Test Definition: AMLFA

Adult Acute Myeloid Leukemia Panel, FISH, Varies

Overview

Useful For

Detecting, at diagnosis, recurrent common chromosome abnormalities associated with acute myeloid leukemia (AML) in patients aged 31 and older using a laboratory-designated probe set algorithm

As an adjunct to conventional chromosome studies in patients with AML

Evaluating specimens in which chromosome studies are unsuccessful

This test **should not be used** to screen for residual AML

Testing Algorithm

This test includes a charge for the probe application, analysis, and professional interpretation of results for 3 probe sets (6 individual fluorescence in situ hybridization [FISH] probes). Additional charges will be incurred for all reflex testing or additional probe sets performed. Analysis charges will be incurred based on the number of cells analyzed per probe set. If no cells are available for analysis, no analysis charges will be incurred.

This test is performed as panel testing only using the following analysis algorithm.

The initial **diagnostic** adult acute myeloid leukemia (AML) FISH panel includes testing for the following abnormalities using the FISH probes listed:
t(8;21)(q21.3;q22) or *RUNX1::RUNX1T1* fusion, RUNX1T1/RUNX1 probe set
t(11q23;var) or *KMT2A* rearrangement, KMT2A break-apart probe set
t(15;17)(q24;q21) or *PML::RARA* fusion, PML/RARA probe set
inv(16) or t(16;16) or *CBFB::MYH11* fusion, MYH11/CBFB probe set

If testing is ordered concurrently with a chromosomal study (CHRBM / Chromosome Analysis, Hematologic Disorders, Bone Marrow; or CHRHB / Chromosome Analysis, Hematologic Disorders, Blood), secondary testing will not be performed. Appropriate secondary FISH probes will be recommended if the chromosome results are informative.

If a chromosome study is **not** ordered concurrently, the following secondary panel of FISH probes will be performed when the initial panel is negative or demonstrate nonclassical abnormalities:
inv(3) or t(3;3) or *GATA2::MECOM* fusion, GATA2/MECOM probe set
-5/5q-, D5S630/EGR1 probe set
t(6;9)(p22.3;q34) or *DEK::NUP214* fusion, DEK/NUP214 probe set
-7/7q-, D7Z1/D7S486 probe set
t(9;22)(q34;q11.2) or *BCR::ABL1* fusion, ABL1/BCR probe set
t(11p15.4;var) or *NUP98* rearrangement, NUP98 break-apart probe set
-17/17p-, TP53/D17Z1 probe set

Appropriate ancillary probes may be performed at consultant discretion to render comprehensive assessment. Any additional probes will have the results included within the final report and will be performed at an additional charge.

In the following situations, additional (reflex) testing may be performed at the laboratory's discretion and may be influenced by available karyotype results or other FISH testing.

When a *KMT2A* rearrangement is identified, 1 or more dual-fusion FISH (D-FISH) probe sets may be used in an attempt to identify the translocation partner for the following abnormalities:
t(4;11)(q21;q23) or *KMT2A::AFF1* fusion, AFF1/KMT2A probe set
t(6;11)(q27;q23) or *KMT2A::AFDN* ;fusion, AFDN/KMT2A probe set
t(9;11)(p22;q23) or *KMT2A::MLLT3* fusion, MLLT3/KMT2A probe set
t(10;11)(p12;q23) or *KMT2A::MLLT10* fusion, MLLT10/KMT2A probe set
t(11;16)(q23;p13.3) or *KMT2A::CREBBP* fusion, KMT2A/CREBBP probe set
t(11;19)(q23;p13.1) or *KMT2A::MLLT1* fusion, KMT2A/ELL probe set
t(11;19)(q23;p13.3) or *KMT2A::ELL* fusion, KMT2A/MLLT1 probe set

When an extra or atypical RARA signal is identified in the absence of *PML::RARA* fusion, the RARA break-apart probe set may be used to identify a potential variant translocation involving *RARA,* t(17;var)(q21;?).
When an extra CBFB signal is identified in the absence of *CBFB::MYH11* fusion, the CBFB break-apart probe set may be used to evaluate for the presence or absence of a potential variant translocation involving *CBFB*, t(16;var)(q22;?).

When an extra RUNX1 signal is identified in the absence of *RUNX1::RUNX1T1* fusion, the RUNX1 break-apart probe set may be used to evaluate for the presence or absence of a potential variant translocation involving *RUNX1,* t(21;var)(q22;?).

When an extra GATA2 signal is identified in the absence of *GATA2::MECOM* fusion, the PRDM16/GATA2 probe set may be used to identify a potential t(1;3)(p36;q21).

When an extra MECOM signal is identified in the absence of *GATA2::MECOM* fusion, the break-apart MECOM probe set may be used to identify a potential variant translocation involving *MECOM*, t(3;var)(q26.2;?).

When an extra ABL1 signal is identified in the absence of *BCR::ABL1* fusion, the ABL1 break-apart probe set may be used to evaluate for the presence or absence of a potential variant translocation involving *ABL1*, t(9;var)(q34;?).

For more information see:
Acute Promyelocytic Leukemia: Guideline to Diagnosis and Follow-up
Acute Leukemias of Ambiguous Lineage Testing Algorithm
Acute Myeloid Leukemia: Testing Algorithm

Special Instructions

- Acute Promyelocytic Leukemia: Guideline to Diagnosis and Follow-up
- Acute Leukemias of Ambiguous Lineage Testing Algorithm
- Acute Myeloid Leukemia: Testing Algorithm

Method Name

Fluorescence In Situ Hybridization (FISH)

NY State Available

Yes

Specimen

Specimen Type

Varies

Ordering Guidance

This test is only performed on specimens from patients with acute myeloid leukemia (AML) who are age 31 years or older.

If acute promyelocytic leukemia is probable and expedited PML/RARA results are needed, order AMLMF / Acute Myeloid Leukemia (AML), Specified FISH, Varies. If *PML::RARA* fusion is identified in AMLFA, the laboratory will automatically expedite analysis. Results will not be provided until the complete panel testing is finalized. The laboratory is unable to provide preliminary results.

This test **should NOT be used** to screen for residual AML.

Minimal residual disease (MRD) monitoring in patients with AML known to have either t(15;17) with *PML::RARA* fusion, inv(16) or t(16;16) with *CBFB::MYH11* fusion, t(8;21) with *RUNX1::RUNX1T1* fusion, or t(9;22) with *BCR::ABL1* fusion should be performed by quantitative reverse transcriptase polymerase chain reaction and **not** by fluorescence in situ hybridization (FISH) testing.

It is recommended that MRD monitoring in AML patients be performed by AML-MRD flow cytometry rather than FISH testing using individual FISH probe sets. This is particularly true for the deletion/monosomy probe sets (5, 7, 17) which have cutoffs that exceed 10% of nuclei.

If targeted AML FISH probes are preferred, order AMLMF / Acute Myeloid Leukemia (AML), Specified FISH, Varies and request specific probes for targeted abnormalities.

This test is intended for instances when the entire AML FISH panel is needed for an **adult** patient.

If this test is ordered on a patient aged 30 years or younger, this test will be canceled and automatically reordered by the laboratory as AMLFP / Pediatric Acute Myeloid Leukemia Panel, FISH, Varies.

If this test is ordered and the laboratory is informed that the patient is age 30 years or younger **and** is on a Children's Oncology Group protocol, this test will be canceled and automatically reordered by the laboratory as COGMF / Acute Myeloid Leukemia (AML), Children's Oncology Group Enrollment Testing, FISH, Varies.

If either (or both) BALAF / B-Cell Acute Lymphoblastic Leukemia/Lymphoma (ALL), FISH, Adult, Varies; or TALAF / T-Cell Acute Lymphoblastic Leukemia/Lymphoma (ALL), FISH, Adult, Varies, is ordered concurrently with this test, the laboratory may cancel this test and automatically reorder as AMLMF / Acute Myeloid Leukemia (AML), Specified FISH, Varies with the following FISH probes: RUNX1T1/RUNX1, PML/RARA, MYH11/CBFB, GATA2/MECOM, DEK/NUP214, D5S630/EGFR1, D7Z1/D7S486, TP53/D17Z1, and NUP98 3'/5'. If an abnormality is identified that would result in reflex testing in this test, the same reflex testing will be performed in the AMLMF. This cancellation is necessary to avoid duplicate testing. The break-apart KMT2A probe set will still be performed as part of either the adult B-cell acute lymphoblastic leukemia (B-ALL) or T-ALL FISH panel.

For testing paraffin-embedded tissue samples from patients with AML/myeloid sarcoma, order MSTF / Myeloid Sarcoma, FISH, Tissue. If a paraffin-embedded tissue sample is submitted for this test, this test will be canceled and MSTF will be added and performed as the appropriate test.

Additional Testing Requirements

At diagnosis, conventional cytogenetic studies (CHRBM / Chromosome Analysis, Hematologic Disorders, Bone Marrow) and this fluorescence in situ hybridization panel should be performed. If there is limited specimen available, only this test will be performed.

Shipping Instructions

Advise Express Mail or equivalent if not on courier service.

Necessary Information

1. A reason for testing must be provided. If this information is not provided, an appropriate indication for testing may be entered by Mayo Clinic Laboratories.
2. A flow cytometry and/or a bone marrow pathology report should be submitted with each specimen. The laboratory will not reject testing if this information is not provided but appropriate testing and interpretation may be compromised or delayed.

Specimen Required

Submit only 1 of the following specimens:

Preferred
Specimen Type: Bone marrow
Container/Tube:
Preferred: Yellow top (ACD)
Acceptable: Green top (sodium heparin) or lavender top (EDTA)
Specimen Volume: 2 to 3 mL
Collection Instructions:
1. It is preferable to send the first aspirate from the bone marrow collection.
2. Invert several times to mix bone marrow.
3. Send bone marrow in original tube. **Do not aliquot.**

Acceptable
Specimen Type: Whole blood
Container/Tube:

Preferred: Yellow top (ACD)
Acceptable: Green top (sodium heparin) or lavender top (EDTA)
Specimen Volume: 6 mL
Collection Instructions:
1. Invert several times to mix blood.
2. Send whole blood in original tube. **Do not aliquot.**

Specimen Minimum Volume

Bone marrow: 1 mL; Whole blood: 2 mL

Reject Due To

All specimens will be evaluated at Mayo Clinic Laboratories for test suitability.

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Ambient (preferred)		
	Refrigerated		

Clinical & Interpretive

Clinical Information

Acute myeloid leukemia (AML) is one of the most common adult leukemias, with almost 10,000 new cases diagnosed per year. AML also comprises 15% of pediatric acute leukemia and accounts for the majority of infant (<1 year old) leukemia.

Several recurrent chromosomal abnormalities have been identified in AML with associated clinical significance. The most common chromosome abnormalities associated with AML include t(8;21), t(15;17), inv(16) or t(16;16), and rearrangements of the *KMT2A* gene at 11q23. The most common genes juxtaposed with *KMT2A* through translocation events in AML include *MLLT3*- t(9;11), *AFDN*- t(6;11), *MLLT10*- t(10;11), and *ELL*- t(11;19p13.1).

Other recurrent chromosome abnormalities associated with AML include inv(3) or t(3;3), t(6;9), t(9;22), and abnormalities of the *NUP98* gene at 11p15.4. In addition, AML can also evolve from myelodysplasia (MDS). Thus, the common chromosome abnormalities associated with MDS can also be identified in AML, which include: -5/5q-, -7/7q-, and 17p-. Overall, the recurrent chromosome abnormalities identified in patients with AML are observed in approximately 60% of diagnostic AML cases.

Conventional chromosome analysis is the gold standard for identification of the common, recurrent chromosome abnormalities in AML. However, some of the subtle rearrangements can be missed by karyotype, including inv(16) or t(16;16) and *KMT2A* rearrangements.

Fluorescence in situ hybridization analysis of nonproliferating (interphase) cells can be used to detect the common diagnostic and prognostic chromosome abnormalities observed in patients with AML.

Reference Values

An interpretive report will be provided.

Interpretation

A neoplastic clone is detected when the percent of cells with an abnormality exceeds the normal reference range for any given probe set.

The absence of an abnormal clone does not rule out the presence of an acute myeloid leukemia clone or another neoplastic disorder.

Cautions

This test is not approved by the US Food and Drug Administration, and it is best used as an adjunct to clinical and pathologic information.

Fluorescence in situ hybridization (FISH) is not a substitute for conventional chromosome studies because the latter detects chromosome abnormalities associated with other hematological disorders that would go undetected in a targeted acute myeloid leukemia FISH panel test.

Bone marrow is the preferred specimen type for this FISH test. If bone marrow is not available, a blood specimen may be used if there are circulating myeloblasts in the blood specimen (as verified by a hematopathologist).

If no FISH signals are observed post-hybridization, the case will be released indicating a lack of FISH results.

Clinical Reference

1. Swerdlow SH, Campo E, Harris NL, et al, eds. WHO Classification of Tumour of Haematopoietic and Lymphoid Tissues. 4th ed. IARC Press; 2017
2. Dohner H, Estey E, Grimwade D, et al. Diagnosis and management of AML in adults: 2017 ELN recommendations from an international expert panel. Blood. 2017;129(4):424-447. doi:10.1182/blood-2016-08-733196
3. Pollyea DA, Bixby D, Perl A, et al. NCCN Guidelines Insights: Acute Myeloid Leukemia, Version 2.2021. J Natl Compr Canc Netw. 2021;19(1):16-27. Published 2021 Jan 6. doi:10.6004/jnccn.2021.0002

Performance

Method Description

This test is performed using commercially available and laboratory-developed fluorescence in situ hybridization (FISH) probes. Deletion or monosomy of chromosomes 5, 7, and 17 are detected using enumeration strategy probes. Rearrangements involving *MECOM*, *ABL1, NUP98, KMT2A*, *CBFB, RARA*, and *RUNX1* are detected using a dual-color break-apart (BAP) strategy probe. Dual-color, dual-fusion FISH (D-FISH) strategy probe sets are used to detect t(1;3), inv(3) or t(3;3), t(6;9), t(8;21), t(9;22), t(15;17), inv(16) or t(16;16), and in reflex testing when rearrangements of the *KMT2A* gene are detected. For enumeration and BAP strategy probe sets, 100 interphase nuclei are scored; 200 interphase nuclei are scored when D-FISH probes are used. All results are expressed as the percent abnormal nuclei.(Unpublished Mayo method)

PDF Report
No

Day(s) Performed
Monday through Friday

Report Available
7 to 10 days

Specimen Retention Time
4 weeks

Performing Laboratory Location
Mayo Clinic Laboratories - Rochester Main Campus

Fees & 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 account representative. 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. It has not been cleared or approved by the US Food and Drug Administration.

CPT Code Information
88271x6, 88275x3, 88291x1-FISH Probe, Analysis, Interpretation; 3 probe sets

88271x2, 88275x1-FISH Probe, Analysis; each additional probe set (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
AMLFA	Adult-AML panel, FISH	102101-3

Result ID	Test Result Name	Result LOINC® Value
622381	Result Summary	50397-9
622382	Interpretation	69965-2
622383	Result Table	93356-4
622384	Result	62356-1
GC147	Reason for Referral	42349-1
GC148	Specimen	31208-2
622385	Source	31208-2

622386	Method	85069-3
622387	Additional Information	48767-8
622388	Disclaimer	62364-5
622389	Released By	18771-6