Overview

Useful For
Supporting the diagnosis of endometrial stromal tumors when used in conjunction with an anatomic pathology consultation

Reflex Tests

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<th>Always Performed</th>
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Testing Algorithm
This test does not include a pathology consultation. 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.

Unless otherwise indicated, JAZF1 FISH testing will be performed:

- If a JAZF1 rearrangement is not identified by FISH, reflex testing for PHF1 and YWHAE rearrangement will be performed.

- If JAZF1 FISH testing was previously performed, reflex testing for PHF1 or YWHAE may be ordered separately.

- If testing was not performed at Mayo Clinic, provide a copy of the JAZF1 FISH report.

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: Six 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
Three 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 by Mayo Clinic Laboratories for test suitability.

Specimen Stability Information

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<th>Time</th>
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Clinical and Interpretive

Clinical Information
Endometrial stromal tumors (EST) arise from the uterus and include the benign endometrial stromal nodule (ESN) and infiltrative endometrial stromal sarcoma (ESS). These tumors are characterized by a translocation that fuses JAZF1 at 7p15 to JJAZ1 at 17q21 or a variant 6;7 translocation involving JAZF1 and PHF1. Published literature
employing FISH and reverse transcriptase PCR (RT-PCR) suggests rearrangement of JAZF1 occurs in approximately 76% of ESN and approximately 58% of ESS. JAZF1 is not generally considered to be involved in the genetic mechanism of the high-grade undifferentiated endometrial sarcoma (UES), although rarely some cases of UES are positive for JAZF1, which may reflect the presence of an ESS component.

For PHF1 disruption, a study of 94 EST demonstrated the following:

- **PHF1/JAZF1** fusion in 4 primary ESS
- **PHF1/EPC1** fusion in 2 primary ESS and 1 extrauterine ESS
- **PHF1** rearrangement without a known partner in 6 primary or metastatic ESS and 1 extrauterine ESS

JAZF1/JJAZ1, PHF1/JAZF1 and PHF1/EPC1 fusions were mutually exclusive in individual patients. No rearrangement of PHF1 was found in ESN, UES, or non-EST tumors in the differential diagnosis. These results indicate that PHF1 can rearrange with both known and unknown partners in addition to JAZF1 and is potentially specific for ESS.

In high-grade ESS, a recurrent t(10;17)(q22;p13) resulting in fusion of YWHAE (also called 14-3-3epsilon at 17p13.3 with either FAM22A or FAM22B was identified. In contrast, JAZF1 rearrangements are typically observed in low-grade ESS. JAZF1 and YWHAE rearrangements are mutually exclusive and have distinct gene expression profiles. YWHAE rearrangement is potentially specific for high-grade ESS as no YWHAE disruption has been reported in other uterine or nonuterine mesenchymal tumors.

The clinical utility of identifying JAZF1 rearrangement is mainly to address the differential diagnostic dilemma that occurs when ESS are present as metastatic lesions or exhibit variant morphology. In JAZF1-negative EST cases, reflex genetic analysis to identify PHF1 or YWHAE rearrangement increases the diagnostic sensitivity for EST. In addition, confirmation of YWHAE rearrangement may have prognostic implications as YWHAE defines a distinct, clinically more aggressive and histologically higher grade subgroup of ESS compared to those with JAZF1 rearrangements.

**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 any given probe.

Detection of an abnormal clone likely indicates a diagnosis of an endometrial stromal tumor of various subtypes.

The absence of an abnormal clone does not rule out the presence of a neoplastic disorder.

**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 (e.g., 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

JAZF1:

FISH analysis was performed on 101 formalin fixed paraffin-embedded tissue specimens. These included 37 endometrial stromal tumors (EST) (8 endometrial stromal nodule [ESN], 20 primary or metastatic endometrial stromal sarcoma [ESS], and 9 primary or metastatic undifferentiated endometrial sarcoma [UES]), 38 histologic mimics of EST including cellular leiomyoma and 26 noncancerous control specimens. The normal controls were used to generate a normal cutoff for this assay. No rearrangements of JAZF1 were identified in the 26 normal controls or 37 of the histologic mimics. A single histologic mimic was positive with a complex pattern. Rearrangement of JAZF1 was identified in 17 of 37 (46%) EST specimens (3 ESN, 13 ESS, and 1 UES).

PHF1 and YWHAE:

FISH analysis was performed on 91 formalin fixed paraffin-embedded tissue specimens, including 20 EST (5 ESN, 7 primary or metastatic ESS, and 8 primary or metastatic UES), 35 histologic mimics of EST and 26 non-cancerous control specimens. Also included were 6 known JAZF1-positive cases and 4 clinical cases that were negative for JAZF1 rearrangement. The normal controls were used to generate a normal cutoff for this assay. No rearrangements of PHF1 or YWHAE were identified in the 26 normal controls or 35 of the histologic mimics. One primary UES failed to hybridize and yielded no results for either probe. Rearrangement of PHF1 was identified in 1 of 19 (5%) of EST specimens (primary ESS) and in 1 of 6 (17%) of known JAZF1-positive cases (metastatic ESS). Rearrangement of YWHAE was identified in 2 of 19 (11%) of EST specimens (primary ESS and metastatic UES).

Clinical Reference

Performance

Method Description
The test is performed using laboratory-developed dual-color break-apart strategy probes (BAP) for JAZF1, PHF1, and YWHAE. Formalin-fixed, paraffin-embedded tissues are cut at 5 microns and mounted on positively charged glass slides. The selection of tissue and the 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 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 for each probe set) with the results expressed as the percent 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.-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 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
88271x2, 88291 Å¢â„¬â€œ DNA probe, each (first probe set), Interpretation and report
88271x2 Å¢â„¬â€œ DNA probe, each; each additional probe set (if appropriate)
88271x1 Å¢â„¬â€œ DNA probe, each; coverage for sets containing 3 probes (if appropriate)
Test Definition: ESTUF
Endometrial Stromal Tumor, FISH, Ts

88271x2  Åcâ¬àë¢œ DNA probe, each; coverage for sets containing 4 probes (if appropriate)
88271x3  Åcâ¬àë¢œ DNA probe, each; coverage for sets containing 5 probes (if appropriate)
88274 w/modifier 52  Åcâ¬àë¢œ Interphase in situ hybridization, <25 cells, each probe set (if appropriate)
88274  Åcâ¬àë¢œ Interphase in situ hybridization, 25 to 99 cells, each probe set (if appropriate)
88275  Åcâ¬àë¢œ Interphase in situ hybridization, 100 to 300 cells, each probe set (if appropriate)

LOINC® Information

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