# Acute Myeloid Leukemia Molecular Genetic Testing

Acute myeloid leukemias (AMLs) are a heterogeneous group of disorders characterized by the clonal expansion of myeloid precursors in the peripheral blood, bone marrow, and/or other tissues, which results in impaired hematopoiesis and bone marrow failure.[1,2,3,4] AML is the most common acute leukemia in adults (~80% of leukemia cases) and accounts for the largest number of annual deaths from leukemia in the United States.[2,4] Gene alterations, along with translocations and inversions, carry prognostic importance in AML. In addition to large chromosomal rearrangements, molecular changes have also been implicated in the development of AML. A comprehensive evaluation of several molecular markers, including *FLT3*, *NPM1*, *CEBPA*, *KIT*, *IDH1*, and *IDH2*, is important for risk assessment and prognostication in certain patients with AML, and may guide treatment decisions.[2]

For more information on next generation sequencing testing for AML, see Myeloid Malignancies Mutation Panel by Next Generation Sequencing. For information on cytogenetic testing related to AML, see Acute Myeloid Leukemia with Myelodysplastic Syndrome (MDS) or Therapy-Related MDS Panel by FISH.

## Testing Strategy

At diagnosis, the minimum AML workup includes a bone marrow aspirate for morphology, flow cytometric immunophenotyping, cytogenetics (eg, karyotyping and fluorescence in situ hybridization [FISH]), and appropriate molecular genetic testing.[1,2,3]

## Disease Overview

### Incidence

>20,0000 cases/year in the U.S.[4]

### Age of Onset

Median is 67 years[2]

### Symptoms

- Symptoms resulting from thrombocytopenia, neutropenia, and anemia due to the accumulation of blasts in the marrow[2]
- Morphologic hallmark: excessive accumulation of blasts (typically >20%) and other defined immature cells which affect one or more myeloid lineage[2]

## Test Interpretation

For more detailed information on the prognostic significance of molecular markers in AML, see the ARUP Consult Acute Myeloid Leukemia topic.

## Tests to Consider

**Detect and quantitate gene alterations/translocations/ inversions. Use for minimal residual disease (MRD) and relapse risk monitoring.**

CBFB-MYH11 inv(16) Detection, Quantitative 2011114

**Method:** Reverse Transcription Quantitative Polymerase Chain Reaction

NPM1 Mutation Detection by RT-PCR, Quantitative 3000066

**Method:** Quantitative Reverse-Transcription Polymerase Chain Reaction

PML-RARA Detection by RT-PCR, Quantitative 2002871

**Method:** Reverse Transcription Polymerase Chain Reaction

RUNX1-RUNX1T1 (AML1-ETO) t(8;21) Detection, Quantitative 2010138

**Method:** Quantitative Reverse Transcription Polymerase Chain Reaction

**Use for diagnosis, prognosis, and management. Not intended for MRD monitoring.**

FLT3 ITD and TKD Mutation Detection 3001161

**Method:** Capillary Electrophoresis

IDH1 and IDH2 Mutation Analysis, exon 4 2006444

**Method:** Polymerase Chain Reaction/Sequencing

**Use for prognostication in cytogenetically normal AML (CN-AML).**

CEBPA Mutation Detection 2004247

**Method:** Polymerase Chain Reaction/Sequencing

**Use for prognostication in core-binding factor-related (CBF) AML.**

KIT Mutations in AML by Fragment Analysis and Sequencing 2002437

**Method:** Capillary Electrophoresis

| Recurrent Genetic Abnormalities in AML[2,4,5] | |
|---|---|
| **Molecular Genetic Alteration** | **Prognostic Significance** |
| *CEBPA* | Biallelic mutations confer favorable prognosis |
| *FLT3* ITD and TKD | Poor prognosis associated with *FLT3* ITD mutation; *FLT3* TKD mutation has unclear prognosis |
| *IDH1* and *IDH2* | Unfavorable prognosis; targeted therapy is available for AML with *IDH1* or *IDH2* mutation |
| *KIT* | Poor prognosis, usually associated with core binding factor leukemias; increased risk of relapse |
| *NPM1* | Favorable prognosis in patients with normal karyotype and without *FLT3* ITD mutation; poor prognosis if present with *FLT3* ITD and *DNMT3A* mutations |
| *CBFB-MYH11*[a] | Usually associated with high rate of CR and long-term, disease-free survival when treated with intensive consolidation therapy |
| *PML-RARA*[a] | More favorable prognosis than any other AML cytogenetic subtype when treated appropriately |
| *RUNX1-RUNX1T1*[a] | Usually associated with a high rate of CR and long-term, disease free survival when treated with intensive consolidation therapy |

[a]These fusions can initially be screened by FISH but are also useful in monitoring for MRD.

CR, complete remission

Sources: NCCN, 2019[2]; De Kouchkovsky, 2016[4]; WHO, 2017[5]

**Related Next Generation Sequencing Tests**

Acute Myeloid Leukemia Mutation Panel by Next Generation Sequencing 3002714

**Method:** Massively Parallel Sequencing

Myeloid Malignancies Mutation Panel by Next Generation Sequencing 2011117

**Method:** Massively Parallel Sequencing

Sensitivity/Specificity

| Gene | Methodology | Analytical Sensitivity | Analytical Specificity (%) |
|---|---|---|---|
| *CEBPA* | PCR/sequencing | 40% mutated cells | 100 |
| *FLT3* ITD and TKD | PCR/CE | Signal ratio of 0.05 for ITD and 0.05 for TKD D835 | 100 |
| *IDH1* and *IDH2* | PCR/sequencing | 40% mutated cells | 100 |
| *KIT* | PCR/fragment analysis/sequencing | 30% mutated cells for exon 17<br>5% mutated cells for exon 8 | 100 |
| *NPM1* | Quantitative reverse transcription PCR | 1:100,000 | 100 |
| *CBFB-MYH11*[a] | Quantitative reverse transcription PCR | 1:10,000 | 100 |
| *PML-RARA*[a] | Quantitative reverse transcription PCR | 1:10,000 | 85 |
| *RUNX1-RUNX1T1*[a] | Quantitative reverse transcription PCR | 1:100,000 | 100 |

| Gene | Methodology | Analytical Sensitivity | Analytical Specificity (%) |
|---|---|---|---|

[a]These fusions can initially be screened by FISH but are also useful in monitoring for MRD.

CE, capillary electrophoresis; PCR, polymerase chain reaction

## Limitations

- Variants outside the targeted regions or below the limit of detection will not be identified
- Results must always be interpreted within the patient's clinical context and in conjunction with other relevant data and should not be used alone for a diagnosis of malignancy

## 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. PubMed

2. National Comprehensive Cancer Network. NCCN clinical practice guidelines in oncology: acute myeloid leukemia, Version 3.2020. [Last update: Dec 2019; Accessed: Sep 2020]

3. Weinberg OK, Sohani AR, Bhargava P, et al. Diagnostic work-up of acute myeloid leukemia. Am J Hematol. 2017;92(3):317-321. PubMed

4. De Kouchkovsky I, Abdul-Hay M. 'Acute myeloid leukemia: a comprehensive review and 2016 update'. Blood Cancer J. 2016;6(7):e441. PubMed

5. Swerdlow S, Campo E, Jaffe E, et al. WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues, Revised 4th ed. International Agency for Research on Cancer, 2017.


# Related Information

Acute Myeloid Leukemia - AML


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Content Review October 2019 | Last Update February 2021