

TECHNICAL PROCEDURE MANUAL

COMPLETE BLOOD COUNT

NEOGEL E . FERNANDEZ
BSMT 1

TABLE OF CONTENTS

| TITLE | PAGE # |
|----------------------------|--------|
| PRINCIPLE | 1 |
| SPECIMEN REQUIREMENTS | 2 |
| MATERIALS NEEDED | 3 |
| CALIBRATION | 4 |
| QUALITY CONTROL | 6 |
| PARAMETERS | 7 |
| STEP-BY-STEP INSTRUCTIONS | 8 |
| LIMITATIONS | 10 |
| SAMPLE PROCESSING | 10 |
| RESULTS AND INTERPRETATION | 11 |
| TROUBLESHOOTING | 14 |
| POST ANALYSIS PROTOCOL | 16 |
| REFERENCES | 18 |
| LABORATORIAN'S PROFILE | 19 |

I. PRINCIPLE

A complete blood count (CBC) is a blood test that providers use to monitor or diagnose health conditions. It can give your provider information about how medications or medical conditions are affecting your body, and about the health of your immune system. It can detect blood cancers, anemia, infections and other conditions.

For a CBC blood test, a healthcare provider takes a sample of your blood and sends it to a lab. The lab measures the amount of red blood cells, hemoglobin (the protein that carries oxygen in your red blood cells), white blood cells and platelets. They also measure the size of your blood cells. Along with a CBC, your provider might order a peripheral blood smear, which gives them more information about how your blood cells look under a microscope.

II. SPECIMEN REQUIREMENTS

Type of specimen: Whole blood

Collection container: EDTA tube (lavender top).

• Volume required: 2-4 mL of whole blood.

Storage and stability: Specimens should be

processed within 2 hours of

collection. Store at 4°C if

delayed but do not exceed

24 hours.

Rejection criteria: Clotted samples.

Hemolyzed samples.

Insufficient quantity.

Samples older than 24

hours.

III. MATERIALS NEEDED

NAME

1. Tourniquet

2. Alcohol swabs

22 gauge)

- 3. Sterile needles (20–
- 4. Blood collection tubes (Lavender top, containing EDTA)
- Gauze and adhesive bandage
- Syringe or vacutainer
- 7. Hematology analyzer
- 8. Labels and requisition forms
- Biohazard waste container
- Centrifuge (if needed)

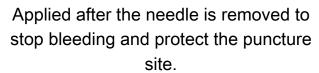
OBJECTIVE

Applied to the upper arm to make veins more prominent for easier blood collection.

Used to clean the skin at the puncture site to prevent infection.

Used to draw blood from the vein.

Prevents blood from clotting, preserving the sample for analysis.



Devices used to draw and hold the blood during collection.

Processes the blood sample to measure components like red and white blood cells and platelets.

Ensure the blood sample is correctly identified and linked to the patient for accurate analysis.

Used to safely dispose of contaminated materials like needles and swabs.

Sometimes used to separate different blood components for further testing.

IMAGE















IV. CALIBRATION

For hematology analyzers, the calibration process is performed at the time of installation and per the manufacturer's recommendations and minimally every six months as per compliance requirements. Measured parameters are evaluated in the calibration process. White blood cell differentials are not calibrated in the calibration process. Required adjustments for white blood cell differentials must be made by manufacturer representatives.

Additional conditions when the analyzer should be calibrated include:

- Following major preventative instrument maintenance or replacement of critical operating parts.
- 2. Quality control material with an unexplainable trend or outside of acceptable limits that cannot be corrected by maintenance procedures or troubleshooting the instrument.
- 3. Upon the advice of technical support or service representatives.

Calibration can be accomplished by using whole blood samples that have been assayed by reference methods or a calibrator material assayed with system-specific values traceable to a reference method. When assaying the calibrator material, replicate analyses are performed on individual whole blood calibrated hematology analyzers.

IV. CALIBRATION

Precision evaluation is incorporated into the calibration process and must be completed with successful results before performing calibration. A fresh, normal patient sample or a fresh vial of a commercial product that reflects normal patient values is evaluated. The sample is tested on the instrument 11 times. The first run is eliminated. The mean, standard deviation (SD), and coefficient of variation (CV%) are calculated, and the results are compared to the limits in the performance specifications for the analyzer. If any parameter's CV% exceeds the limits, perform troubleshooting and repeat the precision check.

The calibration procedure is performed following the manufacturer's instructions. Following an appropriate number of test runs, the mean is calculated. Compare the calculated mean to the assay target. If the results are within the assayed tolerance limits, no change in instrument settings is required. Accept the new calibration factors if the obtained calculations are close to the tolerance limits. This process and adjusting the calibration factors tell the analyzer how to read the analyte concentrations.

Following completion of the calibration process, evaluate quality control. Verify acceptable results. If post-calibration quality control is not acceptable, perform troubleshooting and repeat calibration if necessary.

Calibration verification is performed each day of testing. Successful testing of at least two levels of quality control serves as verification of calibration for automated cell counters. Achieving results within range for the quality control material verifies calibration.

V. QUALITY CONTROL

Commercial Controls

- Three levels (low, normal, high)
- Values stored in instrument computer Levey-Jennings graph generated and stored for each parameter

Mode to Mode QC

- Most automated hematology instruments have a primary and secondary mode of sample aspiration.
- Controls must be run on BOTH and correlate.
- Primary Automated or Closed Secondary Manual or Open

Delta Checks

- When the Laboratory Information System (LIS) and the instrument are interfaced (connected) delta checks are conducted by the LIS on select parameters.
- Current values compared to most previous result Differences greater than the limits set within the LIS are flagged

VI. PARAMETERS

| PARAMETER | UNIT OF REPORTING | COMMON METHOD OF DETERMINATION |
|-----------|--|---|
| WBC | X 10³ /μL | Impedance count X calibration (cal) factor |
| RBC | X 10 ⁶ /μL | Impedance count X calibration factor |
| HGB | g/dL | Colorimetric absorbance in proportion to hemoglobin |
| MCV | fL | From RBC histogram, #of RBCs X size of RBCs X cal constant OR Calculated : <u>HCT X 10</u> RBC |
| НСТ | % | Calculated: <u>RBC X MCV</u> 10 |
| MCH | Pg | Calculated: <u>HGB X 10</u> RBC |
| МСНС | g/dLor % | Calculated: <u>HGB X 100</u> HCT |
| RDW | % | Impedance (from histogram) |
| Platelet | X 10³/μL | Impedance count X cal factor |
| WBC Diff | Absolute: X10³ /μL Percent of WBC : % | Light Scatter , flow cytometry |

VII. STEP-BY-STEP INSTRUCTIONS

A. Patient Preparation:

- 1. Verify Patient Information:
- Confirm the patient's identity using two identifiers (e.g., name and date of birth).
- Ensure the test has been ordered and all information is correct.
- 2. Explain the Procedure:
- Briefly explain the CBC test to the patient, including the process of drawing blood, to ease any anxiety.
- 3. Position the Patient:
- Have the patient sit or lie down comfortably with the arm extended.

B. Sample Collection:

- 1. Prepare the Equipment:
- Place all materials within reach.
- Apply gloves and prepare the blood collection tube with EDTA anticoagulant.
- 2. Wrap the Tourniquet:
- Wrap the tourniquet 2–3 inches above the puncture site to make veins more prominent.
- Ask the patient to make a fist.

VII. STEP-BY-STEP INSTRUCTIONS

3. Select the Venipuncture Site:

- Prefer the antecubital vein (median cubital vein).
- Clean the area with an alcohol swab in a circular motion, starting from the center outward.

4. Perform Venipuncture:

- Insert the needle into the vein at a 15-30° angle and bevel up.
- Once the blood starts flowing, attach the collection tube.

5. Collect the Blood Sample:

- Allow the tube to fill the indicated line (typically 2–5 mL).
- Gently invert the tube 8–10 times to mix with the anticoagulant and prevent clotting.

6. Remove the Needle and Apply Pressure:

- Once the collection is complete, remove the needle, release the tourniquet, and apply gauze pressure immediately.
- Dispose of the needle in a receptacle designated for sharps.

7. Label the Sample:

- Label the tube with the patient's information and the time of collection.
- Place the tube in a transport bag.

8. Provide Aftercare:

- Apply a bandage to the puncture site.
- Instruct the patient to clean the area and avoid heavy lifting for a few hours.

VIII. LIMITATIONS

- Clotted or hemolyzed samples may give inaccurate results.
- Samples stored at room temperature for more than 24 hours may lead to cell degradation and erroneous counts.
- High WBC counts or platelet clumping may require manual verification.

IX. SAMPLE PROCESSING

1. Transport:

- Transport the labeled blood sample to the laboratory at room temperature.
- Avoid extreme temperatures or delays longer than 24 hours to ensure sample integrity.

2. Preparation for Analysis:

- Inspect the sample for clots. Discard if clotted.
- If needed, centrifuge the sample to separate plasma from cells (most CBCs do not require this step).

3. Loading the Sample into the Analyzer:

- Load the sample into the hematology analyzer according to the manufacturer's instructions.
- The analyzer will measure RBCs, WBCs, hemoglobin, hematocrit, and platelets.

X. RESULTS & INTERPRETATION

A CBC is performed using an automated instrument that measures the numbers of RBCs, WBCs, and platelets per unit volume. In addition, the RBC indices are generated. A standard complete blood count is performed on an automated laboratory instrument that quantitates the amount of Hgb as well as the size, shape, and number of RBCs. A variety of calculations are performed to produce indices that provide information about RBC disorders. The standard indices are:

- RBC Count: The number of RBCs per unit volume is measured directly and given in millions per microliter.
- Mean Corpuscular Volume (MCV): The MCV is measured directly.
 The unit is a femtoliter (fL), or it can be given as cubic millimeters
 (mm3). The MCV measures the size of RBCs and is the most
 important index for classification of anemias into "macrocytic" with
 higher than normal MCV and "microcytic" with low MCV.
- Hemoglobin (Hgb): The Hgb content is measured directly and given in grams per deciliter (g/dL). This value, along with Hct, provides the most useful measure of the oxygen carrying capacity of the blood.
- Hematocrit (Hct) in % = (RBC count in millions X MCV) ÷ 10: The Hct
 is the packed cell volume and is a calculated value and provides a
 measure of the amount of oxygen carrying capacity in relation to
 blood volume.

X. RESULTS & INTERPRETATION

- Mean Corpuscular Hemoglobin (MCH) in pg = (Hgb X 10) ÷ RBC count in millions: The MCH is calculated and gives the average mass of Hgb in an individual RBC; the unit is a picogram (pg).
- Mean Corpuscular Hemoglobin Concentration (MCHC) in g/dL = (Hgb X 100) ÷ Hct: The MCHC is calculated and provides a measure of the concentration of Hgb in the cells in g/dL.
- Red Cell Distribution Width (RDW) = standard deviation of MCV: The RDW is calculated to provide a measure of the anisocytosis, or variation in size of the RBCs.

1. Review Results:

- Ensure that results are within the acceptable range for quality control.
- Any abnormal results should be flagged for further review by a physician.

2. Document Results:

- Record the results in the patient's medical record.
- Ensure timely delivery of the results to the ordering physician.

3. Critical Values:

- Immediately notify the healthcare provider if critical values are detected (e.g., dangerously low platelets or high WBC count).

X. RESULTS & INTERPRETATION

Red Blood Cell (RBC) Components

| Component | Normal Range (Male) | Normal Range (Female) | | |
|--------------------------------------|-----------------------------|-----------------------------|--|--|
| RBC Count | 4.7 - 6.1 million cells/mcL | 4.2 - 5.4 million cells/mcL | | |
| Mean Corpuscular Volume (MCV) | 80 - 95 fL | 80 - 95 fL | | |
| Mean Corpuscular Hemoglobin (MCH) | 27 - 31 pg | 27 - 31 pg | | |
| MCH Concentration (MCHC) | 32 - 36 g/dL | 32 - 36 g/dL | | |
| Red Cell Distribution Width (RDW) | 11 - 15% | 11 - 15% | | |
| Hemoglobin | 13.8 - 17.2 g/dL | 12.1 - 15.1 g/dL | | |
| Hematocrit | 40.7% - 50.3% | 36.1% - 44.3% | | |
| RUPA HEALTH | | | | |

XI. TROUBLESHOOTING

| ISSUE | |
|-------|--|
|-------|--|

ACTION

Hemolysis

Discard and recollect if the sample appears hemolyzed (pinkish).

Clotted Sample

Ensure proper mixing with EDTA; if clotted, discard and recollect.

Analyzer Error

Recalibrate or rerun the sample if there are issues with the machine.

No result or flagged result

Check for clots in the sample. Recollect if necessary.

Inconsistent QC results

Recalibrate the instrument and repeat the QC run.

Low platelet count with no clinical correlation

Check for platelet clumping; consider using citrate as an anticoagulant.

XII. POST-ANALYSIS PROTOCOL

1. Review Results

- Carefully examine the generated report for completeness and accuracy.
- Look for any flagged or abnormal results that require further attention.

2. Document Findings

- Ensure all results are accurately documented in the laboratory information system (LIS) or electronic health record (EHR).
- Include any relevant notes on the sample quality or any anomalies encountered during testing.

3. Communicate Results

- Notify the requesting physician or healthcare provider of critical values immediately.
- Provide a detailed explanation of any abnormal results, especially if they deviate significantly from normal ranges.

4. Storage of Specimens

- Store any remaining specimens as per institutional protocols. Whole blood samples may typically be retained for 24 hours for potential repeat testing.
- Label specimens clearly with patient information, date, and time of collection.

XII. POST-ANALYSIS PROTOCOL

5. Clean Up

- Dispose of used materials (e.g., gloves, pipettes, and slides) in accordance with biosafety guidelines.
- Clean and disinfect work surfaces to maintain laboratory safety and hygiene.

6. Quality Assurance

- Review and log any quality control measures taken during the testing process.
- Ensure that all QC data is recorded and any discrepancies are addressed.

7. Follow-Up Actions

- Prepare the appropriate specimens and reagents if abnormal results necessitate further testing (e.g., reticulocyte count, iron studies).
- Schedule follow-up appointments or consultations with the patient as directed by the healthcare provider.

XIII. REFERENCES

Clinical and Laboratory Standards Institute (CLSI). *Reference and Selected Procedures for the Quantitative Determination of Hemoglobin in Blood; Approved Standard*. CLSI Document H15-A3. Clinical and Laboratory Standards Institute, 2018.

Klatt, E. C., MD. (n.d.). Laboratory Medicine curriculum. Edward C. Klatt MD. https://webpath.med.utah.edu/EXAM/LabMedCurric/LabMed04_01.html

McKenzie, S. B., & Williams, L. S. *Clinical Laboratory Hematology*. 3rd ed., Pearson Education, 2014.

Rodak, B. F., et al. *Hematology: Clinical Principles and Applications*. 5th ed., Elsevier, 2020.

Sysmex Corporation. *Sysmex XN-Series Automated Hematology Analyzer Instructions for Use*. Sysmex, 2021.

World Health Organization (WHO). *Laboratory Manual for the Examination and Processing of Human Semen*. 6th ed., WHO, 2021.

XIV. LABORATORIAN'S PROFILE

NAME: NEOGEL LOURENZE E. FERNADNEZ

COURSE: BACHELOR OF SCIENCE IN MEDICAL TECHNOLOGY

SCHOOL: SILLIMAN UNIVERSITY

SIGN:

DATE: OCTOBER, 14, 2024