Answering your questions

Criteria for identifying bands

Q We are having a difficult time in our facility establishing general consensus of when to call a band a band. We have over-calling and under-calling. Some facilities in the area are eliminating differentiating a band at all. I realize that metas, myelos, and so forth need to be enumerated. Are there any situations in which we would need to differentiate a band from a segmented neutrophil?

A What criteria should be used for identifying a band? The literature contains at least three definitions of the band. The Clinical and Laboratory Standards Institute (CLSI) and College of American Pathologists (CAP) have very similar criteria, which are the most stringent and should be used as standard practice guidelines in the laboratory. The hematology glossary (sent out annually with the CAP survey binders) and CAP’s publication Color Atlas of Hematology serve as good references for defining bands.¹² The nucleus of a band is indented more than half the distance to the farthest nuclear margin, but in no area is the chromatin condensed to a thin filament. A thin filament contains no internal chromatin between two margins and gives the appearance of a solid thread-like dark line. The presence of these thread-like filaments is the most significant feature of segmented neutrophils and is the basis for distinguishing a segmented neutrophil from a band.

The lack of specificity of bandemia and technical problems with band-count methodology limit the test’s usage in clinical diagnosis.

There is generally no problem distinguishing a typical band with an S- or C-shaped nucleus from a typical segmented neutrophil with clear filaments. There are times, however, when the nucleus is twisted or folded, making it impossible to see the filaments. The following rules should be applied:

Assume a hidden filament exists and classify the cells as segmented neutrophils if:

1. the margins of two adjacent lobes are completely separated;
2. the width of either of two adjacent lobes narrows markedly, making it possible that a thin filament could be hidden;
3. the nucleus is extensively folded such that it cannot be determined whether a filament is present (when in doubt, call it a segmented neutrophil).

Assume that no filament exists and classify the cell as a folded band if:

1. an elongated band crosses over itself without evidence of constriction;
2. only the tip of the nucleus is slightly bent back upon itself;
3. the hidden area in a fold between two superimposed lobes is so small and the lobes so wide that a filament could not have been formed.

When do we need to identify the bands? In general, clinicians use bandemia and neutrophilia as an indicator of acute infection. There are, however, several problems with such practice. First of all, bandemia and segmented neutrophil are not specific for infection. Second, different band-reference ranges for bandemia are used among laboratories. Third, enormous interlaboratory variability in distinguishing bands from segmented neutrophils exists in repeated studies of CAP proficiency-testing participants, even with morphologic criteria.

The lack of specificity of bandemia and technical problems with band-count methodology limit the test’s usage in clinical diagnosis. Studies³ have shown that band counts are neither sensitive enough nor specific enough for suspected bacteriologic infection in patients greater than three months of age, appendicitis, and sickling hemoglobin disorders. The white blood cell count and the automated neutrophil count are better diagnostic tests for adults and most children.

Currently, band count is more commonly used in the pediatric population for the following two situations: in infants younger than three months old with suspected bacterial infection, and in newborns with suspected bacterial infection.

Rochester criteria³ are widely used to evaluate the risk of the febrile infant who presents in the emergency department with no obvious source of infection. These young infants lack the ability to communicate and may not show signs of serious or infectious disease. The Rochester criteria include both clinical and laboratory parameters to identify low-risk infants. Laboratory parameters include total white cell count, absolute band count, and/or band-neutrophil ratio.

For newborns, leukocytosis and neutrophilia are unreliable indicators of neonatal sepsis. The reference range changes daily, even hourly, in the first few days of life. In this situation, it is necessary to calculate an immature-to-total neutrophil ratio (I:T ratio) for evaluation of a newborn with suspected bacterial infection, since this ratio may be less influenced by birth weight, gestational age, or maternal or intrapartum complications.

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References

Continues on page 46

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PFA-100 platelet function analyzer

What is the Dade Behring PFA-100 platelet function analyzer? What does it measure? What if the patient is on anti-platelet drugs? What is its application in head trauma injury?

A The Dade Behring PFA-100 platelet function analyzer is used to evaluate primary hemostasis in citrated whole blood. It measures ex vivo platelet binding to collagen/epinephrine (CEPI) or collagen/adenosine diphosphate (CADP). It contains a high-shear flow system to allow blood to pass through a membrane with an aperture. The membrane is coated with collagen fibrils and one of activators epinephrine or ADP (adenosine diphosphate). The platelet activation occurs as blood flows through the aperture. When citrated whole blood mixes with activators, platelets start to form aggregates. The time taken for blood to form a platelet plug that occludes the aperture is an indication of platelet function and is referred to as the closure time (CT). PFA-100 is only a screening test to determine whether abnormal platelet function is present in the patient. There is no critical value at which you can predict whether the patient will bleed in the surgery. If the patient is on an anti-platelet drug, you can only say that the patient’s platelet function is compromised due to drugs, using PFA-100, but you cannot predict the bleeding tendency.

Von Willebrand Factor, or vWF, is the key adhesive protein that mediates platelet adhesion and aggregation in the PFA-100 test cartridge. CT is highly sensitive to von Willebrand disease, or vWD. CT is also highly sensitive to qualitative and quantitative defects to platelet receptors that mediate adhesion and aggregation. Thus, CT is also sensitive to inherited or acquired defects in platelet function. If aspirin is the cause of platelet dysfunction, however, only the CT of collagen/epinephrine will be prolonged but not collagen/adenosine diphosphate.

Other factors affecting the CT include low hematocrit (<35%) or low platelet counts (<150,000/mL). In these situations, the prolonged CT may not reflect abnormal function of platelets. Bleeding time (BT) was designed to evaluate platelet function. BT, however, is very imprecise with a high degree of variation. Currently, PFA-100 has almost replaced BT in evaluating platelet function for pre-surgical screening. Abnormal hemostasis is associated with trauma-related morbidity and mortality. Platelet function is one of the key factors in the maintenance of hemostasis. Severe injury usually results in increased platelet activation and function. One study found that the combination of increased platelet activation with decreased function was associated with increased mortality.

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References

Urine reflex testing

Please advise regarding reflex testing in urinalysis. Our present methodology calls for a microscopic if any of the dipstick chemistry is positive and if the color is not clear. This
results in 60% to 70% of microscopic to be performed. If glucose or ketone or bile is positive, can microscopic not be performed?

A For several years, our institution performed microscopic urinary sediment examinations only on urine specimens with abnormal reagent-strip (dipstick) tests for protein, blood, leukocyte esterase, nitrite, glucose, bilirubin, or specimens with unusual appearance to include unusual color or any lack of clarity. Repeated studies of urinary findings in routine urinalysis by our laboratory showed a very good correlation between reagent-strip results (and physical properties) and the urine sediment findings.

An unpublished study of 800 specimens collected from outpatients at the University of Minnesota outpatient clinics showed 42% of patient specimens had correlating abnormal results, while 55% had correlating negative results. Of the remaining 3%, positive sediment findings of hyaline casts, a rare granular cast, bacteria, or a few PMNs (polymorphonuclear leukocytes) were found with negative reagent-strip results. Patient-chart examination of these exceptions indicated no urinary-tract disease.

At a later date, in-house studies showed that the presence of a positive reagent-strip test for glucose or bilirubin did not require a microscopic examination of the urine.

In summary, a request for a routine urinalysis results in a physical examination and a chemical screen with a multiple reagent strip. The microscopic analysis is performed (as a reflex test) when any of the following show a positive result:

- any abnormal color or any lack of clarity;
- protein;
- blood;
- leukocyte esterase; and
- nitrite.

Specimens with a pH greater than 8.0 or a refractometer specific gravity greater than 1.035 are also examined for sediment abnormalities. Microscopic examinations of the urine sediment are available regardless of the physical and chemical screen, if so requested by the physician.

Since the introduction of the IRIS Urinalysis 500 Workstation in our clinical laboratory at the Fairview University of Minnesota Medical Center, the microscopic examination is performed and reported on all urine specimens regardless of the chemical screen. —Karen M. Ringsrud, MT(ASCP)
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