Overview

Useful For
Supporting the diagnosis of Ewing sarcoma (EWS)/primitive neuroectodermal tumor (PNET), myxoid chondrosarcoma, desmoplastic small, round cell tumor, clear cell sarcoma, and myxoid liposarcoma when used in conjunction with an anatomic pathology consultation

An aid in the diagnosis of EWS when reverse transcriptase-PCR results are equivocal or do not support the clinical picture

Reflex Tests

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<th>Available Separately</th>
<th>Always Performed</th>
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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 FISH test will be ordered and performed at an additional charge.

This test includes a charge for application of the first probe set (2 FISH probes) and professional interpretation of results. Additional charges will be incurred for all reflex probes 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.

See the Method Description for specific details.

Method Name
Fluorescence In Situ Hybridization (FISH)

NY State Available
Yes

Specimen

Specimen Type
Tissue

Shipping Instructions
Advise Express Mail or equivalent if not on courier service.
Necessary Information

A reason for referral and pathology report are required in order for testing to be performed. Send information with specimen. Acceptable pathology reports include working drafts, preliminary pathology or surgical pathology reports.

Specimen Required

Submit only 1 of the following specimens:

Specimen Type: Tissue

Preferred: Tissue block

Collection Instructions: Submit a formalin-fixed, paraffin-embedded (FFPE) tumor tissue block. Blocks prepared with alternative fixation methods may be acceptable; provide fixation method used.

Acceptable: Slides

Collection Instructions: Four consecutive, unstained, 5 micron-thick sections placed on positively charged slides, and 1 hematoxylin and eosin-stained slide.

Forms

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

Specimen Minimum Volume

Two consecutive, unstained, 5 micron-thick sections placed on positively charged slides and 1 hematoxylin and eosin-stained slide

Reject Due To

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

Specimen Stability Information

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Clinical and Interpretive

Clinical Information

Ewing sarcoma (EWS)/primitive neuroectodermal tumors (PNET) are members of the small, round cell group of tumors that are thought to originate in cells of primitive neuroectodermal origin with variable degrees of differentiation. The small, round cell group of tumors also includes rhabdomyosarcomas, desmoplastic small, round cell tumors, and poorly differentiated synovial sarcomas. Although immunohistochemical markers can be helpful in the correct diagnosis of these tumors, recent molecular studies have shown the specificity of molecular markers in differentiating specific subtypes of small, round blue-cell tumors. Accurate diagnosis of each tumor type is important for appropriate clinical management of patients.
Ewing tumors are characterized cytogenetically by rearrangements of the EWSR1 gene at 22q12 with FLI1 at 11q24 (t(11;22)) or ERG at 21q22 (t(21;22)) in 85% and 5% to 10% of Ewing tumors, respectively. Less than 1% of cases may have other fusion partners such as ETV1 at 7p22, E1AF at 17q12, or FEV at 2q33. Detection of these transcripts by reverse transcriptase-PCR (RT-PCR) (EWS, Ewing Sarcoma RT-PCR) that allows specific identification of the t(11;22) and the t(21;22), has greatly facilitated the diagnosis of Ewing tumors. However, if the quality of the available RNA is poor, the results are equivocal, or if a rare translocation partner is present, FISH testing has proven to be useful in identifying the 22q12 EWS gene rearrangement in these tumors.

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 cutoff for the EWSR1 FISH probe set.

A positive result is consistent with a diagnosis of Ewing sarcoma (EWS)/primitive neuroectodermal tumors (PNET).

A negative result suggests that a EWSR1 rearrangement is not present but does not exclude the diagnosis of EWS/PNET.

Cautions
This test is not approved by the U.S. Food and Drug Administration and is best used as an adjunct to existing clinical and pathologic information.

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

Paraffin-embedded tissues that have been decalcified are generally unsuccessful for FISH analysis. The pathologist reviewing the hematoxylin and eosin-stained slide may find it necessary to cancel testing.

Supportive Data
FISH analysis was performed on 38 formalin-fixed, paraffin-embedded tissue samples, including 16 tumors, and 22 noncancerous control specimens. The normal controls were used to generate a normal cutoff for this assay. Rearrangement of EWSR1 was identified in 14 tumor specimens and 2 yielded no results due to poor hybridization.

Clinical Reference
1. World Health Organization Classification of Tumors. Pathology and Genetics Tumours of Soft Tissue and Bone. Edited by CDM Fletcher, KK Unni, F Mertens: IARC Press; Lyon 2002, pp 298-300

Performance

Method Description
This test is performed using a commercially available EWSR1 dual-color, break-apart strategy probe (BAP). 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 is performed by a pathologist. Using the H and E-stained slide as a reference, target areas are etched with a diamond tipped etcher on the back of the unstained slide to be assayed. The probe set is hybridized to the appropriate target areas and 2 technologists each analyze 50 interphase nuclei (100 total) with the results expressed as the percent of abnormal nuclei. (Unpublished Mayo method)

PDF Report
No

Day(s) and Time(s) Test Performed
Samples processed Monday through Sunday.

Results reported Monday through Friday 8 a.m. to 5 p.m.

Analytic Time
7 days

Maximum Laboratory Time
10 days

Specimen Retention Time
Slides and H&E used for analysis are retained by the laboratory in accordance to CAP and NYS requirements. Client provided paraffin blocks and extra unstained slides (if provided) will be returned after testing is complete.

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 using an analyte specific reagent. Its performance characteristics were 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
88271x2, 88291 Ä¢â¬¬â€œ DNA probe, each (first probe set), Interpretation and report
88271x2 Ä¢â¬¬â€œ DNA probe, each; each additional probe set (if appropriate)
88271x1 Ä¢â¬¬â€œ DNA probe, each; coverage for sets containing 3 probes (if appropriate)
88271x2 Ä¢â¬¬â€œ DNA probe, each; coverage for sets containing 4 probes (if appropriate)
88271x3 Ä¢â¬¬â€œ DNA probe, each; coverage for sets containing 5 probes (if appropriate)
**Test Definition: EWSF**

EWSR1 (22q12), FISH, Ts

88274 w/modifier 52 88274 w/modifier 52 88274 w/modifier 52 88274 w/modifier 52

- Interphase in situ hybridization, <25 cells, each probe set (if appropriate)

- Interphase in situ hybridization, 25 to 99 cells, each probe set (if appropriate)

- Interphase in situ hybridization, 100 to 300 cells, each probe set (if appropriate)

**LOINC® Information**

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