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The CBC

click here to continue
Special thanks is given to Ms. Angela Foley, MS, MT(ASCP), Department of Clinical Laboratory Sciences, LSUHSC School of Allied Health in New Orleans, LA for the use of some of her images of blood cells and for her assistance in the art of creating image files.
This is the third module of a 4-part study exercise regarding the CBC. The 4 parts are entitled:

CBC – Part 1  The hemogram
CBC – Part 2  WBC differential & blood morphology
CBC – Part 3  RBC morphology & platelet estimate
CBC – Part 4  Post-test

The review of hematopoiesis and blood cell morphology (i.e., “Tutorial: Blood Cell Morphology” located on the LSUHSC “M Drive” server) is recommended as a prerequisite for the CBC exercise.
Feedback

Feedback as to the quality and usefulness of this exercise is solicited and suggestions for improvement are welcomed. Please forward your remarks by e-mail cwalte@lsuhsc.edu

or via US MAIL:

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The directions for navigating through the exercise are given on the next 3 pages. They are the same as those used in the other modules of this 5-part exercise. Click on:

to visit the directions before continuing with the exercise.

or

to go directly to the Main Menu.
The following directional icons are provided throughout the exercise for your convenience. You can click on:

- in the upper left hand corner of every page to return to the previous page
- in the upper right corner of the page to return to the Main Menu selection.

click here to continue
You can click on:

in the **lower right** corner of the page to continue.

in the **lower right** corner of the **Main Menu** page to **Quit** (i.e., end the exercise).
“Hot points” (symbols, words, phrases) have been inserted on the pages as navigational tools and can be identified by their “gold” color. If it’s “gold”, click on it to move to the next text/data entry. Also, sounds have been added in a few places for emphasis.

**Caution**, failure to follow the structured order of the “hot points” may result in confusion. If you use the mouse without placing the cursor directly on the “hot point”, you may skip over vital information.

Remember, if it’s **gold**, click on it. Try it!
Special Comments

This exercise has numerous images. You may note that, when a page contains images, there may be a rather long delay before you regain control of the cursor. Please be patient. I think you will find the images are worth the wait.

NOTE:

Some animation and/or interactive affects may be lost if you attempt to replay a page by returning to the previous page and then advancing to that page again.

Now, click on the gold to begin.
The Red Blood Cell Morphology and Platelet Estimate
Introduction

CBC-3 is designed to aid in evaluating red blood cell and platelet morphology; differentiating normal and abnormal cells; identifying abnormal and/or immature forms; selecting or interpreting terminology used to indicate the presence of abnormal cell morphology; and estimating the adequacy of a platelet population.
Red Blood Cell (RBC) Morphology
Introduction

Remember, in addition to a differential WBC count, the “diff” will include:

A description of RBC morphology which includes (if any) a description of RBC morphologic alterations and, if present, the number of NRBC per 100 WBC.
Normal mature red blood cells:

Normal mature RBC are biconcave, round discs that are about 6 - 8 μ in diameter, which is only slightly smaller than the normal small mature lymphocyte (about 6-10μ in diameter).

The term used to indicate red blood cells of normal size and shape is normocytic. The term used to indicate a normal color or central pallor (i.e., normal hemoglobin content) is normochromic.
Immature red blood cells in normal peripheral blood:

Polychromatophilic erythrocytes are anucleated slightly immature red blood cells that may be found in small numbers (0.5-1.5%) in normal peripheral blood. They have a faint bluish-grey tint and are usually slightly larger than a mature RBC.

When present in increased numbers, a comment is made as a part of the “diff” report.
Are nucleated red blood cells normally seen in peripheral blood?

Immature nucleated red blood cells (NRBC) are not normally seen in adult blood. However, they may be seen normally in newborns and abnormally in disease.

When present, the number observed per 100 WBC is noted and used to correct the total WBC count.
Abnormal RBC are differentiated and identified as part of the “diff”.

Changes in size, shape, hemoglobin content, and/or appearance of cellular inclusions may occur as a result of a disease process. Such changes are noted as part of the “diff”.

What terminology is used to indicate the presence of abnormal red cells?
**Definitions:**

**Hypochromic** erythrocytes that demonstrate a central pale area that becomes larger and paler as the hemoglobin content diminishes.

- **normochromic**
- **hypochromic**
Anisochromic or dimorphic indicates the presence of both normochromic and hypochromic cells in the same blood film.

Definitions:

- Normochromic: cells with normal coloration.
- Hypochromic: cells with reduced coloration.
- Anisochromic or dimorphic: cells with variable coloration.
Polychromasia and polychromatophilia are interchangeable terms used to indicate the increased presence of non-nucleated immature erythrocytes (polychromatophilic erythrocytes) that contain residual RNA which gives a blue-gray tint to the red cells. These cells, which remain after ejection of the nucleus from the orthochromatic erythroblast, are slightly larger than mature erythrocytes. After exposure to a supravital stain, the cytoplasmic organelles of these cells clump into an easily recognized blue-staining reticulum and the cell is called a reticulocyte.
continued:

**Polychromasia and polychromatophilia**

- **normochromic**
- **polychromasia** (polychromatophilia)
**Definitions:**

**Microcytes** are abnormally small erythrocytes (i.e., less than 6 μm in diameter).

- Lymphocytes (≈ 6-10 μm in diameter)
- Normocytic RBC ≈ 6-8μm diameter
- Microcytic (predominant)
**Definitions:**

**Macrocyes** are abnormally large erythrocytes (i.e., greater than 8 \( \mu \text{m} \) in diameter).

**normocytic RBC** \( \beta \) 6-8\( \mu \text{m} \) diameter

**macrocytic**

**lymphocyte** (6-10 \( \mu \text{m} \) in diameter)
Anisocytosis is a “generic” term used to indicate an abnormal variation in size of erythrocytes.

Definitions:

- **normocytic**
  - RBC β 6-8 μm diameter

- **normocytic microcytic**

- **macrocytic**
Poikilocytosis is a “generic” term used to indicate variation in shape of erythrocytes (e.g., oval, pear-shaped, teardrop-shaped, saddle-shaped, helmet-shaped, sickle-shaped, and irregularly shaped).

Definitions:

- **Poikilocytosis**
- **Normocytic**
- **RBC round biconcave**
- **RBC variable shapes**
Elliptocytes and Ovalocytes are interchangeable terms used to indicate oval-shaped erythrocytes.

Definitions:

normocytic RBC round biconcave

RBC predominantly ovalocytes
Definitions:

**Spherocytes** are nearly spherical erythrocytes which are nearly spherical erythrocytes which usually have a diameter smaller than normal. They lack the central pale area due to their **spherical** shape.

round, biconcave RBC

normocytic
Target cells (leptocytes) are erythrocytes that are thinner than normal which show a peripheral rim of hemoglobin with a dark central hemoglobin-containing area. A pale unstained ring containing less hemoglobin separates the central and peripheral zones and gives the cell a target appearance.

Definitions:
Schistocytes are fragmented red cell segments that are the result of some hemolytic process. The segments can be a variety of shapes but helmet cells and triangularly-shaped cells are particularly characteristic.
Sickle cells (drepanocytes, meniscocytes) are interchangeable terms used to indicate sickle-like forms of erythrocytes (crescent-shaped, irregular spines, filaments, holly-leaf appearance) noted when RBC containing HbS are subjected to reduction in oxygen tension or pH.
Definitions:

**Keratocytes or “Bitocytes”** interchangeable terms used to indicate irregularly contracted erythrocytes which stain densely and have contraction of hemoglobin from a part of the cell membrane, thereby giving the appearance that a “bite” has been taken out of the cell. These cells are thought to be cells from which Heinz bodies have been removed by the spleen.

normocytic RBC, round, biconcave

keratocytes
Definitions:

**Acanthocytes** are irregularly shaped red cells with spiny or thorny projections and dark centers which may be found in severe liver disease, infantile pyknocytosis (with underlying hemolytic process), abetalipoproteinemia, or anorexia nervosa.

- **normocytic**
- **RBC round biconcave**
- **acanthocytes**
Crenated red blood cells are uniformly shrunken red cells with uniform irregular, wrinkled cell membranes. Their presence is an artifact of storage and all red cells in the field are usually affected. (By contrast, ecinocytes are intermixed with normal red cells.)

Definitions:

- Crenated red blood cells
- Normocytic RBC: round, biconcave
- Crenated RBC
Echinocytes are irregularly shaped red cells with spiny projections and preserved central pallor. While their presence is usually an artifactual phenomenon, they may be seen in liver and renal disease, hyperlipidemia, and red blood cell enzymopathies.
Definitions:

Rouleaux formation describes an aggregation of erythrocytes that are aligned one upon the other, resembling stacks of coins, caused by elevated plasma fibrinogen or globulins. This phenomenon causes an increased erythrocyte sedimentation rate. This finding is especially characteristic of paraproteinemia (monoclonal gammopathy).
Definitions:

**Agglutination of red cells** is caused by agglutinins and resembles rouleaux but is more irregular with round clumps rather than linear rouleaux.
**Definitions:**

**Hemoglobin C crystals** are hexagonal crystals that may be found in individuals with HbC syndromes. The crystals may be intracellular or extracellular.

**Normocytic RBC** (round, biconcave, without inclusions)

**Intracellular HbC crystals**

**Extracellular HbC crystal**
Basophilic stippling is the term used to indicate the presence of irregular basophilic granules in the cytoplasm of erythrocytes. The granules are composed of unstable RNA and may be fine or coarse.
**Definitions:**

**Pappenheimer bodies** are intracellular inorganic iron-containing granules that may be observed on Wright’s stained peripheral blood smears in iron-loading disorders. When the inclusion bodies are demonstrated by stains for iron (e.g., Prussian Blue), the cells are called siderocytes.

- **normocytic**
- **Pappenheimer bodies** *(Wright stain)*
- **siderocytes** *(Prussian Blue stain)*

*(image pending)*
Definitions:

Howell-Jolly bodies are intracellular particles which are smooth, round remnants of nuclear chromatin (DNA). Usually, only one per cell is seen but, occasionally, there may be more than one.

Howell-Jolly body (single)

Howell-Jolly body (multiple)

image pending

normocytic
Nucleated red blood cells (NRBC) are precursors of the non-nucleated mature red cells, usually orthochromatophilic erythroblasts when noted in peripheral blood in disease states but earlier forms may also be seen.

Definitions:
- **mature RBC**
- **orthochromatophilic erythroblast**
- **polychromatophilic erythroblast**
- **basophilic erythroblast**
- **proerythroblast**
Reticulocytes are anucleated slightly immature erythrocytes, identified as polychromatophilic erythrocytes on Wright stained smears.

The cells are identified as reticulocytes only after exposure to a supravital stain which causes the cytoplasmic organelles of the cells to clump into an easily recognized blue-staining reticulum.
What is the clinical significance of an abnormal number of reticulocytes?

A decreased number of reticulocytes (i.e., <0.5% of circulating red blood cells) is an indication that the bone marrow is no longer functioning properly and fewer than normal erythrocytes are being placed into circulation (e.g., aplastic anemias).

An increased number of reticulocytes (i.e., >1.5% of circulating red blood cells) or reticulocytosis, is an indication that the bone marrow has responded to an increased demand for release of erythrocytes into circulation (e.g., acute blood loss).
Heinz bodies are erythrocytic inclusions which may be detected only after supravital staining. The inclusions are precipitates of denatured hemoglobin which characteristically adhere to the red cell membrane.

If the inclusions are removed from the cells by the spleen, the cells are then called keratocytes (or “bitocytes”) on Wright’s stained smears.
Can you define the following?

Siderocytes, as previously states, are red blood cells containing intracellular iron granules. When seen on Wright’s stain, they are called normal RBC.

Only after exposure to an iron stain (e.g., Prussian blue) are the cells called siderocytes.
When are increased numbers of siderocytes seen?

Siderocytes may be found in large numbers when hemoglobin synthesis is impaired (as in siderochrestic anemias) and after splenectomy. They may also be seen in iron-loading anemias, hyposplenism, and in hemolytic anemias of various etiologies.
Can you define the following?

**Sideroblasts** are nucleated erythrocytes that contain siderotic (iron) granules which can be demonstrated by special iron stains (e.g., Prussian blue).

When the siderotic granules surround the nucleus (at least two thirds of the circumference), the cell is called a “ring” sideroblast.
When are abnormal numbers of sideroblasts seen?

Decreased numbers of these cells in bone marrow indicate depleted iron stores (e.g., iron deficiency anemia).

Increased numbers indicate increased iron storage which may be associated with defective synthesis of heme and may be inherited (e.g., hereditary sideroblastic anemia) or acquired (e.g., secondary toxin-induced sideroblastic anemia caused by lead poisoning).
Can you ID abnormally shaped RBC?

- Ovalocytes
- Target cells
- Spherocytes
- Tear drop
- Crenated
- Schistocytes
- Keratocytes "bitocytes"
- Acanthocytes "spiculated"
- Sickle cell
Can you ID RBC with inclusions?

- HbC crystals
- basophilic stippling
- Howell-Jolly body
- Pappenheimer bodies
- Heinz bodies
- Cabot rings
End of Red Blood Cell (RBC) Morphology

This concludes the Introduction to the Differential WBC Count. Select one of the following:

Go to Platelet Estimate, the next section, to with the exercise as designed.

OR

Return to the Main Menu and make an alternate selection.
Platelet Estimate
A platelet count is the number of platelets per unit of blood (e.g., per \( \mu L \) or per L) and an absolute reference range has been established to determine decreases or increases in the number of circulating platelets. The count is obtained by an actual count (manual or electronic) of the platelets in a sample of blood.
Platelet Estimate

A platelet estimate is performed to verify abnormal platelet counts and/or when a platelet count is unavailable. It is made by a visual count of platelets seen on a microscopic examination of a Wright’s or Wright-Giemsa stained blood smear.
How is the platelet estimate made?

Provided the blood smear is properly prepared with an even distribution of platelets, an estimate of the number of platelets can be made by determining the number of platelets per oil field (100x objective) in the feather edge section of the smear (where there is a one cell thickness of evenly distributed RBC).
What is a normal platelet estimate?

Each platelet on the blood smear correlates to about 15,000 to 20,000 platelets per μL of whole blood. Generally, an average number of 5 to 25 platelets per oil field is considered an adequate number of platelets. Fewer than 5 is considered decreased. More than 25 is considered increased.

An estimate is valid only if the blood smear has been properly prepared so that the distribution of platelets is consistent throughout the smear without clumping or other interfering factors.
Example Platelet Estimate

If this field is a representative oil field on the blood smear, there is an average of about 5 platelets/field (a borderline low platelet population).
Example Platelet Estimate

If this field is a representative oil field (a patient with megaloblastic anemia), there is an average of > 25 platelets/field or slightly increased with a giant platelet.

The RBCs are fewer on this smear (and spread thinner) because the patient is anemic.
By the way, what feature suggests megaloblastic anemia?

In addition to red cells that look larger than normal when compared to the size of the neutrophil in the field, the neutrophil is hypersegmented (> 5 lobes) with seven lobes noted.

Hypersegmented neutrophils are a characteristic feature of megaloblastic anemias.
Abnormal Platelet Morphology

Manual examination of a stained blood smear is also helpful when platelet abnormalities are present (e.g., morphology, clumping).

Platelet counts will be unreliable when there is invivo or invitro clumping of platelets. The platelets in clumps will not be included in the platelet count. Giant platelets (i.e., those that exceed the size setting for platelets) will also be excluded from the count.
What kind of platelet abnormalities may be seen on blood smears?

Manual examination of a stained blood smear is also helpful when platelet abnormalities are present (e.g.):

- giant platelet
- platelet satellite
- platelet clump
This concludes the Platelet Estimate section. Select one of the following:

Go to **The End** to quit the program.

OR

Return to the **Main Menu** and make an alternate selection.
End of CBC - 3

This concludes the CBC-3 segment of the study tutorial, “The Complete Blood Cell Count (CBC)”.

Click on return to repeat CBC-3.

OR

Click on to quit the exercise.
The Complete Blood Cell Count (CBC) tutorial has 4 parts:

CBC-1   The hemogram
CBC-2   WBC differential & blood morphology
CBC-3   RBC Morphology & Platelet Estimate
CBC-4   Post-Test (self evaluation)

Return to the Lecture folder to select Part 1, 2, or 4 of the CBC tutorial.