ADVANCED BLOOD CELL ID: PERIPHERAL BLOOD FINDINGS IN A CASE OF MALARIA

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. Click on this link for the API ImageViewer™ Instructions.

Learning Outcomes
After completing this exercise, participants should be able to:

- describe morphologic characteristics of peripheral blood lymphocytes and granulocytes.
- identify morphologic features of peripheral blood platelets.
- distinguish malarial parasites from platelets on a peripheral blood smear.

Case Study
A 44 year old female was seen by her physician for fever, headache, and nausea. The CBC results are as follows: WBC=4.6 x 10^9/L, RBC=4.25 x 10^{12}/L, Hgb=11.9 g/dL, Hct=34.2%, MCV=80.5 fL, MCH=28.0 pg, MCHC=34.8 g/dL, RDW=40.7%, Platelet=71 x 10^9/L.

Educational Commentary
The cells and organisms annotated for discussion in this advanced testing event were selected from the blood smear of a patient diagnosed with malaria. Malaria is a worldwide infection caused by any one of four species of Plasmodium. This disorder is usually diagnosed by identifying the organism on stained blood smears.

Cell ABI-15 is a lymphocyte. Lymphocytes vary in size, but this example represents a normal, small cell. Small lymphocytes are generally round or oval and have a thin rim of blue cytoplasm. The nucleus is usually also round or oval with dense and clumped chromatin.

The image selected for ABI-16 shows a platelet. Platelets actually represent fragments from a much larger bone marrow cell, the megakaryocyte. Though anucleated, platelets are nevertheless still considered cells because of their origin from megakaryocytes, which are nucleated. Platelets are often small, although this example is slightly bigger than usual and may be described as appearing large or enlarged. Larger platelets may indicate those that are younger and have been recently released from the bone marrow. Platelets stain a light purple or blue-gray. They are generally round or oval. Often, a
central granular core surrounded by a less dense halo may be seen. In some platelets, such as this example, the cellular margins may appear frayed and uneven.

Image **ABI-17** identifies the ring form of a malarial parasite inside a red blood cell. Sometimes peripheral blood smears must be reviewed carefully to locate malarial inclusions. Note the chromatin dot and fine blue or purple “ring.” The chromatin dot in this particular organism is not as red or distinct as is often seen. Nevertheless, the clear “ring” defines this inclusion as a malarial parasite. Also notice that the infected erythrocyte has retained its normal size and shape.

The red blood cell in **ABI-18** shows another example of the ring form of a malarial parasite. Observe again the delicate ring and chromatin dot that are clearly an abnormal inclusion in this erythrocyte. Although the chromatin circle in this organism is still not as red as would be expected, the dot is more defined than in the organism shown in ABI-17. Variations in coloration of these RBC inclusions most likely represent differences in the stain quality of the smear.

Image **ABI-19** shows a platelet overlying a red blood cell. This superimposed position of platelets can often be mistaken for the ring form of a malarial parasite. However, a careful and systematic microscopic review of a peripheral blood smear provides clues to differentiate such artifacts from true malarial inclusions. Superimposed platelets will focus separately from the RBCs whereas organisms are intracellular and focus with the cell. Also, platelets are granular and light purple or blue-gray, with no clearly defined ring. Scanning the smear can also show extracellular platelets that resemble the superimposed platelet in regards to cell color, size, and shape.

The cell selected for **ABI-20** is a band neutrophil. Band cells are immature precursors of neutrophil maturation and a small percentage (up to about 6%) may normally be seen in the peripheral blood of an adult such as this case study patient. Increased numbers may also be associated with infection. Bands are medium-sized cells that are usually round. Their cytoplasm shows numerous pink, violet, or tan granules, though the granules in this cell appear slightly darker. However, a review of adjacent red blood cells indicates they appear more bluish than usual as well. This observation suggests that the overall stain is more blue than preferred.

Band cells are characterized by a nucleus that is shaped like a band or the letters C or U. The lobes are connected by a bridge of chromatin with no indication of constriction. As bands are nearly mature, it is expected that the nuclear chromatin is dense and clumped.
The last cell chosen in this testing event, **ABI-21**, is a segmented neutrophil. This cell is also medium-sized and round. The cytoplasm contains many small pink, violet, or tan granules. The granules in this cell appear slightly larger and more distinct than usual for a segmented neutrophil. It is possible these granules represent toxic granulation. Likewise, this granulation could suggest a suboptimum stain, as is seen in the band cell in ABI-20. However, in a patient diagnosed with malaria, it would not be unexpected to see toxic granulation, which can be associated with infection. The most characteristic morphologic feature of mature neutrophils is the segmentation of the nucleus. Although this cell only has two lobes, they are clearly connected by a thin strand of chromatin. Segmented neutrophils may normally have 2-5 nuclear lobes with dense and clumped chromatin.

The cell shown in ABI-21 should not be considered a Pelger-Huet neutrophil. Pelger-Huet cells are hyposegmented and the lobes could be connected by a thin wisp of chromatin, as in this example. In addition, some Pelger-Huet cells may also have nuclei that resemble peanuts or dumb-bells. However, a scan of the virtual slide does not show any such pelgeroid forms. The smear does have definite band neutrophils, not only as seen in ABI-20, but in the field of view for ABI-18 as well. Identifying bands in the smear is reason to question whether or not a neutrophil with only two lobes (which is normal) should be classified as a Pelger-Huet cell.

**Malaria**

Malaria is a global disease caused by four species of *Plasmodium*: *P. falciparum*, *P. malariae*, *P. ovale*, and *P. vivax*. The condition is transmitted to humans through the bite of a female *Anopheles* mosquito. Infection with *P. falciparum* generally causes the most severe disease. These microorganisms complete an asexual maturation cycle in humans that results in the presence of ring trophozoites in the peripheral blood. Amoeboid shaped late trophozoites, schizonts, and gametocytes of some species may also be seen in the peripheral blood. Since malaria infects red blood cells, the life span of erythrocytes is shortened and a hemolytic anemia may develop. Additional laboratory results for this case study patient indicate elevated parameters (bilirubin and lactate dehydrogenase levels) that suggest a hemolytic process did occur.

**Summary**

The peripheral blood cells and organisms evaluated in this advanced blood cell identification exercise are from a patient diagnosed with *Plasmodium falciparum*. Visual observation of malarial parasites in the peripheral blood is important. It is also critical that these microorganisms be differentiated from other cells or inclusions, such as superimposed platelets. The laboratory professional must always perform a detailed and methodical review of the peripheral blood smear. Recognizing and identifying inclusions
such as malarial parasites contributes to an accurate diagnosis and timely therapeutic intervention for patients.

References