ADVANCED BLOOD CELL ID: PERIPHERAL BLOOD CELLS AND ARTIFACTS IN A CASE OF ACUTE LYMPHOBLASTIC LEUKEMIA

Educational commentary is provided for participants enrolled in program #259- Advanced Blood Cell Identification. This virtual blood cell identification program includes case studies with more difficult challenges. To view the blood cell images in more detail, click on the sample identification numbers underlined in the paragraphs below. This will open a virtual image of the selected cell and the surrounding fields. If the image opens in the same window as the commentary, saving the commentary PDF and opening it outside your browser will allow you to switch between the commentary and the images more easily. To avoid the need to log in for each image, use the online tool to choose the cell you want to view. Click on this link for the API ImageViewer™ Instructions.

**Learning Objectives**

After completion of this testing event, participants will be able to:
- Describe morphologic characteristics of normal peripheral blood leukocytes.
- Identify morphologic features of immature leukocytes.
- Discuss distinguishing morphologic characteristics of peripheral blood artifacts.

**Case History**

A CBC with differential was ordered on a 23-year-old female seen at the hematology-oncology clinic. Her CBC results are as follows: WBC=28.9 x 10⁹/L, RBC=2.45 x 10¹²/L, Hgb=7.4 g/dL, Hct=22.1%, MCV=90.2 fL, MCH=30.2 pg, MCHC=33.5 g/dL, Platelets=8 x 10⁹/L, RDW-CV=15.6%.

**Educational Commentary**

The patient presented in this exercise is a 23-year-old woman evaluated at the hematology-oncology clinic for a recurrent elevated WBC count, anemia, and thrombocytopenia. It was subsequently determined that she was in relapse from a previously diagnosed T-cell acute lymphoblastic leukemia (ALL). The cells selected for commentary include normal mature white blood cells, but also immature leukocytes and artifacts seen in her peripheral blood smear.

The cell annotated in ABI-15 is a band neutrophil. Band neutrophils are usually medium-sized cells and represent an immature stage of neutrophil development immediately before the segmented neutrophil. Bands are the earliest precursor of neutrophil maturation that can normally be seen in the peripheral blood. Note that this particular band cell is larger than usual. Band nuclei are distinctive in their shape, which may appear like a band, a sausage, or the letters C, U, or sometimes S. The indentation of the nucleus is greater than half the distance of a hypothetically round nucleus. Since bands are maturing cells, the chromatin is dense and clumped. The band selected here is a classic example of this cell.
ADVANCED BLOOD CELL ID: PERIPHERAL BLOOD CELLS AND ARTIFACTS IN A CASE OF ACUTE LYMPHOBLASTIC LEUKEMIA (cont.)

However, sometimes the nucleus is folded and twisted such that it can be difficult to identify the cell as a band. True band cells do not have any nuclear filaments. The cytoplasm in band neutrophils generally has numerous pink or tannish specific granules. This band is also not typical in that several darker purple primary or nonspecific granules have been retained.

**ABI-16** identifies a blast. Blasts should not be seen in the peripheral blood. However, it is expected that this type of cell be observed in a case study situation when the diagnosis is ALL. Blasts are typically large cells with a high nuclear to cytoplasmic ratio and a moderate to scanty amount of blue cytoplasm, features all seen in this cell. Blasts are generally oval or round cells with oval to round nuclei. The nuclear chromatin is loose and open. Some blasts will have one or more prominent nucleoli. Notice the large nucleolus in the upper left area of the nucleus in this blast. It is also important to note that the lineage of this blast was determined using additional testing methods, such as immunophenotyping. Blasts of different cell lines share similar characteristics, so it is often difficult to classify these cells based only on morphology. Morphology, however, is critical in helping to identify at least the presence of immature cells such as blasts.

**ABI-17** is a lymphocyte. Lymphocytes are variable in size and this cell is a small lymphocyte. The cell may be round or oval. The nucleus may also be round or oval or, sometimes, slightly indented. The nuclear chromatin is condensed and clumped. The cytoplasm in small lymphocytes is typically blue and scanty.

The annotation for **ABI-18** is not a cell at all, but a leukocyte remnant called a smudge cell. These artifacts result during the process of blood smear preparation when fragile cells, most often lymphocytes, degenerate or disintegrate. The size of smudge cells varies and may range from about the size of a lymphocyte to much larger. The chromatin has no or little detail, is homogeneous, and does appear spread out or smudged. There is no cytoplasm. Sometimes, strands of chromatin can be seen, as in this cell. In other cases, the wisps of chromatin fan out from the central smudge and these artifacts can be called basket cells. Because no cytoplasm remains, these cells cannot be classified. However, the presence of significant numbers of smudge cells should be reported. Smudge cells are typically seen in conditions associated with increased lymphocytes and lymphocyte fragility, such as in chronic lymphocytic leukemia. But, when scanning this virtual slide with ImageViewer, it is not unexpected to see a few other smudge cells in this case of ALL. Note that smudge cell formation can be prevented by adding a drop of bovine serum albumin to 4 or 5 drops of patient blood before making the slide.

The cell selected in **ABI-19** is a monocyte. Monocytes are the largest cells that can normally be seen in the peripheral blood. The nuclei in monocytes may be round, oval, indented or, as in this cell, lobulated. The chromatin shows only minimal clumping and no nucleolus is visible. Monocytes have abundant,
blue-gray cytoplasm. The cytoplasm typically appears grainy or uneven. Sometimes vacuoles or faint pink or red-purple azurophilic granules are present. This monocyte does have a few vacuoles and just a hint of pinkish granulation above the nucleus.

The annotation in ABI-20 is identifying a pyknotic cell or cell with a pyknotic nucleus. Pyknotic cells are seen when the smear is prepared from an old blood sample or when the cell is in the process of necrosis. Their overall size is variable and they are characterized by one or more various-sized nuclear fragments. These nuclear remnants are dense, compact, round or oval, and completely lack any distinctive parachromatin. In contrast to the smudge cell in ABI-18, pyknotic cells do retain cytoplasm. Sometimes the cytoplasm may give a clue as to the identity of the dying cell, especially if granules are present to suggest a neutrophil. The deep blue cytoplasm of this pyknotic cell and the increase in lymphocytes and lymphoblasts may indicate that this particular cell is lymphoid in origin. However, identifying blast cells is contingent on assessing nuclear features to include chromatin structure and the presence of nucleoli. Therefore, it is appropriate that this particular pyknotic cell not be counted or reported.

The last cell for commentary, ABI-21, is another blast. As with the blast in ABI-16, this cell is large, is slightly oval, and has a large, oval nucleus. The cytoplasm is blue and scanty. Likewise, the nuclear chromatin is loose and open. A nucleolus is also present, though it is not quite as prominent as the one seen in ABI-16.

**Acute Lymphoblastic Leukemia**

Acute lymphoblastic leukemia (ALL) is a malignancy associated with an uncontrolled production of lymphoblasts. The lymphoblasts are usually either T or B cells. The specific lineage of the lymphoblasts cannot be determined morphologically and immunophenotyping is needed to classify the cells. It is important to not only identify lymphoblasts, but also to categorize the blasts as T or B cell because treatment and prognosis vary depending on the type of lymphoblast. Patients with T-cell ALL are typically adolescents or young adults, as in this case study situation. Anemia and thrombocytopenia may also be seen in ALL as demonstrated in this patient. The normal production of red blood cells and platelets is inhibited by the increased number of abnormal cells that overcrowd the bone marrow.

**Summary**

The peripheral blood features discussed in this testing event are from a patient who presented with a relapse of a previously diagnosed ALL. The cells and artifacts annotated for commentary represent both normal and abnormal peripheral blood findings that may be seen in this condition. Although additional laboratory procedures were necessary to establish the original diagnosis and to confirm the patient relapsed with ALL, recognizing blasts in the peripheral blood is an important first indicator that an abnormality is present.
Bibliography
