

Assay Summary

Somatic KIT (C-Kit) and PDGFRA Genotyping

C-Kit Gene (exons 8, 9, 11, 13, 17 and 18) and PDGFRA (exons 12 and 18) Mutation Analysis

Synopsis

The KIT gene encodes the human homolog of the proto-oncogene c-kit, and the PDGFRA gene encodes a cell surface tyrosine kinase receptor for members of the platelet-derived growth factor family. Roughly 75%-85% of gastrointestinal stromal tumors (GIST) have activating mutations in c-kit, and an estimated 5%-7% have activating mutations in the PDGFRA gene¹. Tumors with mutations in exon 9 of the c-kit gene are associated with significantly shorter progression-free survival and overall survival compared to tumors with mutations in exon 11². However, it was found that tumors expressing an exon 9 mutation in c-kit had improved progression-free survival when given a high dose regimen of imatinib, compared to those with an exon 9 mutation who received regular dosing². Mutations in exon 12 and 18 of the PDGFRA gene have also been associated with responsiveness to imatinib. Specifically, two missense changes, D842V and D846V, in the PDGFRA gene are associated with resistance to imatinib¹, and the D561V mutation and deletions in exon 18 of the PDGFRA gene are associated to imatinib sensitivity¹.

Melanomas have also been noted to have activating mutations in the c-kit gene. About 15% of acral and 20% of mucosal melanomas have a c-kit alteration. Case reports have documented dramatic responses to imatinib in melanoma patients with c-kit mutations^{3,4,5}. Recently, an update on a multi-institutional phase II clinical trial of imatinib in melanoma patients at the International Melanoma Congress reported that none of the 10 wild-type c-kit cases had a clinical response, while five of the 10 patients with a c-kit mutation demonstrated a partial response to therapy⁶.

Indications for testing

Testing of the c-kit gene should be considered in individuals with a diagnosis of GIST or melanoma prior to initiating Imatinib therapy. As mutations in the c-kit gene and PDGFRA gene appear to be mutually exclusive in GIST, and the detection rate for c-kit alterations is much higher, it is recommended that a sequential approach to testing be taken with c-kit testing first, followed by PDGFRA testing if c-kit is negative.

Methodology

A fluorescent capillary based sequencing assay for the exons 8, 9, 11, 13, 17, and 18 in the c-kit gene, and exon 12 and 18 of the PDGFRA gene has been optimized, and validated by the Molecular Diagnostic Laboratory (MDL). Testing is performed on micro-dissected cells from formalin fixed, paraffin embedded tissue blocks. We perform complete sequencing of all exons, enabling the detection of any small alteration within these regions, including single base pair changes, small deletions, and small insertions.

Performance

The expected result for this assay is the normal/wild-type sequence, or the presence of any small deviation from the wild-type sequence including single base pair changes, small deletions, and small insertions. The sensitivity of this assay is predicted to detect down to roughly 15-20 in 100 mutation-bearing cells in a micro-dissected area.

Specimen Requirements

We prefer to receive paraffin embedded tissue blocks, and also accept cell blocks obtained from fine needle aspirations. Cell blocks with limited cellularity may not be sufficient for testing. If blocks cannot be sent, please send six slides of tumor sample (5-micron serial sections, five unstained and one H/E stained). Ensure that the slides are clearly labeled with the patient name or identifier and date of birth and type of sample. Place slides in appropriate containers to ensure against breakage. Please include a copy of the Pathology report.

Test Request Form (TRF)

A completed MDL [TRF](#) is required for each specimen. Please submit the completed TRF with the specimen. Complete testing and billing information must be provided before the specimen is processed.

<i>Order Codes</i>	<i>CPT Codes</i>	<i>TAT</i>
C-Kit (C-Kit gene, sequencing (Exons 8, 9, 11,13,17,18))	83890, 88323, 88381, 83892, 83898 (x6), 83894, 83904 (x6), 83912	2 wks
C-Kit-CAS (C-Kit gene, targeted mutation analysis, known mutation)	83890, 83898, 83894, 83904, 83912	2 wks
PDGFRA (PDGFRA gene, sequencing (Exons 12, 18))	83890, 88323, 88381, 83892, 83898 (x2), 83894, 83904 (x2), 83912	2 wks

References

1. Heinrich et al. (2008) J. Clin. Onc. 26 :5360-5367
2. Debiec-Rychter et al. (2006) Eur. J. Can. 42:1093-1103
3. Lutzky et al. (2008) Pig. Cell. Melan. Res 21:492-3
4. Quintas-Cardama et al. (2008) Nat. Clin. Pract. Oncol. 5:737-740
5. Hodi et al. (2008) J. Clin. Oncol. 26 :2046-2051
6. Fisher et al. (2010) Pig. Cell Melan. Res. 23 :14-26

NOTE: This test is performed pursuant to a license agreement with Roche Molecular Systems, Inc.