Myeloid Malignancies Mutation Panel by Next Generation Sequencing

Indications for Ordering

Assess for single gene mutations, including substitutions and insertions and deletions that may have diagnostic, prognostic, and/or therapeutic significance in
- Acute myeloid leukemia
- Myelodysplastic syndromes (MDS)
- Myeloproliferative neoplasms (MPN)
- MDS/MPN overlap disorders such as chronic myelomonocytic leukemia

Test Description

Myeloid Malignancies Mutation Panel by Next Generation Sequencing
- Next generation sequencing (NGS) library construction from genomic DNA
- Enrichment for regions of interest by hybridization
- Massively parallel sequencing

Tests to Consider

Primary tests
- Myeloid Malignancies Mutation Panel by Next Generation Sequencing 2011117
- Myeloid Malignancies Somatic Mutation and Copy Number Analysis Panel 2012182

Related tests
- CEBPA Mutation Detection 2004247
- NPM1 Mutation by PCR and Fragment Analysis 0040174
- IDH1 and IDH2 Mutation Analysis, exon 4 2006444
- WT1 Mutation Detection by Sequencing 2005766
- KIT Mutations in AML by Fragment Analysis and Sequencing 2002437

Disease Overview

Diagnostic issues
- Genetic targets contained in panels are relevant across the spectrum of myeloid malignancies
- Identification of one or more clonal genetic abnormalities may aid in establishing the diagnosis of a neoplasm
- Identification of certain mutations or patterns of mutations may aid in diagnostic subclassification

Prognostic and treatment issues
- Certain mutations or patterns of mutations may have prognostic significance
- Certain mutations may allow for the use of targeted therapies

Genetics

Genes – ASXL1, ASXL2, BCOR, BCORL1, BRAF, CALR, CBL, CEBPA, CSF3R, DNMT1, DNMT3A, EED, ELANE, ETNK1, ETV6, EZH2, FAM5C, FLT3, GATA1, GATA2, HNRNPK, IDH1, IDH2, JAK2, JAK3, KDM6A, KIT, KRAS, LUC7L2, MAP2K1, MLL, MPL, NOTCH1, NPM1, NRAS, NSD1, PHF6, PRPF40B, PRPF8, PTPN11, RAD21, RUNX1, SETBP1, SF1, SF3A1, SF3B1, SMC1A, SMC3, SRSF2, STAG2, SUZ12, TET2, TP53, U2AF1, U2AF2, WT1, ZRSR2

Mutations

A full list of targeted regions within these genes can be found at the ARUP website – Myeloid Panel Coordinates (www.aruplab.com/myeloid-panel-coordinates)
Test Interpretation

Results
- Positive – a somatic mutation in one of the 57 tested genes was detected
  - Clinical relevance (diagnosis, prognosis, therapy) will be correlated, if known
- Negative result – no mutations were detected in the sequenced genes

Limitations
- Mutations may be present below the limit of detection
- Not intended to detect minimal residual disease

Cytogenomic Microarray – Oncology

Indications for Ordering
- Preferred test at time of diagnosis for detecting prognostically important genomic abnormalities in leukemias/lymphomas and solid tumors involving
  - Loss/gain of DNA
  - Loss of heterozygosity (LOH)
- Monitor disease progression and response to therapy

Test Description

Cytogenomic Molecular Inversion Probe Array, FFPE Tissue – Oncology
- Platform – Affymetrix OncoScan
- Contains 220,000 SNP probes across the entire genome
- Average functional resolution – 20 consecutive markers

Cytogenomic SNP Microarray – Oncology
- Platform – Affymetrix CytoScan HD
- Oligo copy number and single-nucleotide polymorphism (SNP) array
- Contains >2.6 million copy number markers
- Includes 750,000 SNP probes
- Detects copy number changes and LOH
- Average marker spacing
  - Intragenic – 880 base pairs (bp)
  - Intergenic (nongene backbone) – 1,700 bp
  - Overall (gene and nongene backbone) – 1,100 bp
- Average functional resolution
  - Deletion of 25 consecutive markers
  - Duplication of 50 consecutive markers

Tests to Consider

Primary tests
- Offer whole genome coverage
- Detect copy number changes and LOH
- Differ in type of specimen and array platform

Myeloid Malignancies Somatic Mutation and Copy Number Analysis Panel 201218
- Panel for myeloid malignancies that combines cytogenomic microarray with a next generation sequencing panel targeting genes with diagnostic, prognostic, and/or therapeutic significance

Related tests
- Fluorescence in situ hybridization (FISH) testing for specific balanced translocations may be considered, based on indication
- For a complete list of ARUP’s oncology FISH tests, including probe targets and genes, see “Oncology FISH” on the ARUP Genetics website (www.aruplab.com/genetics/tests/fish)

Disease Overview

Diagnostic issues
- Gains, losses, and LOH occur in malignancies – identification may be helpful for
  - Diagnosis
  - Prognosis and therapeutic decisions
  - Monitoring disease progression and response to therapy
- Conventional cytogenetic (CC) analysis for detection of genetic abnormalities in oncology is hampered by
  - Lack of tumor cell growth in cell culture
  - Subtle chromosomal abnormalities that are often missed
- FISH
  - Improved rate of detection of clonal abnormalities when compared to CC, but only for the targeted region
  - Detects balanced translocations
  - Limited because only a few loci examined at a time
- Neither conventional karyotyping nor FISH testing can detect copy-neutral events that are associated with hematological malignancies
  - Often due to mutations and subsequent selection of mutant tumor-suppressor genes and oncogenes
- SNP microarray detects many of the chromosomal variants involving gains or losses in chromosomes with complex karyotypes across the genome
Test Interpretation

Results

• Abnormal microarray
  o Well-documented and clinically significant gain or loss or LOH detected
• Copy number change detected, clinical significance unknown
  o Copy number variation detected for which insufficient evidence is available to determine unequivocally the clinical significance
• Normal microarray
  o No clinically significant abnormalities detected based on current knowledge at time of reporting

Limitations

• Low-level mosaicism (<15-20%) may not be detected
• May not be appropriate for individuals with expected lower levels of malignant cells
• FFPE specimens must contain a region with ≥50% tumor
• Not recommended for minimal residual disease
• Does not detect
  o Balanced rearrangements
    ▪ FISH should be used to evaluate specific balanced rearrangements according to indication
  o Base pair mutations and very small deletions/duplications
  o Imbalances of the mitochondrial genome
  o Positional information for chromosome rearrangements
  o Low-level clones