



TECHNICAL NOTICE

SOUTH BEND MEDICAL FOUNDATION, INC.

k-ras/BRAF mutation analysis by multiplex-PCR

Effective Date: September 1, 2011

Performing Department: Molecular Pathology

Clinical Significance: K-RAS (RNA Associated Rat Sarcoma 2 virus gene) is located on short arm of chromosome 12. Recent studies have confirmed that presence of k-ras mutation is highly predicative of resistance to anti-EGFR (epidermal growth factor receptor) inhibitor therapy in metastatic colorectal cancer (MCRC) and non-small cell lung cancer (NSCLC) patients. Presence of k-ras mutation is also indicative of poor prognosis. Prevalence of k-ras mutation in colon cancer and NSCLC is 30-50% and 15-30%, respectively. Only about 15% of patients with metastatic colorectal cancer are responders to targeted therapy with the anti-EGFR therapies panitumumab (Vecibix, Amgen) and cetuximab (Erbix, Bristol-Myers Squibb).

The American Society of Clinical Oncology (ASCO) recommends that all patients with metastatic colorectal cancer in whom anti-EGFR treatments are being considered should be tested for k-ras mutations, at least involving codons 12 and 13. The National Comprehensive Cancer Network (NCCN) guideline with the recommendation that k-ras testing on the primary tumor or a site of metastasis should be part of the pre-treatment work-up for all metastatic colorectal cancer patients. In July 2009, the FDA approved labeling changes to cetuximab and panitumumab indicating that these agents are not recommended for the treatment of colorectal cancer harboring KAS mutations.

BRAF mutations are prognostic indicators of worse outcomes in KRAS wild-type patients. BRAF testing is now permissive but not mandatory. Approx. 3%-12% of metastatic colorectal cancers are characterized by a specific mutation in the BRAF gene (V600E). BRAF mutations are mutually exclusive with KRAS exon 2 mutations. Patients with BRAF mutations also appear not to benefit from anti-EGFR treatment.

Method: DNA from relevant tumor tissue that is identified by a pathologist is extracted, amplified and hybridized directly on target-specific probes by a multiplex PCR reaction. The analysis detects six mutations in each of codons 12 and 13 of KRAS gene and one BRAF mutation in codon 600. This method can detect the following mutations at a detection level of 1-5% mutant alleles.

k-ras codon 12: G12S, G12R, G12C, G12D, G12A, G12V

k-ras codon 13: G13D, G13S, G13R, G13C, G13V, G13A

BRAF: V600E

Use: k-ras/BRAF mutation should be evaluated on patients with metastatic colorectal cancer (MCRC) and non-small cell lung cancer (NSCLC) prior to undergoing treatment of anti-EGFR drugs.

Reference Range: No mutation is identified in codons 12 and 13 of KRAS gene or BRAF V600E.

Specimen Collection Requirements:

Specimen requirement: Paraffin-embedded, formalin-fixed tissue block, or 1 H&E stained and 5 unstained 10 um slides

SOUTH BEND MEDICAL FOUNDATION, INC.

530 N. Lafayette Boulevard • South Bend, IN 46601 • (574) 234-4176

Elkhart (574) 293-8441 • (800) 544-0925

Robert J. Tomec, M.D. • *Medical Director*

Transportation: Ambient temperature. Protected paraffin block from excessive heat and ship in cooled container during summer months.

Unacceptable conditions: No tumor in tissue. Frozen specimens or specimens fixed in fixatives other than formalin

Stability: Ambient: Indefinitely; Refrigerated: Acceptable; Frozen: Unacceptable

Testing Days: Tuesday

Order: CPT: 83890, 83900, 83901 x 2, 83896 x 13, 83912

Please direct any questions, or comments regarding this notice to Deborah H. Sun, Ph.D. (dsun@sbfm.org) or Kevin Maggert, Senior Technologist (kmaggert@sbfm.org) at South Bend Medical Foundation (574) 234-4176 or (800) 544-0925.

SOUTH BEND MEDICAL FOUNDATION, INC.

530 N. Lafayette Boulevard • South Bend, IN 46601 • (574) 234-4176

Elkhart (574) 293-8441 • (800) 544-0925

Robert J. Tomec, M.D. • *Medical Director*