MAYO CLINIC LABORATORIES

Test Definition: NR4A3

NR4A3 (9q22.33) Rearrangement, FISH, Tissue

Overview

Useful For

Identifying *NR4A3* gene rearrangements

Supporting the diagnosis of extraskeletal myxoid chondrosarcoma or acinic cell carcinoma when used in conjunction with an anatomic pathology consultation

Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
_PBCT	Probe, +2	No, (Bill Only)	No
_PADD	Probe, +1	No, (Bill Only)	No
_PB02	Probe, +2	No, (Bill Only)	No
_PB03	Probe, +3	No, (Bill Only)	No
_IL25	Interphases, <25	No, (Bill Only)	No
_I099	Interphases, 25-99	No, (Bill Only)	No
_I300	Interphases, >=100	No, (Bill Only)	No

Testing Algorithm

This test does not include a pathology consult. If a pathology consultation is requested, PATHC / Pathology Consultation should be ordered, and the appropriate fluorescence in situ hybridization (FISH) test will be ordered and performed at an additional charge.

This test includes a charge for the probe application, analysis, and professional interpretation of results for one probe set (2 individual FISH probes). 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.

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.

Method Name

Fluorescence In Situ Hybridization (FISH)

NY State Available

Yes

Specimen

Specimen Type

Tissue

Ordering Guidance

Multiple oncology (cancer) gene panels are also available. For more information see Hematology, Oncology, and Hereditary Test Selection Guide.

Additional Testing Requirements

Confirmation testing by next-generation sequencing to resolve atypical or unbalanced fluorescence in situ hybridization results of this gene region is available, order SARCP / Sarcoma Targeted Gene Fusion/Rearrangement Panel, Next-Generation Sequencing, Tumor.

Shipping Instructions

Advise Express Mail or equivalent if not on courier service.

Necessary Information

1. A pathology report is required for testing to be performed. If not provided, appropriate testing and/or interpretation may be compromised or delayed. Acceptable pathology reports include working drafts, preliminary pathology, or surgical pathology reports.

2. The following information must be included in the report provided:

-Patient name

-Block number - must be on all blocks, slides, and paperwork

-Date of collection

-Tissue source

3. 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.

Specimen Required

Submit only 1 of the following specimens:

Preferred:

Specimen Type: Tissue block

Collection Instructions: Submit a formalin-fixed, paraffin-embedded (FFPE) tumor tissue block. Blocks prepared with alternative fixation methods will be attempted but are less favorable for successful results by FISH testing; provide fixation method used.

Additional Information:

1. Paraffin-embedded specimens can be from any anatomic location (skin, soft tissue, lymph node, etc).
2. Bone specimens that have been decalcified will be attempted for testing, but the success rate is approximately 50%.

Acceptable:

Specimen Type: Tissue slides

Slides: 1 Hematoxylin and eosin stained and 4 unstained

Collection Instructions: Submit 4 consecutive unstained, positively charged, unbaked slides with 5 micron-thick sections of the tumor tissue and 1 slide stained with hematoxylin and eosin.

Forms

If not ordering electronically, complete, print, and send an Oncology Test Request (T729) with the specimen.

Specimen Minimum Volume

Slides: 1 Hematoxylin and eosin stained and 2 unstained

Reject Due To

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

Specimen Stability Information

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

Clinical & Interpretive

Clinical Information

The gene *NR4A3* is often altered in extraskeletal myxoid chondrosarcomas (EMC) and acinic cell carcinoma of the salivary gland.(1,2) Rearrangement of the *NR4A3* gene region may be involved with up to 4 partner genes as a pathway to EMC. Fluorescence in situ hybridization analysis allows for the detection of rearrangement of the *NR4A3* gene region.

Reference Values

An interpretive report will be provided.

Interpretation

A positive result with the NR4A3 probe is detected when the percent of cells with an abnormality exceeds the normal cutoff for the probe set. A positive result of NR4A3 suggests inactivating structural alterations of the *NR4A3* gene region at 9q22.33. A negative result suggests no structural alterations of the locus.

NR4A3 will be clinically interpreted as positive or negative.

A neoplastic clone is detected when the percent of cells with an abnormality exceeds the normal cutoff for the NR4A3 probe set.

A positive result is consistent with rearrangement of the *NR4A3* gene and likely reflects *NR4A3* fusion with a partner gene. A positive result may support a diagnosis of extraskeletal myxoid chondrosarcoma or acinic cell carcinoma of salivary gland. The significance of this finding is dependent on the clinical and pathologic features.

A negative result suggests a *NR4A3* gene rearrangement is not present. A negative result does not exclude the diagnosis of extraskeletal myxoid chondrosarcoma or acinic cell carcinoma of salivary gland.

Cautions

This test is not approved by the US Food and Drug Administration and should be used as an adjunct to existing clinical and pathologic information.

This fluorescence in situ hybridization (FISH) assay does not rule out other chromosome abnormalities.

Fixatives other than formalin (eg, Prefer, Bouin's) may not be successful for FISH assays. Non-formalin fixed specimens will not be rejected.

Paraffin-embedded tissues that have been decalcified may not be successful for FISH analysis. The success rate of FISH studies on decalcified tissue is approximately 50%, but FISH will be attempted if sufficient tumor is present for analysis.

Fluorescence in situ hybridization studies will be attempted if sufficient tumor is present for analysis. The pathologist reviewing the hematoxylin and eosin-stained slide may find it necessary to cancel testing if insufficient tissue/tumor is available for testing.

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

Clinical Reference

1. Benini S, Cocchi S, Gamberi G, Magagnoli G, et al. Diagnostic utility of molecular investigation in extraskeletal myxoid chondrosarcoma. J Mol Diagn. 2014;16(3):314-323
2. Haller F, Bieg M, Will R, et al. Enhancer hijacking activates oncogenic transcription factor NR4A3 in acinic cell carcinomas of the salivary glands. Nat Commun. 2019;10(1):368

Performance

Method Description

This test is performed using a laboratory-developed *NR4A3* (9q22.33) dual-color break-apart probe (BAP) strategy. Formalin-fixed paraffin-embedded tissues are cut at 5 microns and mounted on positively charged glass slides. The selection of tissue and the identification of target areas on the hematoxylin and eosin (H and E)-stained slide are performed by a pathologist. Using the H and E-stained slide as a reference, target areas are etched with a diamond-tipped engraving tool on the back of the unstained slide to be assayed. The probe set is hybridized to the appropriate target areas, and 2 technologists independently analyze 50 interphase nuclei (100 total) with the results expressed as the percent of abnormal nuclei.(Unpublished Mayo method)

PDF Report

No

Day(s) Performed

Monday through Friday

Report Available

7 to 10 days

Specimen Retention Time

Slides used for analysis are retained by the laboratory in accordance with regulatory requirements. Client provided paraffin blocks and extra unstained slides (if provided) will be returned after testing is complete.

Performing Laboratory Location

Rochester

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

88271 x 2, 88291-DNA probe, each (first probe set), Interpretation and report
88271 x 2-NA probe, each; each additional probe set (if appropriate)
88271 x 1-DNA probe, each; coverage for sets containing 3 probes (if appropriate)
88271 x 2-DNA probe, each; coverage for sets containing 4 probes (if appropriate)
88271 x 3-DNA probe, each; coverage for sets containing 5 probes (if appropriate)
88274 w/modifier 52-Interphase in situ hybridization, <25 cells, each probe set (if appropriate)
88274-Interphase in situ hybridization, 25 to 99 cells, each probe set (if appropriate)
88275-Interphase in situ hybridization, 100 to 300 cells, each probe set (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
NR4A3	NR4A3, Rearrangement, FISH, Ts	21796-8

Result ID	Test Result Name	Result LOINC® Value
92340	Result Summary	50397-9
92341	Interpretation	69965-2
92342	Result	62356-1
92343	Specimen	31208-2
92344	Source	31208-2
92345	Tissue ID	80398-1
92346	Method	85069-3
92347	Additional Information	48767-8
92348	Disclaimer	62364-5
92349	Released By	18771-6
CG999	Reason For Referral	42349-1