

**Document Title: Complete Count of Whole Blood on the SYSMEX XN-9000
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TITLE: Complete Blood Count of Whole Blood on the SYSMEX XN-9000**I. PRINCIPLE**

The Sysmex XN-9000 is an integrated system that incorporates hematology analytical modules as well as automated slidemaker/stainer(s).

The analytical module is a quantitative automated hematology analyzer for in vitro diagnostic use in determining 31 whole blood diagnostic parameters and 7 body fluid diagnostic parameters. Examination of the numerical and/or morphological findings of the complete blood count by the physician are useful in the diagnosis of disease states such as anemias, leukemias, allergic reactions, viral, bacterial, and parasitic infections.

The analyzer performs hematology analysis according to the hydrodynamic focusing (DC Detection), flow cytometry method (semiconductor laser), and SLS-hemoglobin method.

The XN analyzer counts and sizes RBC and PLT using electronic resistance detection. HCT is measured as the ratio of the total RBC volume to whole blood using cumulative pulse height detection. HGB is converted to SLS-hemoglobin, and read photometrically.

WBC count, differential, RET, NRBC and PLT-F are all evaluated using flow cytometry with a semiconductor laser exploiting the differences in cell size, complexity and RNA/DNA content. Forward scattered light provides information on blood cell size and Lateral scattered light provides information on the cell interior such as the size of the nucleus. Lateral fluorescent light intensity increases as the concentration of the stain becomes higher. By measuring the intensity of the fluorescence emitted, information is obtained on the degree of blood cell staining. Fluorescent light is emitted in all directions. The XN detects the fluorescent light that is emitted sideways.

The Sysmex SP-10 is a fully automated hematology slide preparation and staining system. Whole blood specimens are mixed and aspirated and a wedge type blood smear is prepared using hematocrit information from the Sysmex XN to determine optimum smearing criteria. The dried smear is automatically loaded into an individual slide cassette and is then advanced to the staining area. In the staining area, stain and buffer are dispensed into the cassette at operator-defined intervals.

The system also provides a manual mode operation where pre-made smears may be added to be stained. The unit is self-monitoring and alarms when operation is interrupted.

Slides prepared by the Sysmex SP-10 are used for differentiation and morphologic evaluation of cellular elements of whole blood.

II. SCOPE

To be used by personnel at UR Medicine Labs at Strong Memorial Hospital, Hematology-Chemistry Lab.

III. RESPONSIBILITIES

Department and functional responsibilities are defined in the table below:

Group/Person	Responsibility
Quality Assurance	<ul style="list-style-type: none"> Supports the development of this document
Medical Director	<ul style="list-style-type: none"> Ensures that the procedure is followed. Review and approval of this document.
Supervisor	<ul style="list-style-type: none"> Ensures that the procedure is followed. Review and approval of this document.
End User	<ul style="list-style-type: none"> Follows the procedure.

IV. SPECIMENS

A. Required specimen

- Whole blood anti-coagulated with K2 EDTA preferred
- Sodium Citrate may be used when EDTA platelet clumping or platelet satellitism is noted on the EDTA specimen. Use Sodium Citrate results for platelet counts only. Multiply instrument PLT result by 1.1 to correct for anticoagulant dilution.
- Serous and synovial fluids should be collected in EDTA-2K anticoagulant.
- The use of anticoagulant with CSF specimens is neither required nor recommended.

B. Specimens volumes required

- A minimum of 1 mL of whole blood is required for analysis, in auto closed mode.
- Manual analysis - whole blood mode
 - Closed tube-1ml
 - Open tube-300ul
 - Open microtainer-250ul
- Manual analysis –SP-10
- Closed tube smear and staining – 1 mL is optimal, 200µL is aspirated.

C. Unacceptable specimens including those listed below must be redrawn:

- Clotted samples are unacceptable for analysis

Note: Samples with fibrin strands are unacceptable for platelet counts.

2. Grossly hemolyzed samples
3. Samples drawn above an IV line

D. Specimen Handling and Storage

1. Stored at 4-8°C, EDTA blood samples with normal results may be analyzed up to 48 hours without significant loss of differential stability.
2. Sample stability at room temperature is 24 hours. Samples stored at room temperature may exhibit an increase in MCV after 24 hours, which may be minimized by refrigeration.
3. Allow refrigerated specimens ample time (approximately 15 minutes) to come to room temperature, 18°–28°C (64°–82°F), and mix well prior to analysis.

II. SPECIAL SAFETY PRECAUTIONS

- A. All patient specimens should be considered potentially infectious and must be handled with precautions used for human blood, as described in CDC (Center for Disease Control) recommendations and in compliance with the Federal OSHA (Occupational Safety and Health Administration) Blood-borne Pathogen Standard, 29 CFR (Code of Federal Regulations) part 1910.1030. Follow specimen handling as outlined by Laboratory Safety Policy, (SH.CP.AU.gen.0005).
- B. The human blood used in *XN CHECK*, *XN CHECK BF*, *XN CAL* and *XN CAL PF* (Sysmex hematology control material) is non-reactive for Hepatitis B Surface Antigen and negative for antibodies to HIV-1, HIV-2, and Hepatitis C Virus using FDA specified techniques. However, no current tests can assure the absence of these pathogens. All the above mentioned XN materials should be considered potentially infectious and must be handled with precautions used for human blood as described in CDC recommendations and in compliance with the Federal OSHA Blood-borne Pathogen Standard, 29 CFR, 1910.1030.
- C. Clorox Ultra: Avoid acidification or contact with ammonia containing products which can generate hazardous chlorine gas. Clorox Ultra contains a strong oxidizing agent that could cause substantial but temporary eye injury, may irritate skin and may cause nausea and vomiting if ingested. Exposure to vapor or mist may irritate nose, throat and lungs.
- D. If the instrument emits an abnormal odor or smoke, turn off the power switch immediately and disconnect the power plug from the wall socket.
- E. Take care not to spill blood or reagent, or drop wire staples or paper clips into the instrument.
- F. Do not touch the electrical circuits inside the front cover of the Main Unit.
- G. Use specified tools and parts when working on instrument, if applicable.

- H. Methanol is flammable and poisonous. Potential human carcinogen. May be fatal if ingested. Harmful if inhaled. Causes irritation to eyes, skin and respiratory tract. Methanol is used for the SP-10 and is a component for the stain used on the SP-10.

III. MATERIALS

- A. Equipment:
1. Sysmex XN-10 analyzers
 2. Sysmex XN-10 Information Processing Units (IPU)
 3. Sysmex SP-10 slide maker/stainers
 4. RU-20 Reagent Unit
 5. TS-10 tube sorter
 6. Pneumatic Unit
 7. HP LaserJet P3005 Printer
- B. Supplies:
1. Deionized water
 2. Gauze
 3. Lint-free plastic lined lab wipes
 4. 12 x 75 mm test tube, for dilutions.
 5. Calibrated pipette
 6. CELLCLEAN® AUTO
 7. Commercial control; XN CHECK, XN CHECK BF
 8. Becton, Dickinson Vacutainer® potassium ethylene diamine tetra-acetic acid. (K2 EDTA), reference # 367861
 9. Microscope slides, frosted with rounded/clipped corners 76x26mm;
0.9 – 1.2 mm thick.
 10. Alcohol prep pads, isopropyl. Used to clean SP-10 spreader glass
- C. Reagents
1. Sysmex Reagents and CELLCLEAN AUTO are used on the Sysmex XN-Series modules.
 2. All reagents are used at room temperature and are to be used within the manufacturer's expiration date on each container.
 3. Record date received and date opened on container.
 4. All reagents are azide free and are intended for in vitro diagnostic use only.
Do not ingest.

Reagent	Abbreviation	Open Expiration
Cellpack DCL	DCL	60 days
Cellpack DST	DST	60 days
Cellpack DFL	DFL	60 days
Sulfolyser	SLS	60 days
Lysercell WNR	WNR	90 days
Fluorocell WNR	WNR	90 days
Lysercell WDF	WDF	90 days
Fluorocell WDF	WDF	90 days
Fluorocell PLT	PLT	90 days
Fluorocell RET	RET	90 days

4. SP10 reagents consist of Stain, Buffer, methyl alcohol and CELLPACK DCL

Reagent	Vendor	Open Expiration
STAIN WRIGHT 1 GAL 4L	Sysmex	90 days
Sysmex 6.8 Buffer	Sysmex	90 days
METHANOL, 1 GAL	VWR	1 year
Cellpack DCL	Sysmex	60 days

D. XN Reagent Replacement

1. When the reagent runs out during analysis, the analysis is paused and an error message appears in the analyzer area of the Control menu.
2. Display the [Reagent Replacement] dialog box to replace the reagent.
 - a) Select the help button on the control menu
 - b) Select [Execute]
 - c) Remaining Reagent Volume indicator appears
3. Replacing a new diluent / hemolytic agent
 - a) Display the [Reagent Replacement] dialog box
 - b) Input the reagent code (barcode)
 - c) Place the cursor in the reagent code field
 - d) Scan the reagent code on the outer box of the new reagent with the hand-held barcode reader or manually enter the reagent code
 - e) Select [OK]
 - f) Remove the cap from the new reagent container
 - g) Confirm the reagent has not expired
 - h) Remove the cap from the old reagent container.
 - i) Pull out the dispensing set straight up.
 - j) Insert the dispensing set straight into the new container.
 - k) Close the cap.
 - l) Select [Execute]
 - m) Reagent replacement starts. When complete, the dialog box closes automatically.

4. Replacing CELLPACK DST with an RU-20
 - a) Display the RU-20 Maintenance menu.
 - b) Select [Replace Reagent]
 - c) Input the reagent code (barcode)
 - d) Place the cursor in the reagent code field.
 - e) Scan the reagent code on the outer box of the new reagent with the hand-held barcode reader or manually enter the reagent code.
 - f) Select [OK]
 - g) Remove the cap from the new reagent container.
 - h) Confirm that reagent has not expired
 - i) Remove the cap from the old reagent container
 - j) Pull out the dispensing set straight up.
 - k) Insert the dispensing set straight into the new reagent container.
 - l) Close the cap
 - m) Select [Execute]
 - n) Reagent replacement starts. When complete, the dialog box closes automatically.
5. Replacing Dye
 - a) Display the [Reagent Replacement] dialog box.
 - b) Prepare the new reagent cartridge.
 - c) Confirm the reagent has not expired.
 - d) Open the top front cover.
 - e) Pull up the cover from the reagent that is to be replaced.
 - f) When the dye solution cover is pulled up, a Help dialog box appears in the IPU screen.
 - g) Remove the old reagent cartridge from its holder
 - h) Install the new reagent cartridge into the holder
 - i) Make sure the color of the label on the new reagent cartridge matches the color of the dye cover and install. Analyzer will beep as confirmation of new reagent installation.
 - j) If the wrong reagent is installed, the analyzer beeps repeatedly and the Help dialog box appears in the IPU screen.
 - k) Pull down the cover on the reagent until you hear a click.
 - l) When the cover is pulled down, the Help dialog box closes automatically.
 - m) The ID of the new reagent is read automatically and the information is registered.
 - n) Close the top front cover.
 - o) Reagent replacement starts.
 - p) When complete, the reagent replacement window closes automatically.
6. SP-10 Reagent Replacement
The following is a list of replacement messages and the reagent requiring replacement:

Message

*DCL not filled
*Stain 1 not filled in Chamber 1
*Stain 1 not filled in Chamber 2
*Stain 2 not filled
*Rinse water not filled
(internal chamber not filled)
Replace Rinse water
(external container empty)
Replace buffer
Replace methanol

Reagent

CELLPACK DCL
Stain
Stain
2nd stain (if using 2 stain method)
Deionized water

Deionized water

Buffer
Methanol

* Reagents with internal chambers. Other reagents use bottle sensors.

All reagent changes need to be properly documented. The received date of the reagent needs to be documented on the reagent log.

IV. CALIBRATION/PRECISION

Initial calibration is performed during installation by the Sysmex Field Service Representative. Perform calibration as needed, e.g., when QC data is fluctuating. However, if the abnormality in the QC analysis data was caused by an error in the analyzer, degradation of the reagent, or degeneration of the control blood, do not perform calibration. Calibrators traceable to reference methods are used in the calibration of the analyzer.

The laboratory must verify calibration every six months or on an "as-needed" basis to ensure accuracy of system. Calibration verification is also required if one or more of the following occur:

- Critical parts are replaced.
- Controls show an unusual trend or are outside of acceptable limits and cannot be corrected by maintenance or troubleshooting.
- When advised by Sysmex Field Service Representative.

Calibration verification may be performed by review and documentation of commercial control and X-BarM QC data, proficiency testing results and patient control testing results. The operator may calibrate the following parameters using XN CAL and XN CAL PF calibrator: WBC, RBC, HGB, HCT, PLT, PLT-F and RET.

Before calibration, ensure that the XN is both clean and precise.

Calibration verification and precision checks will be done after annual PM by Sysmex and six months after by SMH staff while on the phone with Sysmex TAC for assistance and direction.

- A. Precision Check
1. Perform routine maintenance on the analyzer and perform a background count to ensure counts are within acceptable limits.
 2. Verify that there is sufficient volume of all reagents. Precision and Calibration procedures will be aborted if the XN runs out of reagent.

3. Obtain a sample of fresh normal whole blood. **Do not** use commercial controls or calibrators for precision. The blood donor specimen should:
 - a) Be from a healthy person who is not taking any medication
 - b) Have morphologically and numerically normal CBC.
 - c) Be drawn in a potassium EDTA anticoagulant tube using proper collection technique.
 - d) Have a minimum of 2.5 mL of sample.
4. On the main unit, check the Status indicator LED. Confirm the LED is green indicating the analyzer is Ready
5. If the tube holder has not ejected out, press the mode switch
6. Select the Change Analysis Mode button on the control menu and select Whole Blood
7. Select [OK] to close the dialog box
8. Select the Analyzer menu button on the control menu
9. Select [Calibration] – [Precision Check]
10. Mix the vial containing the sample – 10 end-over-end inversions confirming cell button is dispersed
11. Place the vial in the sample tube holder
12. Press the start switch on the analyzer
 - a) The analysis is automatically performed 11 times consecutively with the tube holder pulled into the analyzer
 - b) The tube holder will slide out when analysis is complete
13. The results are displayed in the [Precision Check] analysis dialog box.
 - a) If the analysis results do not satisfy conditions for normal results, or if results are outside acceptable limits, the test numbers of the tests that must be repeated are displayed. Select and redo the manual analysis.
14. When all analysis results satisfy the conditions, select [OK] in the dialog box.
15. Select [Yes] to record passing precision results in the precision check history.

NOTE: If an error occurs during analysis and the analysis can no longer continue, stop precision check. Once the error is cleared, redo the manual analysis.

- B. Calibration –For XN CAL or XN CAL PF
1. On the main unit, check the Status indicator LED. Confirm the LED is green indicating the analyzer is Ready
 2. If the tube holder has not ejected out, press the mode switch
 3. Select the Change Analysis Mode button on the control menu and select Whole Blood
 4. Select [OK] to close the dialog box
 5. Select the Analyzer menu button on the control menu
 6. Select [Calibration] –if performing XN CAL select [Calibrator Calibration]; if performing XN CAL PLT-F select–[Calibrator Calibration (PLT-F)].
 7. Mix the vial containing the calibrator according to package insert
 8. Place the vial in the sample tube holder

9. Press the start switch on the analyzer
 - a) The analysis is automatically performed 11 times consecutively with the tube holder pulled into the analyzer
 - b) The tube holder will slide out when analysis is complete
10. The results are displayed in the [Calibrator Calibration] or [Calibrator Calibration (PLT-F)] analysis dialog box.
11. If the analysis results do not satisfy conditions for normal results, or if results are outside acceptable limits, the test numbers of the tests that must be repeated are displayed. Select and redo the manual analysis.
12. When all analysis results satisfy the conditions, select [Calibration] in the dialog box.
13. Select [OK] to display results in the [Calibrator Calibration] or [Calibrator Calibration (PLT-F)] execution dialog box.
14. Select the check box to include the calibration parameter in the calibration exercise or calibration (PLT-F) exercise, clear the check box to exclude the parameter in the calibration exercise. If a parameter meets all of the following criteria, the check box will automatically be selected:
 - a) $80\% < \text{New Rate} < 120\%$
 - b) $\text{New Rate} - \text{Current Rate} < +5$
 - c) $\text{Range Value} < \text{Max Range}$
 - d) $\text{Acceptable Limit} < \text{Delta Percent} < \text{Service Limit}$
15. If a parameter meets all of the conditions and the Delta Percent is less than the Acceptable Limit, it is excluded from calibration as there is no need for calibration.
16. If a parameter does not meet all of the conditions and the Delta Percent is greater than the Acceptable Limit, the calibration cannot be performed. Calibration is performed with the parameter excluded.
17. Selecting the check box enables you to manually enter a value in [New Rate (%)]. A range of 80% to 120% may be entered.
18. Select [OK] to update the compensation rates. The calibration process is logged in the calibrator calibration history.

V. **QUALITY CONTROL**

WARNING: POTENTIALLY INFECTIOUS MATERIAL

The human blood used in XN CHECK/XN CHECK BF is non-reactive for Hepatitis B Surface Antigen and negative for antibodies to HIV-1, HIV-2, and Hepatitis C Virus using FDA specified techniques. However, no current tests can assure the absence of these pathogens. XN CHECK/XN CHECK BF should be considered potentially infectious and must be handled with precautions used for human blood as described in CDC recommendations and in compliance with the Federal OSHA Bloodborne Pathogen Standard, 29CFR, 1910.1030.

A. When to Run Quality Control.

1. Run controls as follows:

Each shift is responsible for running XN Check on all 3 XN analyzers.

The supervisor or designee reviews commercial and XbarM charts daily.

2. Controls must be run after a calibration has been performed.
3. Controls must be run after specified service procedures are performed or for troubleshooting purposes.
4. Controls must be run when new lots of Quality Control are received, a statistical workup is required, prior to its official use in the laboratory. Please refer to procedure, QC Management for the SYSMEX XN-9000 (SH.CP.AU.hem.0129).
5. To QC the SP-10, examine a stained smear from the SP-10 for smear and stain quality on a daily basis. Document results on appropriate log.

B. XN CHECK Commercial Controls Instructions for Use

1. Remove vials from refrigerator and allow them to come to room temperature (18-25°C), for approximately 15 minutes.
2. Mix vials by gentle end-to-end inversion until the cell button in the bottom of the vial is completely suspended.
3. XN CHECK Analysis
 - a) Auto analysis: Place the QC vials in the Red QC rack and place on the line feeder. The rack will be sent to each XN module.
 - b) Off- line analysis: To run QC on one XN at a time press the mode switch on the conveyor of the analyzer you want to use. Place the rack in the designated area of the analyzers conveyor. Transport will begin automatically. Remove rack when analysis is complete.
 - c) Manual analysis: Press the mode switch to eject the tube holder. Select the analyzer menu button on IPU. Select QC analysis. Select QC file to be analyzed from given list. Place thoroughly mixed vial in tube holder and press start switch.
4. Auto analysis or offline mode results will be plotted on the L-J Chart and Chart for review. In manual mode results will be displayed and the operator needs to accept or cancel the run. Once accepted the results will transfer to the L-J and Radar Charts.
5. Reviewing Quality Control Results
 - a) QC File screen
 - 1) Allows for review of the latest QC results in Radar Chart format the QC file that is selected in the list.
 - 2) Any point exceeding the upper or lower limit is marked with a red "X".
 - 3) Print a hard copy of the radar chart.
 - b) QC Chart screen
 - 1) Allows for review of detailed graph data of all QC runs for selected File.
 - 2) Analysis data is plotted cumulatively and displayed in the chart area as a line graph.

- 3) Any point exceeding the upper or lower limit is marked with a red "X".
- 4) User must scroll up and down through the chart to view all Parameters for each run.
- 5) QC charts may be overlaid on top of each other for comparison.
 - a. Select [Compare QC Files] to view QC charts registered to a single analyzer. This will compare the new lot with the current lot.
 - b. Select [Compare Analyzers] to compare QC files for the same material registered to different analyzers.
6. Handling QC failures.
 - a) Any QC failure is unacceptable and is to be documented in the QC Rejection log.
 - b) A first failure of a control, remix and rerun the failed QC level.
 - c) If that same level and parameter fail a second time obtain a new vial, warm, mix and rerun. If the control is within acceptable limits finish any additional controls and resume specimen testing. If the new vial fails the same parameter(s) troubleshoot the analyzer.
 - 1) View the Levy-Jennings (L-J) chart to check for a steady shift of the failing parameter. If there is shifting seen, consult your supervisor or senior tech.
 - d) Subsequent failures of different parameters should be treated as initial failures.
 - e) Failures of the same parameter that occur on multiple control levels definitely indicate an analyzer problem, troubleshoot the analyzer.

C. X-barM (moving averages) monitoring and failures

1. From the menu screen click the QC File button.
2. Use the tabs near the bottom of the screen to select an analyzer.
3. Scroll to the bottom of the file list to find XM files.
4. Double click a file to view the L-J graph. Each point represents the average of a batch of normal patient samples (refer to SH.CP.AU.hem.0129 – QC Management – Sysmex XN-9000).
5. Any point exceeding the upper or lower limit is marked with a red "X".
6. X-barM failures should be handled as follows:
 - a) On a first failure document it in the QC rejection log and continue running samples, no further action is required.
 - b) If two consecutive batches fail stop running the analyzer. Run XN-check level 2 control. If the control passes, continue sample processing and monitor the next batch update. If the control fails the same parameter(s) and the XM failure troubleshoot the analyzer. *If the control fails a parameter not related to the XM failure, rerun the control. Should the control continue to fail after troubleshooting or X-barM fails a third*

consecutive batch stop running the analyzer, notify a supervisor and call Sysmex TAC.

7. Document all steps taken in the QC rejection log.

D. SP-10 Daily QC Slide review

1. Review the blood smears macroscopically for acceptability:
 - a) Smears are sufficient length (greater than half the length of the unfrosted portion of the slide). The feathered edge becomes gradually thinner without streaks, holes, or tails.
 - b) Even, consistent staining of blood smear.
2. Review the blood smears microscopically for acceptability:
 - a) Relatively even distribution of cellular elements.
 - b) Acceptable morphology within the working area. None or very little artifact of the cell morphology, (e. g., “punched-out” RBC’s, smashed WBC’s).
 - c) None, or very little stain precipitate or debris.
 - d) The staining is consistent and imparts the characteristic cytoplasmic color differences and distinct nuclear chromatic patterns of the whole spectrum of blood cells. Acceptable stains will display the following characteristics:
 - 1) RBC’s should be pink to orange. There should be good differentiation between normochromic, hypochromic, and polychromatic cells.
 - 2) Lymphocytes will display dark purple nuclei with varying shades of blue cytoplasm.
 - 3) Neutrophils will display dark purple nuclei, with light pink cytoplasm and lilac granules.
 - 4) Monocytes will show lighter purple nuclei. The cytoplasm of the monocytes will be gray-blue with reddish granules.
 - 5) Eosinophils show bright orange granules in the cytoplasm.
 - 6) Basophils display dark blue granules in the cytoplasm.
 - 7) Platelets will be violet to purple.
 - e) If smear quality is unsatisfactory, clean, or if necessary, replace the spreader glass. If smear/stain quality is still unsatisfactory, refer to the SP-Series Implementation Manual troubleshooting section. If troubleshooting does not resolve the issue notify a supervisor or call Sysmex TAC.

VI. MAINTENANCE

A. XN Daily Maintenance

1. Press the mode switch on the conveyor in front of the analyzer you want to perform maintenance on.
2. Click the [Analyzer Menu] button on the instrument control menu.
3. Click on Maintenance.
4. Click Cleaning

5. If the tube holder is not ejected, press the mode switch on the analyzer. Place the CELLCLEAN AUTO in the sample tube holder.
6. Press the start switch on the analyzer. Cleaning will take 20 minutes.
7. When cleaning is complete remove the CELLCLEAN AUTO and press the mode switch to return to auto sampler mode.
8. Run QC.
9. Once QC has passed press the mode switch on the conveyor to have the analyzer back on line.
10. Restart the CT-90 (refer to SH.CP.AU.jad.0105 Restarting the CT-90).

B. XN Weekly Maintenance

It is required the IPU and conveyors of the XN-9000 be powered off once a week. Powering off the IPU will shut down all the analyzers.

1. SHUTDOWN

- a) Make sure all the racks have been returned to the collector and the system is at "Ready". There should not be any samples in process.
- b) Press and hold the green start-up switch on the front of the BT-40 until an audible beep is heard. Once the sound is heard release the button.
- c) The status LED on the conveyors will flash green. After approx. 75 seconds all of the conveyors and the CT-90 IPU will turn off and the XN's will put themselves into Manual Mode.
- d) Click [Exit IPU] on the menu screen of the XN software.
- e) Click [Yes] to confirm.
- f) From the Windows desktop, select [Shutdown] from the start menu. This will turn off the XN-IPU and the connected XN analyzers.
- g) After 30 seconds, perform Start-up.

2. START-UP

- a) Press and quickly release the green master start-up switch at the front of the BT-40.
- b) Each conveyor will start and the corresponding LED will be green. All attached analyzers will turn on and begin start-up.
- c) The XN software will display the logon screen. Click OK.
- d) Logon name: xn
- e) Password: xn
- f) The XN's and SP's will perform self-checks. The XN will do a Background Check (it will attempt up to 3 times before requiring Intervention)

XN Acceptable Background Counts	
Parameters	Acceptable Limit
WBC-N	0.10 x 10 ³ / μL
WBC-D	0.10 x 10 ³ / μL
RBC	0.02 x 10 ⁶ /μL
HGB	0.1 g/dL

PLT-I	10 x 10 ³ / μL
PLT-F	3 x 10 ³ / μL

- g) When the background check has passed the analyzers will be in ready mode as indicated for the Green LED on the analyzer and the Green light on the analyzer menu of the IPU.
- h) Click the [Sampler analysis button] on the Analyzer area menu of the IPU. Uncheck [Diff], [Retic], and [Plt-F]. Only CBC should be set for default testing. Do this for all 3 XN analyzers.

C. SP-10 Maintenance

Daily the spreader glass is to be cleaned, the slide towers are to be filled and a Shutdown 2 will be performed. Refer to current maintenance form for guidance (SH.CP.AU.frm.0226, .0227 and .0228).

1. Daily

- a) Clean Spreader Glass: Power must be on to perform this maintenance – may be performed prior to Shutdown, or after Start-up.
 - 1) Press [Maint.] on the main screen. (Maintenance button is not available during routine operation.)
 - 2) Press [Spreader Glass] and the “Spreader Glass Replace” screen displays.
 - 3) Press [OK] to move the smear unit forward.
 - 4) Open the top cover and remove the left tower for easier access to the spreader glass.
 - 5) Wipe the spreader gauze moistened with DiH₂O.
 - 6) Replace the tower so that the frosted end of the slides is towards the back of the analyzer.
 - 7) Press [OK] to return the smear unit to the home position.
 - 8) Press [OK] to reset the spreader glass cycle counter or [CANCEL] to allow the cycle count to continue.
 - 9) Press [RETURN].
- b) Clean Single Cassettes
 - 1) Place cassettes in a bin with open end up.
 - 2) Pour methanol over the cassettes, filling them.
 - 3) Swish the methanol and pour off into designated container for reuse.
 - 4) Invert cleaned cassettes on absorbent material to dry.
- c) Shutdown 2
 - 1) Press the mode switch on the conveyor in front of the SP-10 to have it off line or press the [Conv. Int.] on the main menu, then [Interrupt] then [return].
 - 2) Press [**SHUTDOWN**] on the main screen.
 - 3) Press [**Shutdown 2**].

- 4) The shutdown screen displays the number of cassettes and amount of methanol required for the shutdown process. Ensure that required amounts are available.
- 5) Place a tube of CELLCLEAN AUTO in position 10 of a Sysmex rack.
- 6) Place the rack so that the tube is lined up with tube gripper.
- 7) Press **[OK]**.
- 8) When the process completes, the SP-10 turns off automatically.
- 9) To restart the SP-10, press the green button on the right side.
- 10) After the SP-10 has been off for at least 30 seconds it can be powered back on. Press and hold the green start switch on the unit until "Sysmex" appears on the touchscreen.

2. Weekly

- a) Clean DI water containers
 - 1) If re-usable containers for deionized water are used, empty weekly.
 - 2) Rinse with methanol and allow to air dry.
 - 3) Fill a new container with fresh deionized water.

3. As Needed Maintenance

- a) Refer to the XN-9000 *Instructions for Use* for detailed and illustrated instructions for performing as needed maintenance.

VII. PATIENT SAMPLE PROCESSING

A. System Analysis (sampler analysis):

1. Make sure the analyzer and the sampler are in READY state
2. Check that tube holder has retracted into the analyzer, press mode button if necessary
3. Place barcoded sample(s) in rack(s) in the feeder.
4. Rack(s) will be automatically pushed forward and routed to BT.
5. Samples will run, results will be displayed in the IPU.
6. On-Board IPU rules or Sysmex WAM will determine repeat or reflex testing
7. Rack will run in reverse to perform repeat or reflex testing on the same XN.
8. If smear is required, rack will be transported to SP-10 via feeder line and samples will be aspirated by SP-10.
9. If no smears are required, rack will be transported via collector line to the collector and will not be routed to SP-10.
10. Remove the rack(s) when analysis is completed.

B. Manual Analysis – XN:

1. Check the status of the analyzer. Confirm the analyzer is ready.
2. Press the mode switch to eject the tube holder.
3. Select the Change Analysis Mode button on the control menu
4. Select analysis mode
 - a) [Whole blood] is selected when whole blood is being analyzed

- b) [Low WBC] Select this to perform low WBC analysis on whole blood
5. Select [OK]
6. Select Manual Analysis button on the control menu
7. Input sample ID or select [Read ID]
8. Make sure [Cap Open] is checked.
9. Select [OK]
10. Properly mix the specimen, remove the cap and place in the tube holder
11. Press the start switch on the analyzer
 - a) The tube holder will slide in and the sample will be aspirated
 - b) When the analysis is complete, the tube holder slides out
12. Remove the sample, repeat steps for additional samples
13. Review results in IPU or WAM to determine whether repeat or reflex testing is required. Rerun sample if required. Make smear if required.

C. Off-line analysis:

The conveyor for the analyzer, or the conveyor for the SP-10 is separated from the transport line of the overall system and operated as a standalone device

1. Press mode switch on the conveyor
2. Verify conveyor is in READY state
3. Place the rack in the designated area in the right pool of the conveyor for the analyzer that you wish to use.
4. Transport begins automatically
5. Remove the rack after analysis is complete
6. Press the mode switch on the conveyor to return to on-line analysis.

D. SP-10 Manual Mode – Smear and Stain, Stain Only, Smear Only:

Refer to SP-10 Instructions for use Guide: Chapter 6 - Smear Preparation.

VIII. PROCEDURAL NOTES/LIMITATIONS

- A. The XN-Series analyzers will generate a POSITIVE when an IP Message is present. ERROR will be generated when there is an analysis error. These judgments indicate the possibility of sample abnormality. These results should be reviewed carefully and may require further examination. IP messages and flags from the XN will transfer to WAM triggering any rules that may apply. The operator will follow the OP alerts in WAM for instruction on handling the results (refer to Sysmex XN Series Flagging Guide and SH.CP.AU.jad.0125 - WAM rules) All analyzer flags and results must be interpreted together and in consideration of the patient's clinical condition prior to results being reported from the laboratory. Protocols for comparison of current results to previous results (delta checking) as well as critical value alerts are also useful for identifying potentially erroneous results prior to reporting to the clinician. Abnormal results will have one or more of the following indicators;

Indicators that appear after the data

Indicator	Reason
@	Data is outside linearity
*	Data is unreliable
+, -	Data is outside reference limits
----	Analysis impossible value cannot be displayed
++++	Data exceeds display limit. Out of range
!	Exceeds upper panic limit/below lower panic limit. Exceeds upper acceptable background check value limit

XN-Series - Summary of Linearity

Parameter	Range	Units
WBC	0-440.0	$\times 10^3/\mu\text{L}$
RBC	0-8.60	$\times 10^6/\mu\text{L}$
HGB	0-26.0	g/dL
HCT	0-75.0	%
PLT, PLT-F	0-5000	$\times 10^3/\mu\text{L}$
RET%	0-25	%
NRBC%	0-600	/100 WBC

- B. Parameters that exceed these limits are flagged with @ beside the result. The sample must be diluted, rerun and multiplied by the dilution factor (refer to SH.CP.AU.jad.0122 - XN Linearity).
- C. Note the use of dilution for linearity on the patient report.
- D. Cold agglutinins produce spurious macrocytosis, elevated MCH's & MCHC's, falsely decreased RBC counts and HCT's. Rare, warm agglutinins produce the same spurious results as a cold agglutinin. (Refer to SH.CP.AU.jad.0100)
- E. Severely icteric or lipemic samples may falsely elevate the HGB value and related indices (Refer to SH.CP.AU.jad.0100).
- F. MCV delta needs to have the integrity of the sample verified. An increase by in the MCV of at least six should to be evaluated for glucose contamination. (Refer to SH.CP.AU.jad.0128 – MCV Delta/Low MCHC)
- G. Low MCHC needs to be evaluated for sample integrity. (Refer to SH.CP.AU.jad.0128 – MCV Delta/Low MCHC).
- H. Umbilical cord samples (UCCBC): Refer to job aid.
- I. Reporting Abnormal Results to Physicians
 Follow SH.CP.AU.gen.0020, Results Reporting Procedure Critical Value Notification.

Critical Values

WBC >150
HCT < 20
PLT < 20
Bands >10%

IX. POSSIBLE SAMPLE INTERFERENCES

- A. Specimens must be free of clots and fibrin strands.
- B. Marked changes in plasma constituents, (e.g., low sodium, extremely elevated glucose) may cause cells to swell or shrink. The blood to anticoagulant ratio is important.
- C. Red cell fragments, microcytic RBC's, or white cell cytoplasmic fragments may interfere with automated platelet counts. A fluorescent platelet may be performed to avoid this interference.
- D. Cold agglutinins produce spurious macrocytosis, elevated MCH's & MCHC's, falsely decreased RBC counts and HCT's. Rare, warm agglutinins produce the same spurious results as a cold agglutinin.
- E. Extremely elevated WBCs may cause turbidity and falsely increase the hemoglobin, in addition to RBC and HCT values.
- F. Severely hemolyzed samples (*in vitro*) falsely decrease RBC and hematocrit. Recollect hemolyzed specimens.
- G. Giant platelets and clumped platelets may falsely elevate the WBC count and falsely decrease the platelet count. Platelet clumping and/or "platelet satellitism" can occur in specimens collected in EDTA. This may falsely elevate the WBC count and falsely decrease the platelet count.
- H. Abnormal paraproteins found in Multiple Myeloma patients can falsely increase the HGB.
- I. Rocking specimen excessively may affect the WBC differential.
- J. Megakaryocytes may falsely increase WBC counts on automated hematology analyzers.

X. CALCULATIONS N/A

XI. INTERPRETATION N/A

XII. RESULT REPORTING**A. Reference Intervals**

1. Reference intervals (Normal Population Reference Ranges) were developed for the XN-9000 using normal individuals.
2. The range for each parameter is calculated for 95% confidence intervals.
3. Table below outlines the Normal Population Reference Ranges determined for the XN-9000.

Normal Adult Population Reference Ranges

Parameter	Range for Females n = 133	Range for Males n = 182	Units
WBC x	3.98 - 10.04	4.23 - 9.07	X 10 ³ /μL
Neut%	34.0 - 71.1	34.0 - 67.9	%
Lymph%	19.3 - 51.7	21.8 - 53.1	%
Mono%	4.7 - 12.5	5.3 - 12.2	%
Eo%	0.7 - 5.8	0.8 - 7.0	%
Baso%	0.1 - 1.2	0.2 - 1.2	%
Neut#	1.56 - 6.13	1.78 - 5.38	10 ³ /μL
Lymph#	1.18 - 3.74	1.32 - 3.57	10 ³ /μL
Mono#	0.2 - 0.9	0.30 - 0.82	10 ³ /μL
Eo#	0.04 - 0.36	0.04 - 0.54	10 ³ /μL
Baso#	0.01 - 0.08	0.01 - 0.08	10 ³ /μL
NRBC%	0 - 0.2	0 - 0.2	/100WBC
NRBC#	0 - 0.012	0 - 0.012	10 ³ /μL
RBC x	3.93 - 5.22	4.63 - 6.08	10 ⁶ /μL
HGB	11.2 - 15.7	13.7 - 17.5	g/dL
HCT	34.1 - 44.9	40.1 - 51.0	%
MCV	79.4 - 94.8	79.0 - 92.2	fL
MCH	25.6 - 32.2	25.7 - 32.2	pg
MCHC	32.2 - 35.5	32.3 - 36.5	g/dL
RDW-CV	11.7 - 14.4	11.6 - 14.4	%
RDW-SD	36.4 - 46.3	35.1 - 43.9	fL
RET%	0.5 - 1.7	0.51 - 1.81	%
RET# x	0.0164 - 0.0776	0.026 - 0.095	10 ⁶ /μL
IRF	3.0 - 15.9	2.3 - 13.4	%
PLT x	160 - 370	150 - 330	10 ³ /μL
MPV	9.4 - 12.3	9.4 - 12.4	fL

XIII. TRAINING

Personnel	Training Required
Management	Develop training checklist
End User	Read SOP Perform web based training Perform Skills Assessment

IX. REFERENCES

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- V. Alert Value Notification, SH.CP.AU.gen.0020
- W. Quality Control, SH.CP.AU.gen.0024.
- X. SH.CP.CT.jad.0100 MCHC interpretation guide/flowchart
- Y. SH.CP.CT.jad.0125 Sysmex WAM rules
- Z. Sysmex 1166-LSS, Rev. 1, XN-Series Flagging Interpretation Guide