

## Overview

### Useful For

Evaluation of acute myeloid leukemia (AML) using a focused 11-gene panel at the time of diagnosis or possibly at relapsed/refractory disease, to assist in appropriate classification, prognosis, and therapeutic management of patients

Evaluation to determine if a different gene mutation profile is present at the time of AML relapse

### Genetics Test Information

This test includes next-generation sequencing to evaluate for the following 11 genes: *CEBPA*, *DNMT3A*, *FLT3*, *IDH1*, *IDH2*, *KIT*, *KRAS*, *NPM1*, *NRAS*, *RUNX1*, and *TP53*.

### Highlights

Next-generation sequencing detection of somatic gene mutations, including type, pattern, and distribution, has diagnostic, prognostic, and potential therapeutic implications for patients with hematologic cancers, such as acute myeloid leukemia (AML).

This test enables more accurate classification and prognostic assessment of acute myeloid leukemias (AML).

### Testing Algorithm

See Targeted Genes Interrogated by Next-Generation Sequencing, Acute Myeloid Leukemia, 11-Gene Panel in Special Instructions for a list of the genes and exons targeted by this assay.

### Special Instructions

- [Hematopathology Patient Information](#)
- [Targeted Genes Interrogated by Next-Generation Sequencing, Acute Myeloid Leukemia, 11-Gene Panel](#)

### Method Name

Somatic Mutation Detection by Next-Generation Sequencing (NGS), Hematologic Neoplasms

### NY State Available

Yes

## Specimen

### Specimen Type

Varies

### Advisory Information

This test is a subset of the NGS<sup>SM</sup> / OncoHeme Next-Generation Sequencing for Myeloid Neoplasms test and focuses more specifically on the gene mutations that are most prevalent and clinically significant in acute myeloid leukemias (AML). If a wider gene mutation analysis is desired, or the indication is for a myeloid malignancy other than AML, then NGS<sup>SM</sup> / OncoHeme Next-Generation Sequencing (NGS), Hematologic Neoplasms should be considered.

### Shipping Instructions

**Bone marrow and peripheral blood specimens must arrive within 14 days of collection.**

**Necessary Information**

The following information is required:

1. Clinical diagnosis
2. Pertinent clinical history, including disease phase (diagnostic, remission, relapse/refractory) and therapy status (especially if patient has received a hematopoietic stem cell transplant).
3. Clinical or morphologic suspicion
4. Date of collection
5. Specimen source

**Specimen Required**

Submit only 1 of the following specimens:

**Specimen Type:** Bone marrow aspirate (preferred)

**Container/Tube:**

**Preferred:** Lavender top (EDTA) or yellow top (ACD)

**Acceptable:** Green top (heparin), but not preferred

**Specimen Volume:** 2 mL

**Collection Instructions:**

1. Invert several times to mix bone marrow.
2. Send specimen in original tube.
3. Label specimen as bone marrow.

**Specimen Stability:** Ambient (preferred)/Refrigerate

**Specimen Type:** Peripheral blood

**Container/Tube:**

**Preferred:** Lavender top (EDTA) or yellow top (ACD)

**Acceptable:** Green top (heparin), but not preferred

**Specimen Volume:** 3 mL

**Collection Instructions:**

1. Invert several times to mix blood.

2. Send specimen in original tube.

3. Label specimen as blood.

**Specimen Stability:** Ambient (preferred)/Refrigerate

**Specimen Type:** Extracted DNA from blood or bone marrow

**Container/Tube:** 1.5-2 mL tube with indication of volume and concentration of the DNA

**Specimen Volume:** Entire specimen

**Collection Instructions:** Label specimen as extracted DNA and source of specimen

**Specimen Stability:** Frozen (preferred) /Refrigerated/Ambient

**Forms**

1. Hematopathology Patient Information (T676) in Special Instructions.

2. If not ordering electronically, complete, print, and send a [Hematopathology/Cytogenetics Test Request](#) (T726) with the specimen.

**Specimen Minimum Volume**

Blood, Bone Marrow: 1 mL

Extracted DNA: 100 mcL at 20 ng/mcL concentration

**Reject Due To**

Hemolysis	Mild OK; Gross reject
Lipemia	Mild OK; Gross OK
Icterus	NA
Other	Bone marrow biopsies, slides, paraffin shavings or frozen tissues and paraffin-embedded tissues, paraffin-embedded bone marrow aspirates, or moderately to severely clotted

**Specimen Stability Information**

Specimen Type	Temperature	Time
Varies	Varies	14 days

**Clinical and Interpretive**

**Clinical Information**

Next-generation sequencing (NGS) is a comprehensive molecular diagnostic methodology that can interrogate multiple regions of genomic tumor DNA in a single assay. Many hematologic neoplasms, including acute myeloid leukemia (AML), are characterized by morphologic or phenotypic similarities, but can have characteristic somatic

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mutations in many genes. In addition, many cases of AML lack a clonal cytogenetic finding at diagnosis (normal karyotype) and can be better classified according to gene mutation profile. The presence and pattern of gene mutations in AML can provide critical prognostic information and may help in guiding therapeutic management decisions by physicians, particularly if targeted therapies are available.

### Reference Values

An interpretive report will be provided.

### Interpretation

Mutations (gene alterations) identified, if present, using human reference genome build GRCh37 (hg19). An interpretive report will be provided.

### Cautions

This test is a targeted next-generation sequencing (NGS) (panel) assay that encompasses 11 genes with variable full exon, partial region (including select intronic or non-coding regions), or hot spot coverage (depending on specific locus). Therefore, this test will not detect other genetic abnormalities in genes or regions outside the specified target areas. The test detects single base substitutions (ie, point mutations), as well as small insertion or deletion type events, but it does not detect gene rearrangements (ie, translocations), gene fusions, copy number alterations, or large scale (segmental chromosome region) deletions and complex changes.

This assay does not distinguish between somatic and germline alterations in analyzed gene regions, particularly with variant allele frequencies (VAF) near 50% or 100%. If nucleotide alterations in genes associated with germline mutation syndromes are present and there is also a strong clinical suspicion or family history of malignant disease predisposition, additional genetic testing and appropriate counseling may be indicated. Mutation cells detected between 5% and 10% VAF may indicate low-level (ie, subclonal) tumor populations, although the clinical significance of these findings may not be clear. A low incidence of gene mutations associated with myeloid neoplasms can be detected in nonmalignant hematopoietic cells in individuals with advancing age (clonal hematopoiesis of indeterminate potential, CHIP) and these may not be clearly distinguishable from tumor-associated mutations. Some apparent mutations classified as variants of undetermined significance (VUS) may represent rare or low frequency polymorphisms.

Prior treatment for hematologic malignancy could affect the results obtained in this assay. In particular, prior allogeneic hematopoietic stem cell transplant (HSCT) may cause difficulties in resolving somatic or polymorphic alterations, or in assigning variant calls correctly to donor and recipient fractions, if pertinent clinical or laboratory information (eg, chimerism engraftment status) is not provided.

Correlation with clinical, histopathologic and additional laboratory findings is required for final interpretation of these results. The final interpretation of results for clinical management of the patient is the responsibility of the managing physician.

### Clinical Reference

1. Patel JP, Levine RL: How do novel molecular genetic markers influence treatment decisions in acute myeloid leukemia? *Hematology Am Soc Hematol Educ Program* 2012;2012:28-34
2. Patel JP, Gonen M, Figueroa ME, et al: Prognostic relevance of integrated genetic profiling in acute myeloid leukemia. *N Engl J Med* 2012;366:1079-1089
3. Ley T, Miller C, Ding L, et al: Genomic and epigenomic landscapes of adult de novo acute myeloid leukemia. *N Engl J Med* 2013;368(22):2059-2074
4. Amatangelo MD, Quek L, Shih A, et al.: Enasidenib induces acute myeloid leukemia cell differentiation to promote clinical response. *Blood* 2017;130(6):732-741

5. Stone RM, Mandrekar SJ, Sanford BL, et al.: Midostaurin plus Chemotherapy for Acute Myeloid Leukemia with a FLT3 Mutation. N Engl J Med 2017;377:454-464

## Performance

### Method Description

Next-generation sequencing is performed to test for the presence of a mutation in targeted regions of the following 11 genes: *CEBPA*, *DNMT3A*, *FLT3*, *IDH1*, *IDH2*, *KIT*, *KRAS*, *NPM1*, *NRAS*, *RUNX1*, and *TP53*. See Targeted Genes Interrogated by Next-Generation Sequencing, Acute Myeloid Leukemia, 11-Gene Panel in Special Instructions for details regarding the targeted gene regions identified in this test. This is a laboratory-developed test using Research Use Only reagents. Extracted DNA from the clinical specimen is fragmented, adapter ligated, and a sequence library of fragments is prepared using a custom capture hybridization method. Individual patient samples are indexed ("bar-coded") for identification and the library is sequenced on an Illumina platform. Sequence data are processed through the Mayo Clinic Clinical Genome Sequencing Lab bioinformatics pipeline and a variant call file is generated for final analysis and reporting. (Unpublished Mayo method)

### PDF Report

No

### Day(s) and Time(s) Test Performed

Monday, Wednesday, Friday

### Analytic Time

14 days

### Maximum Laboratory Time

21 days

### Specimen Retention Time

DNA 3 months

### Performing Laboratory Location

Rochester

## Fees and Codes

### Fees

- Authorized users can sign in to [Test Prices](#) for detailed fee information.
- Clients without access to Test Prices can contact [Customer Service](#) 24 hours a day, seven days a week.
- Prospective clients should contact their Regional Manager. For assistance, contact [Customer Service](#).

### Test Classification

This test was developed and its performance characteristics determined by Mayo Clinic in a manner consistent with CLIA requirements. This test has not been cleared or approved by the U.S. Food and Drug Administration.

### CPT Code Information

81450

### LOINC® Information

Test ID	Test Order Name	Order LOINC Value
NGAML	Next Gen Sequencing, AML, 11 Gene	In Process

Result ID	Test Result Name	Result LOINC Value
MP038	Specimen Type	31208-2
43554	NGAML Result	No LOINC Needed
43488	Pathogenic Mutations Detected	41103-3
43487	Interpretation	69047-9
43489	Clinical Trials	82786-5
43490	Variants of Unknown Significance	82939-0
43491	Additional Notes	48767-8
43492	Method Summary	49549-9
43493	Disclaimer	62364-5
43494	AML Panel Gene List	36908-2
43495	Reviewed By	19139-5