

LABORATORY TEST ALERT
REGIONAL PATHOLOGY SERVICES
UNIVERSITY OF NEBRASKA MEDICAL CENTER

July 2009

KRAS MUTATION DETECTION

SPECIMEN REQUIRED

Collect 5 Unstained slides 10µm thick with ≥50% tumor (with one adjacent H&E) and/or paraffin embedded formalin fixed tissue block.

Minimum volume: 5mm X 5mm area if tissue is 100% tumor;
10mmX10mm if tissue is 50% tumor

Please include submitting facility's surgical pathology report.

Transport Paraffin block; Ambient temperature (send on cold packs during summer months)
Fresh Tissue; Send snap frozen on dry ice.

Unacceptable Condition Paraffin blocks with ≤10% tumor.
Tissue fixed in heavy metal fixative, decalcified tissue.

REFERENCE INTERVAL

Interpretation by pathologist.

NOTES

The assay is designed to detect mutations in codons 12 and 13 of the KRAS gene. Specifically, one primer set amplifies a 122 bp fragment which contains codons 12 and 13. Amplified products are subjected to pyrosequencing analysis to quantify mutations in these codons.

The human *KRAS* gene (v-Ki-ras2 Kirsten rat sarcoma viral oncogene homolog) is located at chromosome 12p12, and encodes a 21-kD protein (p21ras or K-Ras). K-Ras functions as a binary molecular switch that controls intracellular signaling networks in a number of signaling pathways, including the signal transduction pathway involving Epidermal Growth Factor Receptor (EGFR).

Certain mutations in *KRAS* result in constitutively active KRAS, leading to uncontrolled cell proliferation. Such oncogenic activation has been suggested to be involved in many aspects of the development and progressions of cancer, including abnormal cell growth and differentiation, as well as increased invasiveness and metastasis. Consequently, mutations in the *KRAS* gene are found in many types of human cancers, with the most common mutations being found in codons 12 and 13. Choosing more effective therapy up front can save 30-60% of medication costs incurred in treating colorectal cancer patients whose tumors contain *KRAS* gene mutations.

Human genomic DNA is extracted from fresh, frozen or paraffin embedded tissue. Standard PCR is performed, generating biotinylated products, which are then immobilized on streptavidin-coated Sepharose beads. Sequencing primers are added and pyrosequencing is performed.

METHODOLOGY

DNA Amplification/Pyrosequencing

PERFORMED

Molecular Diagnostics/Molecular Oncology/ The Nebraska Medical Center Laboratories
Monday - Friday.

REPORTED

Results in 5-7 days.

CPT codes:

83907, 83890, 83898, 83904, 83912

REFERENCES

Pyromark Q24 KRAS v2.0 kit package insert

Call Brian Lenz or Randi Nelson at Regional Pathology Services 559-6420 or 1-800-334-0459 for information about sending this test to the UNMC Laboratory.