**Topic:** **Initial Diagnostic Workup of Acute Leukemia**

**Date:** February 22, 2017

**Why is this guideline needed?**
The initial workup and evaluation of acute leukemia (AL) has become increasingly complex over the last several years. This is due in large part to the availability of new laboratory techniques—particularly genetic studies—that have led to better characterization of AL and to diagnostic classification schemes with improved clinical and scientific relevance. Importantly, however, in addition to facilitating the diagnosis of AL, these laboratory studies have identified antigens and/or genetic abnormalities that may be targets for more specific therapy, prognostic factors that allow for better risk stratification of patients, and markers that can be used to monitor therapy and detect minimal residual disease. Therefore, the collection of the appropriate initial diagnostic specimens, along with their proper distribution, utilization, and conservation, is of paramount importance. This guideline draws on recent literature and the experience of pathologists and clinicians active in the field of leukemia to arrive at recommendations that can be routinely followed for the collection and evaluation of specimens to allow optimal patient care and treatment in an efficient and cost-effective manner.

**What should be done when certain tests are evaluated at another facility?**
It is expected that upon implementation of this guideline, there will be uniformity in the collection of specimens across all institutions. If a patient is transferred from one institution to another, the institution in which the patient was initially evaluated should provide the receiving institution with the complete medical record, and assure that all laboratory results, pathology slides, flow cytometry data (PDF or list mode files if possible), and results of cytogenetic and other genetic testing is sent as well. In addition, a list of any pending studies should be provided, and the results of those forwarded as soon as they are completed.

**When is next generation sequencing (NGS) indicated?**
The literature review for this guideline tended to focus on the significance of individual genes. However, particularly in acute myeloid leukemia (AML), but also in acute lymphoblastic leukemia (ALL) and mixed phenotype acute leukemia (MPAL), numerous mutations as well as submicroscopic genetic abnormalities have been described—often occurring as multiple abnormalities in the same patient—that have significant effects on selection of therapy and overall prognosis. In the initial workup of a patient suspected to have AL it is best to assume that at some time in the course of the patient's illness, NGS will provide important information and collection of specimens (eg, fresh blood and/or marrow cells, cryopreserved cells or non-decalcified, formalin-fixed marrow clot sections) that can be used for NGS studies, either initially or at a later date, is recommended. In cases of relapse, NGS performed on the relapse specimen may identify additional new mutations that may prove to be targets for therapy.

**How did the panel determine which tests to include in the statements that name mutational analysis?**
The mutations included in the guideline are those that were found, in the systematic review of the literature, to have statistically proven importance for the diagnosis, prognosis, and/or therapy of acute leukemia. These database searches were supplemented with recommendations from the expert panel, but in all cases, the evidence for inclusion of the mutations was reviewed by a methodologist to assure its quality. Still, this field is quickly evolving and with more widespread utilization of next generation sequencing, additional mutations of clinical significance are likely to be discovered, necessitating continual review and update of this list.

**For patients with suspected acute promyelocytic leukemia (APL), should treatment be delayed while waiting on the results of rapid PML-RARA testing?**
No, treatment should not be delayed. APL is often associated with a life-threatening coagulopathy. If the diagnosis of APL is suspected for any reason, the patient should be immediately started on All-trans retinoic acid (ATRA), and, if clinically indicated, essential blood components such as platelets and/or cryoprecipitate should be administered. If the diagnosis of APL is not confirmed, these steps will not interfere with the treatment of other types of acute leukemia or related disorders.

**The therapeutic and prognostic value of global/gene specific methylation and miRNA expression is still uncertain. Why should laboratories perform these tests?**
The guidelines do not make a specific recommendation regarding these tests. Although deregulated gene expression, miRNA expression and global/gene specific methylation have been reported to impact outcome in AML, these are currently non-standardized and experimental studies that have not been widely incorporated into clinical practice. As advances in the technical aspects of this testing take place, and a larger body of evidence supports their clinical relevance, then more laboratories may begin to investigate incorporation of these studies into their workup of AL.

**Statement 23 suggests that if a patient is going to need immediate transfer to another institution with expertise in treating acute leukemia, that the primary institution defers invasive procedures. Won't deferring invasive procedures delay diagnosis and possibly interfere with patients being admitted into certain treatment centers?**
In hospitals and clinics where it is standard practice to transfer AL patients to tertiary treatment centers, it is recommended to defer invasive procedures, if possible, to the tertiary center. Not uncommonly, treatment protocols used in referral centers require uniform, often specialized testing procedures to establish the morphologic, immunophenotypic and genetic "fingerprint" of the leukemic cells. This information is used by the treating physician to choose therapy for the patient, including therapies that may target specific genetic abnormalities or cellular antigens that are tailored to take various prognostic parameters into consideration. The initial bone marrow sample is often the best source of leukemic cells for thorough analysis, but the specialized studies necessary to decide the best treatment may not be available in hospitals or clinics that do not routinely treat acute leukemia. This may result in the necessity to repeat a bone marrow aspiration and biopsy at the tertiary center. Because these are invasive and costly procedures that may cause some discomfort and/or apprehension for the patient, every effort should be made to avoid duplication of such procedures if possible. If the patient is stabilized at the initial institution and severe cytopenia(s), coagulation abnormalities, or other life-threatening complications are appropriately addressed, the diagnostic bone marrow sample may be safely delayed until after the patient is transferred. See however, the important caution regarding patients with APL in the FAQ above.

**Molecular testing is moving at a high speed. When might more tests be added to the recommendations?**
Clearly there has been an explosion of information regarding the clinical significance of molecular abnormalities in AL. However, despite the rapidity of publication of this data, it often takes some time for the information to mature and be proven to have valid clinical utility. There is a commitment by CAP and ASH to review the guideline every four years. However in the event of publication of substantive and high-quality evidence that necessitates a change in this guideline, the expert panel will reconvene to consider altering the recommendations.

**How will the guideline be enforced? What happens if a laboratory doesn't follow the guideline?**
As with any clinical evidence-based guideline, following the recommendations is not mandatory. These recommendations may be incorporated into future versions of the CAP Laboratory Accreditation Program (LAP) checklist; however, they are not currently required by LAP or any

regulatory or accrediting agency. It is only highly encouraged that laboratories adopt these recommendations. The hope is that over time, laboratories and clinicians will follow the guideline.

## REFERENCES

1. Arber DA, Borowitz MJ, Cessna M, et al. Initial diagnostic workup of acute leukemia: guideline from the College of American Pathologists and the American Society of Hematology. *Arch Pathol Lab Med.* 2017;141(10):1342-1393. doi: 10.5858/arpa.2016-0504-CP