City of Hope

Dennis D. Weisenburger, MD
Chairman, Department of Pathology, CLIA #05D0665695

Clinical Molecular Diagnostic Laboratory
1500 East Duarte Road
Northwest Building, Second Floor, Room 2236
Duarte, CA 91010-3000
Phone 888-826-4362 Fax 626-301-8142
cmdl@coh.org http://cmdl.cityofhope.org

Assay Summary

KRAS Mutation Analysis

Colorectal cancer
Non-small cell lung cancer

Synopsis

KRAS somatic mutations are found in 30-40% of colorectal cancers (CRC)[1,2]. Most often, these mutations occur at codons 12 and 13, but mutations at other codons (including 61, 117, and 146) have also been detected. Hotspot KRAS mutations have been associated with poor responsiveness to EGFR monoclonal antibody therapies (cetuximab, panitumumab) in CRCs[3, 7, 8]. KRAS mutations are also detected in 10-30% of non-small cell lung carcinomas (NSCLC)[4] and studies have reported decreased sensitivity to EGFR tyrosine kinase inhibitors [5, 6]. Determining the KRAS status of a newly diagnosed CRC or NSCLC tumor prior to initiating anti-EGFR therapy may be useful in selecting candidates for monoclonal antibody (MAB) therapy and tyrosine kinase inhibitor (TKI) therapy, respectively. Additionally, testing should be considered for patients who are already on anti-EGFR therapy, but are demonstrating resistance.

Indications for testing

Newly diagnosed CRC or NSCLC prior to initiating anti-EGFR therapy, or a history of CRC or NSCLC and demonstrated resistance to anti-EGFR therapy.

Genomic DNA (gDNA) is extracted from micro-dissected cells from formalin-fixed, paraffin-embedded tissue. A targeted DNA library is generated using the Ion AmpliSeq™ Cancer Hotspot Panel v2 Kit, and sequenced by semiconductor-based next-generation sequencing technology on an Ion Torrent PGM. This test targets 63 hotspot mutations in exons 2, 3, and 4 of the KRAS gene.

Performance/Limitations

The gene is not sequenced in its entirety; only the regions including the targeted mutations are analyzed. The method will not detect gross genetic alterations including large deletions, duplications, and inversions. The minimum detectable mutant allele ratio is approximately 10%.

Specimen Requirements

Formalin-fixed, paraffin-embedded (FFPE) tissue blocks or slides.

The tissue sample should be large enough to provide at least 3000 tumor cells and at least 30% of tumor cells within the tissue. We prefer to receive FFPE tissue blocks (unused portion will be returned), but slides are also accepted. For slides:

- 10 slides, 10 micron serial sections, unstained, without coverslip.
- 1 representative H&E slide, 4 micron section, with a coverslip.
- If the sample is a needle biopsy or has very little tumor, please send 5 additional slides.

- Slides or blocks should be labeled with the accession number and patient name and accompanied by a copy of the corresponding pathology report.
- Place slides in appropriate container(s) to ensure against breakage.

Test Request Form (TRF)

A completed CMDL (TRF) must be submitted with each specimen. Complete testing and billing information must be provided before the specimen is processed

Order Codes	***CPT Codes***	***TAT***
KRAS-NGS (Targeted analysis for 63 cancer hotspot mutations in the KRAS gene (including codons 12, 13, 61, 117, 146) by next generation sequencing)	81275, 88381, G0452	7-14 days

References

1. Lièvre, A. et al., (2008) J Clin Oncol; 26:374-9
2. Amado, R. G. et al., (2007) Eur J of Can Supp (5):6
3. Di Fiore, F. et al., (2007) Br J Cancer; 96:1166-9
4. Eberhard et al. (2005) J Clin Oncol; 23:5900-9
5. Han. et al., (2006) Clin Can Res; 12:2538-44
6. Pao. et al., (2005) PLoS Med; 2:e17
7. Janakiriman et al. Cancer Res. 2010 Jul 15;70(14):5901-11
8. Loupakis et al. British Journal of Cancer (2009) 101, 715–721