rinse chamber as exterior of sampling probe is rinsed
- Discarding 3 µL aliquot of aspirated sample helps ensure sample integrity
- After rinsing the outside of the sampling probe, three aliquots of the aspirated whole-blood sample are distributed to make the dilutions for analysis

Phase 3 Make the First Dilution



- Horizontal traverse assembly positions the sampling probe over DIL1/HGB (first dilution/Hgb) bath
- Vertical traverse assembly moves the probe downward into the bath; probe tip is positioned to produce a tangential flow when the sample and diluent are simultaneously dispensed into the bath
- 10 µL of the whole-blood partitioned for making the first dilution is delivered to the DIL1/HGB bath using a tangential flow of 1.7 mL of diluent
- Tangential flow of reagent mixes the sample and the diluent; mixing bubbles enter the bath to make a uniform suspension of cells
- This 1:170 dilution commonly referred to as the first dilution

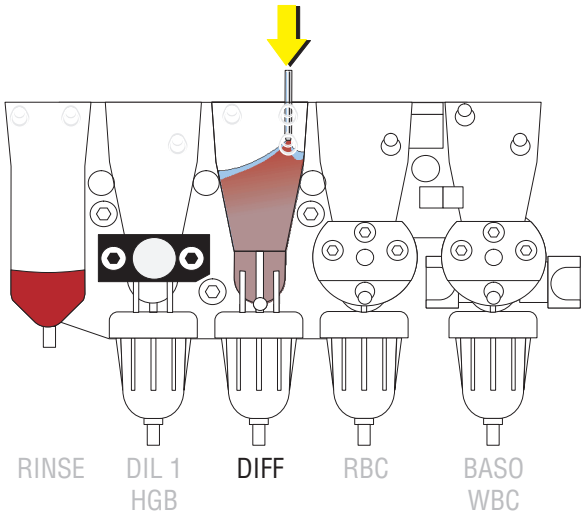
Phase 4 Make the WBC/BASO Dilution



- Horizontal traverse assembly positions sampling probe over the WBC/BASO bath
- Vertical traverse moves the probe downward into the bath; tip of the probe is positioned so that a tangential flow occurs as the 10 µL of the whole-blood sample and 2.0 mL of WBC Lyse are simultaneously dispensed into the bath
- Tangential flow of reagent mixes the sample and reagent; mixing bubbles enter the bath to make a uniform suspension of cells
- WBC Lyse destroys the red blood cells and the specific lytic action on the white blood cells differentiates the basophils from other WBCs
- This final 1:200 dilution is used to determine the WBC count; WBC/BASO histogram, from which the BASO % is determined, is also generated from this dilution.

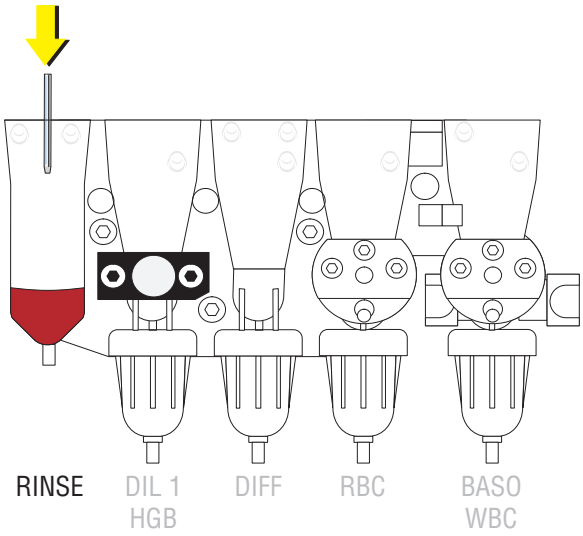
WBC/BASO Bath Dilution	
Whole-blood volume	10 µL
Volume of A^{C}•T 5diff WBC Lyse reagent	2000 µL
Dilution ratio	1:200

Phase 5 Make the DIFF Dilution



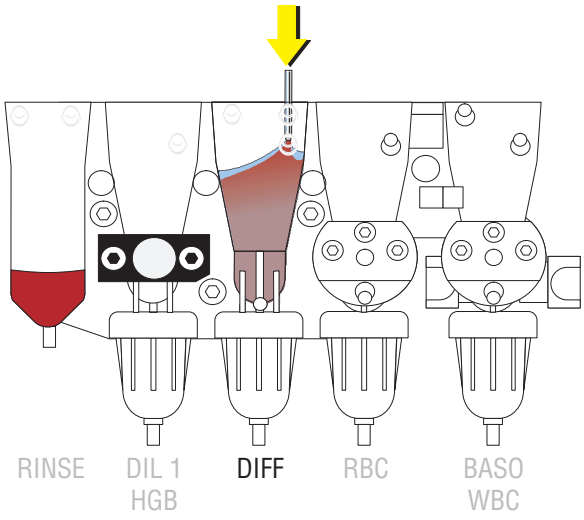
- Horizontal traverse assembly moves sampling probe over DIFF bath
- Vertical traverse assembly moves probe downward into the bath
- Tip of probe is positioned so that a tangential flow occurs as 25 µL of the whole-blood sample and 1.0 mL of Fix reagent are simultaneously dispensed into the bath
- Tangential flow of reagent mixes the sample and the Fix reagent; mixing bubbles enter the bath to make a uniform suspension of cells
- Fix reagent lyses the red blood cells, stabilizes the WBCs in their native form, and differentially stains the lymphocytes, monocytes, neutrophils, and eosinophils, with the eosinophils staining most intensely

Phase 6 Wash the Sampling Probe



- Horizontal traverse assembly moves sampling probe over the rinse chamber
- Double rinse (interior and exterior) of sampling probe flushes all traces of residual whole-blood sample from the probe into rinse chamber
  - Rinsing probe's interior removes whole-blood sample that was aspirated but not used in making the dilutions:
    - In CBC/DIFF mode, 5 µL is discarded in rinse chamber
    - In CBC mode, 7 µL is discarded in rinse chamber
  - Rinsing probe's exterior removes all residual whole-blood from probe's surface which minimizes carryover

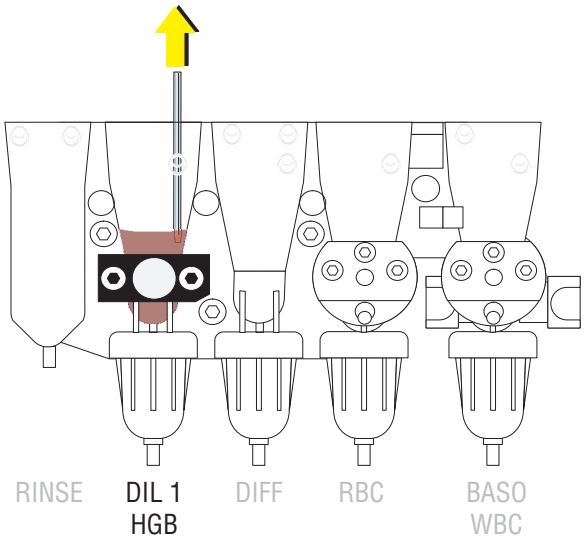
Phase 7 Stop the DIFF Reaction



- After 25 µL of whole-blood sample and 1.0 mL of Fix reagent incubate 12 seconds, staining process inside DIFF bath is completed by adding another 1.0 mL of diluent which stops the cytochemical reaction.
- This final 1:80 dilution is sent to flow cell for generation of the DiffPlot
- Neutrophil %, lymphocyte %, monocyte %, and eosinophil % are determined from this DiffPlot

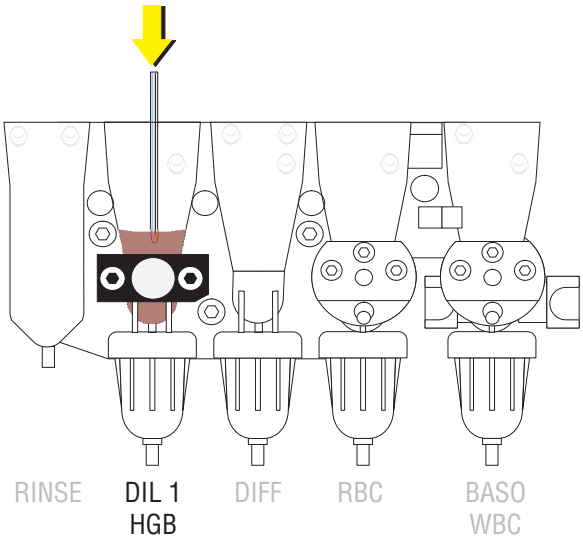
DIFF Bath Dilution	
Whole-blood volume	25 µL
Volume of A^{C}•T 5diff Fix reagent	1000 µL
Volume of A^{C}•T 5diff Diluent	1000 µL
Final dilution ratio	1:80

Phase 8 Aspiration from the First Dilution



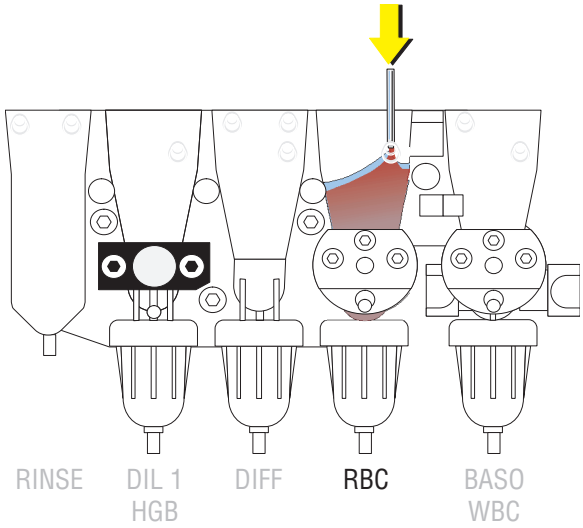
- Horizontal traverse assembly moves sampling probe over the DIL1/HGB bath
- Vertical traverse assembly moves probe downward into the bath
- 42.5 µL of the 1:170 first dilution is aspirated into the sampling probe

Rinse the Outside of the Sampling Probe



- While still inside the DIL1/HGB bath, exterior of sampling probe is rinsed with 0.4 mL of diluent
- Vertical traverse assembly moves probe up out of the bath

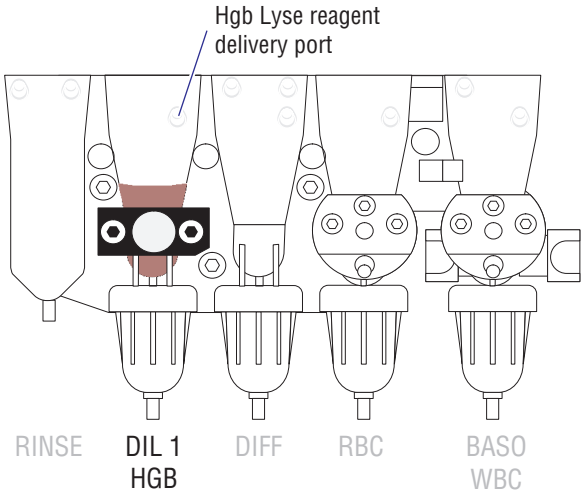
Phase 9 Make the RBC/Plt Dilution



- Horizontal traverse assembly moves sampling probe over the RBC bath
- Vertical traverse assembly moves probe downward into the RBC bath
- Tip of the probe is positioned so that a tangential flow occurs as the 42.5 µL of 1:170 dilution obtained from the first dilution in the DIL1/HGB bath and 0.5 mL of diluent are simultaneously dispensed into the bath
- An additional 2.0 mL of diluent is dispensed through the probe
- This final 1:10,000 dilution is analyzed to determine RBC count and Plt count; RBC and PLT histograms are also generated from this dilution

RBC Bath Dilution	
Volume 1:170 dilution from DIL1/HGB bath	42.5 µL
Volume of A^{C}•T 5diff Diluent reagent	2500 µL
Final dilution ratio	1:10,000

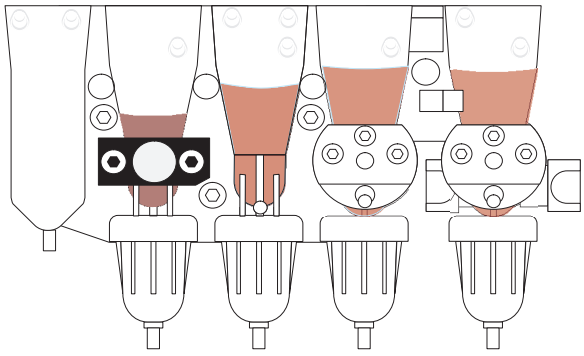
Phase 10 Make the Hemoglobin Dilution



- 0.4 mL of Hgb Lyse is added to the bath
- Hgb Lyse reagent rapidly destroys the red blood cells and converts a substantial proportion of the hemoglobin to a stable pigment so a hemoglobin value can be determined
- Mixing bubbles enter bath to ensure a uniform dilution
- Hemoglobin value is photometrically determined from this final 1:250 dilution.

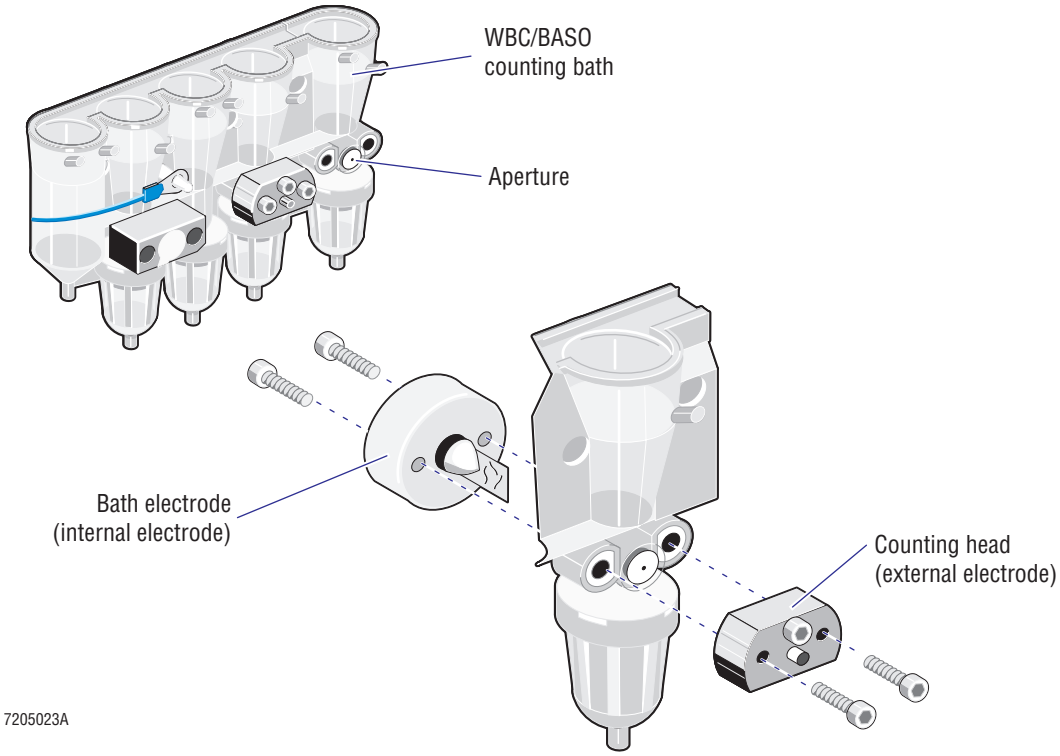
DIL1/HGB Bath Dilution	
First dilution	1:170
Volume of first dilution removed	42.5 µL
Volume of A^{C}•T 5diff Hgb Lyse reagent	400 µL
Volume of A^{C}•T 5diff Diluent reagent	400 µL
Final dilution ratio	1:250

Final Dilutions for Analysis

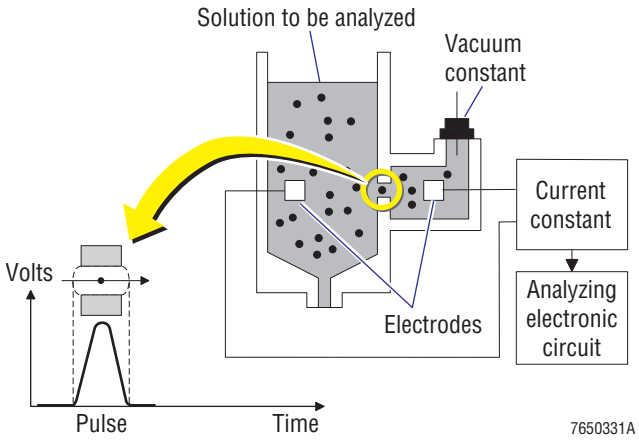


Bath	Dilution	Final Dilution Ratio
DIL1/HGB	Step 1 10 µL whole-blood sample + 1.7 mL diluent Step 2 42.5 µL of the first dilution removed 0.40 mL diluent added during external probe wash Step 3 0.4 mL Hgb Lyse reagent added	1:250
DIFF	Step 1 25 µL whole-blood sample + 1.0 mL Fix reagent Step 2 12 second incubation Step 3 1.0 mL diluent added	1:80
RBC (and Plt)	42.5 µL of the first 1:170 dilution + 2.5 mL diluent	1:10,000
WBC/BASO	10 µL whole-blood sample + 2.0 mL WBC Lyse reagent	1:200

WBC and BASO Counting



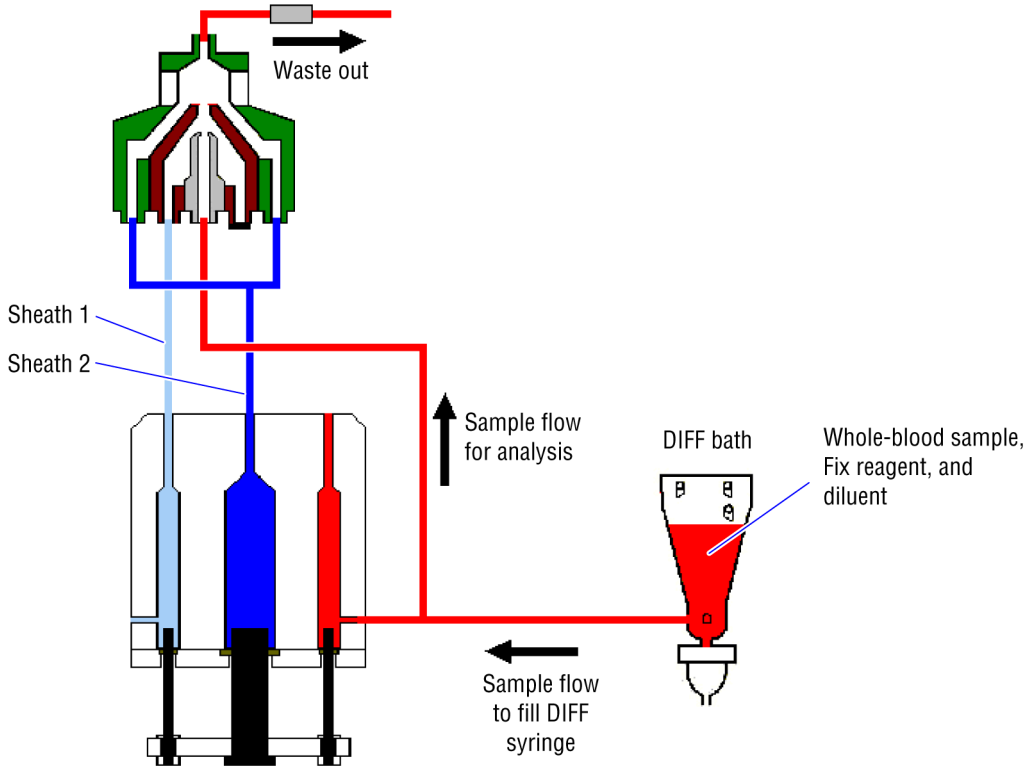
- Final 1:200 dilution inside the WBC/BASO bath is used to:
  - Determine the WBC count, and
  - Develop the WBC/BASO histogram, which is needed to obtain the BASO count
- When 10 µL of whole blood is mixed with 2,000 µL of A^{C}•T 5diff WBC Lyse reagent in the WBC/BASO bath, this reaction lyses the red blood cells and specifically differentiates between basophils and other leukocytes by volume
- It is important that the Analyzer maintains the reagents and reaction at a regulated temperature of 35°C (95°F); therefore, the right side door must remain closed



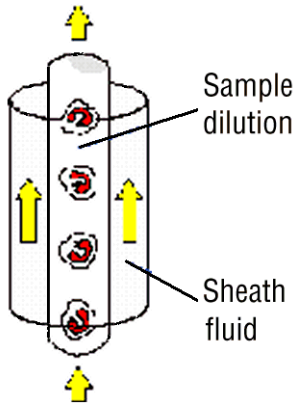
- Count syringe supplies a constant low vacuum to the WBC/BASO bath to pull the diluted sample through an 80 µm aperture
- As each cell passes through aperture:
  - Pulse is generated proportional to the cellular volume
  - Total leukocyte count and basophil percentage are determined by specific thresholds on the WBC/BASO histogram
- WBC count and BASO count determined simultaneously
- WBC count is determined twice using two different methodologies:
  - Count obtained in WBC/BASO bath is the reference WBC count
  - Second WBC count is determined in the flow cell during acquisition of the DiffPlot; dilution analyzed in the flow cell is prepared in the DIFF bath
  - WBC count results from the two methodologies are compared and if results exceed predefined limits, WBC count result is flagged

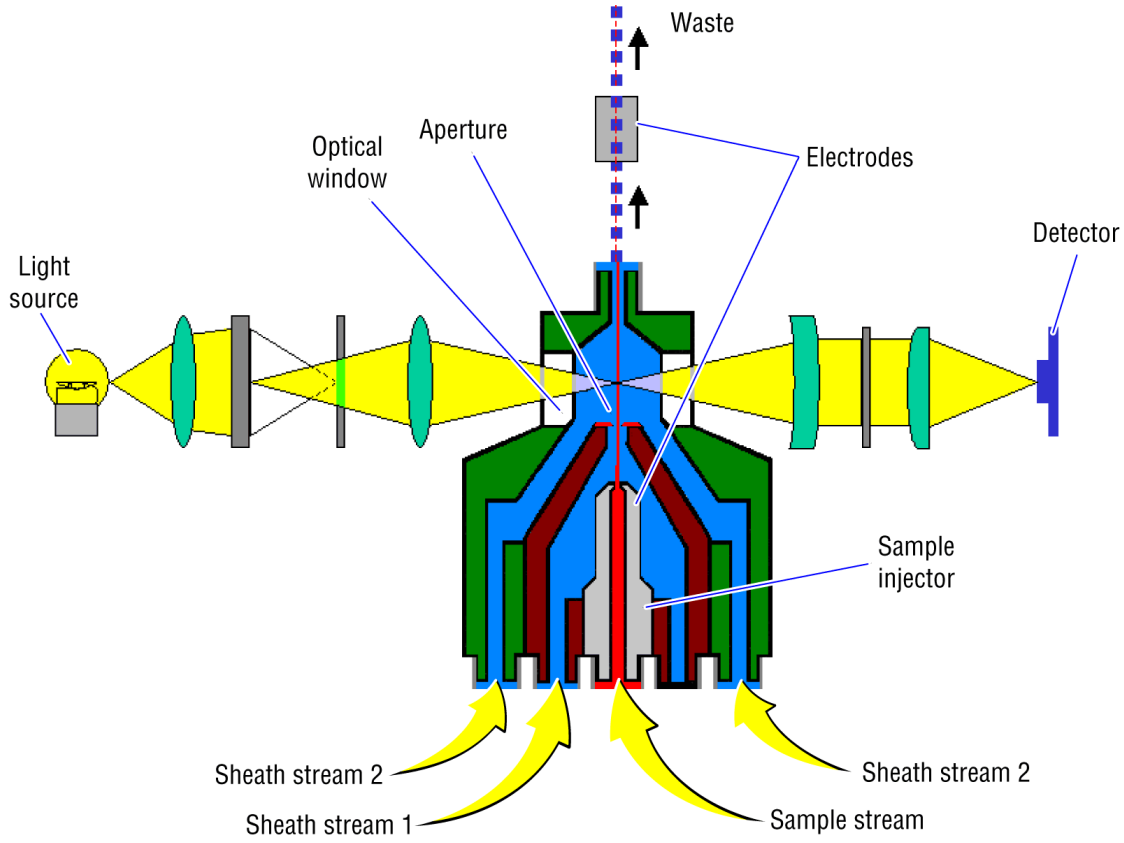
    Note: The comparison between the WBC count from the WBC/BASO bath and the WBC count from the flow cell is not performed when the CBC panel is selected or when this option is disabled in setup.

DIFF Analysis

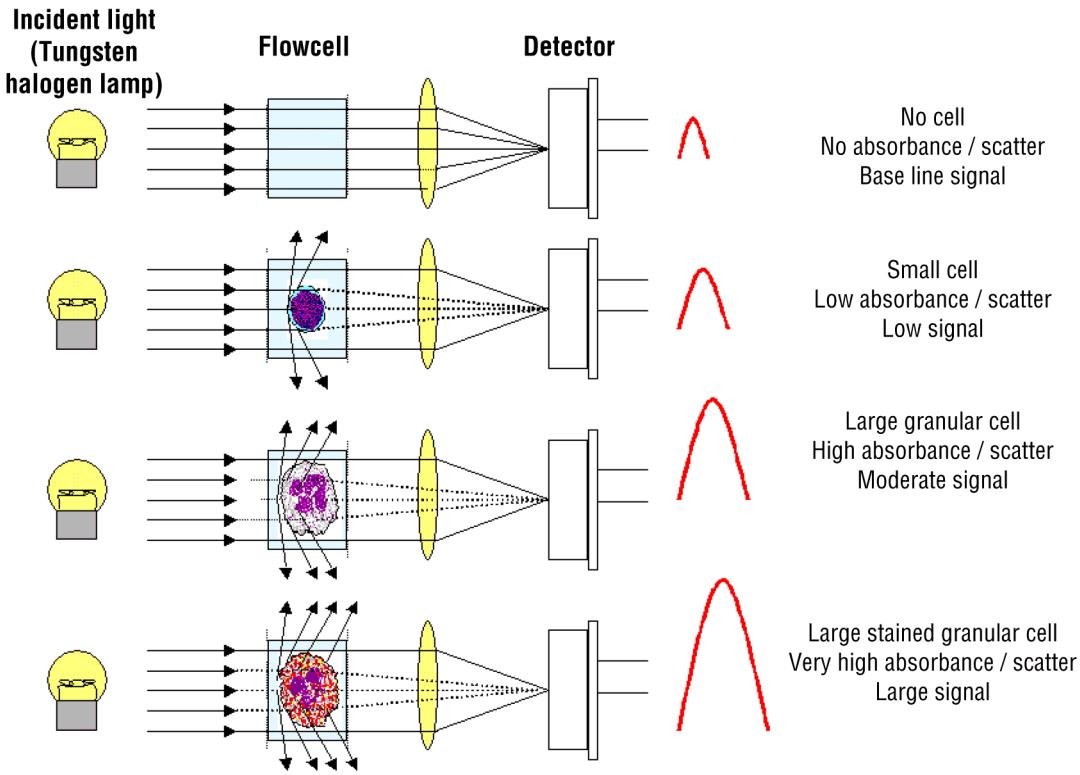


- Final 1:80 dilution in the DIFF bath is sent to the flow cell for generation of the DiffPlot
- Analyzer must maintain reagents and reaction at a regulated temperature of 35°C (95°F); therefore, the right side door must remain closed
- Each stained cell is individually focused by the Dual Focused Flow (DFF) system and transported through the flow cell using sample pressure and diluent sheath flow
- 72 µL of sample from the DIFF bath is injected through the flow cell 15 seconds; for 12 of these 15 seconds, data for developing the DiffPlot is accumulated

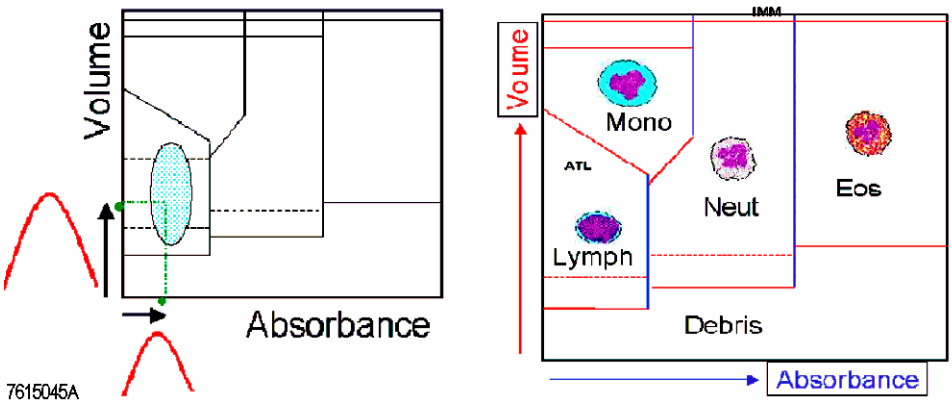




- DFF uses sheath fluid to surround and force cells suspended in diluent to pass one at a time through the center of the flow cell (hydrodynamic focusing process)
  - First sheath flow focuses the sample through the impedance aperture
  - Second sheath flow maintains the focused flow of cells as they exit the aperture into the optical flow cell
- Hydrodynamic focusing in the flow cell enables accurate and rapid cell-by-cell measurements on a large number of individual cells
- Sequential analyses for cell volume (impedance) and light absorbance are performed in the flow cell

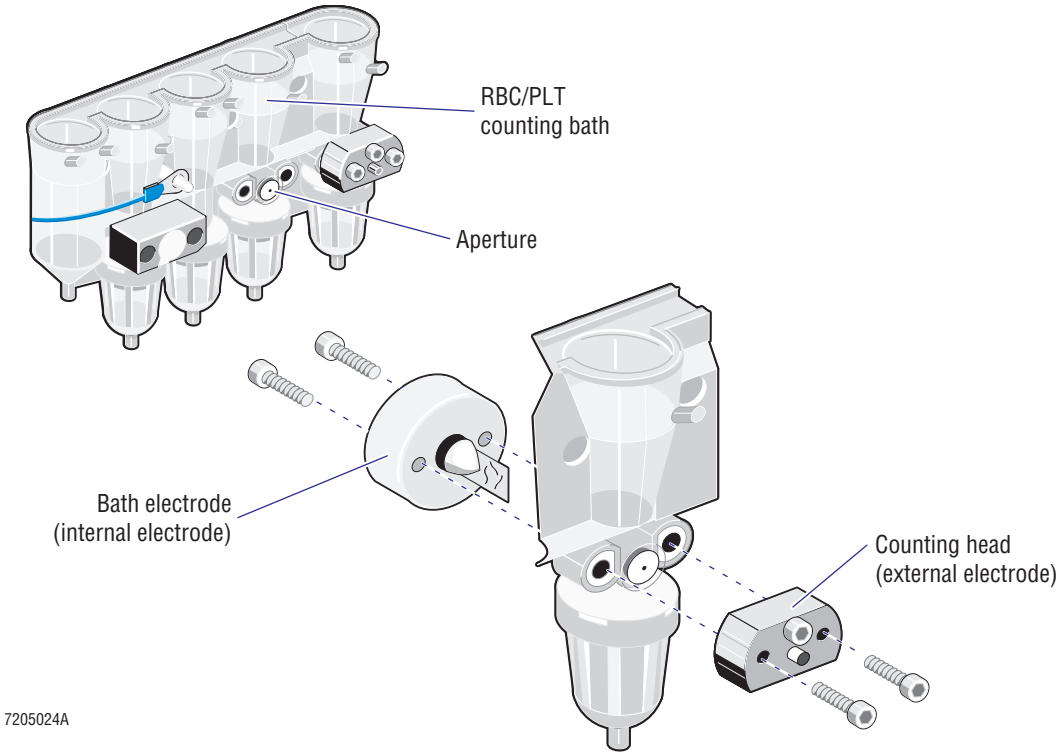


- Lymphocytes, monocytes, neutrophils, and eosinophils each have a unique nuclear and morphology structure and staining intensity
- Light **absorbance** is related to cellular contents (granularity, nuclear content, and so forth) after cytochemical staining
- As a cell passes through the optical portion of the flow cell, light is scattered in all directions
  - Sensor detects only forward scattered light
  - Optical measurement is derived as a function of the amount of light lost due to diffraction and absorbance, as compared to full transmission when no cell is present
  - Collected signals are converted into voltage pulses and are processed
  - Magnitude of the voltage pulses are proportional to the physical and chemical characteristics of the cells being analyzed
  - Measurements provide information for lymphocytes, monocytes, neutrophils, and eosinophils, and their precursors



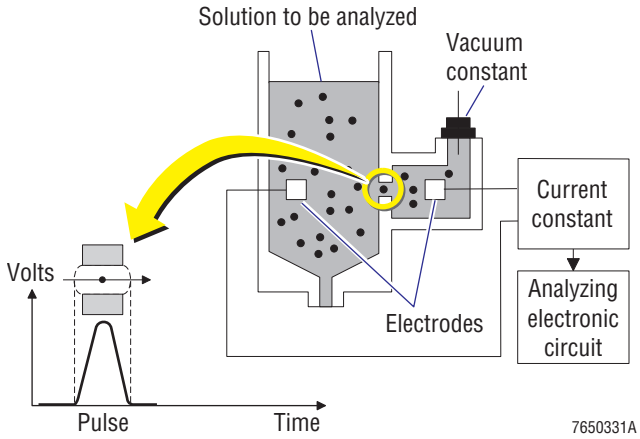
- Focused flow impedance technology measures the electrical resistance of a cell as it passes through the aperture in the flow cell; change in resistance is directly proportional to the **volume** of the cell
- Output signals from the focused flow impedance and the light absorbance measurements are combined to define the WBC differential population clusters

RBC/Plt Counting and Sensing

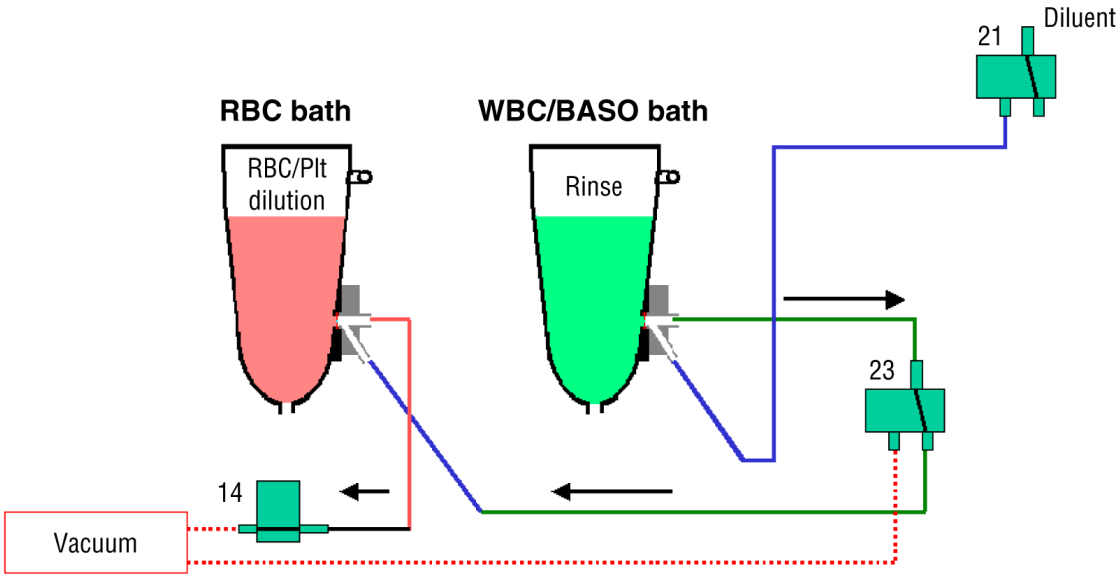


- Final 1:10,000 dilution in RBC bath contains red blood cells, white blood cells, and platelets
- Thresholds are used to separate the platelet pulses, which are much smaller, from the red and white blood cell pulses

  Note: Since white blood cells fall in the red blood cell size range, they are counted and sized as RBCs. The WBCs are not sorted out because any interference is usually insignificant; there are normally very few WBCs (thousands) in comparison to the number of RBCs (millions). Only when the white count is markedly elevated is the red cell count or histogram influenced.

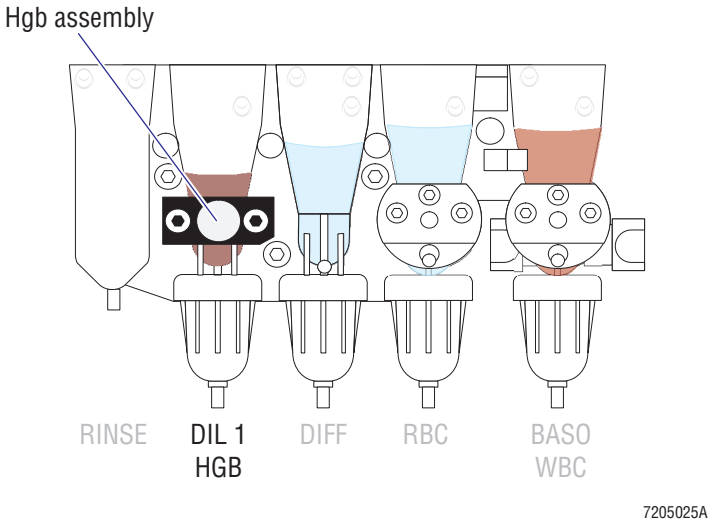


- Count syringe supplies a constant low vacuum to the RBC bath to pull the diluted sample through a 50 µm aperture
- As each cell passes through the aperture, a pulse is generated proportional to the cellular volume
- RBC aperture senses red blood cells and platelets:
  - RBC count and PLT count are determined simultaneously
  - RBC histogram is developed
  - PLT histogram is developed



- Rinse flow is a steady stream of A^{C}•T 5diff Diluent reagent that flows behind the RBC aperture during sensing periods
- Without rinse flow, there is a characteristic swirling of the dilution at the outlet of the aperture so that red (or white) cells caught up in these eddies behind the aperture may reenter the sensing zone and produce small pulses that could be counted as platelets
- With rinse flow, the steady stream of diluent prevents the formation of these eddies

Hemoglobin Sample Reading



- Hemoglobin concentration is determined based on the transmittance of light through the optical part of DIL1/HGB bath using a spectrophotometric technique at a wavelength of 550 nm
- Three one-second readings are collected on the 1:250 dilution

Completion of the Cycle

- The WBC bath drains and fills with A^{C}•T 5diff Rinse reagent
- Hemoglobin blank is read
- Transmittance of hemoglobin sample reading is compared with the transmittance of the reagent blank
- Analyzer calculates the Hgb result based on both the blank and sample readings
- Green LED glows steady; red LED no longer glows

  Note: It is now okay to place another specimen tube in the tube holder and close the cap-pierce door to begin another cycle.
- Main card inside the Analyzer's left compartment sends the sample results to the Workstation

NOTES

BASIC TROUBLESHOOTING TECHNIQUES

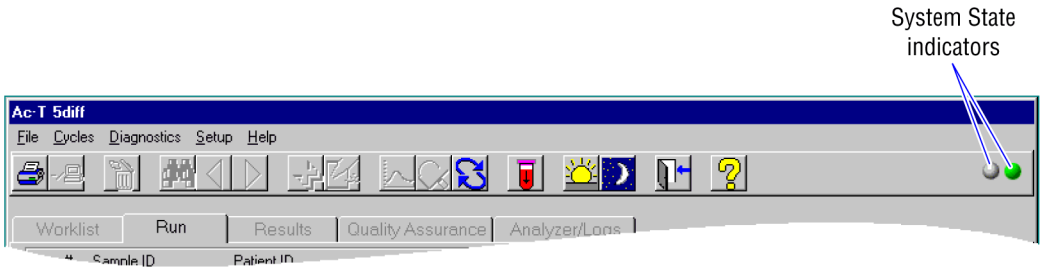
How to Recognize an Instrument Problem

- By the data that is generated
  - Start Up
  - Commercial cell controls
  - XM analysis
  - Patient samples
  - IQAP
- By the way the instrument functions . . . abnormalities in the sample flow
- By the error messages generated and recorded on the Error Log

Error Log

- Click on **Analyzer / Logs** tab ▸▸ **Error Log** tab to view the log
- Shortcut to viewing the Error Log

  Double click on one of the System State indicators



- Maintains most recent 100 entries displaying the most recent log entry at the top of the log and the oldest entry at the bottom of the log
- Rollover is first in, first out when the 101st entry is added
- Log contains the following information fields

  Note: It is necessary to scroll right to see some of these fields.

Information Fields

Field	Function
Date/Time	Displays the date and time the error occurred
Opr	Displays the User name logon information in the Workstation when the error was entered in the log
Category	Displays the category of error that was logged, either related to the Analyzer or Workstation
Description	Displays the error reported
Comment	Displays any comments entered by the operator performing the error:

- Optional entry that provides an opportunity for the operator to type a personal observation or appraisal
- Operator may use the **Add Comments** button to access the Add Comments box, as desired
- If the **Prompt User For Comments** check box is checked (✓), the Add Comments box pops up automatically when an error occurs to prompt operator to enter a comment

  Note: When Add Comments box is displayed, operator cannot access any other window until they either:

  - Enter a comment then click  to save and exit

    or

  - Click  to exit the box without entering a comment

Error Messages

- Error messages with corrective actions are listed in Chapter 11 of the Instructions For Use manual
- To use Online Help to locate these messages:

  1) Click 
  2) Click on **Contents**; table of contents appears in the left frame
  3) Inside the left frame, click on **11 Diagnostics** to list headings in that chapter
  4) Scroll through headings and click on **System Errors**; messages appear inside the right frame
  5) Scroll to locate message that appears in the Error Log

     or

     Print a copy of the messages

     - Right click on the messages (anywhere inside the right frame)
     - Click **Print** on the pop-up menu
     - Click **OK** to initiate printing

Three Steps to More Efficient Troubleshooting

1. Be completely familiar with normal operation
   - Start Up, Sample Analysis, Shut Down
   - Normal Sample Flow
     - Know what should be happening during each part of the cycle
     - Become familiar with the normal sounds of operation
2. Use a logical approach to obtain a clear symptom and isolate the components involved
   - Use the error message information as a starting point
   - Use your knowledge of sample flow
3. Acquire the knowledge and skills necessary to locate and correct problems
   - Use the instrument
   - Read the manuals and become familiar with the kind of information they contain
   - Call your Beckman Coulter Representative

Troubleshooting by Instrument Subsystems

- Three instrument subsystems:
  - Electronic
  - Reagent
  - Fluidic
    - Pneumatics (pressures and vacuums)
    - Hydraulics (liquids)

Electronic Subsystem

- If there's no power,
  - Check to see if one of the indicator lights is on
  - Check power switches
  - Check plug connections
  - Check fuse locations
- If the flow cell lamp is burned out, see Chapter 11 in the Instructions For Use manual

Reagent Subsystem

Level Sensing

- Level sensing is accomplished using two different types of sensors
  - A^{C}•T 5diff Diluent reagent level sensing uses optical sensor
  - Sensing waste container level uses a float-level sensor
- Cycle counters monitor levels for those reagents housed inside the reagent compartment

Replacement Procedures

- To replace a reagent, see the Changing Reagent Summary
- To replace a waste container, see the Replacing a Waste Container Summary
- To replace a stopper assembly, see the Instructions For Use manual

Reagent Related Problems

- May be contaminated
- May be expired
- May have been frozen causing the ingredients to separate
- May be the source of electrical interference due to electrolytic characteristics

Fluidic Subsystem

Key Components

- Tubing
- Valves
- Syringes
- Solenoids

Hydraulics

- Check tubing connections and routing
  - At the reagent container
  - At all associated syringe assembly
  - Throughout the Diluter part of the Analyzer
- Check associated syringes, valves, and solenoids

Pneumatics

- Low vacuum
  - Generated by the count syringe
  - Equivalent to 5.9 inches of mercury
  - Also called aperture vacuum
  - Pulls the dilutions through the RBC and WBC apertures and sweep flow diluent behind the RBC aperture
- Pressure
  - Used for dispensing reagents from pumps, opening pinch valves, and moving air cylinder shafts

Troubleshooting through Cycle Observation

- To successfully troubleshoot this instrument,
  - Become completely familiar with normal sample flow
  - Know what should be happening during each part of the cycle
  - Be familiar with the normal sounds of operation
- This type of knowledge is gained mostly through experience
- To increase your familiarity and your comfort level with the normal sample flow of this instrument, periodically review the Cycle Description in this Training Guide or the Sample Analysis descriptions in Chapter 2 of the Instructions For Use manual
- Key Operator should know normal sample flow well enough to verbally describe the normal operation sequence without the aid of a document

Troubleshooting using Special Procedures

Reproducibility Check

- Check measures how close several results (from the same specimen) are to each other; in other words, this check measures repeatability or precision
- Check may be done using CBC or CBC/DIFF panel
- For best results, a fresh, normal whole-blood specimen must be used
- One normal, fresh whole-blood specimen is cycled at least 5, but no more than 11 times
- CV or Coefficient of Variation is a percentage of deviation from the mean
- Results that exceed the limits appear against a red background
- Reproducibility does not measure accuracy, but true accuracy is not possible unless the instrument is precise
- Preforming a reproducibility check before performing a calibration may catch imprecision problems that may affect the calibration process
- Procedure is located in the Instructions For Use manual, Chapter 11

Carryover Check

- Carryover is the interaction of the previous sample with the current sample
- High to low carryover check verifies the high results of one sample do not affect the low results of the next sample
- Check is done by analyzing a whole-blood specimen with high values followed by a whole-blood specimen with low values; each specimen is run consecutively in triplicate
- Carryover is calculated as follows:

$$\%\ \text{Carryover} = \frac{\text{1st low sample} - \text{3rd low sample}}{\text{1st high sample} - \text{3rd low sample}} \times 100$$

Power Down / Power Up

- When performing certain replacement procedures, it may be necessary to power down the system to prevent personal injury from electric shock
- When the system needs to be powered down, proper sequence must be followed to prevent damage to the system
- When the system is powered up, proper sequence must be followed to ensure proper connection between Workstation and Analyzer

➤ SYSTEM POWER DOWN SUMMARY

1 At the Workstation

- Click  .
- Select **Quit Application**.
- Press Enter or click  .
- Wait while the Workstation closes its program.
- When the Begin Logon box appears, press Ctrl + Alt + Delete simultaneously.
- Click **Shut Down** at the Logon Information box.
- Select **Shutdown** then click **OK** at the Shutdown Computer box.
- When the *It is now safe to turn off your computer* message appears, power the Workstation computer off.

2 At the Analyzer

- Toggle the Power On/Off rocker switch OFF (position O). This rocker switch is located at the base of the left side panel.
- If a replacement or maintenance procedure that requires you to open the Analyzer panels is being performed, unplug the ac power cord. Either remove the cord from the instrument (at the back panel, in the lower right corner) or from the ac wall outlet.

➤ ➤

➤ SYSTEM POWER UP SUMMARY

1 Preliminary Checks

- Check the waste container level.
  - ▸ If the container needs to be replaced, change the container using the Replacing a Waste Container Summary as a guide.
- If the Automatic Startup and Auto-Print functions are enabled, make sure the Printer is ready to print.
  - ▸ If the paper supply is insufficient, add paper.
  - ▸ Make sure the power is ON and ready to print.
  - ▸ Verify the printer is properly configured according to your laboratory's protocol.

2 Power Up the System

At the Analyzer

- If the ac power cord is unplugged, plug the cord into the instrument (at the back panel, in the lower right corner) or the ac wall outlet, as applicable.
- Toggle the Power ON/OFF rocker switch ON (position –). This rocker switch is located at the base of the left side panel.
- Verify the red LED is glowing steady.

At the Workstation

- Power ON the Workstation computer.
- Wait while the computer performs its internal checks.
- When the Begin Logon box appears, press Ctrl + Alt + Delete simultaneously.
- Type the User name and Password then press Enter or click **OK** to log on.
- Type your 3-character (alphanumeric) Operator ID.
- Press Enter or click  .
- Wait for the Reagents window to appear.
- Verify the background behind the lightening bolt is green.

  Note: Analyzer and Workstation should begin communicating within 30 seconds. In the upper right corner, the right circle should also be green.
- If the Automatic Startup function is enabled, Startup automatically activates.
  - ▸ Monitor the *Cycle in progress : Startup* status bar, as desired.
  - ▸ Hard copy of Startup results prints automatically if Auto-Print function is enabled.
  - ▸ Go to step 4 to review results.

3 Prepare the System for Processing Samples

IMPORTANT Risk of inaccurate results if the Analyzer is not properly prepared. Based on the scenarios below, perform either a Startup or Mini Prime to prepare the Analyzer. It is not necessary to do both.

- If the system was Shutdown before the power was turned off, do a Startup.
- If the Analyzer has set idle (not cycled) more than 4 hours, do a Startup.
- If the Analyzer was **not** Shutdown and has set idle (not cycled) less than 4 hours, do a Mini Prime.
- If the Analyzer was powered off then back on again, do a Mini Prime.

Startup

- Click  to initiate a Startup routine.
  - Monitor the *Cycle in progress : Startup* status bar, as desired.
  - Hard copy of Startup results prints automatically if Auto-Print function is enabled.
- Go to step 4 to review results.

Mini Prime

- At the Menu bar, select **Cycles** ▸▸ **Mini Prime**.
- *Cycle in progress : Mini Prime* status bar provides a visual display of how close the routine is to completion.
- When complete, resume normal operation.

4 Review Results

- Once the Startup routine is complete, verify *Passed* is displayed for each parameter.
  - If *Failed* appears for any parameter, click **Startup Log** tab to evaluate numeric results.

    Note: If the **Add Comments** box automatically appears, click  to exit.
  - Click  to initiate another Startup routine.
  - If the Startup continues to fail, contact your Beckman Coulter Representative.
- Save individual or log printout if required by your accreditation agency.

Add Comments

- If the **Add Comments** box appeared automatically, type in your personal observation or appraisal as desired.
- To access the **Add Comments** box, click **Startup Log** tab ▸▸ **Add Comments** button then type your personal observation or appraisal inside the box.
- Click  to save your comments.

5 Perform Quality Control Checks

- Perform quality control checks before running patient specimens according to your laboratory's protocol.

➤ ➤

PREVENTIVE MAINTENANCE

- Proper maintenance of your A^{C}•T 5diff CP hematology analyzer is the best way to ensure proper performance
- Failure to properly execute the maintenance procedures may compromise instrument performance

Maintenance Schedules

- Maintenance procedures you perform are based either on a time schedule or on an as needed basis
  - Startup and Shutdown are daily maintenance procedures
  - Calibration is as needed or when required by your laboratory or regulatory agency
  - Replacing reagents or the rinse drain filter are as needed procedures
  - Extended cleaning is an as needed procedure
- Maintenance performed by your Beckman Service Representative is based on the number of cycles; therefore, the frequency a maintenance procedure needs to be done is driven by your laboratory's workload

Maintenance Log

- Click on **Analyzer / Logs** tab ▸▸ **Maintenance Log** tab to view the log
- Maintains the most recent 100 entries displaying the most recent log entry at the top of the log and the oldest entry at the bottom of the log
- Rollover is first in, first out when the 101st entry is added
- Some maintenance procedures, such as the Extended Cleaning procedure, automatically add entries to the Maintenance Log
- If **Prompt User For Comments** check box is checked (✓), user is prompted to enter a comment
- Log contains the following information fields

  Note: It is necessary to scroll right to see some of these fields.

Information Fields

Field	Function
Date/Time	Displays the time and date the log entry was made, which should correlate with when the maintenance procedure was completed
Opr	Displays the User name logon information in the Workstation when the maintenance was completed

Note: To access the New Entry Log window cited below, you must click 

Action	Displays the maintenance that was performed (entry made by the operator on the New Log Entry window)
Duration	Displays the time it required the operator to complete the maintenance (entry made by the operator on the New Log Entry window)
Comment	Displays any comments entered by the operator performing the maintenance (entry made by the operator on the New Log Entry window)

- Optional entry that provides an opportunity for the operator to type a personal observation or appraisal
- Some maintenance procedures, such as the Extended Cleaning procedure, add entries automatically to the Maintenance Log
- Operator may also use the **Add Comments** button to access the Add Comments box, as desired
- If the **Prompt User For Comments** check box is checked (✓), the Add Comments box pops up automatically to prompt operator to enter a comment

  Note: When Add Comments box is displayed, operator cannot access any other window until they either:

  - Enter a comment then click  to save and exit

    or

  - Click  to exit the box without entering a comment

SERVICE LOG

- Function is similar to that of the Maintenance Log but is used by your Beckman Coulter Service Representative to maintain a record of the service performed on A^{C}•T 5diff CP hematology analyzer in your laboratory
- Must use the Service Password to access

OVERVIEW

What are Summary Pages?

Each summary is an abbreviated reference of essential operating instructions detailed in the Instructions For Use manual. These abbreviated instructions are referred to as either summary pages or operational summaries.

Purpose

Each summary page is an individual, stand-alone document that is designed for use in your laboratory as a training aid and/or as a reference tool for a trained operator.

This chapter contains a master copy of each summary page. These pages may be copied for use during training or as quick reference guides for trained operators while operating the instrument.

Design

These summary pages are designed for ease of use by a newly trained operator as well as an experienced operator. As you examine these summary pages, notice that each summary consists of two levels of detail. The first level states the actual task (usually in bold) and the second level contains abbreviated instructions (usually in a bulleted sequence) that an operator may need to complete that task.

A novice operator is likely to need all this information, but as an operator's skills develop, the bulleted instructions will be needed less and less because the operator already knows how to complete the stated task. An experienced operator is likely to use a summary page more as a quick reference to make sure all required steps are completed in the proper sequence.

An experienced operator may also decide to use the Daily Operations Quick Reference to guide them through familiar daily tasks.

Presentation in This Training Guide

Since these summary pages will be helpful in training other operators in your laboratory and since they are designed to be used as reference tools after training is complete, it is important for you, as the Key Operator, to become proficient in the use of this tool during this training. To eliminate the need to flip back and forth between the topic and the summary masters, the appropriate operational summaries are inserted within each topic. When an operational summary is inserted in the training material, a rotated chevron (➤) appears to the left of the summary page title. For example, at the point of insertion, the Startup Summary would begin with

➤ Startup Summary

Since this is a stand-alone summary for use after training is complete, some of the instructions discussed in the training topic may be repeated in the summary. Two rotated chevrons (➤ ➤) mark the end of the inserted summary.

CONTENTS

Since these summary pages are designed to be used as stand-alone documents, consecutive page numbering is not used on these pages.

Most operational summaries consist of either a single-side or a double-sided page. However, to accommodate the few summaries that are three or four pages long, the footer for each summary shows the total number of pages —for example, 2 of 3 appears on page 2 of a 3-page summary and 1 of 1 appears on a single page summary.

To assist you in locating a desired summary (since the page numbering is not sequential), the summary pages are arranged in the following order.

STARTUP SUMMARY

Before starting this summary, verify both Workstation and Analyzer power are ON. If the system is powered OFF, do not use this summary. Use the System Power Up Summary instead.

1 Preliminary Checks

- If the Workstation screen is blank, power on the monitor.
- Verify the green circle is displayed in the upper right corner.
- Check the waste container level.
  - If the container needs to be replaced, change the container using the Replacing a Waste Container Summary as a guide.
- Make sure the Printer is ready to print.
  - If the paper supply is insufficient, add paper.
  - Make sure the power is ON and ready to print.

2 Perform the Startup

- Click  to initiate a Startup routine.
- Monitor the *Cycle in progress : Startup* status bar, as desired.
- Hard copy of the results prints automatically if the Auto-Print function is enabled.

3 Review Results

- Once the Startup routine is complete, verify *Passed* is displayed for each parameter.
  - If *Failed* appears for any parameter, click **Startup Log** tab to evaluate numeric results.

    Note: If the **Add Comments** box automatically appears, click  to exit.
  - Click  to initiate another Startup routine.
  - If the Startup continues to fail, contact your Beckman Coulter Representative.
- Save individual or log printout if required by your accreditation agency.
- Add comments as desired.
  - If the **Add Comments** box appeared automatically, type in your personal observation or appraisal.
  - To access the **Add Comments** box, click **Startup Log** tab ▸▸ **Add Comments** button then type your personal observation or appraisal inside the box.
  - Click  to save your comments.

4 Perform Quality Control Checks

- Perform quality control checks before running patient specimens according to your laboratory's protocol.

RUNNING COULTER® A^{C}•T™ 5diff CONTROL PLUS SUMMARY

1 Get the Workstation Ready

To run controls with the Control window open

- Click the **Quality Assurance** tab.
- From the Select Control drop-down menu, locate the control file that correlates with the lot number of control you wish to run.

To run controls with the Run window open

- Click the **Run** tab.

  Note: To use this option, the control lot number must be reserved. If you are not familiar with this protocol, use the Control window to run the controls.
- Enter the control lot number as the next Sample ID.

2 Prepare the Control according to the Package Insert

- Warm controls at room temperature for 15 minutes.
- Control levels may be processed in any order.
- Mix the control vial using the 8 x 8 x 8 method.
  - Do not use a mechanical mixer!
  - Inspect the control vial to ensure all cells are uniformly distributed, if not, repeat the 8 x 8 x 8 method of mixing again.

3 Run the Control

- At the tube holder, make sure sample position is in the pierce position (12 o'clock). (In tube holder #1, use sample position 1; in tube holder #2, use sample position 4.)
- Place the well-mixed, closed-vial of control in sample position 1.
- Close the cap-pierce door.

4 Review Results

Note: A hard copy of the results prints automatically if the Auto-Print function is enabled.

- Verify control results are within the acceptable ranges.
- If a result is out-of-limits:
  - Rerun the control after gently remixing the control vial.

    or
  - Refer to When CBC/DIFF Control is Outside Its Expected Ranges that follows.
- Compare the results of this run with previous runs, as desired
  - To evaluate stored numerical data or evaluate the graphs for the presence of shifts or trends, the Control window must be displayed.
  - Click **Quality Assurance** tab to display the Control window, if needed.

When CBC/DIFF Control is Outside Its Expected Ranges

1. Check for a control problem.
   - Ensure the control material was properly mixed. If not, mix it according to the package insert.
   - If the control was processed in the Control window, make sure the control file with the correct lot number was selected.
   - If the control was processed in the Run window, make sure the lot number was entered correctly and verify the lot number is reserved.
   - Unless you have established your own running mean and/or expected ranges, make sure the setup information (assigned values and expected ranges) matches the control package insert. If they do not, change the control setup information to match the package insert.
   - If any of the above problems exist, rerun the control; otherwise, proceed to the next check.
2. Rerun the control to determine if the out-of-limit result is a statistical outlier. If the rerun reflects the same parameter(s) as still being out-of-control, proceed to the next check.
3. Ensure the control material was not contaminated by running another vial or level of control. Follow the instructions on the package insert for proper handling. If the results are still out, proceed to the next step.
4. Perform the Extended Cleaning procedure in the Instructions For Use manual, Chapter 11.

   Use the Online Help to locate this procedure:

   a. Click  .

   b. Click on **Contents**. The table of contents appears in the left frame.

   c. Inside the left frame, click on **11 Diagnostics** to list the headings in that chapter.

   d. Scroll through the headings and click on **Extended Cleaning**. The cleaning procedure appears inside the right frame.

   e. Complete the procedure.
      - Follow the instructions on the Workstation screen.

        or
      - Print a copy of the instructions.
        - Right click on the procedure (anywhere inside the right frame).
        - Click **Print** on the pop-up menu.
        - Click **OK** to initiate printing.

        Note: All the information under the primary heading is printed. Locate the Extended Cleaning instructions and complete the procedure as written.

   f. Rerun the control.
      - If the results are acceptable, run other control levels that need to be processed then resume normal operation.
      - If the results are still out, call your Beckman Coulter Representative to help you troubleshoot.

PN 4277205BA

RUNNING PATIENT SAMPLES USING THE WORKLIST SUMMARY

Note: Use this summary only if you want to use the Worklist. If you do not want to run samples using the Worklist, use the Running Patient Samples Without Using the Worklist Summary.

1 Get the Workstation Ready

- Click **Results** tab to verify only the active archive is open (background is white).
  - If the background is green, an old archive is open and must be closed.
  - At the Menu bar, select **File ▸▸ Close archive** to close the old archive.
- If, according to your laboratory protocol, it is time to create a new archive, select **File ▸▸ New archive**.
- Click **Worklist** tab.

If the Worklist is Set Up Manually

Autonumbering OFF

- Click  .

Inside the Add/Edit Worklist box:

- Enter sample ID:
  - Click on the Sample ID field.
  - Manually type the ID number at the keyboard.
    or
    Scan the specimen tube's bar-code label with the bar-code wand.
  - Verify the ID number inside the Sample ID field is correct.
- CBC/DIFF is the default panel selection; or select CBC from the drop-down box.
- Flagging set currently selected as default is entered; select another flagging set from the drop-down box, if desired.
- Enter comment or other patient demographic information as desired. (If enter gender and/or age and a flagging set is not manually selected, the flagging set automatically changes accordingly.)
- Click  to save entries and clear the box for entering the identifiers and demographics for another specimen.
- When the last entry to the Worklist is complete, click  to save and exit.
- Click **Run** tab.

Autonumbering ON

- Click  .

Inside the Add/Edit Worklist box:

- Sample ID entry is automatic.
  To override the autonumber with another sample ID:
  - Highlight the autonumber in the Sample ID field.
  - Manually type the desired sample ID number at the keyboard.
  - Verify the ID number inside the Sample ID field is correct.
    Note: Autonumbering automatically begins again with the autonumber that was overwritten.
- CBC/DIFF is the default panel selection; or select CBC from the drop-down box.
- Flagging set currently selected as default is entered; select another flagging set from the drop-down box, if desired.
- Enter comment or other patient demographic information as desired.(If enter gender and/or age and a flagging set is not manually selected, the flagging set automatically changes accordingly.)
- Click  to save entries and clear the box for entering the identifiers and demographics for another specimen.
- When the last entry to the Worklist is complete, click  to save and exit.
- Click **Run** tab.

If the Worklist is Downloaded from a Host Computer

- Sample ID and patient demographics are automatically downloaded from the host computer. (Information downloaded from the host computer cannot be edited.)
- Click **Run** tab.

2 Process the Sample

- Check the Sample ID Next field for the next specimen to be processed.

  Note: If you want to run another specimen, manually type the sample ID or scan the bar-code label on the specimen tube. The system automatically matches the desired sample ID with the panel and demographic information previously entered on the Worklist.
- Verify the tube holder is appropriate for the specimen tube being analyzed. If not, change the tube holder.
- Mix the specimen gently but thoroughly according to your laboratory's protocol.
- If the specimen tube does not have a pierceable stopper, remove the stopper.
- Place the well-mixed specimen tube in the sample position that best matches the tube. Rotate the specimen tube to the pierce position (12 o'clock position), as needed.
- Close the cap-pierce door to initiate the cycle.
- Remove the specimen tube when the cap-pierce door automatically opens after aspiration is complete. Notice the red LED is still glowing, the Analyzer is busy processing the sample.
- Overlap sample processing, if desired.
  - Repeat step 2 to get ready for the next cycle.
  - When the cycle is complete (green LED glows steady), go to step 3.

3 When the Cycle is Complete

- Verify the sample results appear in the Run window.
- Verify the sample ID entry and results before reporting.
- Click  for a hard copy of the results.

  Note: A hard copy prints automatically if the Auto-Print function is enabled.
- Repeat steps 2 and 3 until all specimens are analyzed.

RUNNING PATIENT SAMPLES WITHOUT USING THE WORKLIST SUMMARY

Note: Use this summary only if you do not want to use the Worklist. If you want to run samples using the Worklist, use the Running Patient Samples Using the Worklist Summary.

1 Get the Workstation Ready

- Click **Results** tab to verify only the active archive is open (background is white).
  - If the background is green, an old archive is open and must be closed.
  - At the Menu bar, select **File ▸▸ Close archive** to close the old archive.
- If, according to your laboratory protocol, it is time to create a new archive, select **File ▸▸ New archive**.
- Click **Run** tab.

Autonumbering OFF

- Enter Next Sample ID:
  - Click on the Sample ID Next field.
  - Manually type the ID number at the keyboard.
    or
    Scan the specimen tube's bar-code label with the bar-code wand.
  - Verify the ID number inside the Sample ID Next field is correct.
- CBC/DIFF is the default panel selection; or select CBC from the drop-down box.
- Enter a Patient ID if desired.

Autonumbering ON

- Next Sample ID entry is automatic.

  To override the autonumber with another sample ID:
  - Highlight the autonumber in the Sample ID Next field.
  - Manually type the desired sample ID number at the keyboard.
  - Verify the ID number inside the Sample ID Next field is correct.

    Note: Autonumbering automatically begins again with the autonumber that was overwritten.
- CBC/DIFF is the default panel selection; or select CBC from the drop-down box.
- Enter a Patient ID if desired.

2 Process the Sample

- Verify the tube holder is appropriate for the specimen tube being analyzed. If not, change the tube holder.
- Mix the specimen gently but thoroughly according to your laboratory's protocol.
- If the specimen tube does not have a pierceable stopper, remove the stopper.
- Place the well-mixed specimen tube in the sample position that best matches the tube. Rotate the specimen tube to the pierce position (12 o'clock position), as needed.
- Close the cap-pierce door to initiate the cycle.
- Remove the specimen tube when the cap-pierce door automatically opens after aspiration is complete. Notice the red LED is still glowing, the Analyzer is busy processing the sample.
- Overlap sample processing, if desired.
  - Go to step 3 to get ready for the next cycle.
  - When the current cycle is complete, go to step 4.

3 Prepare for the Next Cycle

- Enter information for the next specimen while the current sample is being analyzed.

Autonumbering OFF

- Enter Next Sample ID:
  - Click on the Sample ID Next field.
  - Manually type the ID number at the keyboard.
    or
    Scan the specimen tube's bar-code label with the bar-code wand.
  - Verify the ID number inside the Sample ID Next field is correct.
- CBC/DIFF is the default panel selection; select CBC from the drop-down box, if desired.
- Enter a Patient ID if desired.
- If you are overlapping, go to step 2.

  If you are not overlapping, when the cycle is complete (green LED glows steady), go to step 4.

Autonumbering ON

- Next Sample ID entry is automatic.

  To override the autonumber with another sample ID:
  - Highlight the autonumber in the Sample ID Next field.
  - Manually type the desired sample ID number at the keyboard.
  - Verify the ID number inside the Sample ID Next field is correct.

    Note: Autonumbering automatically begins again with the autonumber that was overwritten.
- CBC/DIFF is the default panel selection; select CBC from the drop-down box, if desired.
- Enter a Patient ID if desired.
- If you are overlapping, go to step 2.

  If you are not overlapping, when the cycle is complete (green LED glows steady), go to step 4.

4 When the Current Cycle is Complete

- Verify the current sample results appear in the Run window.
- Verify the sample ID entry and results before reporting.
- Click  for a hard copy of the results.

  Note: A hard copy prints automatically if the Auto-Print function is enabled.
- Repeat steps 2, 3, and 4 until all specimens are analyzed.

PN 4277205BA

Verify the Desired Archive is Being Used

1. Click the **Results** tab to display the list of sample results.
2. Is this the archive you wish to use?
   - If you wish to use the current, active archive:
     - A white background indicates you are viewing the current, active archive.
     - A light green background indicates you are viewing a previously closed archive that has been opened. At the Menu bar, select **File ▸▸ Close archive**.
   - If you wish to use a previously closed archive:

     Note: Do not analyze patient samples when a previously closed archive is open.
     - A white background indicates you are viewing the current, active archive. At the Menu bar, select **File ▸▸ Open archive** then select the date for the archive you wish to use. When the archive opens, a light green background indicates you are viewing a previously closed archive.
     - A light green background indicates you are viewing a previously closed archive that has been opened. If you are not sure that you are viewing the archive you wish to use, at the Menu bar:
       - Select **File ▸▸ Close archive** to close this open archive.
       - Select **File ▸▸ Open archive**, the table of closed archives appears.
       - Select the date for the archive you wish to use.
3. You may choose to sort, view, print, transmit, or delete selected or all results.
4. If you open an archive, make sure you close the archive when your work is completed.

To Sort the Results List

At the Results window, you can sort information in ascending or descending order using any column except a parameter result or comment column. In other words, you can sort by Sample ID but not by an analysis parameter, such as WBC, RBC, Hgb, Plt, and so forth.

1. Click the title of the column you want sorted. For example, if you wish to sort by sample ID, click the Sample ID column title.
   - >> appearing next to the title indicates an ascending (low to high) sort order.
   - << appearing next to the title indicates a descending (high to low) sort order.
   - Repeating sequence for sort functions:
     - Click once, sort ascending with a >> symbol in the column header
     - Click again, sort descending with a << symbol in the column header
     - Click again, defaults back to original order
     - Click again, start again with sort ascending
   - Sample IDs sort alphanumerically by character. For example, 10 will appear before 4 because it is being sorted by the “1”.
   - Numbers appear in a sorted list before letters. For example, Sample ID 482 will appear before Sample ID N482 (unless sorted in descending order).

To Search for a Specific Sample

1. Scroll through the list of results to locate the sample ID.

   or

2. Use the search feature:
   - Click the **Search Results** tab at the bottom of the Results window.
   - Click **Current Archive** or **Closed Archive**.
   - Click your choice of search criteria: **Sample ID**, **Patient ID**, or **Patient Name**.
   - Type the chosen identifier.
   - Click the Search button.

To View Sample Results

1. To view numeric results, scroll right.
   - You may choose to print or transmit the numeric sample results.
2. To view detailed results (results and histograms in the Run window format):
   - Double-click the desired result.

     or
   - Click  to view the selected results.

     Note: Click  to review the previous result or  to review the next result, as desired.
   - You may choose to print or transmit the detailed sample results.
   - Click  to return to the Results List.

PN 4277205BA

To Output Sample Data: Print and Reprint / Transmit and Retransmit

To Print a Single Sample Data Set

1 At the selected set of sample results, click  to display the print menu.

2 Select the desired print format:

- **Print Summary List For Selected Rows** (prints numeric results only)

  or

- **Print Patient Report For Selected Rows** (prints Run view of results and histograms)

ATTENTION: Beckman Coulter recommends that you do not perform simultaneous batch printing and batch transmission while running the system with the Auto-Print and Auto-Transmit functions enabled. Performing all these activities with a large active archive may affect system processing.

To Batch Print a Group of Samples

1 Display the list of sample results.

2 While pressing the Ctrl key, click on each sample you wish to print. A black dot appears in the far left column and the sample row is highlighted to indicate the sample is selected.

3 Click  to display the print menu.

4 Select the desired print format:

- **Print Summary List For Selected Rows** (prints numeric results only)

  or

- **Print Patient Report For Selected Rows** (prints Run view of results and histograms)

ATTENTION: Beckman Coulter recommends that you do not perform simultaneous batch printing and batch transmission while running the system with the Auto-Print and Auto-Transmit functions enabled. Performing all these activities with a large active archive may affect system processing.

To Batch Transmit a Group of Samples

Note: The transmit option also allows an operator to transmit the last result and all results.

1 Click the **Results** tab if you have not already done so.

2 While pressing the Ctrl key, click on each sample you wish to transmit. A black dot appears in the far left column and the sample row is highlighted to indicate the sample is selected.

3 Click  to display the transmit menu.

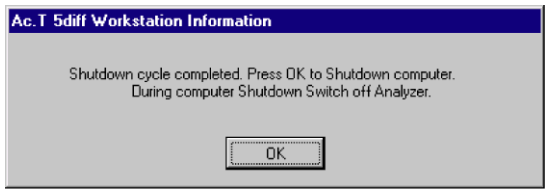
4 Select **The selected results** from the transmit menu.

1 Perform Instrument Shutdown

- Click  to initiate a Shutdown routine.
- Monitor the *Cycle in progress : Shutdown* status bar, as desired.

2 Shut Down Computer then Power Off Analyzer and PC

When Shutdown is complete, a message box appears:



- Click **OK** to shut down the computer.
- Turn off the Analyzer power switch.
- Wait for the *"It is now safe to turn off computer"* message to appear.
- Turn off the PC power switch.

Notes:

- Allow A^{C}•T 5diff Rinse reagent to remain in the instrument a minimum of 30 minutes.
- Wait at least 30 seconds before performing the Power Up and Log On procedures.
- After doing Shutdown, you must do a Power Up and Startup before running patient specimens or controls.

SYSTEM POWER DOWN SUMMARY

1 At the Workstation

- Click  .
- Select **Quit Application**.
- Press Enter or click  .
- Wait while the Workstation closes its program.
- When the Begin Logon box appears, press Ctrl + Alt + Delete simultaneously.
- Click **Shut Down** at the Logon Information box.
- Select **Shutdown** then click **OK** at the Shutdown Computer box.
- When the *It is now safe to turn off your computer* message appears, power the Workstation computer off.

2 At the Analyzer

- Toggle the Power On/Off rocker switch OFF (position O). This rocker switch is located at the base of the left side panel.
- If a replacement or maintenance procedure that requires you to open the Analyzer panels is being performed, unplug the ac power cord. Either remove the cord from the instrument (at the back panel, in the lower right corner) or from the ac wall outlet.

SYSTEM POWER UP SUMMARY

1 Preliminary Checks

- Check the waste container level.
  - If the container needs to be replaced, change the container using the Replacing a Waste Container Summary as a guide.
- If the Automatic Startup and Auto-Print functions are enabled, make sure the Printer is ready to print.
  - If the paper supply is insufficient, add paper.
  - Make sure the power is ON and ready to print.
  - Verify the printer is properly configured according to your laboratory's protocol.

2 Power Up the System

At the Analyzer

- If the ac power cord is unplugged, plug the cord into the instrument (at the back panel, in the lower right corner) or the ac wall outlet, as applicable.
- Toggle the Power ON/OFF rocker switch ON (position –). This rocker switch is located at the base of the left side panel.
- Verify the red LED is glowing steady.

At the Workstation

- Power ON the Workstation computer.
- Wait while the computer performs its internal checks.
- When the Begin Logon box appears, press Ctrl + Alt + Delete simultaneously.
- Type the User name and Password then press Enter or click **OK** to log on.
- Type your 3-character (alphanumeric) Operator ID.
- Press Enter or click  .
- Wait for the Reagents window to appear.
- Verify the background behind the lightening bolt is green.

  Note: Analyzer and Workstation should begin communicating within 30 seconds. In the upper right corner, the right circle should also be green.
- If the Automatic Startup function is enabled, Startup automatically activates.
  - Monitor the *Cycle in progress : Startup* status bar, as desired.
  - Hard copy of Startup results prints automatically if Auto-Print function is enabled.
  - Go to step 4 to review results.

3 Prepare the System for Processing Samples

IMPORTANT Risk of inaccurate results if the Analyzer is not properly prepared. Based on the scenarios below, perform either a Startup or Mini Prime to prepare the Analyzer. It is not necessary to do both.

- If the system was Shutdown before the power was turned off, do a Startup.
- If the Analyzer has set idle (not cycled) more than 4 hours, do a Startup.
- If the Analyzer was **not** Shutdown and has set idle (not cycled) less than 4 hours, do a Mini Prime.
- If the Analyzer was powered off then back on again, do a Mini Prime.

Startup

- Click  to initiate a Startup routine.
  - Monitor the *Cycle in progress : Startup* status bar, as desired.
  - Hard copy of Startup results prints automatically if Auto-Print function is enabled.
- Go to step 4 to review results.

Mini Prime

- At the Menu bar, select **Cycles** ▸▸ **Mini Prime**.
- *Cycle in progress : Mini Prime* status bar provides a visual display of how close the routine is to completion.
- When complete, resume normal operation.

4 Review Results

- Once the Startup routine is complete, verify *Passed* is displayed for each parameter.
  - If *Failed* appears for any parameter, click **Startup Log** tab to evaluate numeric results.

    Note: If the **Add Comments** box automatically appears, click  to exit.
  - Click  to initiate another Startup routine.
  - If the Startup continues to fail, contact your Beckman Coulter Representative.
- Save individual or log printout if required by your accreditation agency.

Add Comments

- If the **Add Comments** box appeared automatically, type in your personal observation or appraisal as desired.
- To access the **Add Comments** box, click **Startup Log** tab ▸▸ **Add Comments** button then type your personal observation or appraisal inside the box.
- Click  to save your comments.

5 Perform Quality Control Checks

- Perform quality control checks before running patient specimens according to your laboratory's protocol.

PN 4277205BA

MAKING CHANGES TO A CONTROL FILE CURRENTLY IN USE SUMMARY

Use this summary only if you need to make a change in a control file that is currently in use and only if you are changing an individual item:

- To correct a typographical error.

  or

- To edit an assigned value (obtained from the Table of Expected Results) to a running mean (established by the laboratory).

If you need to make a set up change (such as changing the lot number), use the Control File Management Summary.

1 Select the File

- Click the **Quality Assurance** tab ▸▸ **Control** tab to display the Control window.
- Choose the existing file you want to change from the Select Control drop-down menu.

2 Access the Setup Control Window

- Click the **Setup Control** button.

3 Make the Changes

- Enter the new information.

4 Verify All Changes

- Check all entries are correct.
- Click  .
  - If control runs are stored in this file:
    - When the *Do You Want The Existing Results To Be Saved?* message appears, click on the desired action.
    - Selected action makes the requested changes then exits the window.
  - If no control runs are stored in this file, requested changes are saved before exiting the window.
- Control file statistics are recalculated based on the changes.

Review the Numeric Control Data and Graphs, if applicable

1 Locate the Control File

- Click the **Quality Assurance** tab to display the Control window.
- Choose the specific file you want to review from the Select Control drop-down menu.

2 Review the Graphs

- Double-click on any thumbnail graph you wish to review.
- Look for any skewed or out-of-limit plot points .
- Check for a shift or a trend.

3 Review Numeric Data

- Review % CV of all parameters (press → to see additional parameters).
- If a CV is high, scroll through the rows and examine each run for results mistakenly run into the file (the graph review should correlate).
- May review and manage the data by shifts, if desired.
- Exclude any mistakes from the control calculations.
  - Locate the control that needs to be excluded from the calculations.
  - Click the ☑ to the left of that control (inside the Include column).
  - ☐ (an empty check box) indicates those control results are no longer being included in the control calculations. N = is decreased by 1.
  - Repeat as needed.

Print the Control File Data

- Click  .
- **Print All Rows** is preselected but if a hard copy of the graphs is also desired select the **Print All Rows and L J Graphs** option.
- If you are submitting data for IQAP evaluation, print a second copy.

Delete the Control File Data

- Click  .
- Choose **Erase All row** from the drop-down menu.

  Note: All the control results are deleted. Target values and +/- limits remain in the control file and may not need to be entered if the same type and level of control is set up again in that same file.

Set Up the New Lot Number of Commercial Control

IMPORTANT Risk of misleading control results. Old control runs remaining in a newly set up control file are included in the statistical data for the new file. Delete all controls runs before starting to run the new control.

Note: Before starting this set up, obtain the control disk for the controls you plan to set up.

1 Select the File

- Click the **Quality Assurance** tab to display the Control window.

  Note: If the Control window is not displayed, click the **Controls** tab at the bottom.
- Choose a CBC/DIFF file from the Select Control drop-down menu.

2 Access the Setup Control Window

- Click the **Setup Control** button.

3 Download the Control Data from the Disk

- Insert the control disk into the floppy drive.
- Select the **Download Values** button.
- Select the control level then click  .
- Select Commercial as the source of control material (if it is not already displayed).
- Print if desired then click  .

4 Reserve the Control Lot Number

- Click **Setup ▸▸ Quality Assurance**.
- Type the password then click  .
- Click the **General** tab, if necessary.
- Click the box in the Reserved column that corresponds to the desired control lot.
- Click  .

5 Set Up Another Level of Control, if applicable

IMPORTANT Risk of misleading control results. Old control runs remaining in a newly set up control file are included in the statistical data for the new file. Delete all controls runs before starting to run the new control.

- Review, print, and delete other files, as applicable.
- Choose another empty, inactive file from the Select Control drop-down menu.
- Repeat steps 2, 3, and 4.

PN 4277205BA

Note: If **all** reagents are being changed, see Chapter 11 of the Instructions For Use manual.

1 Verify a New Reagent Container is Needed

- Display the Reagents window by choosing **Analyzer/Logs** tab ▸▸ **Reagents** tab.
- Check reagent levels to determine which reagent triggered the *REAGENT LOW LEVEL* message.
  - Check all levels because more than one may be low.
  - Obtain a new unopened container of each reagent that according to your laboratory's protocol requires the reagent to be changed.

2 Replace the Reagent Container

- Open the new reagent container.
- Remove the stopper assembly from the old container then transfer it directly to the new container and tighten to ensure an adequate seal.

  Note: If the lower part of the assembly touches you or anything outside the container, flood that lower part of the assembly with distilled water then wipe it with a lint-free tissue.
- Put the cap from the new container onto the empty container.
- Properly dispose of the empty bottle according to your laboratory's protocol.

3 Record the New Reagent Information

- At the Reagents window, click **Change Reagent**.
- Choose the reagent that was changed from the drop-down menu.
- Type the new lot number.

  Note: Today's date automatically appears in the Opened Date box.
- Choose the Expiration Date (or open container stability) from the drop-down calendar.
  - For all reagents except A^{C}•T™ 5diff Hgb Lyse reagent, select the date printed on the reagent container.
  - For the A^{C}•T 5diff Hgb Lyse reagent which has a 90 day open container stability, select the date that is 90 days from today.
- Click  to save the information and return to the Reagents window.

4 Wait for the Reagent Lines to Prime

- System automatically primes the reagent and updates the level indicator.

  Note: Reagent level may not be displayed at 100% due to this priming.
- Once priming is complete, continue normal operation.

REPLACING A WASTE CONTAINER SUMMARY

Note: If the waste container is not full and the waste alarm is chirping (beeping) at regular intervals, replace the 9 Volt alkaline battery in the waste sensor alarm.

1 Power Off the System

At the Workstation

- Click  .
- Select **Quit Application**.
- Press Enter or click  .
- Wait while the Workstation closes its program.
- When the Begin Logon box appears, press Ctrl + Alt + Delete simultaneously.
- Click **Shut Down** at the Logon Information box.
- Select **Shutdown** then click **OK** at the Shutdown Computer box.
- When the *It is now safe to turn off your computer* message appears, power the Workstation computer off.

At the Analyzer

- Toggle the Power On/Off rocker switch OFF (position O). This rocker switch is located at the base of the left side panel.

2 Replace the Waste Container

- Clearly mark or apply a waste label to an empty 20 L container.
- Using biohazardous precautions:
  - Carefully remove the cap (with waste sensor attached) from the full waste container.
  - Transfer the waste assembly directly to the empty waste container and tighten.

3 Neutralize and Disinfect the Waste Before Capping the Container

- For 20 liters of waste liquid, add the following to the waste container:
  - 250 mL of sodium hypochlorite solution (if 12% available chlorine) **or** 500 mL of sodium hypochlorite solution (if 6% available chlorine) to disinfect waste.
  - 50 mL of 200 g/L sodium hydroxide solution to prevent gas from forming if the container is being capped.
- Dispose of the biohazardous waste according to your laboratory's protocol.

4 Power Up the System

At the Analyzer

- Toggle the Power ON/OFF rocker switch ON (position –). This rocker switch is located at the base of the left side panel.
- Verify the red LED is glowing steady.

At the Workstation

- Power ON the Workstation computer.
- Wait while the computer performs its internal checks.
- When the Begin Logon box appears, press Ctrl + Alt + Delete simultaneously.
- Type the User name and Password then press Enter or click **OK** to log on.
- Type your 3-character (alphanumeric) Operator ID.
- Press Enter or click  .
- Wait for the Reagents window to appear.
- Verify the background behind the "lightening bolt" is green.

  Note: Analyzer and Workstation should begin communicating within 30 seconds. In the upper right corner, the right circle should also be green.

5 Prepare the System for Processing Samples

- If the Automatic Startup function is enabled:
  - Startup routine was automatically activated when the power was toggled back ON.
  - *Cycle in progress : Startup* status bar provides a visual display of how close the routine is to completion.
  - When complete, resume normal operation.
- If the Automatic Startup function is not enabled:
  - At the Menu bar, select **Cycles ▸▸ Mini Prime**.
  - *Cycle in progress : Mini Prime* status bar provides a visual display of how close the routine is to completion.
  - When complete, resume normal operation.

PN 4277205BA

ICON QUICK REFERENCE


Print


Transmit


Delete


Find


Previous


Next


Add


Edit


Results / List


Validate


Rerun Sample


Load Sample


Startup


Shutdown


Logout or Exit

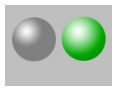

Help


Cancel

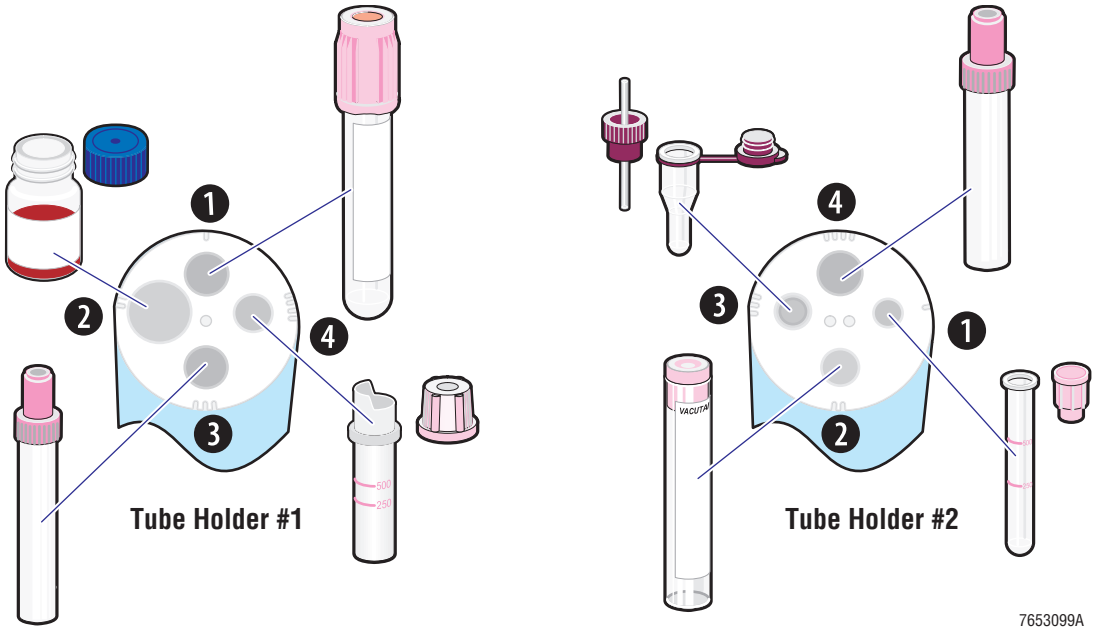

OK


Save


Restore Defaults


System State
(Double-click to quickly access the Error Log)

TUBE HOLDERS QUICK REFERENCE



This is a simplified overview of tube holders and the approved collection devices and control vials they accommodate. For a comprehensive list with specific details, see Appendix D in the Instructions For Use manual.

Tube Holder #1	Types of Collection Devices or Control Vials
Position ❶	✓ Most 13 mm x 75 mm evacuated specimen tubes containing either K_3EDTA or K_2EDTA for collecting whole-blood volumes of 2 to 5 mL ✓ COULTER® A^C•T™ 5diff Control Plus control tubes
Position ❷	✓ COULTER® A^C•T™ 5diff Cal Calibrator vial
Position ❸	✓ Sarstedt Monovette® 11.5 mm x 66 mm specimen tube collecting 2.7 mL of whole-blood
Position ❹	✓ Becton-Dickinson Microtainer for collection of 0.25 to 0.50 mL of whole-blood
Tube Holder #2	
Position ❶	✓ Becton-Dickinson Microtainer for collection of 0.25 to 0.50 mL of whole-blood
Position ❷	✓ Becton-Dickinson 10.25 mm x 64 mm Vacutainer® for collecting 3 mL of whole-blood
Position ❸	✓ RAM Scientific microcollection device for collecting 125 µL of whole-blood
Position ❹	✓ 13 mm x 75 mm specimen tube with mulitiple labels ✓ COULTER® A^C•T™ 5diff Control Plus control tubes

ONLINE HELP SYSTEM QUICK REFERENCE

Accessing Online Help

ATTENTION: In the event that you cannot access the A^{C}•T 5diff Cap Pierce Online Help System, contact your Beckman Coulter Representative.

Using the COULTER® A^{C}•T™ 5diff CP Workstation

- Click  to display the Help screen

Using Your COULTER A^{C}•T 5diff CP Hematology Analyzer Operator Manuals CD-ROM

ATTENTION: Do not use this CD-ROM on your A^{C}•T 5diff CP Workstation.

1. Turn on your personal or office desktop PC and allow it to boot up.
   Refer to the PC manual for instructions, as needed.
2. Insert the CD-ROM into the CD-ROM drive.
   - If Autorun is enabled on your PC, the PC automatically launches the program.
   - If Autorun is disabled on your PC, do steps 3 through 4.
3. Locate the **Start.HTM** file (*CD-ROM drive letter*:\Start.HTM):
   - Click the Windows **Start** button.
   - Click **Run**.
   - Click **Browse** and locate the CD-ROM drive.
   - Double-click the CD-ROM drive letter (usually **D** or **E**).
4. Double-click **Start.HTM** to launch the program.
5. Click **For Viewing** (or **For Printing**) as desired.

Moving or Resizing the Help Screen

1. Access online Help.
2. To move the Help window:
   - Click the title bar and hold down the left mouse button.
   - Move the mouse to drag the window to a new location.
   - Release the mouse button when the window is located as desired.
3. To resize the Help window:
   - Move the cursor over the window border until the arrows appear.
   - Click the left mouse button and drag the border to resize the window.
   - Release the mouse button when the window is the desired size.

Opening / Closing the Topics Pane

1. Access online Help.
2. Click the arrows to close the topics pane.
3. Click **Contents**, **Index**, **Illustrations**, **Tables**, or **Search** to re-open the topics pane.

Viewing Help Topics

1. Access online Help.
2. Navigate through Help as needed:
   - Click **Contents** to browse through topics by heading.
   - Click **Index** to see a list of index entries.
   - Click **Tables** to see a list of tables.
   - Click **Illustrations** to see a list of illustrations.
   - Click **Search** to search for words or phrases.
   - Click **Home** to return to main Help screen.
3. Move through topics as needed:
   - Click  to display the previous topic in the sequence.
   - Click  to display the next topic in the sequence.
   - Click  to display the previous link.
4. View referenced or related topics:
   - Click the highlighted words in a topic body to link directly to the referenced topic.
   - Click a word in the highlighted hierarchy – at the top of the topic – to change topic levels. The current topic's position also appears in the hierarchy.
5. To view information not visible in the Help window, scroll through the window by using the scroll bar.

Using the Contents Option

1. Access online Help.
2. Click **Contents** to browse through topics by heading.
3. Select a top-level heading to expand and view its sub-topic headings.
4. Select a sub-heading to display information about that topic in the Content pane.
5. Use the scroll bar to scroll through the information.

PN 4277205BA

Using the Index Option

1. Access online Help.
2. Click **Index**.
3. Click the letter corresponding with the list of index entries you want to see.
4. Scroll through the topics as needed.
5. Click a number after the index entry to display information about that topic.

Using the Tables Option

1. Access online Help.
2. Click **Tables** to display the list of tables.
3. Scroll through the tables to locate the table you want to view.
4. Click the table to display it on the screen.

Using the Illustrations Option

1. Access online Help.
2. Click **Illustrations** to display the list of illustrations.
3. Scroll through the illustrations to locate the table you want to view.
4. Click the illustration to display it on the screen.

Using the Search Option

1. Access online Help.
2. Click **Search** to display the search field.
3. Type the word you want to locate.
4. Click **Search**. If the word you typed appears in the document, each instance where it appears will be shown.

Printing Operator Manuals

To Print the Instructions For Use Manual from the Workstation

Note: Prints with the same page breaks and document structure as the printed document that is available by order.

1. At the Menu bar, click **Help ▸▸ Contents ▸▸ Print Manual**.
2. Click **Print**.
3. Define what pages you want to print. You can print all or part of the document.
4. Follow the instructions on the screen.

To Print Operator Manuals from the CD-ROM

Note: Prints with the same page breaks and document structure as the printed document that is available by order.

ATTENTION: Do not use this CD-ROM on your A^{C}•T 5diff CP Workstation.

1. Turn on your personal or office desktop PC and allow it to boot up.
   Refer to the PC manual for instructions, as needed.
2. Insert the CD-ROM into the CD-ROM drive.
   - If Autorun is enabled on your PC, the PC automatically launches the program. (The contents of this CD-ROM are not installed on your PC.)
   - If Autorun is disabled on your PC, do steps 3 through 4.
3. Locate the **Start.HTM** file (*CD-ROM drive letter*:\Start.HTM):
   - Click the Windows **Start** button.
   - Click **Run**.
   - Click **Browse** and locate the CD-ROM drive.
   - Double-click the CD-ROM drive letter (usually **D** or **E**).
4. Double-click **Start.HTM** to launch the Home page.
5. Select a language.
6. Under the name of the manual you wish to print, click **For Printing**.
7. Define what pages you want to print. You can print all or part of the document.
8. Follow the instructions on the screen.

PN 4277205BA

OFFICIAL TRAINING DOCUMENTATION

(for COULTER® A^{C}•T™ 5diff CP Hematology Analyzer Training)

Laboratory _____

Address _____

_____

Phone _____

A^{C}•T 5diff CP Serial Number _____

Key Operator _____

Training Dates _____

Instructions

The overall objectives for COULTER A^{C}•T 5diff CP Hematology Analyzer Key Operator training are listed below. Each objective is a comprehensive statement of what an operator should be able to do when training is complete. Please review these objectives and place a check mark (✓) next to any objectives that both the Trainer and Trainee agree are completed either during this training or through previous knowledge. Write N/A (not applicable) inside the block if the objective does not apply to this Trainee.

Checklist

Your signature at the end of this checklist indicates agreement that the Trainee is able to . . .

_____ Locate important procedures in the Instructions For Use manual including:

- _____ Startup and Shutdown procedures
- _____ Quality Assurance and Calibration procedures
- _____ Sample Analysis procedures
- _____ Reagent replacement procedures
- _____ Troubleshooting procedures

_____ Identify basic instrument components and describe their functions

_____ Correctly perform Startup and Shutdown procedures

_____ Explain the importance of the Startup and Shutdown procedures and the frequency with which they should be performed

_____ Perform appropriate Quality Assurance checks

_____ Review, delete, and set up control files

_____ Prepare IQAP data for submission, if applicable

_____ Perform all preliminary procedures required for calibration

_____ Perform calibration with COULTER® A^{C}•T™ 5diff Cal Calibrator and document the results according to your laboratory's protocol

_____ Perform Sample Analysis using their preferred workflow pattern (with or without Worklist)

_____ Look up samples in the current, active archive

_____ Explain how to open a closed archive

_____ Understand the basics principles of parameter derivation and state if a parameter result is directly measured, determined using A^{C}V technology, derived from a histogram, or computed

_____ Review data and recognize if a DiffPlot is normal or abnormal

_____ Review data and recognize if an RBC and/or Plt histogram is typical or atypical

_____ Explain what a particular flag indicates when provided with an example

_____ Recognize an instrument problem based on abnormal sample results, abnormal Startup, abnormal control results, and/or error messages

Comments

_____

_____

_____

_____

_____

_____

_____

_____ _____

Trainee Signature Trainer Signature

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Documentation
COULTER® AC• T™ 5diff CP Hematology Analyzer Documentation

▪ **Instructions For Use** PN 624021	Use and Function • Operation Principles • Specifications/Characteristics • Precautions/Hazards • Running Samples • Reviewing Results • Calibration • Diagnostics • Instrument Setup • Log Sheets • Manual Calibration • References • Glossary • Abbreviations • Index
▪ **Host Transmission Specification** PN 4277065	Defines requirements for interfacing the system with a host computer.
▪ **Training Guide** PN 4277205	Provides training information for using the CP system.
▪ **Daily Operations Quick Reference** PN 4277315	Provides abbreviated procedures for the experienced operator.

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