**Sysmex XP-300™**

**Automated Hematology Analyzer**

****

**CLSI Procedure**

##### COMPLETE BLOOD COUNT (CBC)

##### ON THE SYSMEX XP-300™ AUTOMATED HEMATOLOGY ANALYZER

# PRINCIPLE

The Sysmex XP-300 is a quantitative automated hematology analyzer for *in vitro* diagnostic use in determining 17 hematological parameters. Examination of the numerical and/or morphologic findings of the complete blood count may be useful in diagnosis by a physician of such disease states as anemias, leukemias and allergic reactions as well as viral, bacterial, and parasitic infections. The Sysmex XP-300 analyzer directly measures the WBC, RBC, HGB, HCT, and PLT, LYM #, MXD # and NEUT #. The remaining parameters are calculated or derived: MCV, MCH, MCHC, MPV, RDW-CV and RDW-SD, and differential percentages LYM%, MXD% and NEUT%.

The XP-300 counts and sizes red blood cells (RBC) and platelets (PLT) using Direct Current (DC) detection method. Hematocrit (HCT) is measured as the ratio of the total RBC volume to whole blood using cumulative pulse height detection. Hemoglobin (HGB) is a non-cyanide based method read photometrically at 555nm.

White blood cells (WBC) are analyzed by direct current and discriminated into a 3-part differential using Particle Distribution Analysis (PDA). The resulting WBC histogram is discriminated into lymphocyte, neutrophil and mixed cell populations. The mixed cell population contains monocytes, basophils and eosinophils.

# SPECIMEN

1. Required specimen:
2. Whole blood anticoagulated with a salt of EDTA-2K, EDTA-3K or EDTA-2Na is preferred.
3. Specimen volumes required:
4. Optimal draw is a tube filled to capacity. The collection tube should be filled per the tube manufacturer instructions. A minimum of 1 mL is recommended.
5. An EDTA micro-container filled above the 250 μL line is adequate for testing in the whole blood mode. 500 μL is recommended.
6. Aspirated sample volume in whole blood mode is 50 μL.
7. Unacceptable specimens include the following:
8. Clotted samples or those containing clots, fibrin strands, or platelet clumps. All specimens will be checked visually for obvious clots prior to sampling by the analyzer.
9. Grossly hemolyzed samples.
10. Samples drawn above an IV.
11. Specimen characteristics that may affect test results are lipemia, icterus, cold agglutinins, RBC fragments, lyse resistant RBCs, giant platelets, abnormal plasma proteins, clumped platelets and fibrin strands.
12. Stored Specimen Stability
13. EDTA blood samples stored at 2 - 8° C may be analyzed up to 24 hours after draw without significant loss of differential stability.
14. Sample stability at room temperature is 4 hours. Samples stored at room temperature may exhibit an increase in MCV, RDW-SD, HCT and MPV, and a decrease in MCHC and total WBC over time. These changes may be minimized by refrigeration.
15. Allow refrigerated samples to come to room temperature (minimum 15 minutes) and mix well before analysis.
16. **Do not** place samples on a mechanical rocker. Constant rocking may alter white cell membranes, which may result in inappropriate flagging.

**WARNING**: All patient specimens 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, 29 CFR part 1910.1030. Follow specimen handling as outlined by laboratory safety policy.

**Recommended**: Wear personal protective equipment. Wear safety glasses if there is a risk of splash.

# SUPPLIES AND REAGENTS

1. Supplies
2. Deionized water
3. Lint-free, plastic-lined lab wipes
4. “Filler” pipette supplied with the unit or a 5 cc syringe
5. Clorox® bleach at 5% solution (use when CELLCLEAN® is indicated)
6. Sysmex reagents
7. Tri-level commercial control, EIGHTCHECK®-3WP X-TRA
8. SCS-1000TM Calibrator
9. Sysmex Reagents
10. A diluent, a hemoglobin/WBC lyse, and bleach are used on the Sysmex XP-300 analyzer.
11. Reagents and bleach are stored at room temperature and are to be used within the manufacturer’s expiration date on each container.
12. Record date received and date opened on container. Record the lot, date opened and expiration date on the Reagent Replenishment Record.
13. The reagents are azide free, and are intended for *in vitro* diagnostic use only. **Do not** ingest.

## CELLPACK® is a reagent for measuring the quantity and size of RBCs and PLTs by Direct Current detection.

CELLPACK Ingredients

Sodium Chloride 6.38 g/L

Boric Acid 1.00 g/L

Sodium Tetraborate 0.20 g/L

EDTA-2K 0.20 g/L

CELLPACK Storage

1. Store at a controlled temperature of 15-30oC.
2. **If frozen**, thaw, mix thoroughly, and allow bubbles to disperse before use.
3. CELLPACK is clear and colorless. If there are signs of contamination, instability or color change, **do not** use.

CELLPACK Stability

1. Unopened, 18 months after the date of production labeled on the container.
2. Opened, CELLPACK is stable for 60 days.

CELLPACK Hazard Risk

## The OSHA Hazard Communication Standard of 29CFR part 1910.1200 requires Chemical manufacturers and importers to obtain or develop a Safety Data Sheet (SDS) for each hazardous chemical they produce or import. Hazardous chemical means any chemical which is classified as a physical hazard or a health hazard, a simple asphyxiant, combustible dust, pyrophoric gas, or hazard not otherwise classified. CELLPACK does not have ingredients with those characteristics.

## STROMATOLYSER-WH™ lyses RBCs for accurate measurement of HGB, and enumeration of the WBC count and three differential populations.

###### STROMATOLYSER-WH Ingredients

 Organic quaternary ammonium salt 8.5 g/L

 Sodium chloride 0.6 g/L

STROMATOLYSER-WH Storage

1. Store at controlled room temperature of 2-35oC.
2. **If frozen, do not use.**
3. STROMATOLYSER-WH is a clear, odorless liquid. If there are signs of contamination, instability, or color change, **do not** use.

###### STROMATOLYSER-WH Stability

1. Unopened, 12 months from the date of production labeled on the container.
2. Opened, STROMATOLYSER-WH is stable for 90 days.

STROMATOLYSER-WH Hazard Risk

The OSHA Hazard Communication Standard of 29CFR part 1910.1200 requires Chemical manufacturers and importers to obtain or develop a Safety Data Sheet (SDS) for each hazardous chemical they produce or import. Hazardous chemical means any chemical which is classified as a physical hazard or a health hazard, a simple asphyxiant, combustible dust, pyrophoric gas, or hazard not otherwise classified. STROMATOLYSER-WH does not have ingredients with those characteristics.

## 5% solution of Clorox bleach (when CELLCLEAN is indicated) is recommended for use in cleaning and shutdown of the XP-300 analyzer. Scented or splashless varieties of Clorox bleach must not be used at any time. Refer to the label of the product in use for the sodium hypochlorite concentration. Dilute to 5% as appropriate.

Clorox Ingredients

Sodium Hypochlorite

Clorox Storage

Stable under normal use and storage conditions

**WARNING**: When using Clorox avoid acidification or contact with ammonia-containing products that can generate hazardous chlorine gas.

Clorox Health Risk

Can be a respiratory irritant if mist or vapor is inhaled, and cause nausea and vomiting if ingested. May irritate skin. Contact with eyes can cause severe, but temporary injury.

**WARNING**: Clorox contains a strong oxidizing agent. Causes substantial but temporary eye injury. May irritate skin. May cause nausea and vomiting if ingested. Exposure to vapor or mist may irritate nose, throat and lungs.

**Recommended:** Wear personal protective equipment for protection.

Commercial Control

**EIGHTCHECK-3WP X-TRA** is a tri-level whole blood commercial control for use with the Sysmex XP-300 hematology analyzer.

EIGHTCHECK-3WP X-TRA Ingredients (formulation)

EIGHTCHECK-3WP X-TRA consists of stabilized human erythrocytes, human and simulated leukocytes and a platelet component in a plasma-like fluid. Each vial contains 2.0mL of control material.

EIGHTCHECK-3WP X-TRA Storage:

1. After opening, vials should be stored in the upright position at 2 - 8°C.
2. **Do not** freeze or expose to excessive heat.

EIGHTCHECK-3WP X-TRA Stability:

1. Unopened and properly stored, EIGHTCHECK-3WP X-TRA is stable until the expiration date stated on the vial.
2. Open vial stability is 14 days when promptly refrigerated after each use.
3. Record the date on each vial upon opening.
4. Heat or freezing can damage EIGHTCHECK-3WP X-TRA without gross visible changes. Moderate hemolysis can be normal. Deterioration is suspected when the mean of the control results is not within the assay expected ranges after appropriate troubleshooting.
5. If deterioration is suspected, call the Sysmex Technical Assistance Center at 1-888-879-7639 (1-888-8SYSMEX).

**WARNING: POTENTIALLY INFECTIOUS MATERIAL**

The human blood used in **EIGHTCHECK-3WP X-TRA** 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.  **EIGHTCHECK-3WP X-TRA** 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, 29 CFR, 1910.1030.

SCS-1000™ Calibrator

**SCS-1000** is a secondary whole blood calibrator for use with the Sysmex XP-300 hematology analyzer. Assay values for primary parameters are traceable to reference methods.

SCS-1000 Ingredients (formulation)

SCS-1000 consists of human red and white blood cells with a platelet component suspended in fluid medium. Each vial contains 2.0 mL of calibrator material.

SCS-1000 Storage

Store vials as packaged, at 2-8° C. **Do not** freeze or expose to excessive heat.

SCS-1000 Stability

1. Unopened and properly stored, SCS-1000 is stable until the expiration date stated on the vial.
2. Open vial stability is 4 hours.
3. Storage outside of 2-8o C can damage SCS-1000 causing deterioration that risks inaccurate calibration. If deterioration is suspected, call the Sysmex Technical Assistance Center at 1-888-879-7639 (1-888-8SYSMEX).
4. Use of the product at environmental temperatures that exceed 30°C (86°F) can reduce calibration accuracy.

**WARNING: POTENTIALLY INFECTIOUS MATERIAL.**

The human blood used in SCS-1000 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. SCS-1000 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, 29 CFR, 1910.1030.

## REAGENT REPLACEMENT

When the reagent level is low, the analyzer will automatically stop and an error dialog will appear: [Replace CELLPACK] or [Replace STROMATOLYZER]. Analyzer will be in a ‘Not Ready’ state.

**CAUTION**: Wear personal protective equipment when replacing reagents.

1. Retrieve new reagent.
2. Open the new reagent container. Document the lot number in use per your laboratory protocol.
3. Using care to not contaminate the reagent line, remove the spout kit from the empty container by lifting straight up and place it directly into the new container.
4. Tighten the spout cap on the new container.
5. Record the open date on the new reagent container.
6. Press **[MENU]** on the Main screen.
7. Press **[HELP]**
8. Press the **[Execute]** button.
9. Press the **[Chg.Reag]** button to display the reagent information screen.
10. Press the **[CELLPACK]** button to replace CELLPACK, press the **[S.LYSER]** button to replace STROMATOLYSER-WH.
11. The reagent barcode entering screen will appear.
12. Enter the reagent barcode information either manually with the alphanumeric keypad or with the handheld barcode reader.
13. Press **[OK]** and **[Execute]**

## CALIBRATION

Initial calibration is performed during installation. Calibration compensates for any bias inherent to the pneumatic, hydraulic, and electrical system that may affect the accuracy of results. Calibrators traceable to reference methods are used in the calibration of the analyzer.

Calibration verification is performed according to the laboratory Standard Operating Procedure and accreditation agency requirements to ensure accuracy of the system.

Calibration is also required if one or more of the following occur:

* Critical parts, such as manometers, apertures or detector circuit boards 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 accomplished by review and documentation of commercial controls run each day patient specimens are run and proficiency testing results.

The operator uses SCS-1000 calibrator to calibrate WBC, RBC, HGB, HCT, and PLT. Before calibration, ensure that the XP-300 maintenance procedures and precision run have been performed.

* 1. Precision Check
1. Perform routine daily 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 XP-300 runs out of reagent.
3. Obtain a sample of fresh normal whole blood. The sample should be analyzed within six hours of collection and stored at room temperature. **Do not** use commercial controls or calibrators for precision. The blood donor specimen should:
4. be free from medication and interfering substances such as lipemia, icterus, hemolysis, platelet clumps etc.
5. have morphologically and numerically normal CBC such as WBC >4.00, RBC >4.00, PLT >100.
6. be drawn in EDTA anticoagulant tube using proper collection technique.
7. be a minimum of 2 mL.
8. Precision Check Specimen Analysis
9. Press **[Menu]** while at Ready status.
10. Press **[Calib]**. Enter password if applicable.
11. Press **[Calibrator Cal]**. The Precision Check screen will appear.
12. Analyze the sample 11 times in the whole blood mode. The precision is determined on the last 10 replicates, excluding the first analysis. If an error occurs during analysis, an error message appears and the result is masked with “---.-”. The cursor will not move to the next line and the next sample is analyzed using the same number.
13. Press **[Next]**
14. If the CV%’s for all five parameters are within the limit% shown on the screen, select **[Quit]** and proceed to Calibration using the SCS-1000 calibrator. If the CV% exceeds acceptable limits (results will be highlighted on the screen), press **[Back]** to display the precision check completion screen or **[Top]** to display the precision check quit confirmation screen.
	1. Calibration
15. After the Precision Check is completed, press **[Quit]** to proceed to Calibration. The target values screen will appear.
16. Press the button for the ‘target’ column.
17. Using the numeric keys, enter the Assay Target values from the Sysmex SCS-1000 calibrator product insert for WBC, RBC, HGB, HCT, and PLT on the “Analyzing Calibrator” screen. Press **[ENTER]** after each value. Press **[C]** to clear any entry errors. Confirm all values are correctly entered.
18. Press **[Next]** after all five values have been entered correctly.
19. Press **[OK]** to set the targets and display the analyzing calibrator screen.
20. Analyze the SCS-1000 calibrator in the open mode six times. If an error occurs during analysis, the result is masked with “---.-“, and the next sample result replaces the error results. The first analysis data is not included in the calculations.
21. After all six analyses are displayed, press **[Next]** to display the analyzing calibrator screen 2.
22. If the Range values exceed the MaxRange values, the Range value will be displayed in red. If the Delta% values exceed SERV LMT, the Delta% values will be displayed in red. Calibration will not be allowed. Troubleshooting may be required.
23. When:
24. Range V. is greater than MaxRange or
25. Delta %” is greater than “SERV LMT,” or
26. Delta %” is less than or equal to the “ACPT LMT,” no adjustment to calibration is necessary.
27. If Other than above, New = (Target/MeanV.) x Current
28. Press **[Quit]**. New compensation values are calculated and the Main screen returns. Press **[Back]** to return to the analysis screen without updating the calibration value. Press **[Top] to** return to the Main screen without updating the calibration value.
29. Analyze the commercial controls.
30. Document the date of calibration and new calibration factors.

## QUALITY CONTROL

* 1. EIGHTCHECK-3WP X-TRA Instructions for Use.
1. Remove EIGHTCHECK-3WP X-TRA 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. **Do not** use a mechanical rocker.
3. Wipe residual blood off the cap and screw threads of the vial before replacing the cap.
4. Return vials to the refrigerator promptly after use.

**WARNING: POTENTIALLY INFECTIOUS MATERIAL.**

The human blood used in **EIGHTCHECK-3WP X-TRA** 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.  **EIGHTCHECK-3WP X-TRA** 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, 29 CFR, 1910.1030.

* 1. Frequency of Control Use and QC Data Review

 EIGHTCHECK-3WP X-TRA levels: L, N, H will be run on Day shift.

 EIGHTCHECK-3WP X-TRA levels: L, N, H will be run on Night shift.

The supervisor reviews commercial and patient control charts every 30 Days.

* 1. QC Analysis
1. From the Ready status, Press **[QC]**.
2. Press the display column of the file number to be analyzed.
3. Place the well mixed open QC vial up to the sample probe and press the Start Switch. When the XP-300 beeps, remove the vial. **Do not** wipe the sample probe.
4. When analysis is complete, press either **[🡪]** or **[🡨]** to scroll through the screen pages and review results. For parameters whose values are above the upper limit, a **+** is displayed in the **[Judgment]** column and if the values are below the lower limit, a **–** is displayed.
5. **[IP]** to Print.
6. **[NG]** to reject results, or
7. **[OK]** to accept results.
8. Repeat steps for each level of control.
9. Press **[Top]** to exit QC program.
	1. Viewing QC Charts
10. From the Main Screen, press **[QC]**.
11. Press **[QC Chart]**.
12. Enter the QC file number to be viewed in the **[File]** field to select the file and enter the lot number in the **[Lot ID]** field.
13. Press **[🡪or🡨]** to review the ten (10) screens of data.
	1. Starting a New Lot of Controls

Parallel test new controls by analyzing the three levels of control a minimum of twice a day for 5 days prior to expiration of the previous lot. After a minimum of 10 data points are accumulated and values are running within assay ranges, the lot may be placed into production. The new lot will be validated prior to the current lot expiration.

* 1. Interlaboratory Quality Assurance Program (IQAP):

Complete this section with your laboratory’s account number, analyzer serial number(s), preferred mode for data submission, and who is responsible for completing/reviewing results.

The Sysmex Quality Control Program Lab ID number is: 33095.

The XP-300 serial # is B8061.

Lead Technologist is responsible for gathering the data and submitting it by the submission due date.

Record the daily EIGHTCHECK-3WP X-TRA values on the ***Daily Input forms/ Summary Data forms/control file printouts.***

Sysmex Quality Control Program phone: 1-888-879-7639 (US)

email: Insight@sysmex.com

* 1. Corrective Action for Out of Range QC Results

***Complete this section with your laboratory’s QC action plan for out of range commercial and patient controls.***

* 1. Recording and Storage of QC Data

***Complete this section with your laboratory’s policy for documenting and retaining commercial and patient control.***

# OPERATING PROCEDURE

* 1. Start-Up
1. Check for fluid in the pneumatic trap chamber on the left side of the main unit.
2. If fluid is present, unscrew the chamber counterclockwise, empty and dry.
3. Replace the chamber taking care to make a good seal. Vacuum errors occur if not sealed tightly.
4. Check paper supply. For replacement procedure of IP thermal paper, see Sysmex XP-300 *Instructions for Use*.
5. Power on the XP-300 main unit, and allow the analyzer to perform its automatic microprocessor tests, motor check, Autorinse and a background count. A background check is performed on the third rinse. Two additional rinses occur if the background count was unacceptable. Autorinse cycles are identified by the sample ID# of zero.
6. When the background counts of all parameters are acceptable, the analyzer is “Ready”.
7. If the counts are unacceptable “Background Error” displays and the alarm sounds briefly. Repeat the Autorinse.
8. Press **[SELECT]**.
9. Press **[5]** Autorinse.

|  |
| --- |
| **XP-300 Acceptable Background Counts** |
| **Parameter** | Count |
| WBC | 0.3 x 103/ μL or less |
| RBC | 0.02 x 106/ μL or less |
| HGB | 0.1 g/ dL or less |
| PLT | 10 x 103/ μL or less |

1. If scheduled maintenance is due, an alarm sounds and a reminder to perform maintenance displays.
2. Press **[OK]** to silence the alarm.
3. Perform the maintenance indicated by following the instructions on the screen.
4. If the maintenance is to be performed at a later time, select **[Cancel].**
5. Analyze commercial controls.
6. If the XP-300 remains in the “Ready” mode without operation for 15 minutes, the pneumatic unit shuts down and the LCD displays “PU Sleeping”. Press the Start switch to bring the unit back up to “Ready”.
7. If the vacuum and pressures are not acceptable, error messages display.
8. **Press [Maintenance]**, and **[Status Display]** to view the vacuum or pressures. Refer to the XP-300 *Instructions for Use*.
	1. Patient Sample Processing

**WARNING**: Potential biohazard exposure exists when handling open patient specimens. Follow Standard precautions outlined by laboratory safety guidelines.

**Recommended:** Wear personal protective equipment. Use plastic-lined lab wipes when opening.

1**. Whole Blood Mode (50 μL sample volume)**

1. Confirm the status display indicates “READY”.
2. The analyzer defaults to Whole Blood Mode immediately after powering on.
3. If the analyzer is not in WB Mode, press **[WB]** to select the “Whole Blood” mode.
4. Press **[SAMPLE ID]** and scan the barcode on the patient sample tube using the hand held barcode reader or using the alpha/numeric keypad, enter the patient ID number (up to 15 digits), and then press **[ENT].**

***Note****: Do not use “0” for a sample ID number, as the results are not judged against any criteria and will not print.*

1. Mix the specimen tube well, remove the stopper and hold the tube up to the sample probe.
2. Press the Start switch.
3. When the XP-300 beeps twice, remove the sample from the sample probe. Do not wipe the sample probe. Results print on the designated printer after 60 seconds.

2. **Pre-dilute Mode (200 μL sample volume of a 1:26 dilution)**

a. Prepare a 1:26 dilution using CELLPACK diluent.

1. Pipette 500 μL of CELLPACK into a test tube.
2. Add 20 μL of whole blood and mix well.

**Note:** *Because the dilution factor reduces the reliability of the differential, differential results are suppressed on capillary samples.*

1. To change to the Pre-Dilute (PD) mode, press **[PD]**.
2. Press **[SAMPLE ID.]** and scan the barcode on the patient sample tube using the hand held barcode reader or using the alpha/numeric keypad, enter the patient ID number (up to 15 digits), and then press **[ENT].**

***Note****: Do not use “0” for a sample ID number, as the results are not judged against any criteria and will not print.*

1. Mix the diluted specimen well, remove the stopper and hold the tube up to the sample probe.
2. Press the Start switch.
3. When the XP-300 beeps twice, remove the sample from the sample probe. Do not wipe the sample probe. Results will be multiplied by the dilution factor of 26 and print on the designated printer after 60 seconds.
4. Exit the Pre-Dilute mode.
	1. **Daily Shutdown.** *Approximately 5 minutes*

The Shutdown program cleans the transducer chambers and the diluted sample line. Perform Shutdown at the end of daily operation or at least once every 24 hours.

1. Press **[SHUTDOWN]**. The message **“Aspirate CELLCLEAN. It will take approx. 5 minutes”** displays.
2. Place the prepared tube of 5% Clorox up to the sample probe. Press the Start switch. Remove the tube of bleach when the unit beeps.
3. Once the Shutdown program finishes, “Turn off the power” appears.
4. Power off the XP-300.
5. Record the Shutdown on the Daily Maintenance Log.

## ROUTINE MAINTENANCE

This section includes written procedures for performing weekly, monthly, and quarterly maintenance. Refer to the Sysmex XP-300 *Instructions for Use* for detailed, illustrated procedures. During start-up of the XP-300, any maintenance that is due, based on cycle count, is displayed on the LCD.

Perform the maintenance when prompted on the LCD, and the cycle counter is reset, or press **[Cancel]** to bypass performing the maintenance temporarily. The counter does not reset and the maintenance reminder continues to reappear on start-up.

**WARNING FOR ALL MAINTENANCE**: Clorox contains a strong oxidizing agent and can cause substantial but temporary eye injury if it gets into the eyes. May irritate skin. May cause nausea and vomiting if ingested. Exposure to vapor or mist may irritate nose, throat and lungs. If contact with eyes, flush with copious amounts of water. Potential biohazard exposure exists when performing maintenance on the XP-300.

**Recommended:** Wear personal protective equipment for protection.

**IMPORTANT NOTE FOR ALL MAINTENANCE**

Clorox bleach is available with different concentrations of sodium hypochlorite. Refer to the label of the product in use for the sodium hypochlorite concentration.The Sysmex XP-300 *Instructions for Use* recommends using a 5% sodium hypochlorite solution. Clorox must be diluted to the appropriate concentration of sodium hypochlorite prior to use. The example below describes how to make a liter of 5% sodium hypochlorite solution from Clorox (6% sodium hypochlorite concentration). Store prepared 5% bleach in a dark place for up to one week to prevent solution degradation from exposure to light.

**One Liter of 5% from 6% Sodium Hypochlorite:**

(Conc. 1) x (Vol. 1) = (Conc. 2) x (Vol. 2)

(6%) x (Vol 1) = (5.00%) x (1 liter, 1000 mL)

V1 = 5.00/6 x 1000 mL

V1 = 833 mL bleach and 167 mL of distilled water will make one liter of 5% sodium hypochlorite solution.

* 1. Weekly Maintenance
		+ 1. **Clean the SRV (sample rotor valve) Tray.**

 **(See Sysmex XP-300 *Instructions for Use* for details)**

1. Open the front cover of the main unit.
2. Slide the SRV tray toward you to remove, taking care not to loosen the sample probe fixing screw.
3. Wash the tray clean with tap water and dry.
4. Replace tray.
5. Close XP-300 front cover.
6. Document on the maintenance log.

* 1. Monthly Maintenance
1. **Clean the Waste Chamber. Approximately 15 minutes.**

 **(See Sysmex XP-300 *Instructions for Use* for details)**

1. Press **[Maint]**.
2. Press **[Clean W. Chamber].**
3. Place 5% Clorox up to the sample probe.
4. Press the Start Switch to aspirate the bleach.
5. Remove the bleach tube after the beeps end.
6. After the automated cleaning and background count, the counter is automatically reset.
7. Document on the maintenance log.
8. **Clean the WBC and RBC Transducers. Approximately 7 minutes.**

 **(See Sysmex XP-300 *Instructions for Use* for details)**

1. Open the front cover of the XP-300.
2. Press **[SELECT]**
3. Press **[7]** Maintenance.
4. Press **[2]** Clean Transducer.
5. Open the transducer cover.
6. Using the “filler” provided or a syringe, place 1mL (cc) of 5% Clorox into each of the transducers. **Do not** overfill.
7. Close the transducer cover and XP-300 front cover.
8. Press the Start switch. The XP-300 automatically performs a background check, and resets the counter when the cleaning cycle is complete.
9. Document on the maintenance log.
	1. Quarterly Maintenance
* **Clean SRV once every 3 months or when “Clean SRV” appears every 4500 cycles.**

**(See Sysmex XP-300 *Instructions for Use* for details)**

* + - * 1. Prepare fresh 1:10 Clorox solution using one part 5% bleach and nine parts of distilled water.
				2. Switch off the Main unit power. Allow the pressure and vacuum gauges to reach zero.
				3. Open the front cover of the XP-300. Place paper towels beneath the SRV to absorb liquid from the valve.
				4. Remove the SRV tray.
				5. Gently pull down the rinse cup, removing it completely from the sample probe.
				6. Turn the SRV thumbscrew counterclockwise to remove it.
				7. Remove the two front sections of the SRV. **Do not** remove the rear section with all the tubing connections. Fluid “suction” holds the sections together tightly. Gently twist, and slide the sections apart.
				8. Using a lint free tissue moistened with a 1:10 Clorox solution; wipe all surfaces of all sections of the SRV to remove blood, dirt, or fingerprints. **Do not** scratch or chip the SRV sections.
				9. Rinse well with de-ionized water.
				10. Reassemble the wet sections of the SRV, placing the center section on the shaft with the flat edge toward the top of the SRV and the metal knob between the two silver stops and touching the bottom stop.
				11. Replace the front section on the shaft.
				12. Taking care not to pinch the tubing on the rear SRV section, replace the pressure fixing screw by pushing and turning ¼ turn clockwise.
				13. Install the SRV tray.
				14. Gently and slowly pull up the Manual Rinse Mechanism on the sample probe.
				15. Power on the XP-300. Check for leaks during the Autorinse.
				16. Reset the SRV counter when prompted at start up.
				17. Analyze commercial controls.
				18. Document on the maintenance log.
	1. As Needed Maintenance

 Refer to the Sysmex XP-300 *Instructions for Use* for any “As needed” procedures including:

* Cleaning the rinse cup
* Cleaning the transducer apertures
* Replacing waste container
* Replacing thermal paper
* Replacing fuses

## CALCULATIONS

* 1. When using a diluted patient specimen (1:26) and NOT using the pre-dilute mode, multiply the directly measured parameters x 26.
	2. When correcting the HGB or HCT for interfering substances, recalculate and correct the affected indices:
* MCHC = HGB/HCT x 100
* MCH = HGB/RBC x 10
* MCV = HCT/RBC x 10

## REPORTING RESULTS

1. Report results through the Laboratory Information System. Refer to the LIS Procedure Manual for complete instructions. Use the Result Worklist Template called “DIFF” to do manual diffs and verify all. The LIS has delta checks built in the system to monitor system errors. The system flags if the MCHC is >36.5 to prevent erroneous resulting. Soft will consider this result “Absurd”, if it remains above 36.5, add a canned message that it has been “rerun and/or verified to the MCHC.
2. At the LIS PC, go into **“Instrument Menu”,** choose your analyzer **YXP300**.
3. **Do not Post All** if the CBCWD needs to have a MDIFF performed. **“Partial Post”** the results to leave the WBCI, PLT and Diff parameters unverified. See below for criteria for differentials.
4. Review the scatter plot and instrument flags . Perform a smear to scan and/or perform a manual differential if indicated. See procedure *7180-HE-0180 “DIFFERENTIAL LEUKOCYTE COUNT”*.
5. **The XP300 does not autoverify patient results. Cancer Care and FEC will accept a 3 part differential from XP-300 when the main analyzer is down. OP CBC’s will require a manual 5 part differential.**
6. **Cancer Care and FEC will accept a 3 part differential from XP-300 when the main analyzer is down. OP CBC’s will require a manual 5 part differential.**
7. ***In the event that both hematology analyzers are down, specimens will be sent to RCMC for analysis.***
8. **Review the WBC and differential data:**
9. Perform a differential if there are any action flags.
To include: **%LY >60.0, %MO >20.0**
10. Refer to procedure *7180-HE-0470 “WBC ESTIMATE FROM A STAINED BLOOD SMEAR”.* Note your estimate on the printout as you review the smear. Report the WBC count matching your WBC estimate from the peripheral smear.
11. Perform a manual diff if the WBC is < 2.0 or > 25.0x10³/uL.
Dilute any WBC > 99.0 x 10³ cells/µL
Refer to procedure *7180-HE-0465 “DILUTION OF A HIGH WBC”*.
12. Erroneous Hemoglobin results can occur from leukocytosis.
13. Pseudoleukocytosis can be due to: Platelet clumps, giant platelets, unlysed RBC’s, NRBC’s, megakaryocytes, red cell inclusions, cryoproteins or circulating mucin.



**I. Review the RBC parameters:**

1. Review a stained peripheral smear for red cell morphology if there are any “**\***” or “**X**” flags. Scan a smear if HgB is <7.0g/dL. For those specimens that need microscopic review make one smear using the wedge technique. Label them with an LIS label from the **Y04\_YHemo** printer per procedure 7180-HE-0430 “LABELING SLIDES”. Send the slide through the automated slide stainer. Stained smears are kept for a 1week period in the slide storage box. RBC indices should be monitored to check for random errors.
2. Interfering substances such as in-vitro **hemolysis, lipemia, cold agglutinins, rouleaux, microclots and osmotic matrix effects** will alter the RBC parameters and may present with pseudomacrocytosis. Indices can be affected by leukocytosis. Extreme microcytosis can cause a falsely decreased RBC and Hematocrit. ***Hemolysis*** may: falsely lower RBC counts and the Hematocrit, falsely increase the Platelet Count from red cell stromas. ***Lipemia*** may: falsely elevate the Hemoglobin and MCHC ***Cold agglutinins*** may: falsely lower the RBC count falsely increase the MCV and MCHC
See procedures
* *7180-HE-0320 “PLASMA REPLACEMENT FOR INTERFERING SUBSTANCES IN HEMOGLOBIN DETERMINATION”*
* *7180-HE-0135 “COLD AGGLUTININS (Present in CBC)”*
* And for MCHC’s greater than 36.5 *7180-HE-0255 “REPORTING MCHC’s”.*

RBC’s > 7.00 x 106 cells/µL and HGB’s > 25.0 g/dL must be diluted.
Refer to procedure: *7180-HE-0360 “DILUTION OF A HIGH RBC, HGB”.* Linearity limits are posted at the instrument.

1. To improve the ability to define RBC morphology on a CBC with a HGB less than 4.0 grams, you can remove some of the specimen to another tube and spin it. Remove some of that plasma, resuspend cells and make a peripheral smear.

**J. Review the PLT parameters:**

1. Perform a platelet and WBC estimate from a stained peripheral smear if there is an “**\***” or “**X**” flag by the platelet count Refer procedure *7180-HE-0335 “PLATELET ESTIMATE FROM A STAINED BLOOD SMEAR”*.
Note your platelet estimate on the printout as you review the smear.
2. Refer to procedure *7180-HE-0200* “*EDTA PLATELET PHENOMENON”* for cases that look like platelet agglutination caused by EDTA.
3. Dilute any PLT > 999 x 10³/uL. Refer to procedure *7180-HE-0330 “DILUTION OF A HIGH PLT”*, as this is the linearity of the AcT2. All linearities are posted at instrument.
4. Review slide (Scan) to include platelet counts less than 75 and greater than 700, if there is no previous platelet count.

Report abnormal platelet morphology in LIS with the manual differential.

**K. Reportable Ranges:**

|  |  |  |
| --- | --- | --- |
| **Parameter** | **Operating Range** | **Units** |
| WBC | 0.0 - 150 | X 103 cells/µL |
| RBC | 0.00 – 8.0 | X103 cells/µL |
| HgB | 0.00 – 30.0 | g/dL |
| MCV | 50.0 – 150.0 | fL |
| PLT | 0.00 – 3000 | X 103 cells/µL |
| LY% | 0 – 100 | % |
| MO% | 0 – 100 | % |
| GR% | 0 – 100 | % |
| LY# | 0 – 99.9 | X 103 cells/µL |
| MO# | 0 – 99.9 | X 103 cells/µL |
| GR# | 0 – 99.9 | X 103 cells/µL |

|  |  |  |
| --- | --- | --- |
| **Parameter** | **Units** | **Reference Range** |
| WBC | X 103 cells/µL | 2y-10y 4-12, 10y-18y 4.8-10.818y-100y 4.8-10.8 |
| RBC | X 106 cells/µL | 0y-100y male 4.7-6.10y-100y female 4.2-5.4 |
| HgB | g/Dl | 12-18 |
| HCT | Ratio | 37-52 |
| MCV | Fl | 80-99 |
| MCH | Pg | 27-33 |
| MCHC | g/Dl | 32-36 |
| PLT | X 103 cells/µL | 130-400 |
| LY | % | 20-52 |
| MO | % | 2-10 |
| GR | % | 42-75 |

**\*\*NOTES\*\***

**MANUAL DIFFERENTIALS ARE PERFORMED ON ALL CHILDREN, 0 days through 2 years of age.**

**Buffy coat differentials do not need to be performed more than one time in a 24 hour period. Buffy coat differentials do not need to be performed on the weekends or holidays, as long as the CBC is similar to previous results. First time admits with Critical low WBC counts DO need a Buffy coat differential performed.
See Appendix A: Buffy Coat Preparation**

* 1. **ED (*work around for the physicians*):**
1. **Any CBC that has a WBC or PLT flag that requires a smear to be made to verify the WBCIR or Platelet count;**

**Partial Post the results, perform the MDIFF or PLTE/SCAN and then verify the rest.**

## PROCEDURE NOTES

## For troubleshooting specifics, refer to Sysmex XP-300 *Instructions for Use*.

1. When megakaryocytes are present, perform a WBC and PLT estimate.
2. **Do not** place samples on a mechanical rocker. Excessive mixing may induce alter white cell membranes resulting in false interpretive messages.
3. Clorox, a filtered bleach, is recommended for use in cleaning. Scented or splashless varieties of Clorox bleach must not be used at any time. If Clorox is not available, generic bleach must be 5% sodium hypochlorite concentration and be free of particles that may cause background contamination when used on the analyzer.

## LIMITATIONS OF PROCEDURE

1. XP-300 Manufacturer’s Stated Linearity (Whole Blood Mode)

|  |  |  |
| --- | --- | --- |
| **Parameter** | **Range** | **Units** |
| WBC | 1.0 - 99.9  | x103/ μL |
| RBC | 0.30 - 7.00  | x106/ μL |
| HGB | 0.1 - 25.0  | g/ dL |
| HCT | 10.0 - 60.0  | % |
| PLT | 10 - 999  | x103/ μL |

***Verification of linearity should be performed by the laboratory upon implementation of the new analyzer.***

1. Parameters that exceed these limits are flagged with an exclamation point (!) beside the result. The sample must be diluted, rerun and multiplied by the dilution factor, or repeated using the pre-dilute mode.
2. Note the use of dilution for linearity on the patient report.

## Known Interfering Substances

1. Specimens must be free of clots and fibrin strands.
2. Leukocyte aggregation may report a falsely decreased WBC.
3. Falsely increased WBCs may be caused by PLT clumps, cryoprotein, cryoglobulin, fibrin, and giant platelets (PLT > 1,000,000)
4. Falsely decreased RBC’s and HCT may be caused by RBC agglutination / cold agglutinins, microcytic RBC’s, RBC fragments.
5. Falsely elevated RBC may be caused by giant platelets (PLT> 1,000,000).
6. Falsely elevated RBC, HGB and HCT results may be caused by leukocytes > 100,000.
7. Falsely elevated HGB may be caused by lipemia, abnormal proteins.
8. Falsely elevated HCT may be caused by severe diabetes, uremia, spherocytosis.
9. Falsely decreased PLT may be caused by PLT clumps, pseudothrombocytopenia, and giant platelets.
10. Falsely increased PLT may be caused by microcytic RBC’s, RBC fragments, WBC fragments, cryoprotein, and cryoglobulin.
11. Severe lipemia falsely elevates the HGB & MCHC. Perform a plasma replacement or plasma blank procedure.
12. Severely icteric samples may falsely elevate the HGB value and related indices. Make a 1:5 dilution with CELLPACK. Rerun and correct parameter values for the dilution factor.
13. Rocking specimen excessively may affect the WBC differential.
14. The hemoglobin method on this analyzer cannot detect sulfhemoglobin, verdohemoglobin, choleglobin or other unusual degradation products of hemoglobin.

## REFERENCES

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